Comparison of Levels of Heavy Metals in Nasal Concha Tissues in Rural and Urban Regions

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ABSTRACT

***Objectives*:** City centers are areas where the effects of air pollution are most common. Rural areas are regions where air pollution is relatively less. We aimed to show the presence of heavy metals in the breathing air in the nasal concha and investigate whether there is a difference in heavy metal Manganese (Mn), Lead (Pb), Copper (Cu), Cadmium (Cd), and Zinc (Zn) levels in rural areas and city centers.

***Methods*:** The heavy metal determination was performed by the Inductively Coupled Plasma- Optical Emission Spectrometry method in the concha tissues of 67 patients who underwent partial inferior concha resection, 38 people in the rural, and 29 people in the urban group.

***Results*:** Cu (0.24± 0.048 μg/g) and Zn (3.29± 0.69 μg/g) levels in those living in rural areas were significantly higher than Cu (0.06± 0.019 μg/g) and Zn (0.44± 0.14 μg/g) levels in those living in urban areas (**p <0.001)**. The Cd (0.034± 0.014 μg/g) level in the rural area was higher than the Cd (0.032± 0.013 μg/g) level in the urban residents, but it was not statistically significant. The Pb (0.024± 0.009 μg/g) and Mn (0.273± 0.01 μg/g) levels of the urban group were found to be significantly higher than those in the rural ones Pb (0.008± 0.0002 μg/g) and Mn (0.174± 0.05 μg/g) (**p <0.001**).

***Conclusions*:** Indicating the presence of heavy metals in turbinates and measuring their amount may be useful for diagnostic purposes in diseases

whose etiology is attributed to air pollution, but the pathophysiology has not yet been clarified.

***Keywords**: Heavy metals, Nasal Mucosa, Turbinates, Air Pollution, Spectrophotometry*

HIGHLIGHTS

1) Air pollution is more common in urban areas than in rural areas.

2) Heavy metals in the inhaled contaminated air can accumulate in the nasal mucosa and turbinate tissues.

3) Determining heavy metals in the nasal concha can be considered an indicator of air pollution.

INTRODUCTION

Environmental pollution is a current problem of the whole world, which emerged with the beginning of urban life for the first time and gradually increased in parallel with industrial development. Especially in the last half of the previous century, due to the industrial revolution, industrialization, and rapid population growth, environmental pollution has reached a level that threatens the lives of humans and other living things [1, 2]. Air pollution is a growing, global public health problem. It is considered that 4.2 million people die (World Health Organization- 2008) earlier, most of them from cardiorespiratory diseases due to air pollutants each year [3]. If we group the pollutants that cause this problem and disturb the natural balance, organic materials, industrial wastes, petroleum derivatives, synthetic agricultural fertilizers, detergents, radiation, pesticides, inorganic salts, synthetic organic chemicals, and waste heat can be listed as the main reasons for these. All these groups cause air pollution directly and indirectly. Heavy metals are found in almost all of the items listed under the heading, mostly industrial wastes and pesticides.

Elements are present in the human body in pure form or compounded form. Although some of these are necessary for human physiology, such as Lead (Pb), Mercury (Hg), and Cadmium (Cd), they are not useful in human physiology. Generally, intoxication occurs through accumulation with the increase of exposure time. In the pathophysiology of heavy metal poisoning, these metals have been shown to cause enzymatic changes by attaching oxygen, nitrogen, and sulfhydryl group substances in proteins [4]. For example, there are large amounts of thiol ligands in the structure of metalloproteins in our body. These ligands have a high affinity for binding elements such as Cd, Copper (Cu), and Zinc (Zn), and heavy metals are carried in the body through ligands. Factors such as the heavy-metal particle size, the duration of heavy metal exposure, and the age of the person are the factors that determine the degree of intoxication to heavy metals [5, 6].

