

Article

Micro/Nanoplastic-Enhanced Oxidative Potential, Antioxidant Depletion, Inflammation in PM_{2.5} and Cytologic and Metabolomic Shifts

Hasan Saygin ^{1,*}  and Asli Baysal ² 

¹ Application and Research Center for Advanced Studies, Istanbul Aydin University, Sefakoy Kucukcekmece, Istanbul 34295, Türkiye

² Department of Chemistry, Faculty of Science and Letters, Istanbul Technical University, Maslak, Sariyer, Istanbul 34467, Türkiye; asli.baysal@itu.edu.tr

* Correspondence: hasansaygin@aydin.edu.tr; Tel.: +90-2124441428-12910

Abstract

Micro- and nanoplastics (MNPs) are increasingly contaminating atmospheric particulates, yet their influence on PM_{2.5} chemistry and toxicity remains poorly understood. This study investigates how secondary MNPs derived from common products (water bottles, coffee cups, and food plates) alter the properties of PM_{2.5}. We evaluated PM_{2.5} leaching characteristics, oxidative potential, inflammatory activity, and bacterial-based cytological and metabolomic responses after 24 h of exposure to three MNP doses. MNPs markedly altered PM_{2.5} chromophoric composition, with bottle-derived (PET) MNPs inducing the strongest increases in aromaticity, humification, and slope factor, followed by coffee cups (PLA/paper) and food plates (PP). These leaching shifts aligned with polymer-specific redox behaviors: bottle-derived MNPs enhanced antioxidant enrichment at high PM_{2.5}, whereas cup-derived MNPs produced the most pronounced protein-denaturation-based inflammatory activity. *Escherichia coli* assays showed non-linear growth responses, elevated reactive oxygen species, altered carbohydrate secretion, and membrane and protein perturbations that paralleled PM_{2.5} chemical reactivity. FTIR metabolomic fingerprints revealed dose- and polymer-dependent disruptions in polysaccharide, lipid, and protein domains. Overall, the results demonstrate a mechanistic cascade in which MNP exposure reshapes PM_{2.5} chemistry, amplifies oxidative and inflammatory potential, and culminates in measurable cytological and metabolic stress, with polymer identity (PET > PLA/paper > PP) as the dominant driver.

Keywords: chromophoric transformation; redox reactivity; leaching; proteomic; atmospheric microplastics



Academic Editor: Nicolas Kalogerakis

Received: 10 December 2025

Revised: 5 January 2026

Accepted: 14 February 2026

Published: 3 March 2026

Copyright: © 2026 by the authors.

Licensee MDPI, Basel, Switzerland.

This article is an open access article distributed under the terms and conditions of the [Creative Commons Attribution \(CC BY\)](https://creativecommons.org/licenses/by/4.0/) license.

1. Introduction

Globally, air pollution constitutes a critical public health challenge, ranking as the 13th leading cause of death and responsible for approximately 0.8 million fatalities (1.4%) [1]. Air quality is mainly characterised by the levels and constituents of various particulate matter (PM), which are categorised based on their aerodynamic diameter as either thoracic/coarse particles (PM₁₀; <10 µm) or higher-risk respirable fractions, including ultrafine particles (UFP; <0.1 µm) and fine particles (PM_{2.5}; <2.5 µm). PM_{2.5} is widely recognized as a major environmental and public health issue, owing to its adverse impacts on human health, ecosystems, and overall quality of life [2]. Exposure to PM_{2.5} has been consistently linked to

the development and exacerbation of acute and chronic respiratory disorders, specifically respiratory bacterial infections and lung cancer, neurological and cardiovascular diseases in humans [3–5]. Many studies have demonstrated the relationship between PM and health through oxidative potential and inflammation [6]. Since PM_{2.5} is a chemically and biologically heterogeneous mixture originating from diverse natural and anthropogenic sources, comprising inorganic species, redox-active metals, organic compounds, each exhibiting distinct physicochemical properties and incompletely characterized toxicities that collectively drive oxidative stress, inflammation, and increased susceptibility to adverse respiratory and cardiovascular outcomes [7]. Recently, micro- and nanoplastics (MNPs) (less than 5 mm in size) have been identified as a new type of contaminant in PMs that persists and is becoming more common [8,9].

Atmospheric MNPs are receiving growing scientific attention owing to their increasing abundance in the air [10]. Since the global consumption of plastics is projected to exceed 800 million tons by 2050, this exacerbates the environmental crisis, leading to a continuous increase in microplastic and nanoplastic generation [11]. Given the pervasive nature of MNPs in various environmental matrices, their interaction with PM_{2.5} represents a critical and emerging area of concern for human health. In addition, they have been identified in various human body parts (e.g., blood, tissue, and organs) [12,13].

Beyond their presence in the air or their specific and individual toxic effects, MNPs also possess sorption and deformation properties. MNPs can adsorb chemicals in PM_{2.5}, including heavy metals and polycyclic aromatic hydrocarbons, and MNPs can degrade due to biotic and abiotic factors, releasing various substances into the surrounding medium, such as PM_{2.5} [14]. These processes have the potential to alter the physicochemical composition of PM_{2.5}. Furthermore, the leaching of MNPs into the environment could introduce additional stressors that exacerbate these adverse health effects, particularly in the context of compromised metabolic health [15]. Because plastics contain numerous additives (e.g., plasticizers like phthalates, flame retardants, Bisphenol A (BPA), and colorants), recent research indicates that over 10,000 compounds, with 2400 deemed potentially hazardous, can leach from plastics throughout their life cycle, including endocrine-disrupting chemicals [16]. The vast majority of these plastic additives (e.g., plasticizers, flame retardants, antioxidants, colorants) are not chemically bound (covalently linked) to the main polymer chains during manufacturing. Thus, these chemicals can leach out of the MNP structure when exposed to environmental conditions such as air, humidity, temperature, and other pollutants [17,18]. MNPs can release these unpolymerized monomers, which may be more toxic than the bulk polymer itself [19,20]. Moreover, deformed substances have been shown to directly cause inflammation and oxidative stress through cellular uptake and interference with mitochondrial function [21,22]. These leached chemicals can alter the oxidative characteristics of PM_{2.5}, inducing its cytotoxic effects in bacteria and human cells and contributing to chronic inflammation and metabolic dysregulation [23,24]. In addition, the cytotoxic effects of leached chemicals on various cell levels, including bacteria and human cells, as well as changes in the oxidative characteristics of the media due to the leaching of MNPs, can contribute to the development of bacterial biofilms that protect oxidative conditions, which are a primary cause of bacterial resistance. However, there has been scarce research in this field. Moreover, bacterial respiratory infections, which are highly variable and widespread, remain the leading cause of the global disease burden. It is also known that PMs can inhibit antibacterial activity, suppress the innate immunity of the respiratory epithelium, and induce bacterial imbalances, thereby increasing the risk of respiratory pathogen infections [2,25,26]. In addition, a direct relationship has been established between bacterial composition of ambient air and the microbiome in the lungs [27–30]. For example, a recent study found a relationship between the lung/respiratory microbiome and

PM_{2.5} bacterial constituent, determining that exposure to PM_{2.5} respirable particles affects the lungs through inflammation and oxidative stress [28]. Therefore, PM_{2.5} environment serves as a reservoir for bacteria, providing a habitat for bacteria and demonstrating its association with the human microbiome [31,32]. However, few studies have explored the behavior of bacteria under PM conditions.

The complex interactions may also disrupt normal cell behavior, including microorganisms, potentially exacerbating non-communicable diseases through altered metabolic pathways and persistent inflammation [33]. For instance, *Escherichia coli* (*E. coli*) is widely used as a model organism to understand stress responses and toxicological mechanisms [34]. Certain strains can also contribute to gut microbiota and extraintestinal infections, including respiratory complications, demonstrating relevance to lung disease mechanisms [35]. Shared oxidative and inflammatory pathways further link insights from *E. coli* studies to human lung health and chronic respiratory diseases [36,37]. Metabolic shifts in cells can be attributed to alterations in mitochondrial function and cellular bioenergetics, often initiated by reactive oxygen species (ROS) and exacerbated by the presence of MNPs [38,39]. These MNP-induced shifts can manifest as changes in glycolysis, oxidative phosphorylation, and lipid metabolism, ultimately influencing cellular energy homeostasis and contributing to chronic disease pathogenesis [40]. Fourier transform infrared (FTIR) spectroscopy can be employed to characterize the metabolic perturbations in the cells, for example, by identifying specific biochemical markers such as altered lipid profiles or protein modifications indicative of cellular stress and metabolic reprogramming [41]. Such analyses could reveal how MNPs and their leached constituents, in combination with PM_{2.5}, modulate fundamental cellular processes crucial for maintaining metabolic health [42]. However, there is still uncovered the metabolomic impact of MNP leaching on the cells.

Therefore, this study aims to elucidate the mechanistic effects of MNPs on PM_{2.5} by examining the changes induced by MNPs in the leaching chemistry, oxidative potential, inflammatory response, and bacteria-based cytological and metabolomic alterations of PM_{2.5}. PM_{2.5} extracts at three concentrations were exposed to secondary MNPs derived from real-world plastic products at low, medium and high doses for 24 h and then compared with non-exposed PM_{2.5} extracts. Changes in leaching indices (aromaticity-AR, humification-HI and slope factor-SF), oxidative potential (OP), antioxidant capacity and the inflammatory activity of PM_{2.5} when treated with various MNPs were quantified. Surface changes of MNPs after PM_{2.5} treatment were examined using FTIR. Furthermore, the cytological impact of MNP-treated and untreated PM_{2.5} was examined using *E. coli* responses, including growth, carbohydrates, lipid peroxidation (LPO), reactive oxygen species (ROS), and FTIR-based metabolomic fingerprints.

2. Materials and Methods

2.1. MNP Preparation and Characterization

Secondary MNPs were prepared using a ‘real-world approach’ that involved an in-house method obtained from plastic drinking water bottles, coffee cups, and food plates, which were purchased from markets in Türkiye [43,44]. After preparation of the plastic particles, sieving, cleaning, and room-temperature drying steps were conducted. The main characteristics of the MNPs were listed in Table 1 [43–45].

Table 1. General information on MNPs used throughout the study.

Properties	MNPs		
	Plastic Drinking Water Bottles	Cartoon Coffee Cups	Plastic Food Plates
Particle size distribution, nm	295–460 nm	350–890 nm	310–670 nm
Polymer types	Polyethylene terephthalate (PET)	PLA-Cellulose	Polypropylene (PP)
Elemental composition	C, Al, O	C, O, Al, Si, and Ti	C, O, Al
Functional groups	aromatic CH at 1713 cm ^{−1} C-O at 1094 cm ^{−1} and 1241 cm ^{−1} C=O at 720 cm ^{−1}	OH at 3350 cm ^{−1} CH ₂ at 2915–2910 cm ^{−1} C-O at 1750–1740 cm ^{−1} C-O (ester/carboxylic acid) at 1640 cm ^{−1} CH ₃ at 1373 cm ^{−1} C-OH at 1162 cm ^{−1} and 1070 cm ^{−1}	aromatic C-H at 3024 cm ^{−1} C-H stretch at 2847 cm ^{−1} aromatic ring stretch at 1492 cm ^{−1} and 1601 cm ^{−1} CH ₂ 1451 cm ^{−1} aromatic CH bend 1027 cm ^{−1} aromatic CH at 694 cm ^{−1} aromatic ring out-of-plane bend 537 cm ^{−1}
Color	Light blue	White	White

2.2. PM_{2.5} Sampling and Preparation

PM_{2.5} samples were collected in Maslak, Istanbul, Türkiye, using a high-volume sampler AirFlow PM_{2.5} (Analitica Strumenti, Pesaro, Italy) [46–48]. The samples were collected between April 2023 and October 2024 during 96 h sampling period using a quartz filter. The sampling site in Türkiye was situated in the Maslak district of Istanbul, within the Istanbul Technical University campus, approximately 15 m from a heavily trafficked roadway. The geographic coordinates of the site were 41°06.19' N (latitude) and 29°01.25' E (longitude). During the sampling period, average bidirectional vehicular traffic was approximately 4300 vehicles per hour near the Maslak campus. These were then stored in a cold room in the dark at 4 °C until analysis. To compensate for insufficient sample loading and to eliminate the effects of variations in the chemical composition of the samples, 23 sample filters (spring-summer) from the same sampling location were combined and pooled for all analyses [49,50]. The pooled sample application is a frequently used procedure in cytological studies of PM_{2.5}. The mass concentration of PM_{2.5} was quantified by gravimetric analysis, based on pre- and post-weighing of quartz filters using a five-digit ultra-microbalance. Collected PM_{2.5} samples were pooled and extracted in ultrapure water to obtain a stock suspension equivalent to a high mass concentration of 30.0 µg/m³. Based on a total sampled air volume of 2880 m³ and an extraction volume of 1.0 L, an ambient PM_{2.5} concentration of 30 µg/m³ corresponded to an extract concentration of 86.4 µg/mL. From the high-concentration stock, lower (1.0 µg/m³ = 2.9 µg/mL) and medium (10 µg/m³ = 28.8 µg/mL) PM_{2.5} suspensions were prepared using ultrapure water (Milli-Q, Merck, Darmstadt, Germany). The extracts were passed through a 0.2 µm syringe filter [48–52]. This extraction, filtration and analysis protocol is widely applied in metal characterization and cytotoxicity studies [48–52]. In addition, filtration was performed to achieve sterilization and homogenization of the samples and to reduce interference from potential microbial contaminants. This procedure is commonly used in bacterial growth and toxicity studies of environmental samples [48–52]. Blank quartz filters were processed in parallel as procedural controls, and replicate samples were prepared for each concentration. Parallel set-ups were conducted for the low–medium–high PM_{2.5} exposure levels.

2.3. Exposure Experiments Between MNPs and PM_{2.5}

Experimental processes were illustrated in Supplementary Figure S1. To expose the PM_{2.5} to MNPs, three doses of MNPs were used: low: 10 µg/mL (1×), medium: 50 µg/mL (5×), and high: 200 µg/mL (20×) were weighed using an ultra-microbalance. This was based on available MNP cytotoxicity studies and the occurrence of MNPs in

atmospheric samples. The selected MNP doses reflect MNP toxicity doses reported in the literature between 1 and 1000 $\mu\text{g/mL}$ [48]. The MNPs were then dispersed into $\text{PM}_{2.5}$ extracts at three concentrations. MNP-contained and uncontaminated $\text{PM}_{2.5}$ mixtures (controls) were placed under natural illumination with gentle stirring at a 24-h interval. After the treatment duration (24 h) had ended, the MNPs and $\text{PM}_{2.5}$ supernatant were separated. Exposures were conducted in a parallel setup. Considering parallel $\text{PM}_{2.5}$ concentrations, four replicates for each exposure.

The extracts of MNP-treated $\text{PM}_{2.5}$ samples and the controls (filter blank $\text{PM}_{2.5}$ and only $\text{PM}_{2.5}$) were filtered to avoid MNP particles, then examined for UV-Vis spectral (leaching) indices, oxidative inputs, inflammation activity, and bacteria-based cytology and metabolism. All exposures were three replicates; thus total of 12 data sets were obtained for each parameter with three readings. To examine the MNPs' effect on $\text{PM}_{2.5}$, eliminate $\text{PM}_{2.5}$ contaminants and the dilution factor errors between the PM concentration, the results were presented as the ratio between the MNP-treated $\text{PM}_{2.5}$ in each $\text{PM}_{2.5}$ concentration group (i.e., the sample) and the $\text{PM}_{2.5}$ without MNP in each $\text{PM}_{2.5}$ concentration group (i.e., the control).

