

Health Risk Assessment of Heavy Metal Concentration in Spices Obtained from Markets: A Case Study in Bangladesh

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A thesis submitted to the School of Pharmacy in partial fulfillment of the requirements for
the degree of Bachelor of Pharmacy (Hons.)

School of Pharmacy
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October, 2025

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Declaration

It is hereby declared that

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2. The thesis does not contain material previously published or written by a third party, except where this is appropriately cited through full and accurate referencing.
3. The thesis does not contain material which has been accepted, or submitted, for any other degree or diploma at a university or other institution.
4. I have acknowledged all main sources of help.

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Ethics Statement

This study didn't involve any human participants, animal experiments, or identifiable personal data; therefore, formal ethical approval was not required. All procedures related to sampling and laboratory analysis were conducted in accordance with the institutional guidelines and relevant regulatory standards. The study maintained ethical principles of integrity and transparency and objectivity throughout the process of data collection, analysis and reporting.

Abstract

This study investigated the presence of heavy metals in commonly used spices in Bangladesh to evaluate potential health risks. A total of 27 spice samples (10 branded and 17 non-branded) of chili and turmeric were collected from different markets and analyzed using Inductively Coupled Plasma Mass-Spectrometry (ICP-MS). The concentration of heavy metal in the analyzed samples were estimated as As (0.05-0.12ppm), Se (0.04-0.09ppm), Pb (0.38-0.75ppm), Cd (0.06-0.14ppm), Cr (1.08-2.25ppm), Ni (0.38-.087ppm), Cu (2.08-8.30 ppm), Zn (16.05-39.24 ppm), Hg (0.02-0.19 ppm), Mn (15.95-50.52 ppm), Co (0.13–0.22 ppm), Fe (92.81-272.43 ppm), Al (57.53-618.87 ppm), Be (0.00-0.01 ppm). The analysis revealed that aluminum and iron concentrations were higher than other metals detected. Although most concentrations were within acceptable limits, the Total Target Hazard Quotient (TTHQ) values exceeded 1, indicating potential non-carcinogenic health risks to consumers. The findings highlight the need for continuous monitoring and stricter regulation of heavy metal contamination in spices.

Keywords: Heavy metals, Health Risk Assessment, Carcinogenic, non-carcinogenic.

Dedication

This thesis is dedicated to our parents, whose unwavering support, patience, and inspiration have enabled us to pursue and accomplish this research endeavor and our project supervisor, DR. Sharmind Neelotpol miss for her exceptional guidance and constructive feedback. This work would not have been possible without her invaluable support.

Acknowledgement

We would like to express our sincere gratitude to our supervisor, Dr. Sharmin Neelotpol, for her invaluable guidance, constructive feedback, and continuous support throughout the course of this research. Her expertise and insightful suggestions have been instrumental in the successful completion of this thesis.

We are also deeply thankful to Asef Raj, for providing the necessary academic resources and guidance that greatly contributed to the progress of this work. Our appreciation is extended to all the faculty members, whose guidance, knowledge and encouragement have been invaluable throughout our academic journey.

Special thanks to our friend Saiful Islam and seniors Lovely Kamal & Rafid Fayyaz for their support and encouragement.

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List of Acronyms

WHO	World Health Organization
FDA	Food and Drug Administration
EDI	Estimated Daily Intake
THQ	Target Hazard Quotient
TTHQ	Total Target Hazard Quotient
FAO	Food and Agricultural Organization

Chapter 1: Introduction

1.1 Background

Spices and herbs are plant derived substances that add flavor to any dish. Spices come from the plant parts: root, rhizomes, stem, leaf, bark etc. and herbs are non-woody plants. They are also known for their strong antioxidant activity and rich essential oils, are vital components of the human diet. However, concerns arise regarding their safety due to potential metal toxicity. The dried or fresh parts of plants, referred to as spices, have a long-standing history of use in enhancing the flavor, aroma, and color of various foods and beverages. Both spices and herbs play a crucial role in human nutrition because of their essential oils and notable antioxidant properties. Consequently, the risk of metal poisoning raises important questions about the safety of consuming spices and herbs.