The nose is the main entrance gate of polluted air to the body [7]. Many heavy metals can enter the human body through the inhaled air. Especially showing the presence of heavy metals in the nasal concha tissues, it may mean that heavy metals in the ambient air cause accumulation in nasal mucosal and submucosal tissues during filtration of the air. Due to the mucociliary activity of the nasal mucous membranes, inhalation of pollutants in the atmosphere, and absorption of elements into the mucosa and local immune responses can be detrimental to this body part.

This study aimed to determine the heavy metal levels of Manganese (Mn), Pb, Cu, Cd, and Zn in the inferior nasal turbinate tissue of people living in rural and urban areas to show the effect of air pollution on the nasal mucosa.

MATERIALS AND METHODS

Patients and Study Design

The patient groups consisted of 67 patients who applied to the Otorhinolaryngology outpatient clinic with nasal obstruction. Most of the selected patients were those with compensatory nasal concha hypertrophy accompanying nasal septum deviation, and partial resection of the inferior concha was scheduled. According to the residence, the patients were divided into rural (those living in the village) and urban (city center). The patients were required to have lived in their residence (rural or urban) for at least 15 years.

Twenty-nine patients living in city centers and 38 patients living in villages were included in the study. After informed the patients, we obtained verbal and written consent.

We excluded patients with diseases that could potentially affect the presence of heavy metals in the nasal concha. Exclusion criteria;

- Smokers/ex-smokers
- Patients under the age of 18 and over 55,
- Patients with nasal polyps or polyposis,
- Patients with nasal involvement of systemic diseases,
- Patients using medication continuously due to any chronic disease,
- Patients who have undergone any surgical intervention (septoplasty, concha radiofrequency ablation, partial resection) in the nasal cavity before,
- Patients working as workers in the industry sector,

has been excluded.

There were 38 people in the rural group and 29 people in the urban group. Twenty-six (68.4%) of the patients living in rural areas were male, and 12 (31.2%) were female. Of the patients living in urban areas, 17 (58.6%) were male, and 12 (41.4%) were female. The age distribution of the patient group living in rural areas was between 18 and 52 (average 29.3), and the age distribution of the urban patient group was between 18 and 55 (average 29.1).

We performed partial resection of the inferior concha with septoplasty in 58 patients and partial resecion of the inferior concha in nine patients. Bilateral inferior concha partial resection was performed in 62 patients, while unilateral resection was performed in seven patients.

All patients were operated on under general anesthesia by the same surgeon, and a standard procedure was performed. No local anesthetic was applied to the concha tissue before resection. The inferior turbinates were first medialized with the Cottle elevator, and then the lower-medial parts were cut using concha scissors. We washed the resected turbinate pieces with distilled water, and we removed the mucus and attached particles mechanically. The tissues were transferred to the tubes where they will be stored without contacting them anywhere, and transported to the tank where they will be stored at -80oC (Figure 1).

Measurements of Metal Concentrations

We took nasal concha tissues from -80oC, where they were preserved and left to thaw at room temperature. First, conchal bones, if any, were dissected and removed. From the deboned mucosa and submucosal parenchyma tissue, fragments weighing 0.5 g were precisely considered and then transferred to a tube. We added 3 ml of the acid mixture ($HClO4 + HNO3$, 5: 1). The samples were incubated at 95 C for one hour [8]. Solid conchal tissues were hydrolyzed with acid and became liquid at the end of three hours. Solutions were diluted with distilled water to measure heavy metal (Cu, Zn, Cd, Pb, Mn). Spectrophotometric analyses in our study were performed using the Inductively Coupled Plasma- Optical Emission Spectrometry ICP-OES device (Perkin-Elmer, Optima 4300 DV, ICP / OES, Shelton, USA). The amounts of Cu, Zn, Cd, Pb, and Mn were determined in micrograms. Since the conchal tissues we received were 0.5 g, all results were multiplied by two, and we calculated the heavy metal level in 1 g nasal concha tissue in micrograms (μg).