$\text{PM}_{2.5}$ -treated MNPs and control (pristine, no $\text{PM}_{2.5}$ exposed) MNPs were characterized using Fourier transform infrared–attenuated total reflection spectroscopy (FTIR–ATR).

2.4. FTIR Characterization of MNPs After $\text{PM}_{2.5}$ Treatments

The $\text{PM}_{2.5}$ -treated MNPs and control (pristine) MNPs were characterized using FTIR–ATR (FTIR–ATR, Bruker Invenio S, Billerica, MA, USA). FTIR–ATR analysis performed in the range from 4000 to 400 cm^{-1} . All the samples were analyzed in triplicate, and each spectrum was the sum of 64 scans.

2.5. Spectral Analysis of the Treated and Non-Treated $\text{PM}_{2.5}$ for MNP Leaching Characterization

After MNP removal from the $\text{PM}_{2.5}$ mixtures, the $\text{PM}_{2.5}$ samples were analyzed for spectral indices (AR, HI, and SF) to characterize the leaching of MNPs into the $\text{PM}_{2.5}$ [53,54]. This procedure relies on the direct UV-Vis measurement of $\text{PM}_{2.5}$ samples by a UV-Vis microplate reader (Multiskan SkyHigh, Thermo Scientific, Cleveland, OH, USA) and calculates indices using absorbances at specific wavenumbers. Multiple indices were used, including aromaticity demonstrating aromatic ring substitution ($[\text{Abs at } 253 \text{ nm}]/[\text{Abs at } 203 \text{ nm}]$), humification indicating organic solute aromaticity ($[\text{Abs at } 250 \text{ nm}]/[\text{Abs at } 365 \text{ nm}]$) and slope factor reflecting the balance of low-versus high-molecular-weight chromophores ($[\text{Abs at } 275 \text{ nm} - \text{Abs at } 295 \text{ nm}]/[\text{Abs at } 350 \text{ nm} - \text{Abs at } 400 \text{ nm}]$) [53,54]. Absorbance is used as a.u., and in the index calculation, since each index is a ratio, the units cancel out, therefore, the results are unitless. The data for each index is presented as the ratio between the MNP-treated $\text{PM}_{2.5}$ in each $\text{PM}_{2.5}$ concentration group (i.e., the sample) and the $\text{PM}_{2.5}$ without MNP in each $\text{PM}_{2.5}$ concentration group (i.e., the control), to examine only MNP contamination in each $\text{PM}_{2.5}$ concentration and eliminate $\text{PM}_{2.5}$ contaminants and the dilution factor.

2.6. Oxidative Characterization of $\text{PM}_{2.5}$ Due to MNP Exposure

Oxidative indicators in control $\text{PM}_{2.5}$ (the $\text{PM}_{2.5}$ without MNP) in each $\text{PM}_{2.5}$ concentration and MNP-exposed $\text{PM}_{2.5}$ samples were evaluated using the Dithiothreitol (DTT) assay for oxidative potential (OP) and CUPRAC antioxidant activity [55,56]. For the DTT assay, 198 μL of sample and 22 μL of DTT were incubated at 37 $^{\circ}\text{C}$ for 30 min; 50 μL of the mixture was then combined with 50 μL of DTNB, and absorbance was measured at 412 nm. The consumption results are presented as the ratio between the MNP-treated $\text{PM}_{2.5}$ in each $\text{PM}_{2.5}$ concentration group (i.e., the sample) and the $\text{PM}_{2.5}$ without MNP in each $\text{PM}_{2.5}$

concentration group (i.e., the control), to examine only MNP contamination in each PM_{2.5} concentration and eliminate PM_{2.5} contaminants and the dilution factor.

Antioxidant activity was assessed using the CUPRAC method [55] by mixing sample extracts with CuCl₂, neocuproine, and ammonium acetate, followed by 30 min incubation and absorbance measurement at 450 nm. The results are presented as the ratio between the MNP-treated PM_{2.5} in each PM_{2.5} concentration group (i.e., the sample) and the PM_{2.5} without MNP in each PM_{2.5} concentration group (i.e., the control), to examine only MNP contamination in each PM_{2.5} concentration and eliminate PM_{2.5} contaminants and the dilution factor. Related blank applications and Trolox-based calibration for the quality control were applied.

2.7. Inflammatory Activity of PM_{2.5} Due to MNP Exposure

Inflammatory activity was assessed via protein-denaturation inhibition. Briefly, sample suspensions (controls PM_{2.5} and MNP-exposed PM_{2.5}) were mixed with 1.0% BSA, heated at 72 °C for 5 min, and cooled. A BSA–water mixture served as the control. The absorbance (a.u.) was measured at 660 nm [57]. The results are presented as the ratio between the MNP-treated PM_{2.5} in each PM_{2.5} concentration group (i.e., the sample) and the PM_{2.5} without MNP in each PM_{2.5} concentration group (i.e., the control), to examine only MNP contamination in each PM_{2.5} concentration and eliminate PM_{2.5} contaminants and the dilution factor. Related blank applications were applied.

2.8. Bacteria-Based Cytological and Metabolomic Characterization of PM_{2.5} Due to MNP Exposure

To examine cytological and metabolomic changes, *E. coli* obtained from the American Type Culture Collection (ATCC) was used in controls and MNP-exposed PM_{2.5} samples. These were diluted in Typtic Soy Broth (TSB) containing saturated *E. coli* cultures grown in TSB broth (2:1 TSB: sample). For the negative controls, bacteria in TSB without PM_{2.5} were also used. The samples were incubated separately with bacteria and then analysed. All growth experiments were conducted as three replicates. The controls and samples were incubated in a dark oven at 37 °C for 24 h. Then, the optical density (OD), carbohydrate, lipid, and ROS assays were performed on the bacteria in each of the MNP-exposed and control PM_{2.5} samples, as well as the negative control.

The ODs were measured using a UV–Vis microplate reader [43,44]. Unit of the OD is a.u., and the results are presented as normalized values using negative control (only *E. coli*).

Carbohydrate content in the EPSs was determined using the phenol-sulfuric acid method and measured at 480 nm [58]. The absorbances were measured for the Carbohydrate as a.u., and the results are presented as normalized values using a negative control (only *E. coli*). Blank corrections were applied. For quality control, glucose-, fructose-, and mannose-based calibration was applied.

LPO activity was determined using the thiobarbituric acid (TBA) method and read at 532 nm against a blank. The absorbances were measured for the LPO as a.u., and the results are presented as normalized values using a negative control (only *E. coli*). Blank corrections were applied to the calculation.

The DCFH-DA assay was used to determine ROS, with fluorescence recorded at an excitation wavelength of 488 nm and an emission wavelength of 530 nm (Thermo Scientific, Cleveland, OH, USA) [54]. The absorbances were measured for the ROS as a.u., and the results are presented as normalized values using a negative control (only *E. coli*). Blank corrections were applied to the calculation.

For metabolomics, cell suspensions were centrifuged at 3500 × g for 4 min at 4 °C. The supernatant was then removed, and the cell precipitation was measured directly by FTIR-ATR spectrometry (FTIR-ATR, Bruker Invenio S, Billerica, MA, USA). FTIR-ATR analysis

performed in the range from 4000 to 400 cm^{-1} with 64 scans. All the cell suspensions were tested in triplicate in each replicate, and mean values were presented.

2.9. Statistical Analysis

ANOVA with post hoc Tukey with one- and two-tailed was used to analyse the differences between the control and the samples, as well as the differences among the samples, performed using SPSS 19.0 software (IBM, Armonk, NY, USA). Values of $p < 0.05$ were considered statistically significant.

3. Results

3.1. FTIR Characterization of MNP After $\text{PM}_{2.5}$ Treatment

Table 2 and Supplementary Figure S2 shows FTIR spectra and functional groups of plastic drinking water bottles (PET-derived) MNPs after $\text{PM}_{2.5}$ treatment. Compared to bare PET MNPs, all $\text{PM}_{2.5}$ -treated PET samples exhibited consistent changes in functional group intensities. The absorbance at 1701 cm^{-1} (C=O stretching) increased across all treatments, as reflected by the increased carbonyl index (CI), which increased from 0.57 (for bare PET) to between 0.91 and 1.18. Similarly, the hydroxyl index (HI) increased markedly from 0.02 in the untreated drinking water bottle MNPs to 1.63–2.28 in the treated samples. The normalised intensities of aliphatic C–H stretching bands at 2900 and 2987 cm^{-1} decreased relative to the bare form. Minor variations were observed in the C–O stretching region (1066–1250 cm^{-1}), with the C–O index remaining relatively stable (0.92–0.94). Characterization of the leachate via FTIR revealed the presence of carbonyl (C=O), hydroxyl (–OH), and ester-related C–O chemical groups.

Table 2. Functional groups and oxidative indexes of plastic water bottles. Bare MNP: no $\text{PM}_{2.5}$ treatment applied, MNP1× + $\text{PM}_{2.5}$ Low: Treatment between low dose of MNP and low concentration of $\text{PM}_{2.5}$, MNP1× + $\text{PM}_{2.5}$ Medium: Treatment between low dose of MNP and medium concentration of $\text{PM}_{2.5}$, MNP1× + $\text{PM}_{2.5}$ High: Treatment between low dose of MNP and high concentration of $\text{PM}_{2.5}$, MNP5× + $\text{PM}_{2.5}$ Low: Treatment between medium dose of MNP and low concentration of $\text{PM}_{2.5}$, MNP5× + $\text{PM}_{2.5}$ Medium: Treatment between medium dose of MNP and medium concentration of $\text{PM}_{2.5}$, MNP5× + $\text{PM}_{2.5}$ High: Treatment between medium dose of MNP and high concentration of $\text{PM}_{2.5}$, MNP20× + $\text{PM}_{2.5}$ Low: Treatment between high dose of MNP and low concentration of $\text{PM}_{2.5}$, MNP20× + $\text{PM}_{2.5}$ Medium: Treatment between high dose of MNP and medium concentration of $\text{PM}_{2.5}$, MNP20× + $\text{PM}_{2.5}$ High: Treatment between high dose of MNP and high concentration of $\text{PM}_{2.5}$. CI: Carbonyl index, HI: Hydroxyl index, and C–O index.

Wavelength, nm	Control MNP	Plastic Bottle MNP								
		MNP1× + $\text{PM}_{2.5}$ Low	MNP1× + $\text{PM}_{2.5}$ Medium	MNP1× + $\text{PM}_{2.5}$ High	MNP5× + $\text{PM}_{2.5}$ Low	MNP5× + $\text{PM}_{2.5}$ Medium	MNP5× + $\text{PM}_{2.5}$ High	MNP20× + $\text{PM}_{2.5}$ Low	MNP20× + $\text{PM}_{2.5}$ Medium	MNP20× + $\text{PM}_{2.5}$ High
1066	0.92	0.85	0.84	0.85	0.85	0.85	0.84	0.84	0.84	0.85
1250	0.86	0.93	0.93	0.93	0.93	0.93	0.92	0.93	0.93	0.93
1406	0.95	0.93	0.92	0.93	0.93	0.93	0.92	0.92	0.92	0.92
1701	0.90	0.95	0.94	0.95	0.95	0.95	0.95	0.95	0.94	0.95
2900	1.00	0.91	0.90	0.91	0.90	0.91	0.90	0.90	0.90	0.91
2987	0.99	0.89	0.89	0.89	0.89	0.89	0.88	0.88	0.89	0.89
CI	0.57	1.09	1.09	1.11	1.08	1.18	0.91	1.15	1.07	1.02
HI	0.02	2.20	1.93	2.14	2.00	2.23	1.63	2.28	1.86	1.97
C–O index	0.92	0.94	0.94	0.94	0.94	0.94	0.93	0.94	0.93	0.94

Following PM_{2.5} exposure, the functional group intensities of coffee cup (PLA-cellulose) MNPs showed considerable variability (Table 3 and Supplementary Figure S3). The hydroxyl index (HI) increased from 0.20 for bare cellulose to 0.33–3.08, with the highest values observed in low-dose treatments. Similarly, the carbonyl index (CI) increased from 0.17 in bare cellulose to 0.32–1.24 following treatment. The C–O stretching bands (1016–1055 cm^{−1}) exhibited significant intensity fluctuations depending on the concentrations of PM_{2.5} and MNPs. Several treated samples showed decreased aliphatic C–H stretching bands (2900–2987 cm^{−1}) compared to the bare form. As shown in the FTIR spectra and Table 3, the detected leached functional groups included carbonyl-containing species (C=O), hydroxyl-containing species (–OH), and ester-related C–O functionalities.

Table 3. Functional groups and carbonyl index of the plastic coffee cups (cellulose MNP). Bare MNP: no PM_{2.5} treatment applied, MNP1 × + PM_{2.5} Low: Treatment between low dose of MNP and low concentration of PM_{2.5}, MNP1 × + PM_{2.5} Medium: Treatment between low dose of MNP and medium concentration of PM_{2.5}, MNP1 × + PM_{2.5} High: Treatment between low dose of MNP and high concentration of PM_{2.5}, MNP5 × + PM_{2.5} Low: Treatment between medium dose of MNP and low concentration of PM_{2.5}, MNP5 × + PM_{2.5} Medium: Treatment between medium dose of MNP and medium concentration of PM_{2.5}, MNP5 × + PM_{2.5} High: Treatment between medium dose of MNP and high concentration of PM_{2.5}, MNP20 × + PM_{2.5} Low: Treatment between high dose of MNP and low concentration of PM_{2.5}, MNP20 × + PM_{2.5} Medium: Treatment between high dose of MNP and medium concentration of PM_{2.5}, MNP20 × + PM_{2.5} High: Treatment between high dose of MNP and high concentration of PM_{2.5}. CI: Carbonyl index, HI: Hydroxyl index, and C–O index.

Coffee Cups MNP										
Wavelength, nm	Control MNP	MNP1 × + PM _{2.5} Low	MNP1 × + PM _{2.5} Medium	MNP1 × + PM _{2.5} High	MNP5 × + PM _{2.5} Low	MNP5 × + PM _{2.5} Medium	MNP5 × + PM _{2.5} High	MNP20 × + PM _{2.5} Low	MNP20 × + PM _{2.5} Medium	MNP20 × + PM _{2.5} High
896	0.93	0.94	0.98	0.97	0.92	0.98	0.96	0.87	0.98	0.90
1055	0.86	0.83	0.96	0.96	0.81	0.97	0.92	0.70	0.96	0.80
1016	0.84	0.89	0.96	0.96	0.84	0.97	0.92	0.73	0.96	0.79
1701	0.99	0.95	0.99	0.99	0.96	0.99	0.98	0.94	0.99	0.96
2900	0.97	0.89	0.99	0.99	0.90	0.99	0.98	0.88	0.99	0.94
2987	0.98	0.87	0.99	0.98	0.89	0.99	0.98	0.87	0.99	0.96
CI	0.17	1.24	0.46	0.48	0.57	0.46	0.40	0.44	0.32	0.35
HI	0.20	3.08	0.16	0.19	1.38	0.33	0.32	0.90	0.18	0.36
C–O index	0.88	0.94	0.97	0.97	0.89	0.98	0.94	0.80	0.97	0.84

Exposure to PM_{2.5} resulted in moderate but consistent spectral changes in PP MNPs (Table 4 and Supplementary Figure S4). The carbonyl index (CI) increased from 0.21 in the untreated sample to 0.61–0.92 in the treated samples. The hydroxyl index (HI) increased from 0.20 to 1.21–2.79. The normalised intensities of aliphatic C–H stretching bands at 2900 and 2987 cm^{−1} showed minimal variation compared to the untreated form. The C–O index remained close to unity (1.00–1.03) across all conditions. The FTIR analysis of the leachate identified several key functional groups, specifically carbonyl-containing species (C=O), hydroxyl-containing species (–OH), and minor C–O functionalities.

