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Increased intestinal permeability with elevated peripheral blood endotoxin and inflammatory indices for e-waste lead exposure in children

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ABSTRACT

Lead (Pb) entering the body through different channels can damage the function of intestinal mucosal barrier and cause the body stressful inflammatory response to enhance. This study conducted a cross-sectional study to investigate the effects of Pb exposure on intestinal permeability in children by measuring the level of bacterial endotoxin and index of inflammatory cell types in peripheral blood. From November to December 2018, we recruited 187 participants aged 3–6 years by stratified randomization, from an electronic-waste-exposed group (n = 82) and a referent group (n = 105). General demographic information, past history of the digestive system in child, and family situation were informed by children's guardians with questionnaires. Children in the exposed group showed lower weight, height, and body mass index while more diarrhea in a month. Blood Pb and plasma endotoxin were elevated in exposed children than referent children and the positive relationship between them was shown in all children [B (95% CI): 0.072 (0.008, 0.137), P = 0.033]. Peripheral monocyte counts and leukotriene B_4 (LTB_4) levels were significantly increased in the exposed group. Endotoxin levels were positively correlated with neutrophils, monocytes, and LTB_4 [B (95% CI): 0.054 (0.015, 0.093), 0.018 (0.005, 0.031), and 0.049 (0.011, 0.087), respectively, $P < 0.05$]. To sum up, the exposed children showed lower physical growth levels, poorer gut health, and increased intestinal permeability, which was related to high blood Pb and peripheral inflammatory indices. These results suggest the possible adverse impact of environmental Pb exposure on the intestinal health of children.

1. Introduction

Lead (Pb) is a widespread environmental pollutant and children especially those who live in low- and middle-income countries are subject to Pb burden although the use of leaded gasoline and paint has been banned (UNICEF, 2020). Pb exerts toxicity on the immune, nervous, hemopoietic, cardiovascular, and digestive systems (Boskabady et al., 2018; Chen et al., 2021; Mohammadyan et al., 2019; Ramírez Ortega et al., 2021). In the general population, the gastrointestinal tract is the main source of Pb intake and absorption from food and environment (Kim et al., 2014; Mitra et al., 2017). The absorption rate of Pb by ingesting in children ranges 40–50% that is 4–5 times as much as adults (van der Kuijp et al., 2013; WHO, 2019). Moreover, the percentage of Pb in children's bones is 70%, while that in adults is 85–95%, which will allow children to store more Pb in soft tissues (Flora et al., 2012). Pb exposure is a contributor to the changes in diversity, composition, and abundance of gut microorganisms that inhibit the intestinal absorption and tissue accumulation of Pb, which can affect the integrity of intestinal barrier (Yu et al., 2021; Zhai et al., 2019). In addition, several studies found that Pb is capable of damaging the intestinal mucosal barrier function by damaging the structure, inducing cell apoptosis, and impairing immune function in the intestine of animals (Ding et al., 2019; Kou et al., 2019; Liu et al., 2020).

The intestinal mucosal barrier consists of commensal microbiota, mucus layer, epithelial barrier, and lamina propria, and its integrity is essential for avoiding abnormal intestinal permeability (Hanning et al.,

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2021). Especially, the epithelial barrier is composed of a single layer of epithelial cells and intercellular tight junctions that seal the gap of epithelial cells and form a paracellular pathway of substance transport (Jauregi-Miguel, 2021). With this complex system, intestinal mucosa plays a semipermeability barrier that selectively absorbs small molecular nutrients, ions, and water, while limits the translocation of large molecular antigens, pathogens, and bacterial byproducts (Vancamelbeke and Vermeire, 2017). Intestinal permeability as a measurable characteristic for the mucosal barrier, can be increased by the alterations of intestinal mucosa barrier components (Bischoff et al., 2014; Jiang et al., 2020; Wang et al., 2021). Importantly, intestinal mucosal barrier dysfunction is an important factor for symptoms or diseases, such as diarrhea, growth impairment, inflammatory bowel disease, and obesity-associated metabolic diseases (Basil et al., 2021; Bischoff et al., 2014; Dey et al., 2019; Morais and Silva, 2019; Wang et al., 2020b).

Circulating bacterial endotoxin also called lipopolysaccharide (LPS), shed from the outer membrane of the dead bacteria, is recognized as a useful bacterial biomarker of increased intestinal permeability as it can translocate from gut to the systemic circulation through the incomplete intestinal mucosal barrier (Chen et al., 2019; Nier et al., 2020; Schoultz and Keita Å, 2020; Stevens et al., 2018). Endotoxin, as a pathogen associated molecular pattern, can be recognized and combined by host cells with Toll-like receptor-4 (TLR4) and initiates MyD88 and TRIF pathways to induce secretion of pro-inflammatory cytokines (Farhana and Khan, 2021). Immune cells such as monocytes/macrophages, dendritic cells, B cells, and neutrophils can release interleukin (IL)-1β, IL-6, and tumor necrosis factor (TNF)-α when they are activated by endotoxin (Cavaillon, 2018; Farhana and Khan, 2021; Kondo et al., 2019).

Guiyu, a town in southeast China, known as one of the world's largest electronic-waste (e-waste) areas, has experienced over 40 years of impertinent e-waste dismantling and recycling (Wang et al., 2020a). E-wastes include air conditioners, television, computers, and refrigerators that were abandoned and no longer used. Pb has a higher concentration in soil, dust, plants, air, and water in Guiyu due to the combustions of metal chips or circuit boards during the informal recycling activities. In the dust of family workshops, the soil of the e-waste dumpsite, and plant of roadside in Guiyu, Pb levels were 892 mg/kg, 540.9 mg/kg, and 18.74 mg/kg, respectively, which were higher than most heavy metals (Song and Li, 2014). The concentrations of Pb in the soil (213.61 mg/kg vs. 91.92 mg/kg) and road dust (392.58 mg/kg vs. 95.72 mg/kg) in residential, rest and teaching areas in Guiyu were significantly higher than the referent group (Yekeen et al., 2016). To date, there is no direct evidence for the toxic effects of Pb on intestinal barrier function in the human body, especially in children due to their higher Pb absorption in gastrointestinal tract and storage in soft tissues. To address this knowledge gap, we evaluated the impact of Pb exposure on intestinal permeability according to the level of plasma endotoxin, peripheral inflammatory cells, parameters of physical growth, and monthly diarrhea in children in an e-waste area. It was hypothesized that Pb can perturb intestinal mucosal barrier function and increase intestinal permeability in children, which may do harm to children's growth as well as gut health and result in endotoxin release into the blood circulation that subsequently alters peripheral immune responses. Our research helps the public pay attention to the impact of Pb exposure on gut health.

2. Materials and methods

2.1. Study participants

We conducted a cross-sectional study and enrolled 187 healthy children 3–6 years of age with stratified random sampling, from two kindergartens in an e-waste-exposed group (Guiyu, n = 82) and a referent group (Haojiang, n = 105), from November to December 2018. The children were selected from the classes and stratified by age. As small eastern towns in Shantou, China, Guiyu and Haojiang are 31.6 km apart and there are many similarities such as living habits, cultural background, and socioeconomic conditions, except for e-waste dismantling activities and pollution in Guiyu. People in both areas mainly eat rice products, pickled food, and seafood. Guiyu and Haojiang are dominated by marine culture and believe in Buddhism and Taoism. Haojiang mainly develops beach cultivation, food processing industry, and tourism, whereas e-waste dismantling is the major industry in Guiyu (Zeng et al., 2020). Children who suffered from intestinal or infectious diseases, as well as other known medical diseases were excluded from this study. A questionnaire concerning the general demographic characteristics, living habits and health status of the child, family living conditions, monthly family income, and parental education levels was filled in by children's guardians or parents. Before enrollment, all children's guardians signed informed consent. Our study protocol was reviewed and approved by the Human Ethics Committee of Shantou University Medical College, China (SUMC-2015-19). The work in this study has been carried out following The Code of Ethics of the World Medical Association (Declaration of Helsinki) for experiments involving humans.

