The evaluation of liver dysfunction and oxidative stress due to urban environmental pollution in Mexican population related to Madin Dam, State of Mexico: a pilot study

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Abstract

One of the most important causes of disease and premature death in the world is environmental pollution. The presence of pollutants in both water and air contributes to the deterioration of the health of human populations. The Mexico City Metropolitan Area is one of the most populous and affected by air pollution worldwide; in addition, in recent years there has been a growing demand for water, so urban reservoirs such as the Madin dam are vital to meet the demand. However, this reservoir is highly polluted due to the urban settlements around it. Therefore, the aim of the present study was to evaluate oxidative stress in clinically healthy subjects by means of the degree of lipoperoxidation, as well as the modification of serum enzyme levels, such as alanine aminotransferase, aspartate aminotransferase, alkaline phosphatase and lactate dehydrogenase associated with air and drinking water pollutants from three zones of the Mexico City Metropolitan Area, two of them related to Madin Dam. This descriptive cross-sectional study was conducted between March 2019 and September 2021 in 142 healthy participants (age range 18–65 years). Healthy subjects were confirmed by their medical history. The results showed that chronic exposure to air (SO2) and water pollutants (Al and Fe) was significantly associated with elevated levels of lipoperoxidation. There was evidence that contamination from the Madín dam can generate oxidative stress and affect the health status of people who receive water from this reservoir or who consume fish that inhabit it.

Keywords Air pollution · Water pollution · Lipoperoxidation · Liver enzymes · Human health effects · Metals

Introduction

One of the most important causes of disease and premature death in the world is environmental pollution. According Landrigan et al. (2018), it is responsible for approximately 9 million deaths per year, 16% of all deaths worldwide, three times more deaths than those related to AIDS, tuberculosis, and malaria combined. In Mexico, specifically air pollution has been an important issue for the country’s administration, since in addition to having contributed to climate change (Silva Rodríguez de San Miguel 2019), it harms the health of Mexicans through various ways (Calderón-Garcidueñas
et al. 2015; Mamkhezri et al. 2020). The pollution problem can be explained by excessive industrialization in the largest and most densely populated areas of the country (Paquette 2015) and mobile sources (Camacho-García and Flamand 2007). On the other hand, water pollution also produces negative impacts on ecosystems and human health. Palacios Nava et al., 2018, mentions that in the country, approximately 73% of water bodies are polluted; 80% comes from discharges from urban sources and 85% from industrial discharges that do not undergo prior treatment.

The Metropolitan Area of Mexico City is one of the most affected by pollution, it was declared the most polluted city in the world in 1991 and although, in recent decades, the authorities have developed strategies for the reduction of air pollution, the current levels of particulate matter (PM) and ozone are still above what is established by the World Health Organization (Calderón-Garcidueñas et al. 2015; García-Franco 2020). Bell et al. (2006) mentions that there are several studies that have identified the relationship between air pollution and health, for example, particulate matter (PM) has been associated with mortality, including infant mortality, and with heart rate variability in the elderly. On the other hand, ozone levels have been associated with mortality, increased school absenteeism in children, emergency consultations for asthma and other respiratory problems. Likewise, poor management of urban, industrial, and agricultural wastewater leads to the water consumed by millions of people being contaminated with chemical substances and/or pathogenic microorganisms, which can cause severe damage to their health (WHO 2019).

Air pollution is a harmful phenomenon for the environment and human health; it is formed by a mixture of gases and particles in harmful quantities that are released into the atmosphere mainly due to activities of natural or anthropogenic origin (Dodman et al. 2010). Within the gaseous pollutants are inorganic compounds, for example, nitrogen dioxide (NO₂), sulfur dioxide (SO₂), ozone (O₃), carbon monoxide (CO), and carbon dioxide (CO₂), there are also metals such as lead (Pb) and chromium (Cr), as well as volatile organic compounds such as polycyclic aromatic hydrocarbons (PAHs) (Tiotiu et al. 2020). On the other hand, the smallest particulate matter (PM) of a specific size is a complex mixture of dust, soot, smoke and liquid droplets. The size of the particles is very important, as it determines their impact on the respiratory tract. Depending on their diameter, they are divided into three categories: PM₁₀ coarse (2.5 to 10 µm), PM₂.₅ fine (0.1 to 2.5 µm), and PM₀.₁ ultrafine (less than 0.1 µm). Coarse particles are deposited mainly in the nasopharynx or primary bronchi; fine particles may be deposited in the alveoli and terminal bronchioles, and ultrafine particles cross cell membranes and interact directly with cellular structures (Kelly and Fussell 2012). One of the biological mechanisms that may explain the association between air pollution and respiratory disease is oxidative stress. Pollutants can act directly by forming reactive oxygen species (ROS) that diffuse from the airway surface, or indirectly by inducing inflammation (Kim et al. 2019). On the other hand, the liver being an important organ in the metabolism and biotransformation of inhaled air pollutants, liver function could be directly or indirectly affected by them or their metabolites, causing damage to major liver enzymes such as alanine aminotransferase (ALT) and aspartate aminotransferase (AST) (Qiu et al. 2021). In a study by Kim et al. (2015), they found that short-term exposure to air pollutants such as PM₂.₅, NO₂ and O₃ is associated with increased liver enzyme levels in elderly adults.