Statistical analysis

SPSS v.22 computer program was used to analyze the data. We analyzed the compliance of the groups to the normal distribution with the Kolmogorov Smirnow Test. Data are presented as a number, percentage, median, mean and standard deviation. Student T-Test was used in the analysis of age, Cd, Pb, Cu, Mn, and Zn in urban and rural patient groups, and the Chi-square test was used in the comparison of gender. Mann-Whitney U test was used to analyze age by gender, Cd, Pb, Cu, Mn, and Zn in both groups. Pearson Correlation Test was used in the binary correlation analysis of these parameters. The significance level was taken as $p < 0.05$.

Ethics

This study was found to proper with ethical rules by the XXX University Faculty of Medicine Non-Interventional Clinical Research Ethics Committee (Decision No: XXXX.0.01.00 / 56).

RESULTS

The patient distributions in both groups were found to be homogeneous in terms of age and gender. The mean age of the patients in the rural areas was 29.13 (± 9.4), while the mean age of the patient group living in the city center was 29.1 (± 9.8), and there was no statistically significant difference (p: 0.991). Of the rural patients, 26 (68.4%) were male, 12 (31.2%) were female, and the urban patients were 17 (58.6%) male, 12 (41.4%) was female. There was no significant difference between the male and female ratios within the two groups (Table 1).

Cu (0.24± 0.048 µg/g) and Zn (3.29± 0.69 µg/g) levels in those living in rural areas were significantly higher than Cu (0.06± 0.019 µg/g) and Zn (0.44± 0.14 µg/g) levels in those living in urban areas (***p <0.001, p <0.001***, Table 2). The Cd (0.034± 0.014 µg/g) level in the rural area was higher than the Cd (0.032± 0.013 µg/g) level in the urban residents, but it was not statistically significant (p: 0.505, Table 2). The Pb (0.024± 0.009 µg/g) and Mn (0.273± 0.01 µg/g) levels of the urban group were found to be significantly higher than those in the rural ones Pb (0.008± 0.0002 μg/g) and Mn (0.174± 0.05 μg/g) (***p <0.001, p <0.001***, Table 2) (Figure 2).

Our results show that the differences in heavy metal levels in patient groups living in rural and urban areas are independent of age and gender. When we compared the two groups, we saw that the difference between the heavy metal levels occurred due to the residence difference and was independent of age and gender (Table 3).

DISCUSSION

Air pollution is considered a general health hazard. Technologies that facilitate our current world release a wide variety of pollutants into the atmosphere. These technological products increase the concentration of air pollutants in the atmosphere, containing heavy metals harmful to plants, animals, and humans. When specific risk factors such as a person's genetic background, diet, and lifestyle come together with environmental factors, the negative impact of air pollution on human health will increase [9]. For the individual risk assessment of environmental hazards, it is essential to show the physical presence of the substances in the body or to show the negative functional results. The nasal cavity is a usual gateway to the human body for air pollutants and is a well-known target site for toxicity caused by air pollutants [7]. Our study showed heavy metal in nasal conchal tissues and compared their levels in rural and urban residents.

Few studies in the literature examine the relationship between nasal structures and mucous membranes, and heavy metal. Şenvar, in his study [10], comparing patients with atrophic rhinitis and healthy people in terms of serum Cu and Zn levels, found serum Cu levels high and low Zn levels in patients. In another study [11], trace elements on the nasal lower concha and nasal septum were compared. There was an age-related increase in Pb levels in both tissues and decreased Zn levels, but no other significant difference was found. In another study [12], exposure to urban pollution in adult humans in Mexico has been shown to increase the proliferation rate in nasal cells. It has been reported that increased cellular turnover may be a risk factor for the development of neoplasia.

Calderon- Garciduenas et al. found significant differences in biopsies from nasal mucosa of children exposed to outdoor pollutants compared with a healthy control group [13]. Children who were exposed to polluted air had more complaints about upper airways such as epistaxis, nasal dryness and crusting, nasal congestion. As expected in ENT examinations, abnormal nasal mucosal findings such as increased hyperemia in the nasal mucosa, increased bleeding points, purulent mucus, and sinusitis were observed. The authors reported that the histopathological analysis of nasal mucosa biopsies of children exposed to air pollutants showed; basal cell hyperplasia, decreasing the number of ciliated epithelial cells and goblet cells, submucosal neutrophil infiltration, squamous metaplasia, and dysplasia [13].