Table 4. Functional groups and carbonyl index of the plastic food plates (PP MNP). Bare MNP: no PM_{2.5} treatment applied, MNP1× + PM_{2.5} Low: Treatment between low dose of MNP and low concentration of PM_{2.5}, MNP1× + PM_{2.5} Medium: Treatment between low dose of MNP and medium concentration of PM_{2.5}, MNP1× + PM_{2.5} High: Treatment between low dose of MNP and high concentration of PM_{2.5}, MNP5× + PM_{2.5} Low: Treatment between medium dose of MNP and low concentration of PM_{2.5}, MNP5× + PM_{2.5} Medium: Treatment between medium dose of MNP and medium concentration of PM_{2.5}, MNP5× + PM_{2.5} High: Treatment between medium dose of MNP and high concentration of PM_{2.5}, MNP20× + PM_{2.5} Low: Treatment between high dose of MNP and low concentration of PM_{2.5}, MNP20× + PM_{2.5} Medium: Treatment between high dose of MNP and medium concentration of PM_{2.5}, MNP20× + PM_{2.5} High: Treatment between high dose of MNP and high concentration of PM_{2.5}. CI: Carbonyl index, HI: Hydroxyl index, and C-O index.

Plastic Food Plates MNP										
Wavelength, nm	Control MNP	MNP1× + PM _{2.5} Low	MNP1× + PM _{2.5} Medium	MNP1× + PM _{2.5} High	MNP5× + PM _{2.5} Low	MNP5× + PM _{2.5} Medium	MNP5× + PM _{2.5} High	MNP20× + PM _{2.5} Low	MNP20× + PM _{2.5} Medium	MNP20× + PM _{2.5} High
878	0.79	0.94	0.96	0.96	0.96	0.95	0.96	0.95	0.97	0.96
1065	0.97	0.87	0.91	0.88	0.92	0.88	0.89	0.90	0.95	0.93
1250	0.94	0.94	0.96	0.95	0.96	0.94	0.95	0.95	0.97	0.96
1394	0.79	0.93	0.95	0.94	0.96	0.94	0.94	0.94	0.97	0.96
1712	0.98	0.98	0.97	0.98	0.98	0.97	0.98	0.98	0.98	0.98
2900	0.87	0.92	0.94	0.92	0.95	0.92	0.93	0.93	0.97	0.96
2987	0.98	0.90	0.94	0.91	0.94	0.91	0.92	0.93	0.97	0.95
CI	0.21	0.66	0.92	0.68	0.62	0.86	0.66	0.61	0.72	0.77
HI	0.20	2.53	2.09	2.79	1.91	2.62	2.40	2.16	1.21	1.45
C-O index	1.09	1.03	1.01	1.03	1.01	1.03	1.02	1.02	1.00	1.01

3.2. Leaching Effects of MNP on PM_{2.5}: Spectral Indices (Aromaticity, Humification, and Slope Factor) of PM_{2.5}

Figure 1 shows the AR, HI and SF at various PM_{2.5} concentrations with different types and doses of MNP exposures. Plastic water bottle-derived MNPs generated the most pronounced increases across all parameters, including the highest aromaticity values at medium PM_{2.5} concentration (AR = 1.02), the strongest humification at high PM_{2.5} concentration (HI = 1.66), and consistently elevated SFs (up to SF = 1.25). Coffee cup MNPs induced moderate but consistent increases, particularly at the high PM_{2.5} load, where both humification (HI = 1.46–1.56) and slope factor (SF = 1.20–1.33) rose substantially, however, their effects were more variable at low PM_{2.5}. Plastic food plate MNPs produced the largest reductions at low PM_{2.5} concentration, especially in humification (HI as low as 0.34) and slope factor (SF = 0.22). However, at medium and high PM_{2.5} concentrations, food-plate MNPs also supported significant humification (HI up to 1.62) and SF increases (SF up to 1.23).

MNP exposure induced distinct alterations in the optical and chromophoric character of PM_{2.5}, with responses varying by polymer type, dose, and PM_{2.5} concentration (Figure 1). As shown in Figure 1A, the AR remained close to control for all treatments (0.99–1.02), however, at higher PM_{2.5} loads, several conditions exhibited higher AR values than the controls—most prominently the 20× bottle-derived MNP treatment at low and medium PM_{2.5} concentrations. Lower PM_{2.5} concentrations generally showed AR ≤ 1.00.

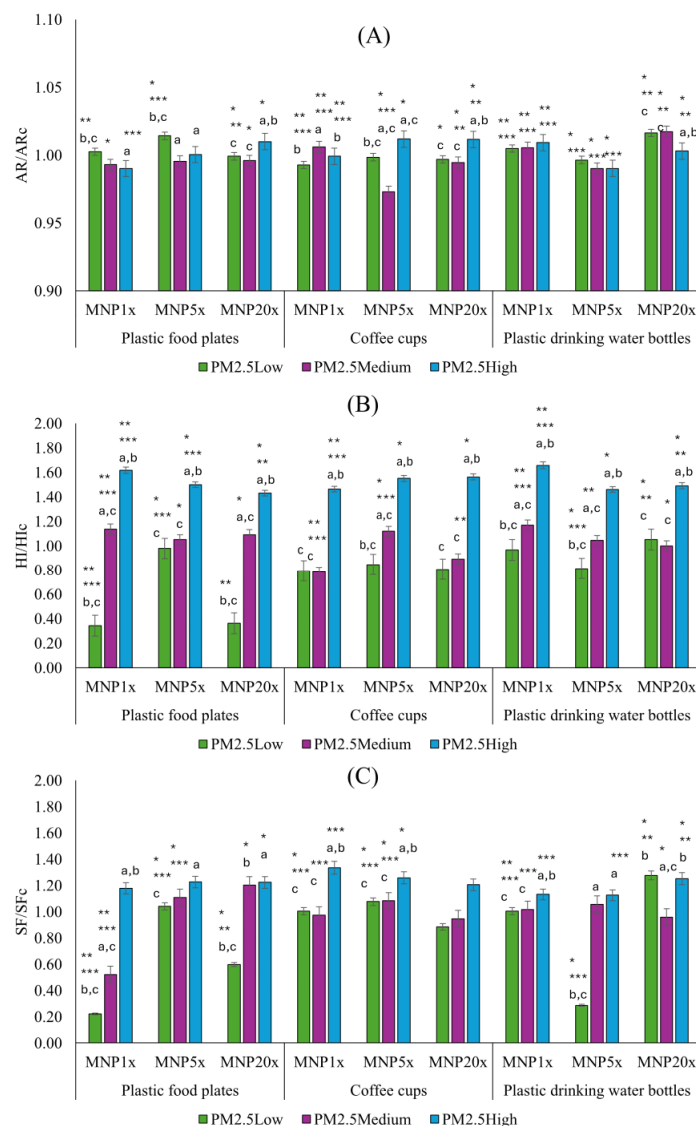


Figure 1. Spectral indices ratio of PM_{2.5} with and without MNP treatment. (A) Aromaticity index (AR) changes in three concentrations of PM_{2.5} samples with various MNP exposures (AR: Aromaticity index in MNP-exposed PM_{2.5}, ARC: Aromaticity index in control PM_{2.5}), (B) Humification index (HI) changes in three concentrations of PM_{2.5} samples with various MNP exposures (HI: Humification index in MNP-exposed PM_{2.5}, HIC: Humification index in control PM_{2.5}), (C) Slope factor (SF) changes in three concentrations of PM_{2.5} samples with various MNP exposures (SF: Slope factor index in MNP-exposed PM_{2.5}, HIC: Slope factor in control PM_{2.5}). The letters and signs above the bars indicated the statistical difference ($p < 0.05$): (a) difference with MNP treatments for PM_{2.5} Low, (b) difference with MNP treatments for PM_{2.5} Medium, (c) difference with MNP treatments for PM_{2.5} High, (*) difference with MNP1x, (**) difference with MNP5x, and (***) difference with MNP20x.

HI demonstrated the strongest MNP-induced responses, spanning a broad range from 0.34 to 1.66 (Figure 1B). Low PM_{2.5} samples consistently showed HI < 1 for all polymer types, however, medium and especially high PM_{2.5} concentrations exhibited marked increases in humification.

As illustrated in Figure 1C, low PM_{2.5} samples showed highly variable SF responses with MNP exposures, including strong reductions (0.22–0.29) and increases (>1.27). In contrast, medium PM_{2.5} concentration remained mostly stable around control when MNP exposures. High PM_{2.5} concentration conditions with MNP exposures consistently exhibited SF > 1.12 for all polymers and doses.

3.3. Effect of MNPs on Oxidative Indicators of PM_{2.5}

Figure 2 illustrates oxidative stress-related indicators, including OP and antioxidant activity in PM_{2.5} with various MNP exposures. As shown in Figure 2A, MNPs derived from plastic food plates induced only marginal deviations from control PM_{2.5}. DTT ratios remained close to all PM_{2.5} loads, ranging from 0.98 to 1.03 at low PM_{2.5} concentrations, from 1.00 to 1.03 at medium PM_{2.5} concentrations, and from 0.98 to 1.02 at high PM_{2.5} concentrations, confirming minimal modulation of OP. In contrast, coffee cup MNPs exhibited a more pronounced response. At low PM_{2.5}, OP ratios increased progressively from 1.02 (MNP1×) to 1.07 (MNP20×), while at medium PM_{2.5}, there were sustained elevations (1.00–1.04). The strongest oxidative stimulation occurred with MNP1× at high PM_{2.5}, where the OP ratio reached 1.20, exceeding that of all other treatments. Plastic drinking water bottle MNPs displayed a biphasic pattern. At low PM_{2.5}, OP ratios increased modestly (to 1.05 with MNP1×), but declined towards or below unity with an increasing MNP dose. This suppressive effect was most evident at high PM_{2.5}, where the OP ratio decreased from 1.00 (MNP1×) to 0.92 (MNP20×), indicating reduced DTT consumption at higher MNP loadings of PM_{2.5}.

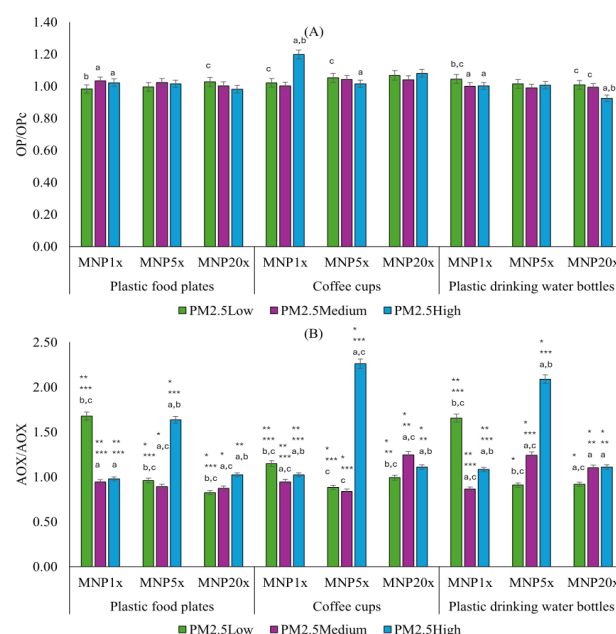


Figure 2. Oxidative indicators for the effect of MNP on PM_{2.5}. (A) Oxidative potential (OP) changes by DTT assay in three concentrations of PM_{2.5} samples with various MNP exposures (OP: Oxidative potential in MNP-exposed PM_{2.5} (au), OPc: Oxidative potential index in control PM_{2.5} (au)), (B) Antioxidant activity changes by CUPRAC method in three concentrations of PM_{2.5} samples with various MNP exposures (AOX: Antioxidant activity in MNP-exposed PM_{2.5} (au), AOXc: Antioxidant activity in control PM_{2.5} (au)). The letters and signs above the bars indicated the statistical difference ($p < 0.05$): (a) difference with MNP treatments for PM_{2.5} Low, (b) difference with MNP treatments for PM_{2.5} Medium, (c) difference with MNP treatments for PM_{2.5} High, (*) difference with MNP1×, (**) difference with MNP5×, and (***) difference with MNP20×.

Figure 2B shows CUPRAC-based antioxidant activity ratios. The antioxidant capacity of PM_{2.5} produced by food plate MNPs increased and decreased depending on the concentration of PM_{2.5} and the dose of MNPs. At low PM_{2.5} levels, the CUPRAC ratio decreased from 1.68 (MNP1×) to 0.82 (MNP20×). At medium PM_{2.5} concentrations, antioxidant activity consistently declined (from 0.87 to 0.94), whereas a marked increase was observed at MNP5× (1.63) at high PM_{2.5} concentrations. Coffee cup MNP contamination generated stronger shifts in PM_{2.5} overall. Antioxidant activity increased moderately at low PM_{2.5}

concentrations (1.15 for MNP1×) and rose sharply at high concentrations, peaking at 2.26 for MNP5×—the highest CUPRAC ratio observed across all treatments. Contamination from plastic drinking water bottles elicited the largest antioxidant responses in PM_{2.5}. At low PM_{2.5}, CUPRAC activity reached 1.65 (MNP1×). Under a high PM_{2.5} burden, antioxidant activity was consistently elevated across all doses, ranging from 1.08 (MNP1×) to 2.09 (MNP5×). This demonstrates a substantial enhancement relative to the control PM_{2.5}.

3.4. Effect of MNPs on Inflammation Response of PM_{2.5}

As schemed in Figure 3, in plastic food plates (PP-Based MNPs), across all PM_{2.5} concentrations, inflammation ratios for food plate MNPs remained close to controls, with only modest increases at 1× doses (approx. 1.11 for PM_{2.5}-Low and High concentrations) and near-neutral or slightly reduced values at 5× and 20× MNP doses. Coffee cup-derived MNPs produced the strongest inflammatory response among all polymers. At PM_{2.5}-Low and PM_{2.5}-High concentrations, the 5× MNP dose resulted in a marked increase (inflammation ratio approx. 1.64), and even at 20× MNPs, values remained elevated (1.28–1.14). Plastic drinking water bottles-derived MNPs generated a moderate but consistent pro-inflammatory effect at PM_{2.5}-Low and High concentrations (ratios approx. 1.05–1.24 across doses) but significantly suppressed inflammation at PM_{2.5}-Medium (0.79–0.86).

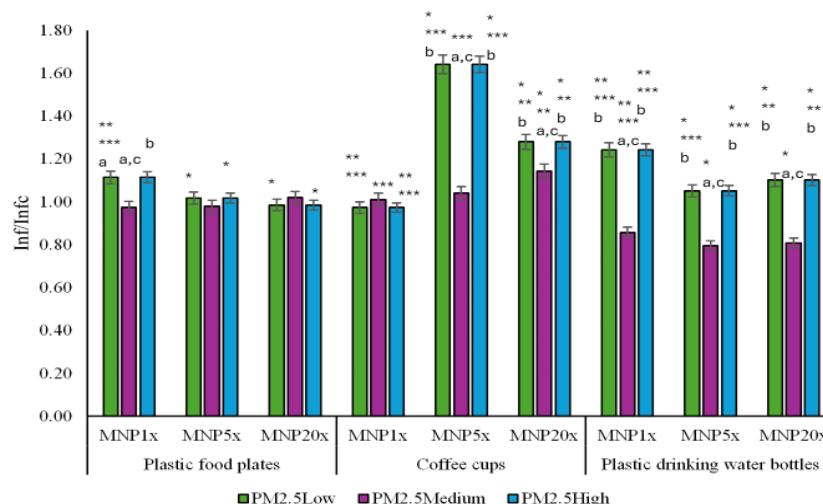


Figure 3. Inflammation (Inf) changes by protein-denaturation assay in three concentrations of PM_{2.5} samples with various MNP exposures (Inf: Inflammation in MNP-exposed PM_{2.5} (au), InfC: Inflammation in control PM_{2.5} (au)). The letters and signs above the bars indicated the statistical difference ($p < 0.05$): (a) difference with MNP treatments for PM_{2.5} Low, (b) difference with MNP treatments for PM_{2.5} Medium, (c) difference with MNP treatments for PM_{2.5} High, (*) difference with MNP1×, (**) difference with MNP5×, and (***) difference with MNP20×.