In relation to water pollution, toxic substances present in water bodies, such as pesticides, emerging pollutants, metals, and POPs, can cause countless damages to human health and affect the balance of ecosystems; they enter through several pathways, including effluents from wastewater treatment plants, untreated wastewater, urban runoff, and leaching from agricultural land and open dumps (Inostroza et al. 2016). There is a strong relationship between contaminants present in water and human health problems, due to the continuous interaction with freshwater supplies impacted by contamination. The adverse effects caused by exposure to the toxics described above are the following: immunological, neurotoxic, endocrine, reproductive, and genetic effects (Larrea Poma 2013). According to the World Health Organization, water pollution contributes to 70% of different diseases and 20% to cancer worldwide (Khan et al. 2021). One of the groups of contaminants have been detected in drinking water are metals, which generally exceed the limits recommended by regulatory systems worldwide. There are a number of adverse effects on human metabolism that has resulted from exposure to drinking water contaminated with metals such as arsenic (As), lead (Pb), nickel (Ni), cadmium (Cd), and mercury (Hg), and the general mechanism of toxicity is through ROS production, oxidative damage and subsequent health effects (Fu and Xi 2020). Metal toxicity is also associated with cancer, dermatitis, cardiovascular disease, neuropathy, diabetes mellitus, renal failure, and liver dysfunction (Islam et al. 2011).

Despite the importance of reservoirs as providers of drinking water for the population, many of them are deteriorated in Mexico, such is the case of the Madín dam (Pérez-Coyotl et al. 2019). This reservoir is located between the municipalities of Atizapán de Zaragoza and Naucalpan de Juárez, its geographical coordinates are 19°31'37" North latitude and 99°15'33" West longitude. The mainstream that feeds the dam is the Tlahnepantla River, which is one of the most important in the Valley of Mexico. Along its course, it receives a series of streams, including the El Muerto and El Polvorín. This reservoir was built to control the flow of the Tlahnepantla River and
convert part of the volume of stored water into drinking water. The reservoir has a maximum capacity of 25 million m³, but generally stores only 13 million m³, in order to anticipate higher flows; a small part of this volume, approximately 600 L/s, is pumped to the Madín water treatment plant for distribution through the municipal water supply. This plant is operated by the National Water Commission (CONAGUA) and uses an aluminum-based treatment to coagulate organic matter. It supplies 2% of the drinking water to some neighborhoods in the municipalities of Naucalpan de Juárez and Atizapán de Zaragoza; the remaining neighborhoods receive drinking water from the Cutzamala system or from wells (Galar-Martínez et al. 2010). Various contaminants have been detected in this water body, such as metals, emerging contaminants, pesticides, and persistent organic pollutants (Pérez-Coyotl et al. 2019). In addition, contaminants in Madín Dam water have been found to induce oxidative stress, genotoxicity, cytotoxicity, and embryotoxicity in aquatic organisms (Galar-Martínez et al. 2010; González-González et al. 2014; Morachis-Valdez et al. 2015; Pérez-Coyotl et al. 2017).

The presence of pollutants in both water and air contribute to the deterioration of the health of human populations; due to this, the objective of the present study was to evaluate clinically healthy subjects the oxidative stress through the degree of lipoperoxidation (LPX), as well as the modification of serum enzyme levels, such as alanine aminotransferase (ALT), aspartate aminotransferase (AST), alkaline phosphatase (ALP), and lactate dehydrogenase (LDH) associated with air and drinking water pollutants in three zones of the Metropolitan Area of Mexico City, two of them related to the Madin Dam.

Materials and methods

Study area and study population

An exploratory descriptive cross-sectional pilot study was carried out between March 2019 and September 2021, in a Mexican population of 142 healthy participants (age range 18–65 years) from the Metropolitan Area of Mexico City. Participants were enrolled in a descriptive study conducted at the Aquatic Toxicology Laboratory of the ENCB, IPN. More than 200 volunteers were invited, among them, 142 subjects participated in this study.

The following inclusion criteria were considered for the study: subjects without diseases, considered apparently healthy as confirmed by their clinical history, as well as by their blood chemistry and blood biometry, with an age range of 18 to 65 years and with 10 years or more of residence in the sites of interest for the present study; who signed the informed consent form.

Exclusion criteria included the following: subjects with chronic degenerative diseases, with a history of having presented SARS-CoV-2 or testing positive for SARS-CoV-2 antigens, chronic smokers and alcoholics, those who use psychoactive substances or chronic use of medications, pregnant women, people who donated or lost blood in a volume greater than or equal to 450 mL during the 2 months prior to sample collection, and subjects who were seriously ill within 90 days prior to sample collection. Those who worked in the chemical industry or were occupationally exposed to chemicals were also excluded.

The study protocol was approved by the Research Ethics Committee of the National School of Biological Sciences, IPN with registration number CEI-ENCB-SH-003–2018. All steps in the methods section were carried out following the relevant guidelines and regulations (SEMARNAT 2002; Secretaría de Salud 2012a, b).

Three study groups were formed, corresponding to zone 1 identified as Nuevo Madín (Atizapán de Zaragoza); this site receives drinking water from wells located in this area and is adjacent to the water body. Group 2 corresponds to the municipality of Naucalpan de Juárez. Some neighborhoods in this area are located near the Madín dam and receive part of their drinking water from the Madín dam and the Cutzamala system. Group 3 is an urban group that works and resides in Gustavo A. Madero (GAM), Mexico City (reference group). This area receives water from the Cutzamala system, as does group 2, and has similar levels of environmental contaminants.

Body weight and height were measured to calculate the body mass index (BMI), which was calculated as weight in kilograms divided by height in meters squared. A questionnaire was applied to learn about the habits of the research subjects, for example: smoking, use of medications, and physical activity, as well as if they consume organisms that inhabit the dam, for example, common carp, or farm animals that consume water directly from the dam. In addition, to verify that the research subjects were not infected during sampling, they were first tested with a rapid test for SARS-CoV-2 antigens, Roche Diagnostics brand. Likewise, a follow-up of COVID-19 was made in the patient’s clinical history, where they were asked if they had suffered from it previously. It should be noted that the participants were quarantined and the sample was taken at the domicile of each one of them.