2.2. Anthropometric measurements and blood sample collection

As described previously, trained personnel conducted anthropometric measurements, including the height and weight of each child (Lu et al., 2018). The children's fasting blood was taken with EDTA anticoagulant containing and anticoagulant-free tubes from a cubital vein in the morning by experienced nurses (Zheng et al., 2019). Finally, two EDTA anticoagulant containing tubes were used to collect 3 mL of whole blood, respectively. One tube of whole blood was used for measurements of peripheral leukocyte counts and blood Pb levels, and the other was centrifugated to obtain plasma for endotoxin analysis. In addition, an anticoagulant-free tube was used to collect 3 mL of whole blood and subsequently was centrifugated to separate the serum for determining serum leukotriene B_4 (LTB_4). Plasma and serum were stored at −80 °C until assayed.

2.3. Blood Pb measurement

To measure the whole blood Pb levels, a graphite furnace atomic absorption spectrophotometer (Jena ZEEnit 650, Germany) was used by an established method in our laboratory (Yang et al., 2013). The main parameters of Pb measurement were: the drying temperatures were 90 °C, 105 °C, and 110 °C, the ashing temperature was 800 °C, and the atomization temperature was 1500 °C. The recoveries of this method were 91.06–101.20% from spiked blood samples by determined 3 times. Analytical procedures of the recovery have been reported in our previous studies (Kim et al., 2019). The detection limit was 0.084 μg/dL by measuring the 0.5% nitric solution as blank. Before measuring the samples, we adopted a five-point calibration curve to ensure the accuracy of the instrument. The correlation coefficient of the standard curve was above 0.99. Each sample was determined for 2 times, then averaged the results and calculated the relative standard deviation that below 15% was qualified.

2.4. Plasma endotoxin measurement

The plasma endotoxin concentrations were determined with commercial ELISA kits based on the manufacturer's instructions (Abbexa Co., Ltd., UK). According to the manufacturer, the sensitivity of the kit was 0.005 EU/mL, and the measuring range was 0.015–1.0 EU/mL. Before formal experiment, we conducted a pre-experiment with 8 samples (4 samples from the exposed group, others from the referent group). The result showed that the levels of the samples were in the range of 0.74–6.38 EU/mL, so the samples were diluted 10 folds in the formal experiment to make the expected concentration fall within the kit's range. The O.D. absorbance was determined using spectrophotometry at 450 nm in a microplate reader (ELx800, BioTek Instruments Inc., USA).

Table 1
Demographic characteristics of the study population.

Characteristics	Referent group (n = 105)	Exposed group (n = 82)	Statistics	*P*-value
Gender (male/female)	64/41	44/38	$\chi^2 = 1.004$	0.316[a]
Age (years) (mean ± SD)	4.93 ± 0.84	4.85 ± 0.90	$t = 0.623$	0.534[b]
Height (cm) (mean ± SD)	109.32 ± 7.30	105.37 ± 7.22	$t = 3.688$	<0.001[b]
Weight (kg) (mean ± SD)	18.70 ± 2.91	16.69 ± 2.80	$t = 4.788$	<0.001[b]
BMI (kg/m^2) (mean ± SD)	15.58 ± 1.11	14.98 ± 1.41	$t = 3.255$	0.001[b]
Diarrhea [n (%)]			$\chi^2 = 6.944$	0.008[a]
None	84 (83.2)	53 (66.2)		
At least once a month	17 (16.8)	27 (33.8)		
Blood Pb levels (μg/dL) [median (IQR)]	4.11 (3.30, 4.90)	4.49 (3.77, 5.66)	$Z = -2.593$	0.010[c]
≥5 μg/dL [n (%)]	22 (21.0)	30 (36.6)	$\chi^2 = 5.605$	0.018[a]
<5 μg/dL [n (%)]	83 (79.0)	52 (63.4)		

BMI, body mass index; SD, standard deviation; IQR, interquartile range. $P < 0.05$ is considered statistically significant.
[a] Analysis by Pearson chi-square test.
[b] Analysis by independent-sample *t*-test.
[c] Analysis by Mann-Whitney *U* test.

2.5. Measurements of other biological substances

Peripheral leukocyte counts in whole blood were measured by an automated hematology analyzer (Sysmex XE-2100, Japan). Following the manufacturer's instructions, commercial ELISA kits were used for measuring serum LTB_4 (R&D Systems Inc., USA). According to the manufacturer, the mean minimum detectable dose of the kit was 8.2 pg/mL (3.7–10.9 pg/mL) and the measuring range was 33.5–564 pg/mL in healthy volunteers.

2.6. Data processing and statistical analysis

Mean and standard deviation (SD), as well as median and interquartile range (IQR), described data with normally distributed and skewed distribution, respectively. Independent-samples *t*-test and Mann-Whitney *U* test were used to compare the differences between the two groups for normal and skewed continuous data, respectively. Moreover, categorical variables were compared by the Pearson chi-square test. To satisfy statistical requirements, some skewed data were converted to approximately normal distribution data using natural logarithmic conversion. We used a cutoff value of 5 μg/dL, based on Pb referent level of the United States Centers for Disease Control and Prevention (CDC), to categorize the high and low level of child blood Pb (Lanphear et al., 2005). The factors associated with Pb exposure were explored by Spearman correlation analyses. Multivariable adjusted linear regression models were used to assess the associations of plasma endotoxin to blood Pb, peripheral leukocyte counts and LTB_4. Based on the previous literature, confounders were composed of gender, age, body mass index (BMI), e-waste contact, hand washing habit, e-waste pollution within 50 m from the house, distance of residence from the road, daily household smoking, parental education levels, and monthly family income (Cai et al., 2019; Lu et al., 2018). All statistical analyses were performed using SPSS 20.0 (SPSS Inc., Chicago, IL, USA) and figures were drawn with GraphPad Prism 8.0 (GraphPad, San Diego, CA). A $P < 0.05$ was considered significant in a two-tailed test.

3. Results

3.1. Demographic characteristics of the participants

The general characteristics and blood Pb levels of 187 participants can be seen in Table 1 and Supplemental Material Table S1. Two groups had a similar gender and age distribution ($P > 0.05$). However, the exposed children showed lower height (105.37 ± 7.22 cm vs. 109.32 ± 7.30 cm, $P < 0.001$), weight (16.69 ± 2.80 kg vs. 18.70 ± 2.91 kg, $P < 0.001$), and BMI (14.98 ± 1.41 kg/m^2 vs. 15.58 ± 1.11 kg/m^2, $P = 0.001$) compared with those in the referent group. Apart from the significant growth differences between the exposed and referent group children, the exposed children exhibited more diarrhea in a month ($P = 0.008$). Diarrhea was defined as at least three loose or liquid stools a day. In addition, the exposed children had unhealthy lifestyle habits and housing conditions compared with referent children, including fewer hands washing, contacting with e-waste, residing within 50 m from an e-waste site and closer to the road ($P < 0.05$). Moreover, the educational levels of children's parents and monthly family income were lower in the exposed group ($P < 0.05$).

The exposed children showed higher blood Pb levels than the referent children (median: 4.49 μg/dL vs. 4.11 μg/dL, $P = 0.010$). Furthermore, 36.6% of children in the exposed group exceeded the U.S. CDC suggested blood Pb threshold in children (5 μg/dL), while 21.0% of children in the referent group exceed it ($P = 0.018$). To sum up, children residing in the e-waste recycling area bear high Pb exposure, have digestive tract symptom as well as lower physical growth and development levels, and live with unhealthy living habits and conditions.

3.2. Factors influencing Pb exposure

To identify whether there were clear factors interrelated to blood Pb levels in participants, Spearman correction analyses were applied. The results indicated that blood Pb levels were positively related to e-waste contact ($r_s = 0.148$, $P = 0.043$) and e-waste pollution within 50 m away from the house ($r_s = 0.169$, $P = 0.021$). Rather, blood Pb levels were negatively associated with hand washing habit ($r_s = -0.178$, $P = 0.015$), distance of residence from road ($r_s = -0.191$, $P = 0.009$), father's education level ($r_s = -0.190$, $P = 0.009$), mother's education level ($r_s = -0.202$, $P = 0.006$) and monthly family income ($r_s = -0.317$, $P < 0.001$). In short, blood Pb levels are influenced by children's daily habits, living conditions, parents' education, and family economic income (Table S2).