Spices are an important part of life as it is used in every dish. They are frequently used as an important component in cooking to provide taste, color and aroma. They also provide antioxidant, antimicrobial and anti-inflammatory properties and are rich in essential nutrients. (Baig et al., 2019; Winiarska-Mieczan et al., 2023).

Uses of spices are related to human civilization and trade routes. It is used for both culinary and nonculinary purposes. Turmeric and chili have both culinary and medicinal properties. Because turmeric is used for healing purposes. Besides, it is also used as an antioxidant and works like a preservative.

However, it is usually suspected that a variety of environmental contaminants such as pesticides and heavy metal are introduced in spices via human activities during growing, processing and harvesting. As a whole, plenty of factors, especially plant type, frequency and other factors, influence the amount of heavy metals in plants. It is also influenced by which

agro chemicals are applied, soil type, pH and organic matter (Tinker, 1981; Lubben, and Sauerbeck, 1991). Through plant absorption trace elements are entered into the food chain. Many studies prove that human consumption of heavy metals through the food chain leads to toxic and health issues (Jarup, 2003; Mahan & Raymond, 2012). They are not eliminated easily from the body and cause many diseases. Heavy metals like arsenic, cadmium, mercury, lead are not that important nutrients for our body. (Kachenko and Singh, 2006, Sharma et al., 2007).

In addition, the elimination rate of heavy metals is very poor and they accumulate in the body and after a certain period of time it affects the endocrine and neurological system which causes reduced organ function, chronic kidney disease and potential organ failure.

1.2 Chili

Chilies, commonly referred to as hot peppers, are among the most extensively used spices worldwide, particularly in South Asia. Originating in the Americas, chilies became a globally significant spice after the 15th century, reshaping culinary traditions and influencing food cultures across continents. Although relatively recent introductions to Asia and Europe, chilies have become deeply integrated into numerous traditional cuisines. Asia, in particular, has emerged as one of the leading producers of chilies due to their high commercial value, vibrant color, and distinctive flavor (Kumar et al., 2006).

The primary origin of chilies lies in native South America, from where they spread through regions such as Mexico and Peru more than 6,000 years ago. In 1493, Christopher Columbus transported chilies to Spain from the New World during his maiden voyage, marking the beginning of a new era in global spice trade and culinary exchange. Subsequently, the Portuguese and Spanish recognized the economic and gastronomic importance of chili peppers and promoted their cultivation and use as an alternative to black pepper.

Hence, they stretch it by their global trade connections. In the 16th century Portuguese traders brought chilies to India, China, South Asia, and Africa by seafaring. There are various types of chilies and their names are different based on the place (Govindarajan & Salzer, 1985). Chilies were quickly adopted and integrated into indigenous cuisine and agriculture in India and now they are one of the world's largest producers and consumers.

Additionally, in China and Thailand, people also adopted chili as one of the main ingredients and made distinctive cuisine using chilies. The Middle East also accepts chili for cooking and medicinal purposes. In addition, chilies are produced at a different quantity throughout the world based on their environment for fertilization. Such as India (37%), China (7%), Bangladesh (5%), Pakistan (5%), Peru (5%), Thailand (4%), Myanmar (3%), Viet Nam (2%), and others (29%). India is the largest producer of chilies, and they are also the largest exporter of them in the whole world. China holds the second position for exporting throughout the world. (Madala n. & Nutakki .K.M, 2020).

A suitable temperature is needed for growing and it is almost 20-30 degree celsius and the production can be hampered if it exceeds more than 30 degree celsius or the temperature falls below 15 degree celsius (Rai et al., 2013).

Chili peppers may be used as a feasible natural treatment for treating cancer. In addition, capsaicin and antioxidants in chili powder can eradicate cancer cells in leukemia and prostate cancer. Therefore, this is mostly attributable to the elevated antioxidant and anti-inflammatory characteristics of chili peppers. In case of prostate cancer, capsaicin inhibits the proliferation of prostate cancer cells by inducing a depletion of the principal cancer cell types and inhibits the growth of cancer cells, leading to the cell death of the cancer cell. Moreover, these comparable effects are observed in breast, pancreatic, and bladder cancers (Madala n. & Nutakki.K., 2020).