Blood sample collection

The participants were asked to have a minimum fasting time of eight hours, then a blood sample was taken by venipuncture. The samples were labeled with an alphanumeric code of one letter and 3 numbers and were transferred to the Toxicology Laboratory of the National School of Biological Sciences for analysis.
Liver enzyme activity

As hepatic biochemical biomarkers, the levels of alanine aminotransferase (ALT), aspartate aminotransferase (AST), alkaline phosphatase (ALP), and lactate dehydrogenase (LDH) were used, these enzymes are commonly used as liver function tests (Kim et al. 2019). The collected samples were centrifuged (2500×g, 15 min) and serum was separated, stored at 2–8 °C, and analyzed within the first 24 h. Biochemical analysis of serum samples from all subjects was performed on a Dimension EXL 200 automatic analyzer (Siemens). The calibration was performed by means of simple linear regressions of three points on Siemens quality controls; subsequently, the equation of the line was obtained, and the reading data were interpolated on the curve; this was done through the equipment’s software. The following levels were considered to define enzyme alteration: ALT > 78 U/L, AST > 37 U/L, ALP > 116 U/L, LDH > 190 U/L.

Determination of lipoperoxidation levels

For the evaluation of oxidative stress, the degree of lipoperoxidation determined by the reaction between N-methyl-2-phenylindole with malondialdehyde (MDA) formed by the peroxidation of cell lipids was used as a biomarker (Estebauer and Cheeseman 1990). Plasma was obtained from the blood samples by centrifugation (2500×g, 15 min) and stored at −70 °C until analysis. To 200 μL of sample, 650 μL of 10.3 mM N-methyl-2-phenylindole solution was added and shaken for 4 s. A total of 150 μL of 37% HCl were added, mixed, and incubated at 45 °C for 60 min. After incubation, it was allowed to cool and centrifuged at 846×g for 15 min and absorbance was determined at 586 nm. The results were expressed as μM of MDA/L. The cut-off value for LPX was defined based on the 90th percentile of young healthy Mexican subjects which is 0.320 µmol/L (Sánchez-Rodríguez et al. 2004).

Environmental contaminants

To evaluate SO₂, NO₂, O₃, CO, and PM₁₀ pollutants in the air, information from the Automatic Atmospheric Monitoring Network (RAMA) of the Metropolitan Zone and the Valley of Mexico (ZMVM) was used. This network operates 29 stations distributed in the ZMVM and the information was taken for the urban reference group of 2 monitoring stations, the GAM station that only measures O₃ and PM₂.₅, and the Camarones (CAM) station that measures the rest of the pollutants (CO, SO₂, NO₂, NO, and PM₁₀). These are located at a distance from the study site of 5.55 and 4.45 km, respectively. In the case of the Nuevo Madín group, the closest station to the site of interest was Atizapán (ATI), located 6 km away, and in the Naucalpan de Juárez group, information was collected from the FES Acatlán (FAC) station, located 4 km away (see Supplementary material, Fig. 1S), for the period January 2019 to September 2021.

Quantification of metals in drinking water and in tissue of fish obtained from Madín dam

The quantification of iron (Fe), aluminum (Al), lead (Pb), cadmium (Cd), and chromium (Cr) was performed according to the guidelines of the Mexican Standard NMX-AA-051-SCFI-2016 at the Central Instrumentation Laboratory of the National School of Biological Sciences (COTEMARNAT 2016). Drinking water samples were directly collected from the tap connected to the municipal water supply in sterile Whirl–Pak bags, and a volume of 500 mL was collected. Three samples were taken from the different study areas and were placed in a cooler to preserve them at a temperature of 4 °C ± 2 °C for subsequent transfer to the Laboratory of Aquatic Toxicology. Samples were adjusted to a pH of 2 with reagent grade concentrated HNO₃ (Tecsiquim). Samples were filtered through a 0.45-µm pore membrane. For digestion, an extraction hood, a heating rack and a 150 mL beaker were placed in an extraction hood, the membrane was transferred and 4 mL of concentrated HNO₃ was added. One filter was included for blank measurement and one for the digestion standard. It was covered with a watch glass and heated until complete digestion of the membrane was achieved. It was heated almost to dryness avoiding the sample to reach its boiling point. The watch glass was cooled and washed with type I water and 3 mL of concentrated HNO₃ was added, covered, and continued heating until a crystalline appearance was obtained. The sample was then evaporated to near dryness (approximately 2 mL), cooled, and washed with type I water, added 10 mL of HCl (1:1) and 15 mL of type I water. It was heated for 15 min to dissolve precipitates. Finally, it was cooled and the walls of the beaker and watch glass were washed with type I water and filtered to remove insoluble material that could clog the nebulizer. The volume was adjusted to 50 mL.

In addition, fish that inhabit the Madín dam and have been consumed by the inhabitants of the Nuevo Madín neighborhood were collected. The organisms were euthanized by immersion in water with clove essence oil. They were frozen with liquid nitrogen and 5 specimens in juvenile stage and 5 in adult stage were macerated. 0.5 g of tissue was taken in duplicate and subjected to acid digestion with concentrated HNO₃. Samples were placed in plastic tubes along with a reagent blank, and 2 mL of concentrated reagent grade HNO₃ (Tecsiquim) was added and placed in an autoclave at 120 °C and 15-lb pressure for 1 h. The samples were cooled to room temperature and filtered with a Whatman membrane of 0.2-µm pore size in a Falcon tube. Finally, they were diluted to 50 mL with type I water.

Subsequently, the metal content was quantified by atomic absorption spectrometry (Agilent Technologies 240 FS AA) by flame and graphite furnace (Agilent Technologies GTA 120).
High Purity Standards ICP-200–7-6-A-500 (200 mg/L) and CCV1 1,722,128 (20 mg/L) of 99.5–99.9% purity were used. For Fe quantification, the method used was by flame. The wavelength was 248.3 nm and the lamp current was 5.0 mA. Seven mL/min of sample was aspirated for 3 to 4 min. A calibration curve of 5 concentrations and a reagent blank was performed, the range of the curve was from 0.16 to 1.0 mg/L.