3.5. Effect of MNPs on PM_{2.5} Cytological and Metabolomic Changes by *E. coli*

Figure 4 depicts cytological and metabolomic indicators of *E. coli* with MNP-exposed PM_{2.5}. As shown in Figure 4A, at low PM_{2.5}, bacterial OD was mostly suppressed (0.69–0.96), except for the coffee-cup 5× condition (1.50). Medium PM_{2.5} induced more divergent patterns, with increases for plastic-plate 1× (1.32) and drinking-bottle 5×–20× (1.15–1.27), but suppression for coffee-cup 5× (0.78). At high PM_{2.5}, OD values converged around 0.85–1.05 across all polymers.

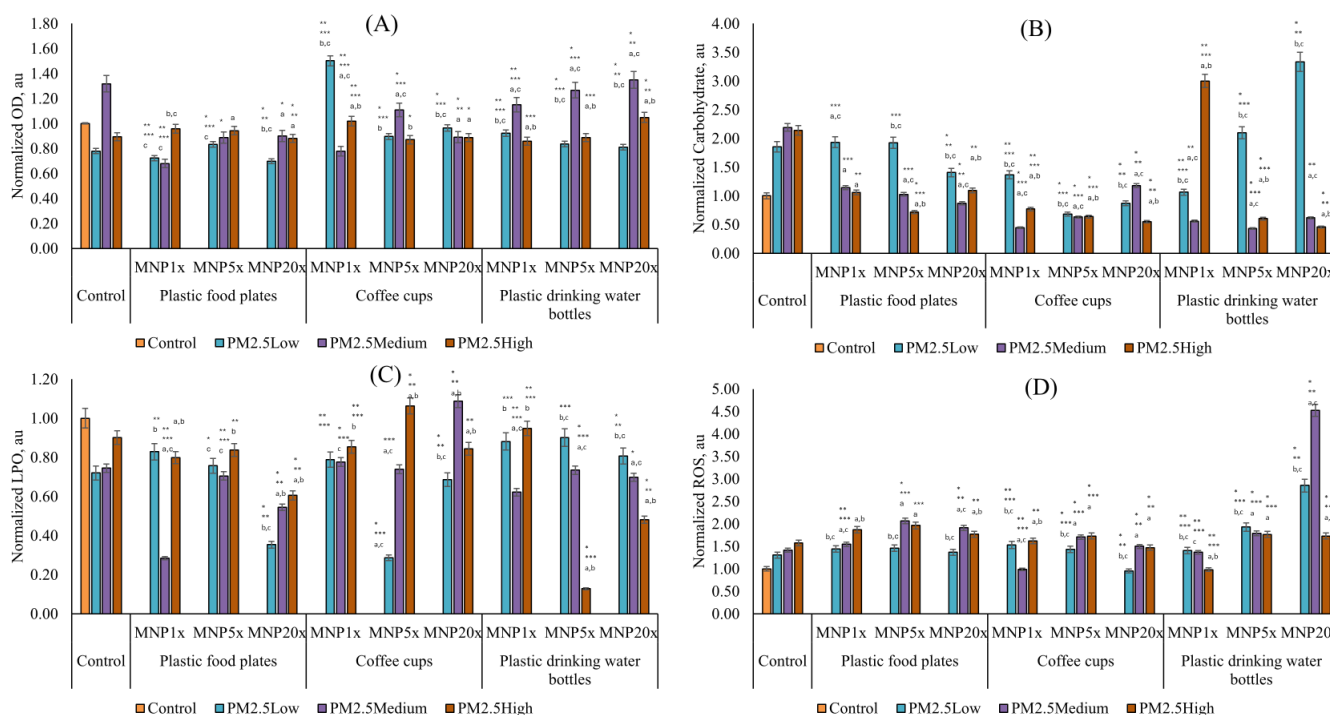


Figure 4. Cytological indicators of *E. coli* with MNP-exposed PM_{2.5} and without MNP-exposed PM_{2.5}. (A) Normalized OD (au) of *E. coli* exposed to without and with MNP-treated PM_{2.5}. (B) Normalized Carbohydrate (au) of *E. coli* exposed to without and with MNP-treated PM_{2.5}. (C) Normalized LPO (au) of *E. coli* exposed to without and with MNP-treated PM_{2.5}. (D) Normalized ROS (au) of *E. coli* exposed to without and with MNP-treated PM_{2.5}. The letters and signs above the bars indicated the statistical difference ($p < 0.05$): (a) difference with MNP treatments for PM_{2.5} Low, (b) difference with MNP treatments for PM_{2.5} Medium, (c) difference with MNP treatments for PM_{2.5} High, (*) difference with MNP1 $\times$, (**) difference with MNP5 $\times$, and (***) difference with MNP20 $\times$.

Carbohydrate levels of *E. coli* presented in Figure 4B showed marked fluctuations with MNP-treated PM_{2.5}. At low PM_{2.5}, higher carbohydrate levels were strongly enhanced across most conditions (1.40–3.33), with the highest value observed for drinking-bottle MNP20 $\times$ (3.33). Medium PM_{2.5} generally reduced carbohydrate levels (0.43–1.18), particularly for coffee-cup 5 $\times$ (0.45) and drinking-bottle 20 $\times$ (0.62). High PM_{2.5} induced mixed responses, including a sharp increase for coffee-cup 5 $\times$ (3.00), while several drinking-bottle treatments remained below 0.65.

LPO activity of *E. coli* remained largely below the control level in most exposure conditions (Figure 4C). At low PM_{2.5}, LPO was strongly reduced for coffee-cup 1 $\times$ (0.35) and 20 $\times$ (0.29). Medium PM_{2.5} maintained suppressed values (0.28–0.88), except for plastic-plate 20 $\times$ (1.09). At high PM_{2.5}, most values remained between 0.60 and 0.95, although drinking-bottle MNP at 20 $\times$ showed a dramatic decline (0.13).

As illustrated in Figure 4D, ROS responses of *E. coli* showed the clearest and most consistent toxicity signal across treatments. At low PM_{2.5}, ROS increased to 1.30–2.85, with the strongest induction observed in drinking-bottle MNP20 $\times$. Medium PM_{2.5} further amplified oxidative stress, particularly for bottle MNP20 $\times$ (4.53). High PM_{2.5} maintained elevated ROS levels (1.57–1.97) across most polymer types.

Across all wavenumbers, the FTIR data show that *E. coli* biochemical fingerprints are subtly but consistently perturbed by PM_{2.5}, and that MNP-treated PM_{2.5} modulates these effects in a polymer-, dose-, and PM_{2.5}-concentration-dependent manner (Figure 5 and Table 5). At the wavenumbers of 1082 and 1240 cm⁻¹, indicating polysaccharides/phosphodiester (cell wall & nucleic acids), the control PM_{2.5} already shows slight

reductions vs. pure *E. coli* (normalized ≈ 0.95 – 0.98). In most MNP exposure conditions, 1082 and 1240 cm^{-1} intensities are further decreased at low $\text{PM}_{2.5}$, especially for plate and cup MNPs (values ~ 0.84 – 0.89 vs. control ~ 0.95 – 0.96). At medium $\text{PM}_{2.5}$, some treatments (e.g., plastic plates 1 $\times$) show values slightly above control (~ 1.01 – 1.04), whereas bottle-derived MNPs tend to keep these bands below control at medium–high $\text{PM}_{2.5}$. At the wavenumber of 1406 cm^{-1} in *E. coli* corresponding to COO^-/CH_2 (lipid and carboxylate environment), carboxylate/ CH_2 deformation at 1406 cm^{-1} is modestly reduced by $\text{PM}_{2.5}$ alone (controls ~ 0.89 – 0.92), and further depressed in most MNP conditions, particularly at medium $\text{PM}_{2.5}$, where normalized values for bottle MNPs stay <0.86 . At the highest MNP dose (20 $\times$) and high $\text{PM}_{2.5}$, intensities tend to approach or slightly exceed control (e.g., plates 0.91).

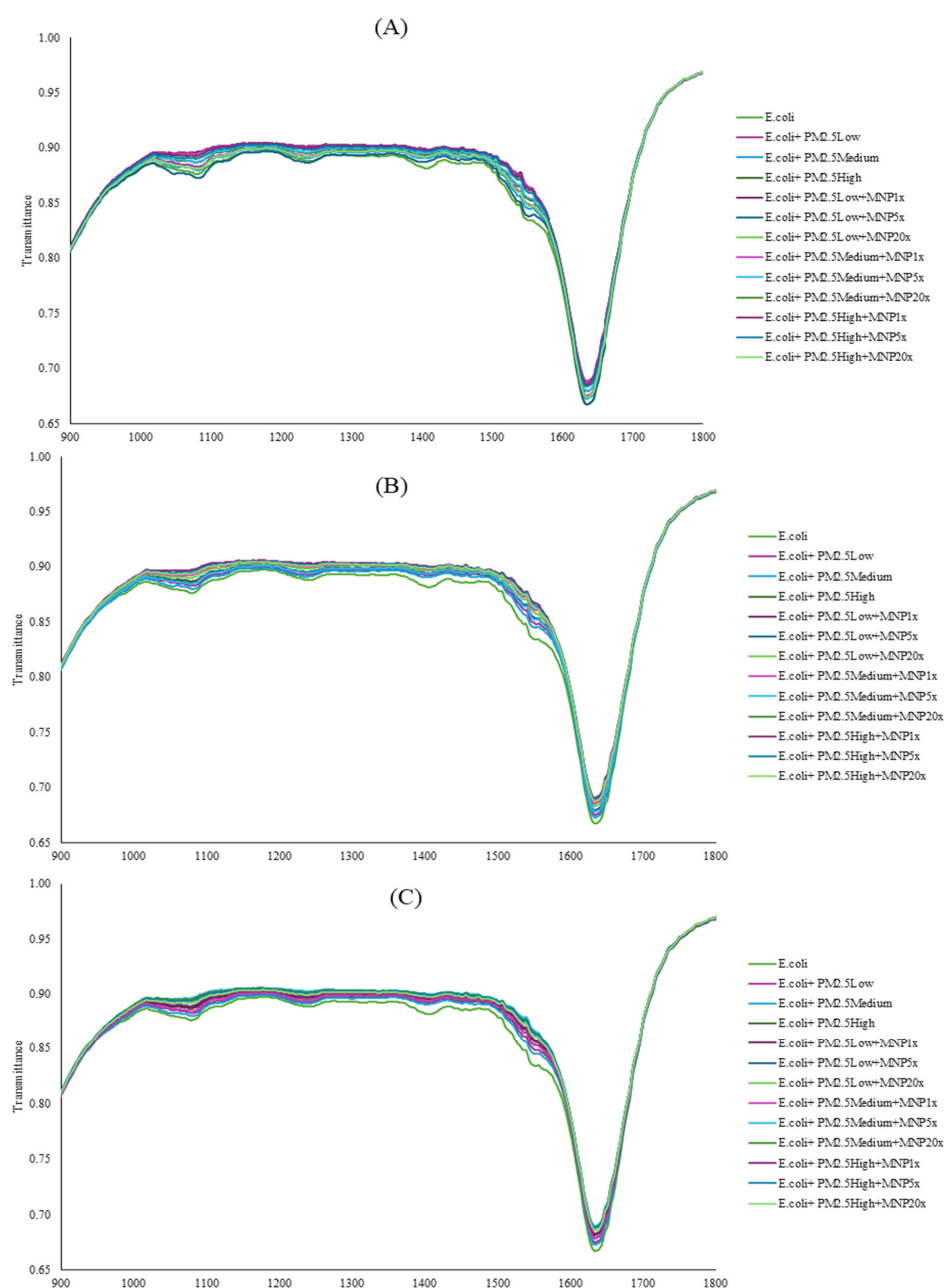


Figure 5. Metabolomic analysis of *E. coli* with and without MNP-exposed $\text{PM}_{2.5}$ using FTIR spectroscopy. (A) FTIR spectrum of *E. coli* with and without plastic food plates MNP-exposed $\text{PM}_{2.5}$, (B) FTIR spectrum of *E. coli* with and without coffee cups MNP-exposed $\text{PM}_{2.5}$, (C) FTIR spectrum of *E. coli* with and without plastic drinking water bottles MNP-exposed $\text{PM}_{2.5}$.

Table 5. Normalized absorbances of functional groups in *E. coli* exposed to MNP-treated PM_{2.5} and control PM_{2.5} (non-MNP-treated PM_{2.5}) at various PM_{2.5} concentrations. (Normalization calculated using pure *E. coli*, SD < 4.6).

Normalized FTIR Functional Group on <i>E. coli</i>			PM _{2.5} Low	PM _{2.5} Medium	PM _{2.5} High
1082	Control		0.952	0.975	0.916
	Plastic food plates	MNP1×	0.841	1.037	0.881
		MNP5×	0.848	0.913	0.949
		MNP20×	0.865	0.886	0.967
	Coffee cups	MNP1×	0.849	0.926	0.889
		MNP5×	0.867	0.933	0.867
		MNP20×	0.830	0.847	0.857
	Plastic drinking water bottles	MNP1×	0.908	0.845	0.871
		MNP5×	0.937	0.835	0.842
		MNP20×	0.895	0.859	0.875
1240	Control		0.960	0.975	0.937
	Plastic food plates	MNP1×	0.881	1.014	0.907
		MNP5×	0.885	0.929	0.947
		MNP20×	0.894	0.908	0.964
	Coffee cups	MNP1×	0.882	0.937	0.910
		MNP5×	0.895	0.945	0.957
		MNP20×	0.866	0.879	0.889
	Plastic drinking water bottles	MNP1×	0.922	0.878	0.897
		MNP5×	0.942	0.871	0.881
		MNP20×	0.915	0.887	0.902
1406	Control		0.909	0.923	0.894
	Plastic food plates	MNP1×	0.854	0.951	0.872
		MNP5×	0.859	0.887	0.847
		MNP20×	0.862	0.872	0.913
	Coffee cups	MNP1×	0.854	0.892	0.874
		MNP5×	0.864	0.898	0.983
		MNP20×	0.843	0.851	0.858
	Plastic drinking water bottles	MNP1×	0.882	0.850	0.864
		MNP5×	0.895	0.846	0.853
		MNP20×	0.877	0.857	0.867
1515	Control		0.909	0.923	0.894
	Plastic food plates	MNP1×	0.854	0.951	0.872
		MNP5×	0.859	0.887	0.899
		MNP20×	0.862	0.872	0.913
	Coffee cups	MNP1×	0.854	0.892	0.874
		MNP5×	0.864	0.898	0.961
		MNP20×	0.843	0.851	0.858
	Plastic drinking water bottles	MNP1×	0.882	0.850	0.864
		MNP5×	0.895	0.846	0.853
		MNP20×	0.877	0.857	0.867
	Control		0.913	0.772	0.747
	Plastic food plates	MNP1×	0.707	0.789	0.721
		MNP5×	0.708	0.736	0.656
		MNP20×	0.825	0.843	0.916

Table 5. Cont.