3.3. Plasma endotoxin concentrations and the associations with blood Pb levels

We used plasma bacterial endotoxin to assess the intestinal barrier function. Compared with referent children, the plasma endotoxin levels in exposed children were increased markedly (median: 2.24 EU/mL vs. 1.85 EU/mL, $P = 0.037$) (Fig. 1A). Similarly, children with higher blood Pb levels had higher plasma endotoxin concentrations compared to those with low blood Pb levels (median: 2.31 EU/mL vs. 1.93 EU/mL, $P = 0.009$) (Fig. 1B).

To understand the association between blood Pb and plasma endotoxin, multivariable adjusted linear regression models were applied (Table 2). The positive relationships between blood Pb levels and plasma natural logarithm-transformed endotoxin (Ln-endotoxin) concentrations could be seen in the results, whether in an unadjusted regression model or model adjusted by potential confounders [unadjusted: B (95% CI) = 0.064 (0.006, 0.123) and adjusted: B (95% CI) = 0.072 (0.008,

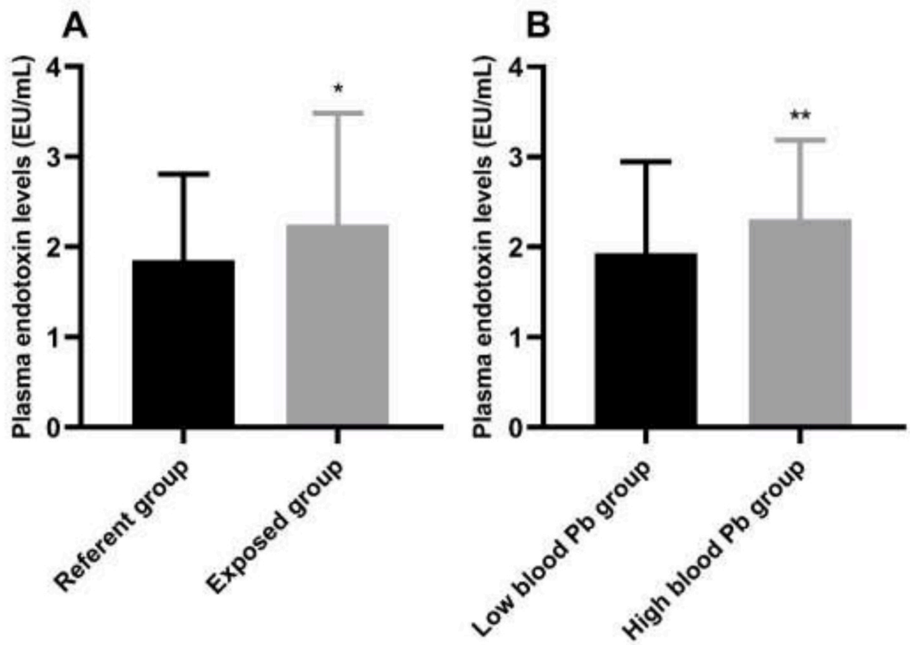

Fig. 1. Levels of plasma endotoxin in children. Data are presented as median (interquartile range) and analyzed by the Mann-Whitney *U* test, $^{*}P < 0.05$, $^{**}P < 0.01$.

Table 2
Associations between blood Pb levels and Ln-endotoxin levels in children.

Blood Pb	Ln-endotoxin		
	B (95% CI)	*β*	*P*-value
Model 1	0.064 (0.006, 0.123)	0.160	0.031
Model 2	0.072 (0.008, 0.137)	0.180	0.033

Model 1: unadjusted.
Model 2: adjusted for gender, age, BMI, hand washing habit, e-waste contact, e-waste pollution within 50 m from the house, distance of residence from road, daily household smoking, parental education levels, and monthly family income.
B, unstandardized coefficient; CI, confidence interval; *β*, standardized coefficient; BMI, body mass index; Ln-endotoxin, ln-transformed endotoxin. $P < 0.05$ is considered statistically significant.

0.137), respectively, $P < 0.05$]. Thus, increased intestinal permeability is exhibited in children living in the e-waste area and has a positive association with higher blood Pb levels.

3.4. *Peripheral leukocyte counts and serum LTB_4 levels*

Peripheral blood leukocyte counts and serum LTB_4 were used to evaluate the peripheral immune state. Comparisons of the counts of WBC, neutrophil, and monocyte between referent and exposed children showed that only monocytes were significantly higher in exposed children (median: 0.43×10^9/L vs. 0.37×10^9/L, $P < 0.001$) (Fig. 2A). WBCs and neutrophils had suggestive increases in the exposed group compared to the referent group, albeit the differences were not statistically significant (median: 7.25×10^9/L vs. 6.57×10^9/L and 3.12×10^9/L vs. 3.05×10^9/L, respectively, $P > 0.05$). In addition, serum lipid mediator LTB_4 levels in exposed children were higher compared with referent children (median: 211.70 pg/mL vs. 184.98 pg/mL, $P = 0.024$)

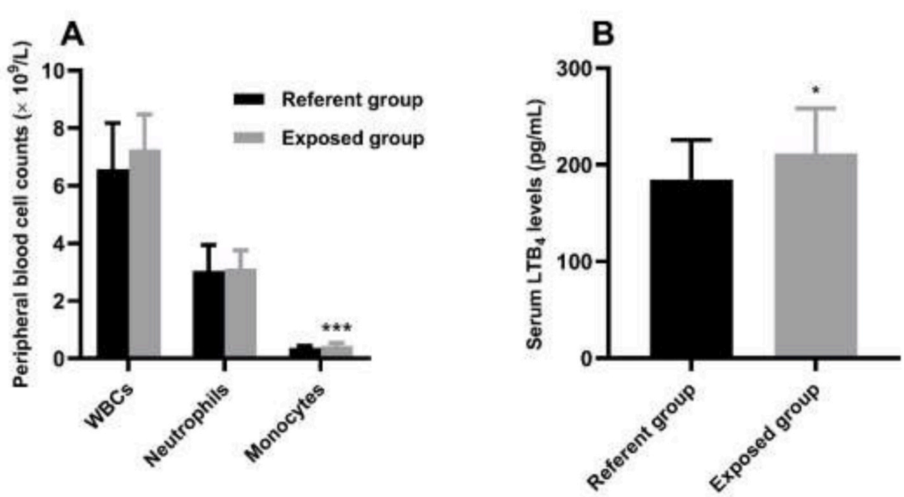

Fig. 2. Peripheral blood cell counts and serum LTB_4 levels in children. WBC, white blood cell; LTB_4, leukotriene B_4. Data are presented as median (interquartile range) and analyzed by the Mann-Whitney *U* test, $^{*}P < 0.05$, $^{***}P < 0.001$.

(Fig. 2B). In brief, exposed children show changes in the state of peripheral inflammation.

3.5. Relationships between endotoxin and peripheral inflammatory indices

For understanding the association between endotoxin and peripheral inflammatory indices, we adopted multivariable adjusted linear regression analysis (Table 3). After adjustment for potential confounders, the results showed that Ln-neutrophils, monocytes, and Ln-LTB_4 were positively associated with endotoxin [B (95% CI) = 0.054 (0.015, 0.093), B (95% CI) = 0.018 (0.005, 0.031), and B (95% CI) = 0.049 (0.011, 0.087), respectively, $P < 0.05$]. Hence, the alteration of peripheral inflammatory response is linked to increased intestinal permeability.

4. Discussion

According to literature study, it is the first time to research the harm of Pb on intestinal mucosal barrier function in children according to plasma bacterial endotoxin, peripheral inflammatory factors, the levels of physical growth, and diarrhea. It was observed that blood Pb, plasma endotoxin, and peripheral inflammatory factors including monocyte counts and serum LTB_4 were increased in children from e-waste recycling area. Results showed a positive association between plasma endotoxin and blood Pb in all participants. In addition, endotoxin levels were related to peripheral monocyte counts, neutrophil counts, and serum LTB_4 levels. Furthermore, we found the exposed group children had lower height, weight, and BMI while more diarrhea a month. The results indicate that Pb may be a potential risk factor for intestinal health, and increased intestinal permeability may promote the elevation of peripheral blood inflammatory indices.