In general, rural areas are the regions with very few air pollutants, except for allergens. If the agricultural industry is developed, particulate matter in airborne pesticides and fertilizers can be considered air pollutants for the rural area [14].

Unlike rural areas, city centers are focal points of air pollution due to motor vehicles, industrial facilities, and factories. In urban regions where the population is much more crowded, the primary source of air pollutant particles is traffic density and exhaust gases from both diesel and gasoline engines [15, 16]. Major outdoor air pollutants include carbon monoxide, sulfur dioxide, ozone, nitrogen dioxide, and lead [14]. The polluted air in the city also contains ultrafine particles in high concentrations, and their effect on human health is not fully known [16, 17].

Glück et al. examined the nasal mucosa of chronically exposed people to diesel engine emissions in their study cytopathological [18]. They showed goblet cell hyperplasia in the nasal mucosa and inflammatory response caused by increased leukocytes and defined this condition as chemical-induced rhinitis. Pb is the heavy metal that increases the most in air pollution [19]. In air pollution caused by exhaust gas, heavy metals mostly enter the body by inhalation [20]. Yılmaz et al. investigated [21] heavy metal levels in the leaves of trees in the city center and trees in the countryside. The amount of Pb in the samples taken from the trees in the city center was significantly higher than the rural ones, and they associated this height in Pb levels with exhaust gas and air pollution. In another study [22], Suchodoller found that the amount of Pb accumulated in the barley and corn they planted next to the road was high. They found that amount of Pb in the plants decreased as they moved away from the road. They reported that the effect of traffic was not noticed 30-40 m away from the road. Our study found that Pb levels in the tissues of the patient group living in the city center were significantly higher than those in the rural ones ($p < 0.001$).

In the study [23] by Youssaf et al. to measure bark suitability as bioindicators for assessing air pollution in downtown Toronto, Canada, Cu, Hg, Mn, Ni, Pb, and Zn concentrations were measured collecting bark from fields with high vehicle traffic and away from roads. They found high Pb and Mn levels in trees on the roadside with heavy traffic. Our study also found that the Mn and Pb values in the urban group to be significantly higher.

People are not only exposed to polluted air outdoor. The best-known effect is cigarette smoke, the air pollutant inside. In an experimental study[24], significant changes were observed in the nasal mucosa of animals exposed to tobacco smoke as an air pollutant. Disruptions in intercellular tight junction complexes, significant structural changes in cell membranes, and increased infiltration of neutrophils on the nasal mucosa surface have been demonstrated in exposed animals. Cigarette smoke or occupationally exposed smoke is inhaled directly into the body along with the heavy metals it contains and causes the levels of heavy metals in the blood to rise [25]. Since our study aimed to compare heavy metal levels by residency, smokers and ex-smokers were excluded.

Heavy metals inhaled with polluted air cause accumulation in the human body. It has been reported that heavy metals trigger oxidative stress in the body and show harmful effects in this way [4, 26, 27]. Oxidative stress mechanisms, on the other hand, play a role in the formation of many diseases. It is a challenge to show the harmful effects of exposure to air pollution on human health with evidence. It is necessary to present the pollutant at the same time above the acceptable concentration in both ambient air and human tissue, which is not an easy application in the normal population.

As the limitations of our study, the absence of simultaneous heavy metal measurements in the patients' blood sample and the absence of indexes showing air pollution in terms of heavy metal in the atmosphere of the urban and rural areas where the patients live can be considered. Another weakness of the study is that passive smoking status was not considered, although active smokers and ex-smokers were excluded from the study.

Demonstration of heavy metals in the nasal mucosa may be useful in cases where we attribute the etiology to air pollution, but the pathophysiology has not yet been elucidated. Also, the fact that we have demonstrated heavy metals in the nasal concha in our study, tissues obtained by nasal mucosal biopsies may be sufficient to prove heavy metals in the body in diseases that we considered these metals in the etiology.