Normalized FTIR Functional Group on <i>E. coli</i>			PM _{2.5} Low	PM _{2.5} Medium	PM _{2.5} High
1540	Coffee cups	MNP1×	0.709	0.742	0.725
		MNP5×	0.704	0.743	0.757
		MNP20×	0.800	0.807	0.817
	Plastic drinking water bottles	MNP1×	0.734	0.708	0.715
		MNP5×	0.742	0.698	0.709
		MNP20×	0.855	0.811	0.837
1552	Control		0.913	0.934	0.886
	Plastic food plates	MNP1×	0.820	0.975	0.845
		MNP5×	0.823	0.879	0.970
		MNP20×	0.833	0.849	0.920
	Coffee cups	MNP1×	0.821	0.884	0.856
		MNP5×	0.839	0.892	0.812
		MNP20×	0.803	0.816	0.827
	Plastic drinking water bottles	MNP1×	0.867	0.815	0.835
		MNP5×	0.888	0.808	0.820
		MNP20×	0.858	0.823	0.839
1634	Control		0.977	0.984	0.965
	Plastic food plates	MNP1×	0.937	1.000	0.949
		MNP5×	0.939	0.963	0.988
		MNP20×	0.945	0.952	0.981
	Coffee cups	MNP1×	0.939	0.966	0.953
		MNP5×	0.946	0.968	0.997
		MNP20×	0.930	0.937	0.940
	Plastic drinking water bottles	MNP1×	0.957	0.936	0.944
		MNP5×	0.967	0.933	0.937
		MNP20×	0.954	0.938	0.944

At the wavenumber of 1540–1552 cm^{−1} in *E. coli* showing amide II (N–H bending/C–N stretching), the amide II region is among the most responsive. PM_{2.5} controls already show a pronounced drop at higher PM_{2.5} (e.g., 1540 cm^{−1} control declining from 0.913 at low to 0.747 at high). MNP-treated PM_{2.5} generally further suppresses amide II at low PM_{2.5} (0.70–0.82). At high PM_{2.5} and high MNP dose (20×), intensities partially recover towards or slightly above control (e.g., 0.916–0.920). In the wavenumber of 1634 cm^{−1} corresponding to amide I (C=O stretching, overall protein secondary structure), amide I intensities remain close to 1.0 in controls and most treatments (typically 0.93–0.98). Slight but consistent reductions with bottle-derived MNPs at medium–high PM_{2.5} were observed.

4. Discussion

4.1. Effect of PM_{2.5} on MNPs Surface Characteristics

Increases in carbonyl and hydroxyl indices detected by FTIR after PM_{2.5} exposure are consistent with previous reports showing that oxidized MNP surfaces release or exchange oxygen-containing organic functionalities with surrounding particulate matrices [59,60]. Atmospheric PM_{2.5} has been shown to act as an effective sink for such polymer-derived oxygenated compounds [61], supporting the FTIR-based evidence of polymer-specific leaching to PM_{2.5} observed in this study.

4.2. Effect of MNPs Leaching/Deformation on PM_{2.5} Spectral Indices

Further insights into the chemistry of PMs, source, and diagenesis characteristics of leaching are given by spectral slopes, including AR, HI, and SF [62]. For example, AR reflects the changes in aromatic skeleton in the medium [53,63,64]. Similar levels in AR with control (PM_{2.5} without MNP exposure) in many treatments may indicate overall subtle modifications in aromatic chromophore exposure. However, high AR values than the controls at higher PM_{2.5} loads with MNP exposures suggest dose-dependent unmasking or enrichment of aromatic moieties, likely driven by MNP–PM_{2.5} surface reorganization processes [53,54,65]. AR ≤ 1.00 levels at lower PM_{2.5} concentrations are consistent with slight aromatic masking or dilution [63,64].

The balance between condensed aromatic fluorophores and simpler aliphatic structures is mimicked by HI [53,66]. HI < 1 levels at low PM_{2.5} samples with MNP exposures indicated depletion or destabilization of higher molecular-weight chromophores at early exposure stages [67,68]. In contrast, higher HI values in multiple high-PM_{2.5} conditions with MNP exposures suggested intensive formation or concentration of humic-like, higher-order chromophores. This concentration dependence indicates that concentrated PM_{2.5} matrices promote the retention, condensation, or structural transformation of chromophores under MNP influence.

Another spectral indicator is the SF, and it reflects the balance between low- and high-molecular-weight chromophores in leachates [62,64]. The increasing SF ratios in PM_{2.5} with MNP exposures indicated greater proportions of low-molecular-weight chromophores and freshly generated optical components [62,64]. High PM_{2.5} concentration conditions consistently exhibited SF > 1.12 for all polymers and doses reveal enhanced leaching, fragmentation, or generation of small aromatic species. The parallel rise of SF and HI in high PM_{2.5} samples with MNP exposures suggests simultaneous formation of low-molecular-weight and humified chromophores, likely reflecting combined degradation–repolymerization pathways facilitated by MNP surfaces.

Overall, the data from AR, HI, and SF indicate that increasing PM_{2.5} mass amplifies MNP-induced chromophoric transformations, transitioning from early-stage depletion and instability at low PM_{2.5} with MNP exposures to moderate reorganization at medium PM_{2.5} with MNP exposures and substantial enrichment of aromatic, humified, and low-molecular-weight components at high PM_{2.5} with MNP exposures compared to PM_{2.5} without MNP exposures.

When assessing the leaching spectral characteristics (AR, HI, and SF) by plastic material, distinct polymer types produced characteristic chromophoric responses in PM_{2.5}. For example, plastic water bottle-derived MNPs generated the most pronounced increases across all parameters in PM_{2.5}, suggesting that plastic water bottle-derived fragments possess surface chemistries or leaching profiles that promote both aromatic unmasking and low-molecular-weight chromophore generation. Coffee cup MNPs induced moderate but consistent increases for HI and SF of PM_{2.5}, particularly at the high PM_{2.5} load, indicating release of additives, adhesives, or oxidized fragments. Since cardboard cups contained various functional groups, including -OH, CH₂, C-O stretch nonionic carboxylate, C-O stretch ester/carboxylic acid, CH₃, C-OH corresponding to cellulose-type polymers and elements (e.g., C, O, Al, Si, and Ti) [43,44]. Their more variable effects at low PM_{2.5}, where chromophoric suppression dominated, indicate stronger interactions during early adsorption phases. Plastic food plate MNPs produced the largest reductions at low PM_{2.5} concentration for HI and SF, suggesting strong initial chromophore depletion or structural disruption. However, at medium and high PM_{2.5} concentrations, food-plate MNPs also supported significant humification and SF increases aligned with the concentration-dependent intensification observed for other polymers. Taken together, bottle-derived MNPs exerted

the strongest enhancing effects, coffee cups showed intermediate, and PM_{2.5} concentration-dependent responses, and food plates demonstrated the most pronounced early depletion followed by high-mass enrichment.

Furthermore, spectral indices of PM_{2.5} were explained with FTIR spectrum results of PM_{2.5}-exposed MNPs to understand and corrected these changes. An increase in the oxygen-containing functional groups on plastic water bottle MNPs occurs alongside increased HI and SF ratios of PM_{2.5}, while AR remains approximately unchanged. The highest values and greatest variability in the hydroxyl and carbonyl indices of coffee cup MNPs coincide with the greatest increases in HI and SF ratios of PM_{2.5}, with minimal change in AR. Moderate increases in the carbonyl and hydroxyl indices of food plates MNPs result in moderate increases in HI and the SF ratios of PM_{2.5}, while AR remains stable.

4.3. Effect of MNPs Leaching/Deformation on PM_{2.5} Oxidative Indicators

MNPs markedly alter the oxidative profile of their surrounding medium by releasing diverse leachates, such as low-molecular-weight oligomers, monomers, plasticizers, and surface-associated additives [69]. Many of these constituents contain redox-active functional groups capable of driving ROS generation through photosensitized pathways, electron-transfer reactions with dissolved organic matter, and catalytic redox cycling mediated by trace metal impurities [70,71]. Degradation-derived carbonyl- and aromatic-enriched fragments from polymers such as PET and PE further amplify superoxide and hydroxyl radical production, collectively elevating the oxidative potential of the medium [72]. These MNP-derived leachates and degradation fragments translate directly into measurable oxidative effects: in the DTT assay, their redox-active functional groups accelerate DTT consumption, indicating elevated oxidative potential [73–75]. At the same time, the increased generation of ROS and radical intermediates depletes or competes with endogenous antioxidant constituents, producing lower CUPRAC responses. Together, these shifts reflect a medium in which MNP-induced redox chemistry intensifies oxidative pressure while diminishing available antioxidant capacity. Evaluation of oxidative stress-related parameters revealed distinct, polymer- and dose-dependent effects of MNPs on PM_{2.5} redox behavior. DTT-based OP ratios showed that higher deviations from control values across all PM_{2.5} concentrations with plastic food plate-derived MNPs indicated minimal capacity to enhance or suppress electron-transfer activity. In contrast, more pronounced responses of coffee cup MNPs at low and medium PM_{2.5} levels suggested enhanced redox cycling likely driven by coating- or filler-derived leachates. MNPs from drinking water bottles displayed a biphasic pattern—low-dose treatments (1×) led to mild elevations in OP, whereas higher doses (especially at PM_{2.5}-High) consistently reduced DTT consumption, indicating a quenching or adsorption-driven attenuation of PM-bound redox-active species [76–78].

CUPRAC-based antioxidant activity ratios further highlighted these polymer-specific trends. Increases and depletions in antioxidant activity with food plate MNPs, depending on dose and PM_{2.5} level, reflected variable release or consumption of reducing agents [54,69]. Coffee cup MNPs generated stronger shifts in antioxidant enrichment at higher PM_{2.5} loads, considering the contribution of PLA-derived degradation products or paper additives with reducing functionality [79]. The largest antioxidant response with plastic water bottle MNPs, particularly under high PM_{2.5}, suggested the release of PET oligomers or metal-interacting species that enhance the overall reducing environment [69]. Collectively, these results demonstrate that MNP–PM_{2.5} interactions modulate oxidative indicators in a complex, matrix-dependent manner, where polymer chemistry, MNP dose, and PM_{2.5} load jointly determine whether the net effect is pro-oxidant, neutral, or antioxidant.

Furthermore, the OP and antioxidant activity of PM_{2.5} after MNP treatment can be explained by FTIR spectra of MNPs following PM_{2.5} treatment. For all polymers, the FTIR functional group data for MNPs after PM_{2.5} exposure were consistent with the oxidative and antioxidant responses of PM_{2.5} following MNP treatment. PET MNPs exhibited increased carbonyl and hydroxyl indices (CI: 0.91–1.18; HI: 1.63–2.28), coinciding with PM_{2.5} DTT OP ratios close to or slightly above unity (0.924–1.045), and antioxidant activity ratios that frequently exceeded unity (0.864–2.089). Cellulose MNPs exhibited the widest ranges and highest values of the hydroxyl and carbonyl indices (HI: 0.16–3.08; CI: 0.32–1.24), alongside consistently elevated PM_{2.5} DTT ratios (1.001–1.199) and the broadest antioxidant activity range (0.839–2.260). In contrast, plastic food plate MNPs displayed moderate increases in the carbonyl and hydroxyl indices (CI: 0.61–0.92; HI: 1.21–2.79), corresponding to smaller deviations of the PM_{2.5} DTT ratios around unity (0.982–1.033) and more limited antioxidant activity ranges (0.824–1.678). Overall, polymers exhibiting larger numerical changes in FTIR-derived oxygen-containing functional groups were associated with broader ranges of PM_{2.5} OP and antioxidant responses, demonstrating internal consistency across the datasets.

4.4. Effect of MNPs Leaching/Deformation on PM_{2.5} Inflammatory Activity

It is well established that ROS generate oxidative stress, which in turn activates inflammatory pathways and contributes to inflammation [80,81]. The inflammation pattern in plastic food plates (PP-based MNPs) for all PM_{2.5} concentrations suggests that food plate MNPs do not substantially modify PM_{2.5} inflammatory behavior. The slight elevation at low doses may correspond to weak protein-denaturing interactions, but the absence of a dose-dependent escalation indicates low intrinsic inflammatory potential of food plates-derived MNPs (PP-derived particles). Coffee cup-derived MNPs produced the strongest inflammatory response among all polymers. These results indicate that coffee cup MNPs amplify PM_{2.5}-driven protein denaturation, suggesting a more reactive surface chemistry or the presence of coating residues, PLA oligomers, or paper fillers capable of promoting protein unfolding [43,44,65]. The biphasic trend in plastic drinking water bottles-derived MNPs suggests that PET's impact depends strongly on PM_{2.5} chemical composition. For example, under PM_{2.5}-Low/High, PET may enhance protein–particle interactions through aromatic or surface-polar groups, promoting mild inflammation. Under PM_{2.5}-Medium, PET appears to attenuate protein denaturation, possibly by binding or masking redox-active or protein-denaturing moieties in the PM fraction [82,83]. Thus, PET does not uniformly promote inflammation; rather, it modulates PM_{2.5}-induced inflammatory potential in a matrix-dependent manner. Inflammation outcomes demonstrate that polymer chemistry is the dominant determinant of PM_{2.5} inflammatory modulation. For example, food plates showed minimal effect due to being chemically inert, low interaction with proteins. Coffee cups (PLA/paper composites) revealed the strongest pro-inflammatory activation, robust across doses and PM burdens by likely due to the additive-rich composition and hydrophilic surfaces enhancing protein destabilization [69]. PET bottles show moderate pro-inflammatory activity in most conditions, but with suppression at PM_{2.5}-Medium, indicating conditional shielding or competitive binding effects. These results collectively show that MNPs can either amplify or mitigate PM_{2.5}-induced inflammation, depending on polymer type, dose, and PM_{2.5} chemical richness. The elevated inflammatory ratios from coffee cup MNPs highlight them as the most potent contributors to protein–denaturation-based inflammatory pathways, while PP remains largely inert and PET demonstrates context-dependent redox and protein-interaction behavior.

For all types of polymer, the inflammation activity of PM_{2.5} after MNP exposure showed numerical consistency with the extent of changes to functional groups on

MNPs that were detected using FTIR following PM_{2.5} treatment. Drinking water bottle (PET-derived) MNPs exhibited increased carbonyl and hydroxyl indices after PM_{2.5} exposure (CI: 0.91–1.18; HI: 1.63–2.28), which coincided with PM_{2.5} inflammation ratios that were predominantly above unity at both low and high PM_{2.5} levels (1.05–1.24). However, values were below unity at medium PM_{2.5} levels. Coffee cup (cellulose-derived) MNPs displayed the widest ranges and highest values of the hydroxyl and carbonyl indices (HI: 0.16–3.08; CI: 0.32–1.24), which corresponded to the highest and most variable PM_{2.5} inflammation ratios observed among all polymers (0.97–1.64). Food plates (PP-derived) MNPs displayed moderate increases in the carbonyl and hydroxyl indices (CI: 0.61–0.92; HI: 1.21–2.79) and smaller deviations of the PM_{2.5} inflammation ratios around unity (0.97–1.11). Overall, polymers that exhibited larger numerical changes in FTIR-derived oxygen-containing functional groups were associated with broader and higher ranges of PM_{2.5} inflammation activity. This demonstrates internal consistency between the FTIR and inflammation datasets.