Consistent with the result of this work, our previous investigations have found that Guiyu children experienced higher Pb exposure than children 0–6 years of age in China (geometric mean: 4.24 μg/dL, from 2012 to 2017), both exceed the blood Pb levels of U.S. children 1–5 years of age (geometric mean: 0.758 μg/dL, from 2015 to 2016) (CDC, 2019; Xu et al., 2020; Xu et al., 2015; Zhang et al., 2019). In addition to children, pregnant women in Guiyu also have higher blood Pb levels than referent groups, which may enhance the Pb levels in their infants' body (Kazi et al., 2014; Kim et al., 2020). Moreover, the proportion of children who had blood Pb levels over 5 μg/dL was elevated in the exposed group. The result shows that children living in e-waste areas still suffer from higher Pb exposure. Similar to the living environment of e-waste-exposed children, workers with e-waste recycling occupations also displayed high blood Pb levels in Nigeria (geometric mean: 11.0 μg/dL) as well as those with battery recycling in Pakistan (median: 25.5 μg/dL) (Baloch et al., 2020; Popoola et al., 2019). The levels of environmental pollutants in the body can be affected by routes and frequencies of exposure, as well as the absorption, distribution, metabolism, and excretion of the poisons (Abass et al., 2017). Therefore, we conducted a questionnaire survey to investigate the demographic characteristics, daily routine, and family environment to find out the factors influencing Pb exposure in children. The results showed that e-waste pollution within 50 m away from residence and experience of e-waste contacting had positive relationships with higher blood Pb levels, whereas times of handwashing, the distance between residence and road, parental education levels, and monthly family income were opposite, which were in accordance with our previous studies (Dai et al., 2017; Zhang et al., 2020). These results suggest that poor living conditions and hygienic habits are potential promoters of Pb exposure. Due to the higher Pb levels in soil or dust in e-waste area and frequent hand to mouth contact, children have more chances to direct or indirect (through the air, food, toys, and dust/soil contaminated by metal Pb) contact Pb when there are e-waste sites near or in his family or live near the roads (Heacock et al., 2016; Li et al., 2020; Zhao et al., 2015). Compared to higher-income families, low-income families may lack the finance to refurbish the older house decorated by Pb paint, and their children had a greater possibility to bear high Pb exposure (Marshall et al., 2020). In addition, people with lower educational level may lack the awareness to protect them from pollutants exposure, which has potential contributions of higher blood Pb and diseases (Abass et al., 2017; Decharat and Kiddee, 2020; Kazi et al., 2015). Therefore, parents, especially those in the exposed group with low education levels may carry out environmental pollutants home from the workplace and do not effectively guide their children to stay away from the pollutants (Cai et al., 2019). To reduce Pb exposure, guardians should strengthen their children's awareness of hygiene and diet, as well as improve living conditions.

To evaluate the impact of Pb on intestinal mucosal barrier function, we chose endotoxin as an indicator of increased intestinal permeability and bacterial translocation (Schoultz and Keita Å, 2020). Endotoxin is derived from the outer membrane of gram-negative bacteria and normally exists in the gut, but it is only very low in healthy individuals' bloodstream (Kell and Pretorius, 2015). We showed circulating bacterial endotoxin was higher in children in the exposed and the high Pb-exposed groups, suggesting children exposed to e-waste or who had higher blood Pb levels suffer from increased intestinal permeability. Elevated circulating endotoxin level is not the essential biomarker of infectious diseases caused by gram-negative bacteria, because gram-positive infections and some non-infectious conditions, such as obesity, trauma, cardiac arrest, and severe autism also are associated with circulating endotoxin (Sturgeon et al., 2019). Additionally, in our study, the higher endotoxin levels were positively correlated with elevated blood Pb levels in different models, indicating Pb has a potential influence on the integrity of the intestinal barrier. Pb exposure can cause oxidative stress by inducing the production of oxidant and disturbing the expression and activity of antioxidants (Jing et al., 2020; Zhao et al., 2020). Pb also has strong immunotoxicity for its capability of changing the immunophenotype of cells and increasing inflammatory cells and mediators (Boskabady et al., 2018). Accumulating evidence suggests that gut microflora dysbiosis, intestinal epithelial cells damage, dysregulated expression of tight junction proteins, and gut inflammation are conducive to increase intestinal permeability (Feng et al., 2019; Kim, 2021; Solis et al., 2020). Several studies have suggested that accompanied immune response and oxidative stress, Pb exposure impairs the ultrastructure of the gut epithelium (damaged nucleus, vacuolized mitochondria, and contracted microvilli) and influences intestinal microbiota homeostasis (changes of the community structure, species diversity, taxonomic compositions, and taxa abundance), which are harmful intestinal barrier function (Kou et al., 2019; Zheng et al., 2021). In addition, *in vitro* and *in vivo* studies show that after Pb exposure in HT-29 cells and colon tissue of mice, the mRNA expression of ZO-1 and occludin are suppressed (Yu et al., 2021). In turn, the injured intestinal barrier may damage itself by LPS stimulation (Bein et al., 2017). In all,

Table 3
Associations among plasma endotoxin, peripheral leukocyte subtypes, and serum LTB_4 in children.

	Endotoxin		
	B (95% CI)	β	*P*-value
Ln-neutrophils	0.054 (0.015, 0.093)	0.202	0.007
Monocytes	0.018 (0.005, 0.031)	0.197	0.008
Ln-LTB_4	0.049 (0.011, 0.087)	0.228	0.012

Adjusted for gender, age, BMI, hand washing habit, e-waste contact, e-waste pollution within 50 m from the house, distance of residence from road, daily household smoking, parental education levels, and monthly family income.
B, unstandardized coefficient; CI, confidence interval; β, standardized coefficient; BMI, body mass index; Ln-neutrophils, ln-transformed neutrophils; Ln-LTB_4, ln-transformed leukotriene B_4. $P < 0.05$ is considered statistically significant.

our findings suggest that Pb exposure may damage intestinal health with increased permeability, which promotes endotoxin translocation.

The intestinal mucosa plays an essential role in the absorption of nutrients, water, and ions, and increased intestinal permeability can have adverse impacts on its absorption function. Increased intestinal permeability promotes intestinal antigenic translocation, thereby activating gut and systemic inflammatory, and is related to various gastrointestinal disorders, including diarrhea, inflammatory bowel disease, food allergy, and small intestinal bacterial overgrowth (Eutamene et al., 2018; Keusch et al., 2014). In our study, we found the parameters of physical growth and development (height, weight, and BMI) were decreased, and monthly diarrhea was increased in exposed children. Similarly, children with environmental enteric dysfunction are linked to increased intestinal permeability, defective absorption of nutrient, and intestinal flora imbalance, contributing to growth impairment (Morais and Silva, 2019). A study showed that the decrease in linear growth of rural Malawian children 2–5 years of age is related to increased barrier permeability (Weisz et al., 2012). In addition, children with innutrition have higher Pb levels in blood and scalp hair compared with well-nourished children (Talpur et al., 2018). Pb exposure is associated with height, weight, BMI, bust, head circumference, and waistline for children (Kuang et al., 2020; Zeng et al., 2019). Consequently, the decline in growth and more diarrhea of children in the exposed group may be due to the adverse effect of Pb exposure on intestinal barrier function.