Regarding the quantification of Al, Pb, Cr, and Cd, the method used was by graphite furnace. The wavelength used was 309.3 nm, lamp current 10 mA; 217.0 nm, lamp current 6 mA; 357.9 nm, lamp current 7.0 mA and 228.8 nm, lamp current 4 mA, respectively. The amount of sample injected was 20 µL. A calibration curve of 5 concentrations and a reagent blank was performed, the range of the curve was from 2.0 to 10.0 µg/L for Al, from 0.33 to 2 µg/L for Pb and Cr, and from 0.16 to 1.0 µg/L for Cd.

Statistical analysis

Data were processed using SPSS 18.0, SigmaPlot 12.3 and RStudio statistical software. Descriptive statistics are presented as means ± standard deviation (SD) and median with their respective interquantile recollection (IQR). Results of liver biomarkers and oxidative stress were analyzed by Kruskal–Wallis analysis of variance (ANOVA) followed by a post-hoc Dunn test, with the exception of LDH levels (ANOVA, post-hoc Student–Newman–Keuls), significant difference was considered when p ≤ 0.05. To establish normality and homoscedasticity of the data, a Shapiro–Wilk test and Bartlett’s test were applied.

An odds ratio (OR) from logistic regression analysis with 95% confidence interval (CI) was established for significant liver biomarkers and LPx levels; these variables were reclassified to a dichotomous variable (altered, normal). Altered subjects were identified when liver enzyme or LPx levels were found to be outside the cutoff values. Additionally, an association analysis between the frequency of subjects with altered or normal enzymes and the consumption of carp and farm animals drinking water from the dam was performed for the inhabitants of Nuevo Madín, establishing the OR with its 95% confidence interval.

Finally, a principal component analysis was performed to establish the correlation between the biomarkers evaluated and the exposure levels of environmental and water contaminants found.

Results

Of the 142 participants, 68.3% were women and the mean age was 37.4 years. None of them claimed to be exposed to chemical agents at work: 26.8% are students, 16.9% are housewives, 29.6% work in offices, 13.4% are engaged in teaching, 2.1% are laborers unrelated to the chemical industry, 8.5% are tradesmen, and 2.8% are unemployed. Among the participants, 57.0% had not smoked before, 57.7% drank alcohol frequently, and 47.9% exercised (Table 1). The mean BMI of the study participants was 26.62 kg/m².

Figure 1 shows the LPX and liver enzyme levels of each study group, where it was observed that for AST and ALT enzymes no significant differences were found between the groups. In the case of ALP and LDH there was a significant difference between the values presented in the research subjects from Nuevo Madín and those from Naucalpan and Gustavo A. Madero (p ≤ 0.05). The ALP of the inhabitants of Nuevo Madín was around 30% with respect to the other two study areas and 20% for LDH. In the LPX levels, a significant difference was shown between the different study groups (p < 0.01), being much higher in the subjects from Naucalpan (288%), followed by Nuevo Madín (53%) compared to the subjects from Gustavo A. Madero (reference group).

To study the association between the different areas studied, and the biomarkers that presented significant differences, a logistic regression model adjusted for the characteristics that were significant (sex, age, tobacco, alcohol, and physical activity) was performed (Table 2). In the logistic regression analysis, LDH showed a positive association in subjects from Nuevo Madín considering the group from Gustavo A. Madero as the reference group. In the case of LPX, there was also a significant association with the groups of interest (Nuevo Madín and Naucalpan).

With respect to the Nuevo Madín group, 57% indicated that they consume carp that are grown in the reservoir, as well as 6% provide water directly from the reservoir to the farm animals they consume. Table 3 shows the association between carp consumption and animals that drink water from Madín Dam, where a significant association (p < 0.01) is observed between the altered values of ALP, LDH, and LPX with carp consumption.

Figure 2 shows the mean trends of the five air pollutants in the regions of interest from January 2019 to September 2021. The median concentrations with their respective IQR of PM10, NO2, SO2, O3, and CO were 48 (33) g/m³, 19 (21) ppb, 2 (3) ppb, 38 (60) ppb, and 0.3 (0.4) ppm for Gustavo A. Madero; 36 (30) g/m³, 16 (16) ppb, 2 (3) ppb, 30 (44) ppb, and 0.3 (0.34) ppm for Atizapán de Zaragoza (Nuevo Madín) and 33 (31) g/m³, 21 (16) ppb, 3 (4) ppb, 33 (51) ppb, and 0.4 (0.41) ppm for Naucalpan de Juárez, respectively. This figure shows that there is a cyclical peak in pollutant levels, with pollutant levels increasing during the first months of the year and decreasing during the summer. In addition, CO levels decreased significantly in 2021 compared to 2020 for the Gustavo A. Madero area. In the case of the Madín dam area (Nuevo Madín, Atizapán), a significant reduction was observed for SO2 and O3.
Gustavo A. Madero and Naucalpan de Juárez areas have higher levels than those observed in the Madín dam area (Nuevo Madín, Atizapán).