4.5. Effect of MNPs Leaching/Deformation on PM_{2.5} Cytological and Metabolomic Responses of *E. coli*

Exposure to MNP-treated PM_{2.5} produced non-linear changes in *E. coli* growth, reflecting polymer-specific and dose-dependent metabolic responses. At low PM_{2.5}, inhibition in bacterial growth showed a notable growth stimulation [84]. Responses at medium PM_{2.5} induced more divergent patterns; for example, OD increases for plastic-plate and drinking-bottle MNP exposed PM_{2.5}, but inhibition for coffee-cup MNP exposed PM_{2.5} indicated that intermediate particulate loads modulate bacterial proliferation depending on the chemical profile of the polymer-derived leachates [85,86]. At high PM_{2.5}, relatively stable OD values across all polymers exposed PM_{2.5} suggested adaptive stabilization or reduced bioavailable toxicity at elevated particulate loading. Overall, the OD results indicate a balance between stress-induced inhibition and hormetic stimulation, driven by polymer–PM_{2.5} interactions. Furthermore, the OD responses of *E. coli* for different polymers-exposed PM_{2.5} reflected the modulation of PM_{2.5}-derived dissolved organic matter and surface deformation by the polymers. Coffee cup (cellulose-based) MNPs induced the strongest growth stimulation, particularly at low PM_{2.5} loading. This is consistent with FTIR evidence of pronounced hydroxyl and carbonyl enrichment, as well as UV–Vis indices indicating relatively low humification. This suggests an enhanced release or mobilisation of readily bioavailable, oxygenated organic substrates. In contrast, food plate (PP-based) MNPs generally suppressed bacterial growth, despite increased leaching by surface deformation/oxidation. This was because their nonpolar matrix favoured the partitioning and complexation of PM-bound organics. Meanwhile, elevated humification and altered slope factor ratios pointed to the enrichment of humic-like, redox-active dissolved organic matter that constrained proliferation. Plastic drinking water bottle (PET-derived) MNPs exhibited intermediate behavior, with moderate leaching by surface oxygenation and dose-dependent OD effects that were attenuated under high PM_{2.5} loading due to strong humification. Taken together, these findings demonstrate that bacterial growth outcomes are governed by the balance between MNP-mediated carbon availability and PM_{2.5}-driven chemical complexity rather than aromaticity alone.

Carbohydrate levels in *E. coli* can be used as a proxy for extracellular polymeric substance (EPS) secretion [87,88]. Fluctuations with MNP-treated at low PM_{2.5} suggested that EPS secretion was strongly enhanced across most conditions, indicating acute stress-induced polysaccharide release [84,87,88]. Reduction in medium PM_{2.5} with MNP exposures is consistent with metabolic exhaustion and depletion of reserve carbohydrates during sustained oxidative pressure. Mixed responses at high PM_{2.5} highlight that EPS production serves as a dynamic stress-buffering mechanism, escalating under acute chemical pressure but diminishing when prolonged exposure exceeds metabolic capacity. Elevated levels of

carbohydrates in *E. coli* can arise from either increased access to oxygenated, labile carbon released by deformed MNP surfaces under conditions of low humification and PM_{2.5}, or from stress-induced carbon storage and EPS production when surface oxidation coincides with highly humified PM_{2.5} organic matter.

Lower LPO activity of *E. coli* compared to the control level in most exposure conditions indicated membrane oxidation at various levels. For example, at low and medium PM_{2.5} with MNP exposed, a reduction in LPO suggested effective shielding by EPS layers or membrane restructuring to mitigate oxidative attack. At high PM_{2.5}, 5–40% lower activities indicated severe membrane compromise. Enhanced LPO in *E. coli* primarily reflects synergistic effects of leaching owing to MNP surface oxidation in PM_{2.5} and highly humified, redox-active PM_{2.5} organic matter that intensify ROS-mediated membrane damage, whereas lower humification and greater availability of labile organics attenuate lipid peroxidation. The overall decrease in LPO despite elevated OP implies that oxidative stress is largely redirected toward extracellular defenses or intracellular antioxidant pathways, reducing direct peroxidative damage to membrane lipids [89]. Since lipid peroxidation is a late-stage oxidative injury event that declines when antioxidant or protective mechanisms intercept ROS before they reach the membrane. In *E. coli* exposed to MNP-treated PM_{2.5}, this redirection is likely achieved through enhanced secretion of carbohydrates and matrix restructuring. Extracellular carbohydrates may act as a physical and chemical barrier that absorbs or neutralizes oxidants, thereby reducing the burden of peroxidative attack on membrane lipids [90,91].

ROS responses showed the clearest and most consistent toxicity signal across treatments. At low PM_{2.5}, ROS increased with drinking-bottle exposed PM_{2.5}, reflecting synergistic pro-oxidant effects between polymer-derived leachates and PM_{2.5}PM_{2.5} surface chemistry [92]. High PM_{2.5} maintained elevated ROS levels with most polymer types, indicating persistent oxidative pressure even when growth and membrane parameters became more stable. ROS generation in *E. coli* is primarily governed by the redox coupling between oxidatively aged MNP surfaces reflecting the leaching and humified PM_{2.5} organic matter, with lower humification and greater labile carbon availability mitigating intracellular oxidative burden. Moreover, ROS responses in *E. coli* when exposed to MNP-treated PM_{2.5} can be explained using leaching characteristics of MNPs surface deformation and solution chemistry of PM_{2.5}. For example, ROS responses associated with drinking-water bottle (PET-derived) MNPs reflect the leaching effect of MNPs in PM_{2.5} (intermediate surface oxidation (moderate CI and HI increases)) that enhanced PM_{2.5} redox interactions without substantial carbon release, resulting in ROS levels that closely track MNP-treated PM_{2.5} humification rather than SF changes. Elevated humification ratios under higher PM loads therefore promote ROS accumulation, while lower humification allows partial metabolic compensation. In addition, ROS decline at MNP20× -treated PM_{2.5} from PM_{2.5} Medium to PM_{2.5} High can be explained by the fact that intermediate surface oxidation at 20× promotes complexation rather than catalytic cycling of PM-derived redox components under PM_{2.5} High, resulting in reduced intracellular ROS relative to PM_{2.5} Medium conditions. For coffee cup (cellulose-based) MNPs treated PM_{2.5}, the strong enrichment of hydroxyl and C–O functional groups promote the release of oxygenated, carbohydrate-like compounds to the PM_{2.5} that support cellular antioxidant responses. This may lead to comparatively moderate ROS levels when humification indices are low. However, when spectral data indicated increased humification, ROS levels increased due to redox-active dissolved organic matter in MNP-treated PM_{2.5}. The generation of ROS in the presence of food plates (PP-derived) MNPs-treated PM_{2.5} is primarily driven by surface oxidation, as evidenced by increased C=O and -OH bands in FTIR. This oxidation has the potential to enhance the adsorption and interfacial redox activity of PM_{2.5}-associated metals and quinone-like

organics. Meanwhile, UV–Vis indices indicating increased humification at higher PM loads suggest the presence of persistent redox-active dissolved organic substances, which sustain ROS formation. However, the limited release of labile carbon from food plates restricts metabolic buffering, resulting in oxidative stress dominating. Overall, ROS production was the most sensitive indicator of MNP–PM_{2.5}-induced stress, and its pattern closely matched conditions associated with reduced growth and depleted carbohydrate reserves [92].

Across all wavenumbers, changes in *E. coli* biochemical fingerprints revealed the metabolomic interaction of PM_{2.5} and MNP-treated PM_{2.5} (Figure 5) [93,94]. The reduction in the polysaccharides/phosphodiester bands (cell wall & nucleic acids) indicated mild attenuation of carbohydrate and phosphate-related bands. In most MNP conditions, further decreases at 1082 and 1240 cm^{−1} intensities can be explained by consistent disturbance or partial loss of cell wall polysaccharides and phosphodiester groups [95]. At medium PM_{2.5}, some MNP treatments (e.g., plastic plates 1×) increased the intensities, suggesting a partial overcompensation or increased contribution from EPS, whereas bottle-derived MNPs tend to keep these bands below control at medium–high PM_{2.5}, implying more persistent damage or rearrangement of surface carbohydrates and phosphate moieties. Changes in the lipid and carboxylate environment by deformation by PM_{2.5} alone, and further depressed in most MNP conditions, particularly for bottle MNPs, can originate from alterations in membrane lipid packing and carboxylated surface groups, compatible with oxidative and structural stress at the cell envelope. However, higher intensities with the highest MNP dose (20×) and high PM_{2.5} indicated some re-ordering or compensatory accumulation of carboxylated components, potentially linked to stress-induced remodeling of membrane lipids or EPS. The amide II region (1540–1552 cm^{−1}—amide II), MNP-treated PM_{2.5} is generally lower than PM_{2.5}, which can be consistent with enhanced protein denaturation, unfolding, or loss from the cell envelope due to MNP exposure [96]. Recovery at high PM_{2.5} and high MNP dose (20×) may reflect stress-induced synthesis/accumulation of proteins (e.g., membrane proteins, stress chaperones, EPS-associated proteins) or tighter packing/aggregation of cells that increases apparent amide II contribution [97]. Relatively stable amide I (C=O stretching, overall protein secondary structure) with the exposures indicated that the global protein backbone structure is relatively preserved, even though amide II and side-chain-related bands are more clearly affected. Slight but consistent reductions with bottle-derived MNPs at medium–high PM_{2.5} suggest subtle changes in α-helix/β-sheet balance or hydrogen bonding, but not massive protein loss. Taken together, decreased 1082/1240 cm^{−1} (polysaccharides/phosphodiester bands) and 1406/1540–1552 cm^{−1} (carboxylates and amide II) bands indicate that PM_{2.5} exposure, amplified by MNP treatment, primarily targets the *E. coli* cell envelope—damaging or rearranging membrane lipids, peptidoglycan, and surface proteins—rather than completely degrading bulk cellular proteins (amide I). The partial recovery or even slight enhancement of some bands at the highest MNP doses and high PM_{2.5} is consistent with adaptive stress responses such as EPS secretion, biofilm-like aggregation, or remodeling of membrane/protein composition rather than a simple monotonic loss of biochemical material.

Cytological and metabolomic responses in *E. coli* closely aligned with the chemical patterns observed in leaching, oxidative potential, and inflammation assays. Conditions that released more aromatic and low-molecular-weight chromophores owing to MNP exposures in PM_{2.5}, showed higher DTT consumption, exhibited stronger protein-denaturation activity, also produced the most pronounced cellular stress—elevated ROS and lipid peroxidation, reduced growth, and FTIR evidence of membrane and protein disruption. Conversely, samples with weaker leaching and lower oxidative potential induced only minor biochemical shifts. Overall, the bacterial responses reflect a coherent cascade in which

MNP-modified PM_{2.5} chemistry amplifies redox reactivity and inflammatory potential, translating directly into measurable cytological and metabolic injury.

5. Conclusions

This study demonstrates that micro- and nanoplastics markedly and differentially modulate the chemical and biological behavior of PM_{2.5}. Across all analyses, polymer type emerged as the dominant determinant of PM_{2.5} transformation, followed by PM_{2.5} concentration and MNP dose. Among the tested materials, plastic water bottle-derived MNPs (PET) produced the strongest overall effects—consistently enhancing aromaticity, humification, slope factor, oxidative potential, and bacterial ROS generation, alongside pronounced alterations in FTIR-based metabolomic fingerprints. Coffee cup MNPs (PLA/paper composites) induced the highest protein-denaturation-based inflammatory activity and also elevated chromophoric and oxidative responses, though in a more PM_{2.5}-dependent manner. In contrast, food plate MNPs (PP) exerted the weakest impacts, showing initial chromophore depletion at low PM_{2.5} and only moderate enrichment or redox activation at higher PM_{2.5} loads.

A clear hierarchical pathway became evident across all treatments, in which MNP exposure first reshaped PM_{2.5} leaching chemistry, subsequently elevating oxidative potential and diminishing antioxidant capacity, which in turn amplified inflammatory activity and ultimately manifested as cytological and metabolomic stress in *E. coli*. Conditions that intensified chromophoric restructuring and oxidative potential produced the most significant biological effects, including elevated ROS, reduced growth, altered carbohydrates, suppressed LPO, and membrane/protein perturbations detected by FTIR.

Overall, the findings reveal that MNP-contaminated PM_{2.5} exhibits polymer-specific toxicity signatures, with PET > PLA > PP in their ability to amplify PM_{2.5} reactivity and biological impact. These results highlight the need to consider both the type and intensity of MNP exposure when evaluating PM_{2.5}-associated health risks and provide a mechanistic framework for understanding how MNP–PM_{2.5} interactions propagate from chemical transformations to biological injury.

Future studies should examine longer exposure durations, broader polymer types, and environmentally aged MNPs to determine how real-world weathering influences PM_{2.5} reactivity. Integrating mammalian cell models, advanced metabolomics, and molecular docking could further clarify the biological pathways triggered by MNP-modified PM_{2.5}. Field-based PM collections with known MNP burdens, coupled with multi-omics and oxidative profiling, will also be essential to assess environmental relevance and to identify chemical drivers of toxicity. Ultimately, linking polymer-specific leaching signatures to biological outcomes will support predictive models for assessing human health risks in MNP-polluted atmospheres.

Supplementary Materials: The following supporting information can be downloaded at: <https://www.mdpi.com/article/10.3390/microplastics5010043/s1>, Figure S1: Scheme of experimental process of the study; Figure S2: FTIR spectra of the plastic water bottles (PET MNP) treated with and without various concentrations of PM_{2.5}. (A) Low dose of MNP treated with low, medium, and high concentration of PM_{2.5}, (B) Medium dose of MNP treated with low, medium, and high concentration of PM_{2.5}, and (C) High dose of MNP treated with low, medium, and high concentration of PM_{2.5}; Figure S3: FTIR spectra of the coffee cups (PLA-cellulose MNP) treated with and without various concentrations of PM_{2.5}. (A) Low dose of MNP treated with low, medium, and high concentration of PM_{2.5}, (B) Medium dose of MNP treated with low, medium, and high concentration of PM_{2.5}, and (C) High dose of MNP treated with low, medium, and high concentration of PM_{2.5}; Figure S4: FTIR spectra of the plastic food plates (PP MNP). (A) Low dose of MNP treated with low, medium, and high concentration of PM_{2.5}, (B) Medium dose of MNP treated with low, medium,

and high concentration of PM_{2.5}, and (C) High dose of MNP treated with low, medium, and high concentration of PM_{2.5}.

Author Contributions: Conceptualization, H.S.; methodology, H.S. and A.B.; formal analysis, H.S.; investigation, A.B.; resources, A.B. and H.S.; data curation, H.S. and A.B.; writing—original draft preparation, H.S. and A.B.; visualization, H.S.; supervision, H.S. and A.B.; project administration, A.B. and H.S.; funding acquisition, A.B. All authors have read and agreed to the published version of the manuscript.

Funding: This work was supported by the Scientific Research Projects Department of Istanbul Technical University. Grand (Project) Number: TED-2024-45420.

Institutional Review Board Statement: Not applicable.

Informed Consent Statement: Not applicable.

Data Availability Statement: Data are contained within the article.

Acknowledgments: The authors also thank Batuhan Tilkili-IAU and Seyda Kalkavan-ITU for their help during exposure experiments.

Conflicts of Interest: The authors declare no conflicts of interest. The funders had no role in the design of the study; in the collection, analyses, or interpretation of data; in the writing of the manuscript; or in the decision to publish the results.