After the translocation to the blood, endotoxin activates immune cells by combining differentiation 14 (CD14) and TLR4, inducing activation of MyD88 dependent and TRIF pathways, leading to the transcription and expression of pro-inflammatory cytokines by immune cells (Kawasaki and Kawai, 2014). Even without infection or diseases, peripheral leukocytes have been used as indicators to assess inflammatory response with the increased counts (Parsons et al., 2020). As the main target cells of endotoxin, peripheral monocytes and neutrophils expressed membrane CD14 and TLR4 can be activated by endotoxin, leading to inflammatory response (Salvesen et al., 2016). In our earlier studies, peripheral leukocytes and pro-inflammatory factors levels were increased in Guiyu children (Dai et al., 2017; Zheng et al., 2019). Here, we also presented higher absolute counts of WBC, neutrophil, and monocyte in exposed children, although only the difference of monocytes between the two groups was significant, suggesting children in Guiyu maintain disrupted immune responses. In addition, plasma endotoxin was positively correlated with increased monocytes and Ln-neutrophils after adjusting for confounders. Consistently, two experimental endotoxemia models have suggested that in humans, intravenous administration of endotoxin can increase peripheral neutrophils and plasma cytokines (Grigoleit et al., 2010; Taudorf et al., 2007). In addition, during intraperitoneal intermittent LPS injection, pro-inflammatory mediators including C-reactive protein, TNF-α, IL-1β, IL-6, and IL-8, as well as peripheral neutrophils, monocytes, and lymphocytes, were elevated in rats, indicating chronic subclinical systemic inflammation (Ranneh et al., 2019). However, the above changes of neutrophils and monocytes were shown in the endotoxemia or systemic inflammation models, whereas the participants in this study were healthy children. The proportion of neutrophil (50–70%) in peripheral blood leukocytes of children is larger compared with monocytes (1–8%), which needs more large changes to make the difference statistically significant. Furthermore, the sample size of this study may not be enough to exhibit the statistical difference of the WBCs and neutrophils. In our present results, LTB_4 levels were elevated in children from the exposed group and related to increased endotoxin levels. LTB_4 as one of the products of arachidonic acid metabolism as well as a pro-inflammatory lipid mediator participates in systemic inflammation with other inflammatory biomarkers (Elango et al., 2013). LPS has been shown to increase secretion of LTB_4 in the bovine mammary gland and porcine endometrial endothelial cells (Czarzasta et al., 2018; Piotrowska-Tomala et al., 2015). According to current research, the chronic inflammatory condition may be a contributor to the development of chronic non-communicable diseases (such as obesity, depression, cardiovascular diseases, and nonalcoholic fatty liver disease) (Bessone et al., 2019; Gasmi et al., 2020; Kiecolt-Glaser et al., 2018; Thomas and Advani, 2006). Therefore, in our study, e-waste-exposed children with elevated circulating endotoxin have increased blood inflammatory biomarkers, which may harm the development and intestinal health of children.

Some limitations of this study cannot be ignored. Firstly, bacterial endotoxin serves as a biomarker of increased intestinal permeability, but it cannot reflect the injury site of the intestinal barrier. We did not measure zonulin, fatty acid-binding proteins, and diamine oxidase to evaluate the injury of intestinal epithelium or tight junction to further find out the possible mechanisms of increased intestinal permeability by Pb exposure (Galipeau and Verdu, 2016; Wang et al., 2015). Secondly, there are a variety of e-waste contaminants in Guiyu, which may not be conducive to accurately assessing the toxicity of Pb exposure to the intestine. Although the intestinal barrier function also may be affected by other pollutants, in our results, higher endotoxin levels were showed in children with higher blood Pb levels, which can provide evidence for the effect of Pb exposure on intestinal permeability. Finally, the cross-sectional study that we conducted cannot conclude that Pb exposure would cause adverse outcomes on the intestinal barrier, and only can show the correlation between them. Further research should be conducted, especially cohort studies, to pay attention to the harm of Pb exposure to intestinal health and immune function.

5. Conclusions

This is the first study to measure the blood bacterial endotoxin, peripheral inflammatory indices, the levels of physical growth, and intestinal health to reflect the impact of Pb burden on intestinal mucosal barrier function in children in an e-waste area. We find that increased plasma bacterial endotoxin is shown in e-waste-exposed children and related to high blood Pb and peripheral inflammatory indices. In addition, e-waste-exposed children show lower levels of growth and more diarrhea a month. In general, this study is consistent with the hypothesis that Pb may harm intestinal mucosal barrier function in children. Pb exposure is still a global health issue, especially for children and this research draws public concerns about the unhealthy gut for the Pb-exposed population.

Credit author statement

Xiuli Luo: Conceptualization, Investigation, Formal analysis, Writing – original draft. Xia Huo: Writing – review & editing, Supervision, Project administration, Funding acquisition. Yuling Zhang: Investigation, Project administration. Zhiheng Cheng: Investigation, Formal analysis. Shuqin Chen: Data curation, Investigation. Xijin Xu: Conceptualization, Resources, Writing – review & editing, Supervision, Project administration, Formal analysis.

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Declaration of competing interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

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Appendix A. Supplementary data

Supplementary data to this article can be found online at https://doi.org/10.1016/j.chemosphere.2021.130862.

References

Abass, K., Koiranen, M., Mazej, D., Tratnik, J.S., Horvat, M., Hakkola, J., Jarvelin, M.R., Rautio, A., 2017. Arsenic, cadmium, lead and mercury levels in blood of Finnish adults and their relation to diet, lifestyle habits and sociodemographic variables. Environ. Sci. Pollut. Res. Int. 24, 1347–1362. https://doi.org/10.1007/s11356-016-7824-5.

Baloch, S., Kazi, T.G., Baig, J.A., Afridi, H.I., Arain, M.B., 2020. Occupational exposure of lead and cadmium on adolescent and adult workers of battery recycling and welding workshops: adverse impact on health. Sci. Total Environ. 720, 137549 https://doi.org/10.1016/j.scitotenv.2020.137549.

Basil, M., Schwartz, D., Dalal, R., Horst, S., Scoville, E., Adams, D., Beaulieu, D., Slaughter, J.C., Higginbotham, T., Vaezi, M., Choksi, Y., 2021. Mucosal integrity testing can detect differences in the rectums of patients with inflammatory bowel disease compared to controls: a pilot study. Dig. Dis. Sci. https://doi.org/10.1007/s10620-021-06888-5.

Bein, A., Zilbershtein, A., Golosovsky, M., Davidov, D., Schwartz, B., 2017. LPS induces hyper-permeability of intestinal epithelial cells. J. Cell. Physiol. 232, 381–390. https://doi.org/10.1002/jcp.25435.

Bessone, F., Razori, M.V., Roma, M.G., 2019. Molecular pathways of nonalcoholic fatty liver disease development and progression. Cell. Mol. Life Sci. 76, 99–128. https://doi.org/10.1007/s00018-018-2947-0.

Bischoff, S.C., Barbara, G., Buurman, W., Ockhuizen, T., Schulzke, J.D., Serino, M., Tilg, H., Watson, A., Wells, J.M., 2014. Intestinal permeability–a new target for disease prevention and therapy. BMC Gastroenterol. 14, 189. https://doi.org/10.1186/s12876-014-0189-7.

Boskabady, M., Marefati, N., Farkhondeh, T., Shakeri, F., Farshbaf, A., Boskabady, M.H., 2018. The effect of environmental lead exposure on human health and the contribution of inflammatory mechanisms, a review. Environ. Int. 120, 404–420. https://doi.org/10.1016/j.envint.2018.08.013.

Cai, H., Xu, X., Zhang, Y., Cong, X., Lu, X., Huo, X., 2019. Elevated lead levels from e-waste exposure are linked to sensory integration difficulties in preschool children. Neurotoxicology 71, 150–158. https://doi.org/10.1016/j.neuro.2019.01.004.

Cavaillon, J.M., 2018. Exotoxins and endotoxins: inducers of inflammatory cytokines. Toxicon 149, 45–53. https://doi.org/10.1016/j.toxicon.2017.10.016.

CDC, 2019. Fourth National Report on Human Exposure to Environmental Chemicals. https://www.cdc.gov/exposurereport/pdf/FourthReport_UpdatedTables_Volume1_Jan2019-508.pdf.

Chen, M., Huang, H., Zhou, P., Zhang, J., Dai, Y., Yang, D., Fan, X., Pan, H., 2019. Oral phosphatidylcholine improves intestinal barrier function in drug-induced liver injury in rats. Gastroenterol Res Pract, 8723460. https://doi.org/10.1155/2019/8723460.

Chen, Z., Huo, X., Zhang, S., Cheng, Z., Huang, Y., Xu, X., 2021. Relations of blood lead levels to echocardiographic left ventricular structure and function in preschool children. Chemosphere 268, 128793. https://doi.org/10.1016/j.chemosphere.2020.128793.

Czarzasta, J., Meller, K., Andronowska, A., Jana, B., 2018. Lipopolysaccharide and cytokines modulate leukotriene (LT)B4 and LTC4 production by porcine endometrial endothelial cells. Reprod. Domest. Anim. 53, 101–109. https://doi.org/10.1111/rda.13077.

Dai, Y., Huo, X., Zhang, Y., Yang, T., Li, M., Xu, X., 2017. Elevated lead levels and changes in blood morphology and erythrocyte CR1 in preschool children from an e-waste area. Sci. Total Environ. 592, 51–59. https://doi.org/10.1016/j.scitotenv.2017.03.080.