Table 4 shows the concentrations of metals found in the water samples from the study areas. It can be seen that the highest levels of metals are found in the drinking water intakes of Naucalpan de Juárez for Fe, Al, and total Cr. The metal with the highest frequency detected is aluminum; however, it is within the permitted limits of metals according to PROY-NOM-127-SSA1-2017 (Secretaría de Salud 2019). In the case of lead, the levels observed in Naucalpan de Juarez and Gustavo A. Madero are similar, however, for Naucalpan de Juarez, the concentration of lead in one of the samples exceeds the maximum limit. For Fe, the values presented in a sample from Naucalpan de Juarez exceed the limits allowed by Mexican regulations. For Nuevo Madín, the levels and frequency of metals quantified were lower than in the rest of the areas studied. Table 5 shows the content of metals in µg per g of total tissue weight, where it is observed that the levels of aluminum and iron are much higher compared to the rest of the metals. The PCA is presented in Fig. 3, where it is observed that for LPX levels there is a correlation with SO₂, Al, Fe, and Cr, with Naucalpan de Juárez being the zone with the highest levels compared to Gustavo A. Madero and Nuevo Madín. In the case of liver enzymes, a correlation was observed between them, with the highest levels in the inhabitants of Nuevo Madín, particularly in ALP and LDH, with no relation to the levels of environmental pollutants and metals in the drinking water.

**Discussion**

A study was conducted to evaluate the alteration of the main liver enzymes (ALT, AST, ALP, and LDH) and the level of LPX associated with the levels of ambient air and
drinking water pollution in the Mexican population related to the Madín dam, State of Mexico. Among the results, it was observed that the areas of Gustavo A. Madero and Naucalpan have higher levels of pollutants (PM$_{10}$, NO$_2$, SO$_2$, O$_3$, and CO) than those observed in the area of Atizapan de Zaragoza (Nuevo Madín). Regarding the drinking water samples, it was observed that the highest levels of metals are found in the drinking water intakes of Naucalpan for Fe, Al, and total Cr. On the other hand, it was obtained that the ALP and LDH enzymes presented significant differences in the research subjects from Nuevo Madín with respect to the subjects from Naucalpan and Gustavo A. Madero. LPX levels showed a significant difference between the different study groups, being much higher in the subjects from Naucalpan. According to the Ministry of the Environment (SEDEMA 2022), the sector that contributes with the highest emission of air pollutants (ozone precursors and particulate matter) is transportation, contributing 40% of PM$_{10}$, 43% of PM$_{2.5}$, 86% of NO$_2$, and 22% of CO. Other important sectors related

![Fig. 1](image_url)

**Table 2** Logistic regression results* for ALP, LDH, and LPX levels by zone of study

| Group            | ALP Estimate | 95% CI         | p-value | LDH Estimate | 95% CI         | p-value | LPX Estimate | 95% CI         | p-value |
|------------------|--------------|----------------|---------|--------------|----------------|---------|--------------|----------------|---------|
| Gustavo A. Madero| 1.00         | –              | –       | 1.0          | –              | –       | 1.0          | –              | –       |
| Nuevo Madín      | 1.93         | 0.29, 12.59    | 0.49    | 21.13        | 1.67, 348.3    | 1.01    | 13.61        | 3.62, 51.30    | 0.00    |
| Naucalpan        | 1.07         | 0.07, 15.26    | 0.95    | 0.13         | 0.00, –        | 1.00    | 244.15       | 50.21, 118,721 | 0.00    |

*Model adjusted for age, sex, smoking, amount of exercise, and alcohol consumption
to the above are industry, urban development, and livestock and agricultural activities. Local pollutant sources in Mexico City contribute about one third of the emissions generated in the Metropolitan Zone of the Valley of Mexico.

With respect to surface water pollution in Mexico, the main sources have been identified as those related to agricultural practices, where agrochemicals are used that can be washed into water bodies by rain and soil erosion; as well as municipal wastewater, which is created from untreated domestic and public waste, in addition to industrial discharges, which are generated by the activities of extraction and transformation of natural resources that at the end of the whole process are used as consumer goods. These discharges may contain synthetic organic substances or metals that do not degrade easily under natural conditions (Ibarrarán Viniegra et al. 2017), leading to the contamination of various water bodies, including aquifers. This could explain the presence of metals in the drinking water received by the urban reference group and the Nuevo Madín group. In particular, the present research focused on metals because the presence of these contaminants in reservoirs that provide water to populations is often a problem, such is the case of the Madín Dam, since water potabilization processes do not eliminate them, leading to a risk to human health (Canpolat et al. 2020).

There are different studies that have shown that contaminants are capable of causing liver dysfunction due to the central role played by this organ in the metabolism of xenobiotics (Lawton et al. 1985; Emmett et al. 1988; Kumar et al. 2014). Classical biochemical tests commonly used in clinical medicine involve the measurement of a number of serum enzymes (ALP, ALT, AST, and LDH) that together indicate the pathophysiological status of the liver (Kumar et al. 2014). We observed that ALP activity increased significantly in the New Madin group with respect to the other groups studied, as did LDH; however, the other two enzymes

| Table 3 | Association between hepatic and oxidative stress biomarkers with the consumption of carp and animals living and drinking water from the reservoir of the settlers of the Nuevo Madín area |
|---------|----------------------------------------------------------------------------------------------------------|
|         | Consumption of carp from the PM | Consumption of animals drinking water from the PM |
|         | OR (95% CI) p* | OR (95% CI) p* |
| AST     | 1.29 (0.14–12.03) 0.82 | 1.03 (1.00–1.06) 0.71 |
| ALT     | 1.56 (0.32–7.63) 0.58 | 1.05 (1.01–1.09) 0.64 |
| ALP     | 2.60 (1.01–6.71) 0.04 | 1.12 (1.06–1.19) 0.48 |
| LDH     | 5.46 (1.87–15.95) <0.01 | 1.10 (1.04–1.15) 0.53 |
| LPX     | 1.97 (1.10–3.54) <0.01 | 0.55 (0.11–2.76) 0.34 |

*Chi-square test, p < 0.05
(AST and ALT) evaluated did not show significant changes between the groups. ALP is mainly produced in the liver and is responsible for the dephosphorylation process. It is distributed throughout the body, being in higher concentration in the kidney, bones and liver, when elevated plasma levels are present it is usually indicative of bile duct obstruction or other liver diseases (Kumar et al. 2014). In recent years, this enzyme has also been associated with cardiovascular disease, diabetes, and metabolic syndrome (Nannipieri et al. 2005; Haarhaus et al. 2017).