References

1. Arca-Lafuente, S.; Núñez-Corcuera, B.; Ramis, R.; Karakitsios, S.; Sarigiannis, D.; García Dos Santos, S.; Fernández-Rodríguez, A.; Briz, V. Effects of urban airborne particulate matter exposure on the human upper respiratory tract microbiome: A systematic review. *Respir. Res.* **2025**, *26*, 118. [\[CrossRef\]](#) [\[PubMed\]](#)
2. Wu, C.H.; Dang, T.T.M.; Mutuku, J.K.; Lin, L.M.; Huang, B.W.; Chang-Chien, G.P. Evaluation of PM_{2.5} bound microplastics and plastic additives in several cities in Taiwan: Spatial distribution and human health risk. *Sci. Total Environ.* **2025**, *959*, 178213. [\[CrossRef\]](#)
3. Zhang, S.; Chen, X.; Dai, C.; Wang, J.; Wang, H. Associations between air pollutants and respiratory infection risk: Patient-based bacterial culture analysis. *Environ. Geochem. Health* **2022**, *44*, 4007–4016. [\[CrossRef\]](#) [\[PubMed\]](#)
4. Huang, X.; Steinmetz, J.; Marsh, E.K.; Aravkin, A.Y.; Ashbaugh, C.; Murray, C.J.L.; Yang, F.; Ji, J.S.; Zheng, P.; Sorensen, R.J.D.; et al. A systematic review with a Burden of Proof meta-analysis of health effects of long-term ambient fine particulate matter (PM_{2.5}) exposure on dementia. *Nat. Aging* **2025**, *5*, 897–908. [\[CrossRef\]](#)
5. Sakthivel, K.; Lin, W.-C.; Lee, Y.-Y.; Huang, B.W.; Chen, Y.L.; Chang-Chien, G.P.; Sheu, J.K. Advancements in electrochemical biosensing of cardiovascular disease biomarkers. *J. Mater. Chem. B* **2024**, *12*, 6305–6327. [\[CrossRef\]](#)
6. Wang, R.; Yang, L.; Guo, M.; Lin, X.Q.; Wang, R.; Guo, S. Effects of microplastic properties and dissolved organic matter on phosphorus availability in soil and aqueous mediums. *Environ. Pollut.* **2023**, *340*, 122784. [\[CrossRef\]](#)
7. Chen, S.; Liu, D.; Huang, L.; Guo, C.; Gao, X.; Xu, Z.; Yang, Z.; Chen, Y.; Li, M.; Yang, J. Global associations between long-term exposure to PM_{2.5} constituents and health: A systematic review and meta-analysis of cohort studies. *J. Hazard. Mater.* **2024**, *474*, 134715. [\[CrossRef\]](#)
8. Pomata, D.; La Nasa, J.; Biale, G.; Barlucchi, L.; Ceccarini, A.; Di Filippo, P.; Riccardi, C.; Buiarelli, F.; Modugno, F.; Simonetti, G. Plastic breath: Quantification of microplastics and polymer additives in airborne particles. *Sci. Total Environ.* **2024**, *932*, 173031. [\[CrossRef\]](#)
9. Solórzano, L.A.; Gavilanes, D.; Cadena, F.; Irusta, L.; González, A.; Aldas, M. A Review of the Sampling, Analysis, and Identification Techniques of Microplastics in the Air: Insights into PM_{2.5} and PM₁₀. *Polymers* **2025**, *17*, 3045. [\[CrossRef\]](#)
10. Chen, Y.; Jing, S.; Wang, Y.; Song, Z.; Xie, L.; Shang, X.; Fu, H.; Yang, X.; Wang, H.; Wu, M.; et al. Quantification and Characterization of Fine Plastic Particles as Considerable Components in Atmospheric Fine Particles. *Environ. Sci. Technol.* **2024**, *58*, 4691–4703. [\[CrossRef\]](#)
11. Yang, Y.; Chen, Y.; Gao, Y.; Jin, Y.; Fang, X. Mapping research frontiers in microplastics-induced oxidative stress: A bibliometric analysis (2010–2024). *Front. Environ. Sci.* **2025**, *13*, 1555341. [\[CrossRef\]](#)
12. Roman, J.; Gondko, D.; Dębiec, P.; Pietrzak, N. The hidden health crisis: Microplastics and their medical consequences. *J. Educ. Health Sport* **2024**, *72*, 51690. [\[CrossRef\]](#)

13. Kumar, S.B.; Pachiyappan, J.K.; Chandrasekar, S.; Reddy Karri, V.V.S.; Krishnamurthy, P.T.; Natarajan, J.; Palanisami, T.; Kuppusamy, G. Microplastics and health hazards: Gastrointestinal risk assessment across a multi-species perspective. *Asian J. Atmos. Environ.* **2025**, *19*, 17. [\[CrossRef\]](#)
14. Rajendran, D.; Chandrasekaran, N. Understanding the kinetics and isotherm models of adsorption reactions of micronanoplastics in the environment. In *Micronanoplastics: From Source to Sink*; Elsevier: Amsterdam, The Netherlands, 2026; pp. 291–319.
15. Zhang, X.; Yu, C.-H.; Wang, P.; Yang, C. Microplastics and human health: Unraveling the toxicological pathways and implications for public health. *Front. Public Health* **2025**, *13*, 1567200. [\[CrossRef\]](#)
16. Aardema, H.; Vethaak, A.D.; Kamstra, J.H.; Legler, J. Farm animals as a critical link between environmental and human health impacts of micro- and nanoplastics. *Microplast. Nanoplast.* **2024**, *4*, 5. [\[CrossRef\]](#)
17. Chang, N.; Chen, L.; Wang, N.; Cui, Q.; Qiu, T.; Zhao, S.; He, H.; Zeng, Y.; Dai, W.; Duan, C.; et al. Unveiling the impacts of microplastic pollution on soil health: A comprehensive review. *Sci. Total Environ.* **2024**, *951*, 175643. [\[CrossRef\]](#)
18. Wright, S.L.; Kelly, F.J. Plastic and human health: A micro issue? *Environ. Sci. Technol.* **2017**, *51*, 6634–6647. [\[CrossRef\]](#)
19. Iftikhar, A.; Qaiser, Z.; Sarfraz, W.; Ejaz, U.; Aqeel, M.; Rizvi, Z.F.; Khalid, N. Understanding the leaching of plastic additives and subsequent risks to ecosystems. *Water Emerg. Contam. Nanoplastics* **2024**, *3*, 5. [\[CrossRef\]](#)
20. Do, A.T.N.; Ha, Y.; Kwon, J.H. Leaching of microplastic-associated additives in aquatic environments: A critical review. *Environ. Pollut.* **2022**, *305*, 119258. [\[CrossRef\]](#)
21. Anih, D.C.; Arowora, A.K.; Abah, M.A.; Ugwuoke, K.C. Biochemical effects of microplastics on human health: A comprehensive review. *Sci. Int.* **2025**, *13*, 27–34. [\[CrossRef\]](#)
22. Souza, A.S.; Ferreira, P.G.; Pereira, P.R.; de Jesus, I.S.; de Oliveira, R.P.R.F.; de Carvalho, A.S.; Rodrigues, L.C.D.; Paschoalin, V.M.F.; Futuro, D.O.; Ferreira, V.F. Micro- and nanoplastics on human health and diseases: Perspectives and recent advances. *Microplastics* **2025**, *4*, 64. [\[CrossRef\]](#)
23. Sun, T.; Timoneda, A.; Banavar, A.; Ovissipour, R. Enhancing food safety and cultivated meat production: Exploring the impact of microplastics on fish muscle cell proliferation and differentiation. *Front. Food Sci. Technol.* **2024**, *4*, 1309884. [\[CrossRef\]](#)
24. Dal, F.; Ahbab, M.A. Mitochondria as a target of micro- and nanoplastic toxicity. *Camb. Prism. Plast.* **2024**, *2*, e6. [\[CrossRef\]](#)
25. Zhou, Z.; Shuai, X.; Lin, Z.; Yu, X.; Ba, X.; Holmes, M.A.; Xiao, Y.; Gu, B.; Chen, H. Association between particulate matter (PM)_{2.5} air pollution and clinical antibiotic resistance: A global analysis. *Lancet Planet. Health* **2023**, *7*, e649–e659. [\[CrossRef\]](#) [\[PubMed\]](#)
26. Ma, J.; Chiu, Y.F.; Kao, C.C.; Chuang, C.N.; Chen, C.Y.; Lai, C.H.; Kuo, M.L. Fine particulate matter manipulates immune response to exacerbate microbial pathogenesis in the respiratory tract. *Eur. Respir. Rev.* **2024**, *33*, 230259. [\[CrossRef\]](#)
27. Wang, S.; Zhou, Q.; Tian, Y.; Hu, X. The Lung Microbiota Affects Pulmonary Inflammation and Oxidative Stress Induced by PM_{2.5} Exposure. *Environ. Sci. Technol.* **2022**, *56*, 12368–12379. [\[CrossRef\]](#)
28. Wang, X.; Yu, D.; Chen, L. Antimicrobial resistance and mechanisms of epigenetic regulation. *Front. Cell. Infect. Microbiol.* **2023**, *13*, 1199646. [\[CrossRef\]](#)
29. Zhao, S.; Li, H.; Yang, F.; Yang, Y.; Zeng, Y.; An, Z.; Li, J.; Wu, H.; Song, J.; Wu, W. Association of short-term PM_{2.5} exposure with airway innate immune response, microbiota and metabolism alterations in human airways. *Environ. Pollut.* **2024**, *345*, 123435. [\[CrossRef\]](#)
30. Li, J.; Hu, Y.; Liu, L.; Wang, Q.; Zeng, J.; Chen, C. PM_{2.5} exposure perturbs lung microbiome and its metabolic profile in mice. *Sci. Total Environ.* **2020**, *721*, 137432. [\[CrossRef\]](#)
31. Jin, L.; Xie, J.; He, T.; Wu, D.; Li, X. Airborne transmission as an integral environmental dimension of antimicrobial resistance through the “One Health” lens. *Crit. Rev. Environ. Sci. Technol.* **2021**, *52*, 4172–4193. [\[CrossRef\]](#)
32. Gao, M.; Zhang, Q.; Lei, C.; Lu, T.; Qian, H. Atmospheric antibiotic resistome driven by air pollutants. *Sci. Total Environ.* **2023**, *902*, 165942. [\[CrossRef\]](#) [\[PubMed\]](#)
33. Krause, S.; Ouellet, V.; Allen, D.; Allen, S.; Moss, K.; Nel, H.A.; Manaseki-Holland, S.; Lynch, I. The potential of micro- and nanoplastics to exacerbate the health impacts and global burden of non-communicable diseases. *Cell Rep. Med.* **2024**, *5*, 101581. [\[CrossRef\]](#) [\[PubMed\]](#)
34. Blount, Z.D. The unexhausted potential of *Escherichia coli*. *eLife* **2015**, *4*, e05826. [\[CrossRef\]](#) [\[PubMed\]](#)
35. Domingue, J.C.; Drewes, J.L.; Merlo, C.A.; Housseau, F.; Sears, C.L. Host responses to mucosal biofilms in the lung and gut. *Mucosal Immunol.* **2020**, *13*, 413–422. [\[CrossRef\]](#)
36. Barnes, P.J. Inflammatory mechanisms in patients with chronic obstructive pulmonary disease. *J. Allergy Clin. Immunol.* **2017**, *138*, 16–27. [\[CrossRef\]](#)
37. Budden, K.F.; Gellatly, S.L.; Wood, D.L.; Cooper, M.A.; Morrison, M.; Hugenholtz, P.; Hansbro, P.M. Emerging pathogenic links between microbiota and the gut–lung axis. *Nat. Rev. Microbiol.* **2017**, *15*, 55–63. [\[CrossRef\]](#)
38. Peng, M.; Vercauteren, M.; Grootaert, C.; Rajkovic, A.; Boon, N.; Janssen, C.; Asselman, J. Cellular and bioenergetic effects of polystyrene microplastic in function of cell type, differentiation status and post-exposure time. *Environ. Pollut.* **2023**, *337*, 122550. [\[CrossRef\]](#)