Decharat, S., Kiddee, P., 2020. Health problems among workers who recycle electronic waste in southern Thailand. Osong Public Health Res Perspect 11, 34–43. https://doi.org/10.24171/j.phrp.2020.11.1.06.

Dey, P., Sasaki, G.Y., Wei, P., Li, J., Wang, L., Zhu, J., McTigue, D., Yu, Z., Bruno, R.S., 2019. Green tea extract prevents obesity in male mice by alleviating gut dysbiosis in association with improved intestinal barrier function that limits endotoxin translocation and adipose inflammation. J. Nutr. Biochem. 67, 78–89. https://doi.org/10.1016/j.jnutbio.2019.01.017.

Ding, Z., Kong, Y., Shao, X., Zhang, Y., Ren, C., Zhao, X., Yu, W., Jiang, T., Ye, J., 2019. Growth, antioxidant capacity, intestinal morphology, and metabolomic responses of juvenile Oriental river prawn (Macrobrachium nipponense) to chronic lead exposure. Chemosphere 217, 289–297. https://doi.org/10.1016/j.chemosphere.2018.11.034.

Elango, N., Kasi, V., Vembhu, B., Poornima, J.G., 2013. Chronic exposure to emissions from photocopiers in copy shops causes oxidative stress and systematic inflammation among photocopier operators in India. Environ. Health 12, 78. https://doi.org/10.1186/1476-069x-12-78.

Eutamene, H., Beaufrand, C., Harkat, C., Theodorou, V., 2018. The role of mucoprotectants in the management of gastrointestinal disorders. Expet Rev. Gastroenterol. Hepatol. 12, 83–90. https://doi.org/10.1080/17474124.2018.1378573.

Farhana, A., Khan, Y.S., 2021. Biochemistry, Lipopolysaccharide. StatPearls. Treasure Island (FL): StatPearls Publishing Copyright © 2021, StatPearls Publishing LLC.

Feng, L., Chen, S., Zhang, L., Qu, W., Chen, Z., 2019. Bisphenol A increases intestinal permeability through disrupting intestinal barrier function in mice. Environ. Pollut. 254, 112960 https://doi.org/10.1016/j.envpol.2019.112960.

Flora, G., Gupta, D., Tiwari, A., 2012. Toxicity of lead: a review with recent updates. Interdiscipl. Toxicol. 5, 47–58. https://doi.org/10.2478/v10102-012-0009-2.

Galipeau, H.J., Verdu, E.F., 2016. The complex task of measuring intestinal permeability in basic and clinical science. Neuro Gastroenterol. Motil. 28, 957–965. https://doi.org/10.1111/nmo.12871.

Gasmi, A., Mujawdiya, P.K., Pivina, L., Doşa, A., Semenova, Y., Gasmi Benahmed, A., Bjørklund, G., 2020. Relationship between gut microbiota, gut hyperpermeability, and obesity. Curr. Med. Chem. https://doi.org/10.2174/0929867327666200721160313.

Grigoleit, J.S., Oberbeck, J.R., Lichte, P., Kobbe, P., Wolf, O.T., Montag, T., del Rey, A., Gizewski, E.R., Engler, H., Schedlowski, M., 2010. Lipopolysaccharide-induced experimental immune activation does not impair memory functions in humans. Neurobiol. Learn. Mem. 94, 561–567. https://doi.org/10.1016/j.nlm.2010.09.011.

Hanning, N., Edwinson, A.L., Ceuleers, H., Peters, S.A., De Man, J.G., Hassett, L.C., De Winter, B.Y., Grover, M., 2021. Intestinal barrier dysfunction in irritable bowel syndrome: a systematic review. Therap Adv Gastroenterol 14. https://doi.org/10.1177/1756284821993586, 1756284821993586.

Heacock, M., Kelly, C.B., Asante, K.A., Birnbaum, L.S., Bergman, A.L., Brune, M.N., Buka, I., Carpenter, D.O., Chen, A., Huo, X., Kamel, M., Landrigan, P.J., Magalini, F., Diaz-Barriga, F., Neira, M., Omar, M., Pascale, A., Ruchirawat, M., Sly, L., Sly, P.D., Van den Berg, M., Suk, W.A., 2016. E-Waste and harm to vulnerable populations: a growing global problem. Environ. Health Perspect. 124, 550–555. https://doi.org/10.1289/ehp.1509699.

Jauregi-Miguel, A., 2021. The tight junction and the epithelial barrier in coeliac disease. Int Rev Cell Mol Biol 358, 105–132. https://doi.org/10.1016/bs.ircmb.2020.09.010.

Jiang, Z., Mu, W., Yang, Y., Sun, M., Liu, Y., Gao, Z., Li, J., Gu, P., Wang, H., Lu, Y., Ba, Q., Wang, H., 2020. Cadmium exacerbates dextran sulfate sodium-induced chronic colitis and impairs intestinal barrier. Sci. Total Environ. 744 https://doi.org/10.1016/j.scitotenv.2020.140844.

Jing, H., Zhang, Q., Li, S., Gao, X.J., 2020. Pb exposure triggers MAPK-dependent inflammation by activating oxidative stress and miRNA-155 expression in carp head kidney. Fish Shellfish Immunol. 106, 219–227. https://doi.org/10.1016/j.fsi.2020.08.015.

Kawasaki, T., Kawai, T., 2014. Toll-like receptor signaling pathways. Front. Immunol. 5, 461. https://doi.org/10.3389/fimmu.2014.00461.

Kazi, T.G., Shah, F., Afridi, H.I., Naeemullah, 2015. Occupational and environmental lead exposure to adolescent workers in battery recycling workshops. Toxicol. Ind. Health 31, 1288–1295. https://doi.org/10.1177/0748233713485883.

Kazi, T.G., Shah, F., Shaikh, H.R., Afridi, H.I., Shah, A., Naeemullah Arain, S.S., 2014. Exposure of lead to mothers and their new born infants, residents of industrial and domestic areas of Pakistan. Environ. Sci. Pollut. Res. Int. 21, 3021–3030. https://doi.org/10.1007/s11356-013-2223-7.

Kell, D.B., Pretorius, E., 2015. On the translocation of bacteria and their lipopolysaccharides between blood and peripheral locations in chronic, inflammatory diseases: the central roles of LPS and LPS-induced cell death. Integr Biol (Camb). 7, 1339–1377. https://doi.org/10.1039/c5ib00158g.

Keusch, G.T., Denno, D.M., Black, R.E., Duggan, C., Guerrant, R.L., Lavery, J.V., Nataro, J.P., Rosenberg, I.H., Ryan, E.T., Tarr, P.I., Ward, H., Bhutta, Z.A., Coovadia, H., Lima, A., Ramakrishna, B., Zaidi, A.K., Hay Burgess, D.C., Brewer, T., 2014. Environmental enteric dysfunction: pathogenesis, diagnosis, and clinical consequences. Clin. Infect. Dis. 59 (Suppl. 4), S207–S212. https://doi.org/10.1093/cid/ciu485.

Kiecolt-Glaser, J.K., Wilson, S.J., Bailey, M.L., Andridge, R., Peng, J., Jaremka, L.M., Fagundes, C.P., Malarkey, W.B., Laskowski, B., Belury, M.A., 2018. Marital distress, depression, and a leaky gut: translocation of bacterial endotoxin as a pathway to inflammation. Psychoneuroendocrinology 98, 52–60. https://doi.org/10.1016/j.psyneuen.2018.08.007.

Kim, H.S., 2021. Do an Altered Gut Microbiota and an Associated Leaky Gut Affect COVID-19 Severity? mBio, vol. 12. https://doi.org/10.1128/mBio.03022-20.

Kim, J., Lee, Y., Yang, M., 2014. Environmental exposure to lead (Pb) and variations in its susceptibility. J. Environ. Sci. Health C Environ. Carcinog. Ecotoxicol. Rev. 32, 159–185. https://doi.org/10.1080/10590501.2014.907461.

Kim, S., Xu, X., Zhang, Y., Zheng, X., Liu, R., Dietrich, K., Reponen, T., Ho, S.M., Xie, C., Sucharew, H., Huo, X., Chen, A., 2019. Metal concentrations in pregnant women and neonates from informal electronic waste recycling. J. Expo. Sci. Environ. Epidemiol. 29, 406–415. https://doi.org/10.1038/s41370-018-0054-9.