In addition, the color of chili indicates that it contains beta carotene, or provitamin A, which is a vital element of the intestinal, respiratory, and urinary systems. Moreover, it also helps to build a strong immune system (Madala n. & Nutakki.K.M, 2020). Hence, it is also used in nasal spray, which contains capsaicin, for preventing congestion or allergies. It also has antifungal properties and has the ability to kill food pathogens.

Moreover, it is also responsible for maintaining good eyesight. Because it contains vitamin A, which is an essential element for rod cells that helps to clear night vision, and lack of chili intake can cause night blindness. Because daily intake of chili fulfills 9% of the recommended vitamin A (Madala n. & Nutakki .K.M , 2020) .

Furthermore, chili peppers contribute to the production of red blood cells because they contain essential minerals such as iron and copper, which play key roles in red blood cell synthesis (Madala & Nutakki, 2020). In addition, chili peppers are beneficial for pregnant women as they contain folic acid, an important nutrient that supports fetal development. A deficiency of folic acid during pregnancy can lead to complications in newborns.

1.3 Turmeric

Curcuma longa is the scientific name of turmeric, and it is native to South Asia, particularly India. It was first used over 4000 years ago. In 2000 BCE, turmeric was utilized in Ayurvedic medicine, Hindu festivities, and culinary applications. Furthermore, it represented purity, fertility and wealth. In 2000 BCE, turmeric was utilized in Ayurvedic medicine, Hindu festivities, and culinary applications. Furthermore, it represented purity, fertility, and wealth. It also has medicinal properties such as wound healing and treatment for digestive disorders, inflammation, and skin conditions. In addition, China and Southeast Asia accepted turmeric as their traditional medicine. In the 8th-9th century the Arab traders established the use of turmeric

in the Middle East and North Africa. In Europe it is inaugurated through trade routes, and it is also known as Indian saffron for its color. The active compound of turmeric is curcumin, which has excellent medicinal properties.

Turmeric helps maintain the blood sugar level and also has an impact on controlling cholesterol levels. It acts as an immunomodulator and is used to control acute and chronic allergies. (Rajanala & Vashi, 2018). Therefore, the controlling ability of acne and psoriasis is also found in turmeric. As it has so many medicinal effects, people often take it with milk or simply mixed in water. In Okinawa, Japan, it is served as an alternative to tea (Rajanala & Vashi, 2018). In our country people use turmeric as a key component of cooking.

Though turmeric has so many benefits, it should be taken cautiously. For example, people with iron deficiency should not take turmeric in large quantities because it can reduce the iron absorption, which is essential for red blood cell formation. Besides, it slows the rate of blood clotting, so people suffering from bladder problems are recommended to not take turmeric (Yadav.P.R & Tarun. G,2016).

1.4 Heavy metal contamination

Despite being essential for cooking and possessing both culinary and medicinal properties, spices like turmeric and chili can also become contaminated in various ways. This contamination occurs naturally through absorption from contaminated soil and through human adulteration. In Bangladesh, heavy metals are present in tanneries, textile and dyeing industries, and shipbreaking yards. These sources are the primary contributors to heavy metal pollution. Industrial waste metals contribute to heavy metal pollution, while atmospheric emissions of heavy metals further increase the levels of these metals found in food (Salim et al., 1993).

Heavy metals are metallic elements characterized by a high atomic number, as well as weights and densities that are at least five times greater than those of water. Many of these elements are crucial for physiological processes in trace amounts but can become toxic at elevated concentrations. Heavy metals such as manganese, cobalt, silicon, mercury, zinc, aluminum, nickel, selenium, and iron significantly influence environmental health and human well-being. While many of these elements are essential in trace amounts, their excesses pose risks, including neurotoxicity, carcinogenicity, respiratory diseases, and ecological disturbances. The persistent nature of heavy metals in the environment necessitates stringent regulation, effective waste management, and ongoing research to mitigate their adverse effects. Recognizing their dual roles as essential nutrients and potential toxins is vital for balancing industrial development with environmental and public health protection.