39. Silva, M.G.; Oliveira, M.M.; Peixoto, F. The impact of micro-nanoplastics on mitochondria in the context of diet and diet-related diseases. *Stresses* **2025**, *5*, 12. [\[CrossRef\]](#)
40. Bora, S.S.; Gogoi, R.; Sharma, M.R.; Anshu, Borah, M.P.; Deka, P.; Bora, J.; Naorem, R.S.; Das, J.; Teli, A.B. Microplastics and human health: Unveiling the gut microbiome disruption and chronic disease risks. *Front. Cell. Infect. Microbiol.* **2024**, *14*, 1492759. [\[CrossRef\]](#)
41. Suman, T.Y.; Rajendiran, K.; Kwak, I. Microplastics and nanoplastics in human health: A comprehensive review of exposure pathways, cellular mechanisms, and toxicological implications. *Arch. Toxicol.* **2025**. [\[CrossRef\]](#)
42. Brander, S.M.; Senathirajah, K.; Fernández, M.; Weis, J.S.; Kumar, E.; Jahnke, A.; Hartmann, N.B.; Alava, J.J.; Farrelly, T.; Almroth, B.C.; et al. The time for ambitious action is now: Science-based recommendations for plastic chemicals to inform an effective global plastic treaty. *Sci. Total Environ.* **2024**, *949*, 174881. [\[CrossRef\]](#)
43. Saygin, H.; Tilkili, B.; Kayisoglu, P.; Baysal, A. Oxidative stress, biofilm formation and activity responses of *P. aeruginosa* to microplastic-treated sediments: Effect of temperature and sediment type. *Environ. Res.* **2024**, *248*, 118349. [\[CrossRef\]](#)
44. Saygin, H.; Tilkili, B.; Karniyarik, S.; Baysal, A. Culture-dependent analysis of bacterial activity, biofilm formation and oxidative stress of seawater with microplastic contamination under climate change consideration. *Sci. Total Environ.* **2024**, *922*, 171103. [\[CrossRef\]](#) [\[PubMed\]](#)
45. Kilincer, M.; Saygin, H.; Ozyurek, M.; Baysal, A. Impact of the in vitro human digestive environment on the interaction between vitamin C and microplastics. *Anal. Lett.* **2025**, *58*, 2087–2102. [\[CrossRef\]](#)
46. Szigeti, T.; Mihucz, V.G.; Óvári, M.; Baysal, A.; Atılgan, S.; Akman, S.; Záray, G. Chemical characterization of PM2.5 fractions of urban aerosol collected in Budapest and Istanbul. *Microchem. J.* **2013**, *107*, 86–94. [\[CrossRef\]](#)
47. Baysal, A.; Saygin, H.; Ustabasi, G.S. Interaction of PM2.5 airborne particulates with ZnO and TiO₂ nanoparticles and their effect on bacteria. *Environ. Monit. Assess.* **2017**, *190*, 34. [\[CrossRef\]](#)
48. Saygin, H.; Baysal, A.; Tilkili, B.; Karniyarik, S. Can micro/nanoplastics influence PM2.5 characteristics? An ex situ investigation by physicochemical indicators of PM2.5 and their bacterial model toxicity. *Microplastics* **2025**, *4*, 103. [\[CrossRef\]](#)
49. Ferraz, E.R.; Rainho, C.R.; Fernandes, A.S.; Felzenszwalb, I. Differential toxicity of an organic PM2.5 extract to human lung cells cultured in three dimensions (3D) and monolayers. *J. Toxicol. Environ. Health A* **2016**, *79*, 221–231. [\[CrossRef\]](#)
50. Tong, X.; Chen, X.-C.; Chuang, H.-C.; Cao, J.J.; Ho, S.S.H.; Lui, K.H.; Ho, K.F. Characteristics and cytotoxicity of indoor fine particulate matter (PM2.5) and PM2.5-bound polycyclic aromatic hydrocarbons (PAHs) in Hong Kong. *Air Qual. Atmos. Health* **2019**, *12*, 1459–1468. [\[CrossRef\]](#)
51. Sattar, A.A.; Good, C.R.; Saletes, M.; Brandão, J.; Jackson, S.K. Endotoxin as a marker for water quality. *Int. J. Environ. Res. Public Health* **2022**, *19*, 16528. [\[CrossRef\]](#)
52. Rasuli, L.; Dehghani, M.H.; Aghaei, M.; Mahvi, A.H.; Mubarak, N.M.; Karri, R.R. Occurrence and fate of bacterial endotoxins in the environment (air, water, wastewater) and remediation technologies: An overview. *Chemosphere* **2022**, *303*, 135089. [\[CrossRef\]](#) [\[PubMed\]](#)
53. Macan, G.P.; Munhoz, D.R.; Willems, L.A.; Monkley, C.; Lloyd, C.E.M.; Hageman, J.; Geissen, V.; Landa, B.B.; Harkes, P. Macro- and microplastics leachates: Characterization and impact on seed germination. *J. Hazard. Mater.* **2024**, *480*, 136013. [\[CrossRef\]](#) [\[PubMed\]](#)
54. Saygin, H.; Baysal, A.; Tilkili, B.; Apaydin, E.; Ozbek, P. Understanding the PET micro/nanoplastics as an environmental stressor on pancreatin enzyme: Leaching and binding characterization by multi-spectroscopic and molecular docking examination, and the resulting impact on *Escherichia coli*. *Nanotoxicology* **2025**, *19*, 508–527. [\[CrossRef\]](#) [\[PubMed\]](#)
55. Apak, R.; Güçlü, K.; Özyürek, M.; Çelik, S.E. Mechanism of antioxidant capacity assays and the CUPRAC (cupric ion reducing antioxidant capacity) assay. *Microchim. Acta* **2008**, *160*, 413–419. [\[CrossRef\]](#)
56. Gao, D.; Mulholland, J.A.; Russell, A.G.; Weber, R.J. Characterization of Water-Insoluble Oxidative Potential of PM2.5 Using the Dithiothreitol Assay. *Atmos. Environ.* **2020**, *224*, 117327. [\[CrossRef\]](#)
57. Douadi, K.; Chafaa, S.; Douadi, T.; Al-Noaimi, M.; Kaabi, I. Azoimine quinoline derivatives: Synthesis, classical and electrochemical evaluation of antioxidant, anti-inflammatory, antimicrobial activities and the DNA/BSA binding. *J. Mol. Struct.* **2020**, *1217*, 128305. [\[CrossRef\]](#)
58. Dubois, M.; Gilles, K.A.; Hamilton, J.K.; Rebers, P.; Smith, F. Colorimetric method for determination of sugars and related substances. *Anal. Chem.* **1956**, *28*, 350–356. [\[CrossRef\]](#)
59. Gewert, B.; Plassmann, M.M.; MacLeod, M. Pathways for degradation of plastic polymers floating in the marine environment. *Environ. Sci. Process. Impacts* **2015**, *17*, 1513–1521. [\[CrossRef\]](#)
60. Luo, H.; Zhao, Y.; Li, Y.; Xiang, Y.; He, D.; Pan, X. Aging of microplastics affects their surface properties, thermal decomposition, additives leaching and interactions in simulated fluids. *Sci. Total Environ.* **2020**, *714*, 136862. [\[CrossRef\]](#)
61. Rummel, C.D.; Escher, B.I.; Sandblom, O.; Plassmann, M.M.; Arp, H.P.H.; MacLeod, M.; Jahnke, A. Effects of Leachates from UV-Weathered Microplastic in Cell-Based Bioassays. *Environ. Sci. Technol.* **2019**, *53*, 9214–9223. [\[CrossRef\]](#)

62. Helms, J.R.; Stubbins, A.; Ritchie, J.D.; Minor, E.C.; Kieber, D.J.; Mopper, K. Absorption spectral slopes and slope ratios as indicators of molecular weight, source, and photobleaching of chromophoric dissolved organic matter. *Limnol. Oceanogr.* **2008**, *53*, 955–969. [[CrossRef](#)]
63. He, X.; Xi, B.; Wei, Z.; Guo, X.; Li, M.; An, D.; Liu, H. Spectroscopic characterization of water-extractable organic matter during composting of municipal solid waste. *Chemosphere* **2011**, *82*, 541–548. [[CrossRef](#)] [[PubMed](#)]
64. Zhao, X.; Wei, Y.; Fan, Y.; Zhang, F.; Tan, W.; He, X.; Xi, B. Roles of bacterial community in the transformation of dissolved organic matter for the stability and safety of material during sludge composting. *Bioresour. Technol.* **2018**, *267*, 378–385. [[CrossRef](#)] [[PubMed](#)]
65. Baysal, A.; Saygin, H.; Soyocak, A.; Kahraman, M.; Apaydin, E.; Ozbek, P. Elucidating the leaching effect of micro/nanoplastics on the binding, structural, and oxidative characteristics of bovine serum albumin and its impact on cytotoxicity and oxidative stress in the human lung cancer cell line A549. *Environ. Sci. Nano* **2025**, *12*, 2091–2113. [[CrossRef](#)]
66. Hautala, K.; Peuravuori, J.; Pihlaja, K. Measurement of aquatic humus content by spectroscopic analyses. *Water Res.* **2000**, *34*, 246–258. [[CrossRef](#)]
67. Lionetto, F.; Esposito Corcione, C. An overview of the sorption studies of contaminants on poly(ethylene terephthalate) microplastics in the marine environment. *J. Mar. Sci. Eng.* **2021**, *9*, 445. [[CrossRef](#)]
68. Chen, L.; Feng, Y.; He, J.; Zhang, Q.; Han, L. Unraveling the effect of continuously accumulating microplastics on the humification of dissolved organic matter in the composting system. *ACS EST Eng.* **2023**, *3*, 811–822. [[CrossRef](#)]
69. Zimmermann, L.; Dombrowski, A.; Völker, C.; Wagner, M. Are bioplastics and plant-based materials safer than conventional plastics? *Environ. Int.* **2020**, *145*, 106066. [[CrossRef](#)]
70. Rummel, C.D.; Plassmann, M.M.; Arp, H.P.H.; MacLeod, M.; Jahnke, A. Effects of leachates from UV-weathered microplastic in cell culture and zebrafish embryos. *Environ. Sci. Technol.* **2019**, *53*, 2138–2146. [[CrossRef](#)]
71. Lithner, D.; Larsson, Å.; Dave, G. Environmental and health hazard ranking and assessment of plastic polymers based on chemical composition. *Sci. Total Environ.* **2011**, *409*, 3309–3324. [[CrossRef](#)]
72. Osman, A.I.; Hosny, M.; Eltaweil, A.S.; Omar, S.; Elgarahy, A.M.; Farghali, M.; Yap, P.S.; Wu, Y.S.; Nagandran, S.; Batumalaie, K.; et al. Microplastic sources, formation, toxicity and remediation: A review. *Environ. Chem. Lett.* **2023**, *21*, 2129–2169. [[CrossRef](#)]
73. Bates, J.T.; Fang, T.; Verma, V.; Zeng, L.; Weber, R.J.; Tolbert, P.E.; Abrams, J.Y.; Sarnat, S.E.; Klein, M.; Mulholland, J.A.; et al. Review of acellular assays of ambient particulate matter oxidative potential: Methods and relationships with composition, sources, and health effects. *Environ. Sci. Technol.* **2019**, *53*, 4003–4019. [[CrossRef](#)]
74. Ni, Z.; Gao, N.; Chen, N.; Zhang, C.; Liu, Z.; Zhu, K.; Sharma, V.K.; Jia, H. Particle-size distributions of environmentally persistent free radicals and oxidative potential of soils from a former gasworks site. *Sci. Total Environ.* **2023**, *869*, 161747. [[CrossRef](#)]
75. Zhang, Y.J.; Huang, C.; Lv, Y.S.; Ma, S.X.; Guo, Y.; Zeng, E.Y. Polycyclic aromatic hydrocarbon exposure, oxidative potential in dust, and their relationships to oxidative stress in human body: A case study in the indoor environment of Guangzhou, South China. *Environ. Int.* **2021**, *149*, 106405. [[CrossRef](#)] [[PubMed](#)]
76. Zhang, Y.; Kang, S.; Allen, S.; Allen, D.; Gao, T.; Sillanpää, M. Atmospheric microplastics: A review on current status and perspectives. *Earth-Sci. Rev.* **2020**, *203*, 103118. [[CrossRef](#)]
77. Turner, A.; Holmes, L.A. Adsorption of trace metals by microplastic pellets in fresh water. *Environ. Chem.* **2015**, *12*, 600–610. [[CrossRef](#)]
78. Sørensen, L.; Rogers, E.; Altin, D.; Salaberria, I.; Booth, A.M. Sorption of PAHs to microplastic and their bioavailability and toxicity to marine copepods under co-exposure conditions. *Environ. Pollut.* **2020**, *258*, 113844. [[CrossRef](#)]
79. Chamas, A.; Moon, H.; Zheng, J.; Qiu, Y.; Tabassum, T.; Jang, J.H.; Abu-Omar, M.; Scott, S.L.; Suh, S. Degradation rates of plastics in the environment. *ACS Sustain. Chem. Eng.* **2020**, *8*, 3494–3511. [[CrossRef](#)]
80. Gupta, A.; Kumar, R.; Ganguly, R.; Singh, A.K.; Rana, H.K.; Pandey, A.K. Antioxidant, anti-inflammatory and hepatoprotective activities of *Terminalia bellirica* and its bioactive component ellagic acid against diclofenac-induced oxidative stress and hepatotoxicity. *Toxicol. Rep.* **2020**, *8*, 44–52. [[CrossRef](#)]
81. Reuter, S.; Gupta, S.C.; Chaturvedi, M.M.; Aggarwal, B.B. Oxidative stress, inflammation and cancer: How are they linked? *Free Radic. Biol. Med.* **2010**, *49*, 1603–1616. [[CrossRef](#)]
82. Hwang, J.; Choi, D.; Han, S.; Choi, J.; Hong, J. An assessment of the toxicity of polypropylene microplastics in human-derived cells. *Sci. Total Environ.* **2019**, *684*, 657–669. [[CrossRef](#)] [[PubMed](#)]
83. Hwang, J.; Choi, D.; Han, S.; Jung, S.Y.; Choi, J.; Hong, J. Potential toxicity of polystyrene microplastic particles. *Sci. Rep.* **2020**, *10*, 7391. [[CrossRef](#)] [[PubMed](#)]
84. Saygin, H.; Baysal, A. Similarities and discrepancies between bio-based and conventional submicron-sized plastics: In relation to clinically important bacteria. *Bull. Environ. Contam. Toxicol.* **2020**, *105*, 26–35. [[CrossRef](#)] [[PubMed](#)]
85. Wu, X.; Pan, J.; Li, M.; Li, Y.; Bartlam, M.; Wang, Y. Selective enrichment of bacterial pathogens by microplastic biofilm. *Water Res.* **2019**, *165*, 114979. [[CrossRef](#)]

86. Auta, H.S.; Emenike, C.U.; Jayanthi, B.; Fauziah, S.H. Growth kinetics and biodeterioration of polypropylene microplastics by *Bacillus* sp. and *Rhodococcus* sp. isolated from mangrove sediment. *Mar. Pollut. Bull.* **2018**, *127*, 15–21. [\[CrossRef\]](#)
87. Flemming, H.C.; Wingender, J.; Szewzyk, U.; Steinberg, P.; Rice, S.A.; Kjelleberg, S. Biofilms: An emergent form of bacterial life. *Nat. Rev. Microbiol.* **2016**, *14*, 563–575. [\[CrossRef\]](#)
88. Sheng, G.P.; Yu, H.Q.; Li, X.Y. Extracellular polymeric substances (EPS) of microbial aggregates in biological wastewater treatment systems: A review. *Biotechnol. Adv.* **2010**, *28*, 882–894. [\[CrossRef\]](#)
89. Metryka, O.; Wasilkowski, D.; Mroziak, A. Insight into the antibacterial activity of selected metal nanoparticles and alterations within the antioxidant defence system in *Escherichia coli*, *Bacillus cereus* and *Staphylococcus epidermidis*. *Int. J. Mol. Sci.* **2021**, *22*, 11811. [\[CrossRef\]](#)
90. Flemming, H.C.; Wingender, J. The biofilm matrix. *Nat. Rev. Microbiol.* **2010**, *8*, 623–633. [\[CrossRef\]](#)
91. Halliwell, B.; Gutteridge, J.M.C. *Free Radicals in Biology and Medicine*, 5th ed.; Oxford University Press: Oxford, UK, 2015.
92. Liu, Z.; Huang, Y.; Jiao, Y.; Chen, Q.; Wu, D.; Yu, P.; Li, Y.; Cai, M.; Zhao, Y. Polystyrene nanoplastic induces ROS production and affects the MAPK-HIF-1/NFκB-mediated antioxidant system in *Daphnia pulex*. *Aquat. Toxicol.* **2020**, *220*, 105420. [\[CrossRef\]](#)
93. Liauw, C.M.; Vaidya, M.; Slate, A.J.; Hickey, N.A.; Ryder, S.; Martínez-Periñán, E.; McBain, A.J.; Banks, C.E.; Whitehead, K.A. Analysis of cellular damage resulting from exposure of bacteria to graphene oxide and hybrids using Fourier transform infrared spectroscopy. *Antibiotics* **2023**, *12*, 776. [\[CrossRef\]](#) [\[PubMed\]](#)
94. Kassem, A.; Abbas, L.; Coutinho, O.; Opara, S.; Najaf, H.; Kasperek, D.; Pokhrel, K.; Li, X.; Tiquia-Arashiro, S. Applications of Fourier transform-infrared spectroscopy in microbial cell biology and environmental microbiology: Advances, challenges, and future perspectives. *Front. Microbiol.* **2023**, *14*, 1304081.
95. Jiang, W.; Yang, K.; Vachet, R.W.; Xing, B. Interaction between oxide nanoparticles and biomolecules of the bacterial cell envelope as examined by infrared spectroscopy. *Langmuir* **2010**, *26*, 18071–18077. [\[CrossRef\]](#)
96. Hlaing, M.M.; Wood, B.R.; McNaughton, D.; Rood, J.I.; Fox, E.M.; Augustin, M.A. Vibrational spectroscopy combined with transcriptomic analysis for investigation of bacterial responses towards acid stress. *Appl. Microbiol. Biotechnol.* **2018**, *102*, 333–343. [\[CrossRef\]](#)
97. Akter, S.; Rahman, M.A.; Ashrafudoulla, M.; Ha, S.D. Biofilm formation and analysis of EPS architecture comprising polysaccharides and lipids by *Pseudomonas aeruginosa* and *Escherichia coli* on food-processing surfaces. *Food Res. Int.* **2025**, *209*, 116274. [\[CrossRef\]](#)

Disclaimer/Publisher’s Note: The statements, opinions and data contained in all publications are solely those of the individual author(s) and contributor(s) and not of MDPI and/or the editor(s). MDPI and/or the editor(s) disclaim responsibility for any injury to people or property resulting from any ideas, methods, instructions or products referred to in the content.