Kim, S.S., Xu, X., Zhang, Y., Zheng, X., Liu, R., Dietrich, K.N., Reponen, T., Xie, C., Sucharew, H., Huo, X., Chen, A., 2020. Birth outcomes associated with maternal exposure to metals from informal electronic waste recycling in Guiyu, China. Environ. Int. 137, 105580 https://doi.org/10.1016/j.envint.2020.105580.

Kondo, Y., Ledderose, C., Slubowski, C.J., Fakhari, M., Sumi, Y., Sueyoshi, K., Bezler, A. K., Aytan, D., Arbab, M., Junger, W.G., 2019. Frontline Science: Escherichia coli use LPS as decoy to impair neutrophil chemotaxis and defeat antimicrobial host defense. J. Leukoc. Biol. https://doi.org/10.1002/jlb.4hi0319-109r.

Kou, H., Fu, Y., He, Y., Jiang, J., Gao, X., Zhao, H., 2019. Chronic lead exposure induces histopathological damage, microbiota dysbiosis and immune disorder in the cecum of female Japanese quails (Coturnix japonica). Ecotoxicol. Environ. Saf. 183, 109588 https://doi.org/10.1016/j.ecoenv.2019.109588.

Kuang, W., Chen, Z., Shi, K., Sun, H., Li, H., Huang, L., Bi, J., 2020. Adverse health effects of lead exposure on physical growth, erythrocyte parameters and school