In addition, following exposure to environmental pollutants, such as persistent organic pollutants (POPs), and arsenic, the levels of this enzyme are increased in plasma (Islam et al. 2011; Kumar et al. 2014). In the case of the Nuevo Madín group, it was found that 17% of the subjects presented abnormal ALP values; however, when estimating the risk of presenting alterations in the normal levels of the enzyme considering as a control group the subjects coming from the area identified as Gustavo A. Madero, no increase in the risk was observed. On the other hand, in LDH there was an increase in risk, 21.13 times more in comparison with the subjects from the Gustavo A. Madero group, since this group presented higher values than the rest of the areas studied. LDH is an enzyme found in a wide variety of tissues; an increase in blood levels of this enzyme is usually indicative of tissue lesions in the heart, kidney, or liver, as well as in a wide variety of cancers (Wu et al. 2021). In addition, it has been proposed as a useful biomarker for early prognosis of organ or tissue damage in people chronically exposed to metals in water and environmental pollutants (Karim et al. 2010; Saeed 2017; Pejhan et al. 2019). On the other hand, there are also reports of changes in this type of enzymatic biomarkers in populations exposed to various environmental pollutants through air, water, or food consumption. Multiple chemical substances have been identified, including metals, hydrocarbons, polychlorinated biphenyls (PCBs), dioxins, particulate matter, and gases such as NOx (Yorita Christensen et al. 2013).

Although the underlying physiological mechanisms that explain the positive association between elevated level of enzymes such as ALP and LDH with various pollutants remain unclear, oxidative stress offers a plausible process for understanding this relationship. Oxidative stress is caused by a disturbance in the balance between ROS production and antioxidant defenses (Sies 2015). There is evidence that various pathophysiological processes, habits and lifestyle can trigger an increase in ROS, but also environmental pollutants can cause oxidative deterioration in organisms (Aslan et al. 1997; Carraro et al. 2018; Rafiee et al. 2021). The elevation of ROS production leads to an increase in the level of lipid peroxidation; during this process, polyunsaturated fatty acids with double bonds react with ROS, particularly

### Table 4 Quantification of metals present in the drinking water supplied in the study areas

| Groups | N. sample | Metals (mg/L) | Iron | Aluminum | Lead | Cadmium | Chromium (total) |
|--------|-----------|---------------|------|----------|------|---------|-----------------|
| Nuevo Madín (Wells) | 1 | <0.16 | 0.02 | 0.002 | 0.0002 | 0.002 |
| | 2 | <0.16 | <0.02 | <0.002 | <0.0002 | 0.002 |
| | 3 | 0.16 | <0.02 | <0.002 | <0.0002 | 0.002 |
| Naucaulan (Madín resevoir + Cutzamala system) | 1 | 0.68 | 0.13 | 0.010 | 0.002 | 0.004 |
| | 2 | <0.16 | 0.19 | <0.002 | <0.0002 | 0.002 |
| | 3 | <0.16 | 0.18 | <0.002 | <0.0002 | 0.003 |
| Gustavo A. Madero (Cutzamala system) | 1 | 0.112 | 0.02 | 0.011 | 0.002 | <0.002 |
| | 2 | <0.16 | 0.06 | 0.006 | 0.0002 | 0.002 |
| | 3 | <0.16 | <0.02 | 0.008 | 0.002 | <0.002 |
| Maximum permissible limit (PROY-NOM-127-SSA1-2017) | | | 0.30 | 0.20 | 0.01 | 0.005 | 0.05 |

### Table 5 Quantification of metals in fish tissues from Madin reservoir

| Stage | Metals (µg/g) |
|-------|---------------|
|       | Iron | Aluminum | Lead | Cadmium | Chromium (total) |
| Juvenile | n 2 | 2.1000 | 8.1000 | 0.0090 | 0.0021 | 0.0130 |
| Mean | 2.0000 | 8.1000 | 0.0090 | 0.0021 | 0.0130 |
| SD | 0.1000 | 0.8000 | 0.0014 | 0.0001 | 0.0099 |
| Adult | n 2 | 1.2000 | 8.1000 | 0.0080 | 0.0012 | 0.0050 |
| Mean | 1.2000 | 8.1000 | 0.0080 | 0.0012 | 0.0050 |
| SD | 0.0000 | 0.1000 | 0.0000 | 0.0004 | 0.0001 |

n replica number, SD standard deviation
the hydroxyl radical (HO) and the reactive nitrogen species peroxynitrite (ONOO), through a chain reaction mechanism. This allows the formation of hydroperoxides that degrade to low molecular weight products, including MDA (Ayala et al. 2014; Morachis-Valdez et al. 2015). In the present study, it could be seen that MDA levels in the subjects from Nuevo Madín and Naucalpan de Juárez presented much higher levels than in the group from Gustavo A. Madero, the risk was significant, particularly for the people from Naucalpan de Juárez. MDA accumulation has been extensively investigated and implicated in many toxic tissue injuries and pathological processes (Ayala et al. 2014). This biomarker presented a correlation between pollutants found in air and water, in particular SO₂ and the metals Fe, Cr, and Al. Ambient air is a complex of various individual pollutants that are either free radicals, such as NO₂, or have the ability to drive the free radical reaction, such as O₃ and particulate matter (Jung et al. 2013). In the case of SO₂, its inhalation reduces the activity of antioxidant enzymes and leads to lipid peroxidation that can damage the brain (Meng 2003). SO₂ pollution is caused by the combustion of fossil fuels, including sulfur, and by pollutants resulting from warming and released from smokestacks (Saygin et al. 2017). The three groups studied are located in the northeastern region of the MCMA, which is characterized by regular SO₂ levels (26 to 110 ppb), with a tendency to present a good category (> 25 ppb), so it has been identified as an area with higher levels of this type of pollutants than in the rest of the Valley of Mexico (Ramos 2019). It is known that exposure to SO₂ is associated with a higher prevalence of respiratory symptoms, such as wheezing and shortness of breath, so it affects the quality of life of populations, mainly in children.