performances for school-aged children in eastern China. Environ. Int. 145, 106130 https://doi.org/10.1016/j.envint.2020.106130.
Lanphear, B.P., Hornung, R., Khoury, J., Yolton, K., Baghurst, P., Bellinger, D.C., Canfield, R.L., Dietrich, K.N., Bornschein, R., Greene, T., Rothenberg, S.J., Needleman, H.L., Schnaas, L., Wasserman, G., Graziano, J., Roberts, R., 2005. Low-level environmental lead exposure and children's intellectual function: an international pooled analysis. Environ. Health Perspect. 113, 894–899. https://doi.org/10.1289/ehp.7688.
Li, M.M., Gao, Z.Y., Dong, C.Y., Wu, M.Q., Yan, J., Cao, J., Ma, W.J., Wang, J., Gong, Y. L., Xu, J., Cai, S.Z., Chen, J.Y., Xu, S.Q., Tong, S., Tang, D., Zhang, J., Yan, C.H., 2020. Contemporary blood lead levels of children aged 0-84months in China: a national cross-sectional study. Environ. Int. 134, 105288 https://doi.org/10.1016/j.envint.2019.105288.
Liu, H.S., Fu, S.L., Zhang, S.S., Ding, M.M., Wang, A.L., 2020. Lead induces structural damage, microbiota dysbiosis and cell apoptosis in the intestine of juvenile bighead carp (Hypophthalmichthys nobilis). Aquaculture 528, 11. https://doi.org/10.1016/j.aquaculture.2020.735573.
Lu, X., Xu, X., Zhang, Y., Zhang, Y., Wang, C., Huo, X., 2018. Elevated inflammatory Lp-PLA2 and IL-6 link e-waste Pb toxicity to cardiovascular risk factors in preschool children. Environ. Pollut. 234, 601–609. https://doi.org/10.1016/j.envpol.2017.11.094.
Marshall, A.T., Betts, S., Kan, E.C., McConnell, R., Lanphear, B.P., Sowell, E.R., 2020. Association of lead-exposure risk and family income with childhood brain outcomes. Nat. Med. 26, 91–97. https://doi.org/10.1038/s41591-019-0713-y.
Mitra, P., Sharma, S., Purohit, P., Sharma, P., 2017. Clinical and molecular aspects of lead toxicity: an update. Crit. Rev. Clin. Lab Sci. 54, 506–528. https://doi.org/10.1080/10408363.2017.1408562.
Mohammadyan, M., Moosazadeh, M., Borji, A., Khanjani, N., Rahimi Moghadam, S., 2019. Exposure to lead and its effect on sleep quality and digestive problems in soldering workers. Environ. Monit. Assess. 191, 184. https://doi.org/10.1007/s10661-019-7298-2.
Morais, M.B., Silva, G., 2019. Environmental enteric dysfunction and growth. J. Pediatr. 95 (Suppl. 1), 85–94. https://doi.org/10.1016/j.jped.2018.11.004.
Nier, A., Huber, Y., Labenz, C., Michel, M., Bergheim, I., Schattenberg, J.M., 2020. Adipokines and endotoxemia correlate with hepatic steatosis in non-alcoholic fatty liver disease (NAFLD). Nutrients 12. https://doi.org/10.3390/nu12030699.
Parsons, C., Roberts, R., Mills, N.T., 2020. Review: inflammation and anxiety-based disorders in children and adolescents - a systematic review and meta-analysis. Child Adolesc Ment Health. https://doi.org/10.1111/camh.12434.
Piotrowska-Tomala, K.K., Bah, M.M., Jankowska, K., Lukasik, K., Warmowski, P., Galvao, A.M., Skarzynski, D.J., 2015. Lipopolysaccharides, cytokines, and nitric oxide affect secretion of prostaglandins and leukotrienes by bovine mammary gland during experimentally induced mastitis in vivo and in vitro. Domest. Anim. Endocrinol. 52, 90–99. https://doi.org/10.1016/j.domaniend.2015.03.001.
Popoola, O.E., Popoola, A.O., Purchase, D., 2019. Levels of awareness and concentrations of heavy metals in the blood of electronic waste scavengers in Nigeria. J Health Pollut 9, 190311. https://doi.org/10.5696/2156-9614-9.21.190311.
Ramírez Ortega, D., González Esquivel, D.F., Blanco Ayala, T., Pineda, B., Gómez Manzo, S., Marcial Quino, J., Carrillo Mora, P., Pérez de la Cruz, V., 2021. Cognitive impairment induced by lead exposure during lifespan: mechanisms of lead neurotoxicity. Toxics 9. https://doi.org/10.3390/toxics9020023.
Ranneh, Y., Akim, A.M., Hamid, H.A., Khazaai, H., Mokhtarrudin, N., Fadel, A., Albujja, M.H.K., 2019. Induction of chronic subclinical systemic inflammation in sprague-dawley rats stimulated by intermittent bolus injection of lipopolysaccharide. Arch. Immunol. Ther. Exp. 67, 385–400. https://doi.org/10.1007/s00005-019-00553-6.
Salvesen, Ø., Reiten, M.R., Heegaard, P.M., Tranulis, M.A., Espenes, A., Skovgaard, K., Ersdal, C., 2016. Activation of innate immune genes in caprine blood leukocytes after systemic endotoxin challenge. BMC Vet. Res. 12, 241. https://doi.org/10.1186/s12917-016-0870-x.
Schoultz, I., Keita Å, V., 2020. The intestinal barrier and current techniques for the assessment of gut permeability. Cells 9. https://doi.org/10.3390/cells9081909.
Solis, C.J., Hamilton, M.K., Caruffo, M., Garcia-Lopez, J.P., Navarrete, P., Guillemin, K., Feijoo, C.G., 2020. Intestinal inflammation induced by soybean meal ingestion increases intestinal permeability and neutrophil turnover independently of microbiota in zebrafish. Front. Immunol. 11, 1330. https://doi.org/10.3389/fimmu.2020.01330.
Song, Q., Li, J., 2014. Environmental effects of heavy metals derived from the e-waste recycling activities in China: a systematic review. Waste Manag. 34, 2587–2594. https://doi.org/10.1016/j.wasman.2014.08.012.
Stevens, B.R., Goel, R., Seungbum, K., Richards, E.M., Holbert, R.C., Pepine, C.J., Raizada, M. K., 2018. Increased human intestinal barrier permeability plasma biomarkers zonulin and FABP2 correlated with plasma LPS and altered gut microbiome in anxiety or depression. Gut 67, 1555–1557. https://doi.org/10.1136/gutjnl-2017-314759.
Sturgeon, J.P., Bourke, C.D., Prendergast, A.J., 2019. Children with noncritical infections have increased intestinal permeability, endotoxemia and altered innate immune responses. Pediatr. Infect. Dis. J. 38, 741–748. https://doi.org/10.1097/INF.0000000000002311.
Talpur, S., Kazi, T.G., Afridi, H.I., Talpur, F.N., Nizamani, S., Lashari, A., Akhtar, A., Khan, M., 2018. Ultrasonically dispersed ionic liquid-based microextraction of lead in biological samples of malnourished children prior to analysis by flame atomic absorption spectrometry. J. AOAC Int. 101, 883–890. https://doi.org/10.5740/jaoacint.17-0269.
Taudorf, S., Krabbe, K.S., Berg, R.M.G., Pedersen, B.K., Moller, K., 2007. Human models of low-grade inflammation: bolus versus continuous infusion of endotoxin. Clin. Vaccine Immunol. 14, 250–255. https://doi.org/10.1128/cvi.00380-06.
Thomas, T.H., Advani, A., 2006. Inflammation in cardiovascular disease and regulation of the actin cytoskeleton in inflammatory cells: the actin cytoskeleton as a target. Cardiovasc. Hematol. Agents Med. Chem. 4, 165–182. https://doi.org/10.2174/187152506776369926.
UNICEF, 2020. The Toxic Truth. https://www.unicef.org/reports/toxic-truth-children-exposure-to-lead-pollution-2020.
van der Kuijp, T.J., Huang, L., Cherry, C.R., 2013. Health hazards of China's lead-acid battery industry: a review of its market drivers, production processes, and health impacts. Environ. Health 12, 61. https://doi.org/10.1186/1476-069x-12-61.
Vancamelbeke, M., Vermeire, S., 2017. The intestinal barrier: a fundamental role in health and disease. Expet Rev. Gastroenterol. Hepatol. 11, 821–834. https://doi.org/10.1080/17474124.2017.1343143.
Wang, K., Qiu, L., Zhu, J., Sun, Q., Qu, W., Yu, Y., Zhao, Z., Yu, Y., Shao, G., 2021. Environmental contaminant BPA causes intestinal damage by disrupting cellular repair and injury homeostasis in vivo and in vitro. Biomed. Pharmacother. 137, 111270 https://doi.org/10.1016/j.biopha.2021.111270.
Wang, L.R., Llorente, C., Hartmann, P., Yang, A.M., Chen, P., Schnabl, B., 2015. Methods to determine intestinal permeability and bacterial translocation during liver disease. J. Immunol. Methods 421, 44–53. https://doi.org/10.1016/j.jim.2014.12.015.
Wang, Q., Xu, X., Zeng, Z., Hylkema, M.N., Cai, Z., Huo, X., 2020a. PAH exposure is associated with enhanced risk for pediatric dyslipidemia through serum SOD reduction. Environ. Int. 145, 106132 https://doi.org/10.1016/j.envint.2020.106132.
Wang, T., Yao, W., Li, J., Shao, Y., He, Q., Xia, J., Huang, F., 2020b. Dietary garcinol supplementation improves diarrhea and intestinal barrier function associated with its modulation of gut microbiota in weaned piglets. J. Anim. Sci. Biotechnol. 11, 12. https://doi.org/10.1186/s40104-020-0426-6.
Weisz, A.J., Manary, M.J., Stephenson, K., Agapova, S., Manary, F.G., Thakwalakwa, C., Shulman, R.J., Manary, M.J., 2012. Abnormal gut integrity is associated with reduced linear growth in rural Malawian children. J. Pediatr. Gastroenterol. Nutr. 55, 747–750. https://doi.org/10.1097/MPG.0b013e3182650a4d.
WHO, 2019. Lead Poisoning and Health. https://www.who.int/news-room/fact-sheets/detail/lead-poisoning-and-health.
Xu, L., Huo, X., Liu, Y., Zhang, Y., Qin, Q., Xu, X., 2020. Hearing loss risk and DNA methylation signatures in preschool children following lead and cadmium exposure from an electronic waste recycling area. Chemosphere 246, 125829. https://doi.org/10.1016/j.chemosphere.2020.125829.
Xu, X., Chen, X., Zhang, J., Guo, P., Fu, T., Dai, Y., Lin, S.L., Huo, X., 2015. Decreased blood hepatitis B surface antibody levels linked to e-waste lead exposure in preschool children. J. Hazard Mater. 298, 122–128. https://doi.org/10.1016/j.jhazmat.2015.05.020.
Yang, H., Huo, X., Yekeen, T.A., Zheng, Q., Zheng, M., Xu, X., 2013. Effects of lead and cadmium exposure from electronic waste on child physical growth. Environ. Sci. Pollut. Res. Int. 20, 4441–4447. https://doi.org/10.1007/s11356-012-1366-2.
Yekeen, T.A., Xu, X., Zhang, Y., Wu, Y., Kim, S., Reponen, T., Dietrich, K.N., Ho, S.M., Chen, A., Huo, X., 2016. Assessment of health risk of trace metal pollution in surface soil and road dust from e-waste recycling area in China. Environ. Sci. Pollut. Res. Int. 23, 17511–17524. https://doi.org/10.1007/s11356-016-6896-6.
Yu, L., Yu, Y., Yin, R., Duan, H., Qu, D., Tian, F., Narbad, A., Chen, W., Zhai, Q., 2021. Dose-dependent effects of lead induced gut injuries: an in vitro and in vivo study. Chemosphere 266, 129130. https://doi.org/10.1016/j.chemosphere.2020.129130.
Zeng, X., Xu, X., Qin, Q., Ye, K., Wu, W., Huo, X., 2019. Heavy metal exposure has adverse effects on the growth and development of preschool children. Environ. Geochem. Health 41, 309–321. https://doi.org/10.1007/s10653-018-0114-z.
Zeng, Z., Huo, X., Wang, Q., Wang, C., Hylkema, M.N., Xu, X., 2020. PM(2.5)-bound PAHs exposure linked with low plasma insulin-like growth factor 1 levels and reduced child height. Environ. Int. 138, 105660 https://doi.org/10.1016/j.envint.2020.105660.
Zhai, Q., Qu, D., Feng, S., Yu, Y., Yu, L., Tian, F., Zhao, J., Zhang, H., Chen, W., 2019. Oral supplementation of lead-intolerant intestinal microbes protects against lead (Pb) toxicity in mice. Front. Microbiol. 10, 3161 https://doi.org/10.3389/fmicb.2019.03161.
Zhang, S., Huo, X., Li, M., Hou, R., Cong, X., Xu, X., 2020. Oral antimicrobial activity weakened in children with electronic waste lead exposure. Environ. Sci. Pollut. Res. Int. https://doi.org/10.1007/s11356-020-08037-3.
Zhang, Y., O'Connor, D., Xu, W., Hou, D., 2019. Blood lead levels among Chinese children: the shifting influence of industry, traffic, and e-waste over three decades. Environ. Int. 135, 105379 https://doi.org/10.1016/j.envint.2019.105379.
Zhao, L., Zheng, Y.G., Feng, Y.H., Li, M.Y., Wang, G.Q., Ma, Y.F., 2020. Toxic effects of waterborne lead (Pb) on bioaccumulation, serum biochemistry, oxidative stress and heat shock protein-related genes expression in Channa argus. Chemosphere 261, 127714. https://doi.org/10.1016/j.chemosphere.2020.127714.
Zhao, W., Ding, L., Gu, X., Luo, J., Liu, Y., Guo, L., Shi, Y., Huang, T., Cheng, S., 2015. Levels and ecological risk assessment of metals in soils from a typical e-waste recycling region in southeast China. Ecotoxicology 24, 1947–1960. https://doi.org/10.1007/s10646-015-1532-7.
Zheng, R., Wang, P., Cao, B., Wu, M., Li, X., Wang, H., Chai, L., 2021. Intestinal response characteristic and potential microbial dysbiosis in digestive tract of Bufo gargarizans after exposure to cadmium and lead, alone or combined. Chemosphere 271, 129511. https://doi.org/10.1016/j.chemosphere.2020.129511.
Zheng, X., Huo, X., Zhang, Y., Wang, Q., Zhang, Y., Xu, X., 2019. Cardiovascular endothelial inflammation by chronic coexposure to lead (Pb) and polycyclic aromatic hydrocarbons from preschool children in an e-waste recycling area. Environ. Pollut. 246, 587–596. https://doi.org/10.1016/j.envpol.2018.12.055.