Fig. 3 Principal component analysis between biomarkers evaluated with air pollutants (a) and metals in drinking water (b) present in the three study areas
(Nascimento et al. 2020). While there are a number of pollutants present in daily life to which people are exposed, the conditions under which the study was conducted (during the COVID-19 pandemic), contributed to participating subjects remaining in their homes 24 h a day, reducing exposure to other substances and leaving a clear geospatial residential exposure of the groups studied. The relationship between air pollutant levels with some effect on human health has been demonstrated in several studies. For example, two prospective cohort studies in Taiwan and Europe found generally positive associations between geospatial residential exposure to PM and nitrogen oxides (NOx) and liver cancer risk (Pan et al. 2016; Pedersen et al. 2017).

On the other hand, metals such as Fe, Al, and Cr can generate oxidative stress; iron is essential for many cellular functions; however, it also has the capacity to gain and lose electrons. This characteristic allows it to participate in reactions that can generate free radicals (RL), producing the OH– radical through the Fenton reaction (Graf et al. 1984); this ROS is reactive and can cause damage to lipids as mentioned above. In the case of Al, the most abundant metal in the earth’s crust; its presence in drinking water is mainly due to water treatment processes, since it is used as a flocculant. Its ability to generate oxidative stress has been widely documented it is known that within its mechanisms is its ability to produce damage in mitochondria, to form colloidal aluminum and bind to pro-oxidant metals (Fe or Cu) and, therefore, promote oxidative events based on pro-oxidant metals, as well as inhibition of the activity of antioxidant enzymes (Kumar et al. 2009; García-Medina et al. 2010; Kumar and Gill 2014). In the case of chromium, two valence states occur in nature (hexavalent chromium and trivalent chromium). Cr (IV) is commonly used in industrial processes such as chromium plating, welding, painting, metal finishing, steel manufacturing, alloying, and cast iron. Also, it has been identified as a known mutagen and carcinogen, its mechanism by which it exerts its toxic effect is not yet well established, but it has been reported to be capable of inducing oxidative stress, DNA damage, cell death, and modification in gene expression in proteins associated with the process of apoptosis (Bagchi et al. 2002; Xu et al. 2018). In contrast, chromium (III) is essential for proper insulin function and is necessary for normal protein, fat, and carbohydrate metabolism, and is recognized as a dietary supplement (Bagchi et al. 2002). Human exposure to this metal is low in drinking water (0.2 to 2 μg Cr (VI)/L), according to the Mexican Official Standard PROY-NOM-127-SSA1-2017 should not exceed 0.05 mg/L total chromium (Secretaría de Salud 2019). The levels presented of metals in drinking water varied, depending on the system that provides drinking water to the three areas studied, noting that the concentrations of metals were lower in the samples from Nuevo Madín, followed by the group from Gustavo A. Madero and finally, the highest levels were found Naucalpan de Juárez, exceeding the limits of the Mexican standard for Fe and Pb (Secretaría de Salud 2019).

For this study, subjects from Gustavo A. Madero were considered as a control group since they are an urban population, just like the subjects residing in Nuevo Madín and Naucalpan de Juárez and are exposed to a similar level of environmental contaminants, particularly with Naucalpan de Juárez, but receive water from different systems. Such is the case of the subjects of Nuevo Madín, which belong to the municipality of Atizapán de Zaragoza, where 75% of the potable water supply comes from three external sources, which are operated by the Comisión de Agua del Estado de México, these are the Cutzamala macrocircuit, the Sistema Barrientos aqueduct, and the Sistema Madín aqueduct. The remaining 25% of the supply comes from 33 deep drinking water wells located in different areas of the municipality; in the case of the Nuevo Madín neighborhood, this receives water mainly from wells (Farrell Baril 2008). With respect to the municipality of Naucalpan de Juárez, of the 3,150 L of water per second supplied, only 28.0% comes from its own sources (40 deep wells), the rest is supplied from sources outside the territory, mainly from the Lerma-Cutzamala System which provides about 66.6% and finally from the Madín Dam which contributes 5.4% (SEDUV 2007). The neighborhoods sampled in this study that belong to this municipality receive drinking water from the Cutzamala System and the Madín Water Treatment Plant. The Cutzamala System is one of the largest drinking water supply systems in the world. It provides 17% of the supply for all uses in the Mexico Valley Basin, calculated at 88 m$^3$/s, which is complemented by the Lerma System (5%), groundwater extraction (68%), rivers and springs (3%) and water reuse (7%). It supplies 11 delegations of Mexico City and 11 municipalities of the State of Mexico and is composed of seven diversion and storage dams, six pumping stations, and a water treatment plant that uses conventional treatment (CONAGUA 2018). Based on the concentrations quantified in the different samples, it can be appreciated that the contamination of the Madín reservoir contributes to the quality of drinking water, and the presence of metals such as Pb, Fe, and Al, the latter two have been reported in several points of the reservoir and usually exceed the maximum allowable limits for the protection of aquatic life (González-González et al. 2014; Pérez-Coyotl et al. 2019).

Finally, it was observed that for Nuevo Madín, the correlation with the levels of contaminants present in the air and water was low; however, an increase in the frequency of subjects with altered values of ALP and LDH enzymes, as well as LPX, was observed. Unlike the other groups studied, the inhabitants of this area usually have direct contact with the reservoir, because some of them consume the carp that inhabit the water body (57%) or animals that drink water.
from the dam (6%). For the consumption of carp from this water body, it was found that there is a 2.6 times greater risk of presenting an alteration in the ALP levels when consuming carp from the reservoir. It is known that aquatic organisms, such as common carp, can bioconcentrate contaminants and be a source of exposure for human populations implying a potential risk for consumers, particularly with metals (Yi et al. 2017). In this sense, Morachis-Valdez et al. (2015) found that the fish present in this reservoir contained high concentrations of metals such as Fe and Al, as well as some drugs found in the different points of the reservoir, so consuming fish coming from this water body may be an important factor in the modification of the enzyme. In addition, the organisms sampled during the study presented high levels of these two metals, thus confirming what was reported by Morachis-Valdez et al. (2015). Similarly, in LDH and LPX, where a risk of 5.46 and 1.97, respectively, was found.

There is evidence from previous studies on liver toxicity and oxidative stress of chemicals that showed an association with the biomarkers evaluated. Among environmental pollutants such as SO₂, an increase in MDA levels of 100% has been found after an eight-week exposure to 15 ppm SO₂ in rat (Zhang et al. 2006), while metals such as Al have been shown to cause significant increases in blood AST, ALT, ALP, LDH, and bilirubin levels, as well as an increase in LPX in rat liver tissue exposed to 30 mg/kg aluminum chloride for 21 days (Al-Kahtani et al. 2020). In the case of chromium, it has been reported that chromium caused a perturbation in biochemical parameters, altering the levels of blood glucose, triglycerides, cholesterol, ALP, ALT, AST, and LDH, as well as an increase in oxidative stress in male rats exposed for 30 days to 15 mg/kg Cr (Saidi et al. 2020). In rats treated with iron (8.3 mg/L FeSO₄ for 60 days), significant increases in MDA and nitric oxide (NO) levels in the liver have been demonstrated (Badria et al. 2015).

Identifying environmental exposures that negatively affect health is an important public health problem (Yorita Christensen et al. 2013); this pilot study was proposed to explore whether the inhabitants related to the Madin dam presented any changes in biomarkers of early damage, before the onset of a disease, due to the environmental problems that have been evidenced in the area. However, being a population that is located north of the metropolitan area of Mexico City, with reports in recent years of high levels of air pollution (Ramos 2019), the contaminants present in the air could not be ruled out. Exposure to multiple environmental chemicals and classes of substances complicates understanding the biological processes that these mixtures trigger, as well as the consequences on human health (Yorita Christensen et al. 2013); however, the results found are a start to continue investigating and at the same time taking measures to avoid more serious consequences on the health of the inhabitants.

To improve the understanding of the role of pollutants on the effects on human health in order to make important changes and reduce the probability of the occurrence of pollution-related diseases, citizens as well as authorities could follow the following suggestions: establish personal education programs regarding environmental pollution issues. Encourage reasoning about environmental concerns in the work and home environment by asking certain questions (Chance 2001), for example: Is waste production minimized? Is waste disposed of properly? Is plastic separated from materials to be incinerated? Are all appropriate materials recycled? Encourage the use of public transportation, as well as the substitution of less polluting vehicles, in addition to restricting traffic, prohibiting the circulation of a private vehicle for one day a week, making use of renewable energies, improving wastewater treatment, reducing the use of plastic, oils and batteries, and generating “green” spaces in the city. Promote healthy lifestyle habits and improving diet, as well as reducing alcohol and tobacco consumption, encouraging regular and moderate exercise, and considering the biomonitoring of some substances periodically.

There are some limitations to this study. Because it is a cross-sectional study, there is no way to evaluate the evolution of the response with the temporality of exposure, on the other hand, the study was performed during the quarantine of the COVID-19 pandemic. Therefore, factors such as diet, alcohol consumption, smoking, and exercise were modified in the population and could affect some of the responses evaluated; however, we adjusted for several important covariates in our analyses. The sample was taken at the home of the individuals, so the collection and transport of the samples to the laboratories for analysis could compromise the sample, although measures were taken to preserve and conserve them, such as temperature control during transport.

Conclusions

This study identified that chronic exposures to ambient air and water pollutants were significantly associated with elevated levels of lipoperoxidation. There was evidence that pollution from the Madin dam can generate oxidative stress and affect the health status of people who receive water from this reservoir or consume fish that inhabit it. These results highlight the need to integrate a policy that allows authorities, residents, and civil society to establish strategies to clean up the area and reduce the emission of pollutants into the dam and thus provide a better quality of life for the organisms that inhabit it and for the human population.

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**Author contribution** García-Medina S., Galar-Martínez M., and Ruiz-Lara K. participated in the study design, material collection, data analysis, and interpretation, and preparation of the manuscript. Parra-Ortega I. and Morales-Balcázar I. participated in sample preparation and analysis. Hernández-Rosas N. provided medical follow-up to the study participants. Hernández-Diaz M., Cano-Viveros S., Olvera-Roldán E., and Gasca-Pérez E. participated in collection of material. Gómez-Oliván L. and García-Medina AL. assisted with data interpretation and participated in manuscript preparation.

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**Data availability** The datasets used and analyzed during the current research are available from the corresponding author on request.

**Declarations**

**Ethics approval and consent to participate** All applicable international, national, and institutional guidelines for human research were followed. All procedures were performed in accordance with the ethical standards of the institution where the studies were conducted. The study protocol was approved by the Research Ethics Committee of the National School of Biological Sciences, IPN with registration number SIP-IPN, Projects 20210491, 6656 as well as the Secretaría de Investigación y Posgrado of the Consejo Nacional de Ciencia y Tecnología (CONACyT, Project h15061152).

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**Competing interests** The authors declare no competing interests.

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