Some heavy metals are naturally occurring, while others result from industrial waste. Arsenic, cadmium, lead, and mercury are examples of naturally occurring heavy metals. The World Health Organization has classified the metalloid arsenic and the metals cadmium and lead as among the top ten chemicals of public health concern (World Health Organization, 2020). Consequently, the WHO, along with countries such as China, Singapore, Malaysia, and the Republic of Korea, has established limits for cadmium and lead in their herbal remedies (World Health Organization, 2007). Conversely, some, like mercury and cadmium, have no known biological function and are inherently toxic. The widespread industrial use, environmental persistence, and bio accumulative nature of heavy metals have raised significant concerns regarding their impacts on ecosystems and human health.

Heavy metals have a toxic effect on the human body and the surrounding environment. This toxicity primarily occurs through the direct ingestion of contaminated food, water, beverages, and polluted air (Adugna et al., 2024). Agricultural land situated near factories, mining sites,

or industrial areas is at a higher risk of contamination compared to other areas. This proximity increases the likelihood of contamination in the food being grown.

In our country, drinking water, consuming food, or using arsenic-contaminated water are common ways to spread arsenic contamination. This contamination causes various diseases, such as skin lesions, hyperkeratosis, and an increased risk of blood, lung, and bladder cancer. Lead is primarily used to enhance the color of products. Some dishonest businessmen and restaurant owners use it to improve the appearance of food, aiming to make it more attractive and authentic. Unfortunately, this practice raises health risks for the population, leading to symptoms like fatigue, irritability, loss of appetite, abdominal pain, and joint pain, which are collectively known as plumbism. Mercury enters the body through the food chain, particularly through fish consumption, and can result in Minamata disease, acrodynia, and erethism (also known as mad hatter disease). Cadmium adversely affects the gastrointestinal tract and the cardiovascular system.

Manganese is an important trace element that is used in enzymatic processes such as the functioning of superoxide dismutase, which protects against oxidative stress (Aschner & Aschner, 2005). However, excessive manganese exposure, particularly via inhalation in occupational settings (e.g. mining, welding), can cause neurotoxicity resembling Parkinson's disease, characterized by tremors, gait disturbances, and cognitive deficits (Guilarte et al., 2012). Chronic manganese poisoning, or manganism, results from accumulation in basal ganglia regions of the brain, leading to neurological impairments.

Elevated manganese levels in water bodies can affect aquatic organisms by disrupting enzymatic processes and inducing oxidative stress (Choi et al., 2017). Manganese bioaccumulation can also enter the food chain, impacting human health.

Cobalt is vital as a component of vitamin B12, necessary for DNA synthesis and neurological functions (Czaja et al., 2014). Nonetheless, high cobalt exposure, especially through inhalation of dust or fumes, can cause respiratory issues, cardiomyopathy, and skin dermatitis (WHO, 2004). Chronic cobalt toxicity may also induce hypothyroidism and cardiotoxicity. Cobalt contamination from mining and industrial discharge can lead to soil and water pollution, adversely affecting plant growth and aquatic life (ATSDR, 2022).

Silicon, primarily in the form of silica (SiO_2), is abundant in the Earth's crust and plays a role in plant structural integrity. In humans, silica is generally considered inert, but inhalation of crystalline silica dust can cause silicosis, a fibrotic lung disease (Leung et al., 2012). The toxicity arises from silica particles inducing inflammation and fibrosis in lung tissue. Silica dust generated during mining or construction can contribute to air pollution and respiratory diseases among workers and nearby populations.

Mercury is a notorious environmental pollutant with significant neurotoxic effects, especially in its methylated form (MeHg), which bioaccumulates in aquatic food chains (Clarkson & Magos, 2006). Methylmercury exposure impairs neurological development, leading to cognitive deficits and motor dysfunction, particularly in fetuses and children (Grandjean et al., 2010). Mercury poisoning can also cause renal and cardiovascular issues.

Mercury released from artisanal gold mining, industrial processes, and coal combustion contaminates water bodies, where microbial activity converts inorganic mercury to methylmercury, magnifying its toxicity in aquatic ecosystems (Selin, 2009).

Zinc is essential for our body's immune function, enzyme activity, and cellular metabolism (Vallee & Falchuk, 1993). However, excessive zinc intake can cause gastrointestinal disturbances, interfere with copper absorption, and suppress immune function (Maret & Sandstead, 2006). Zinc toxicity often results from industrial exposure or ingestion of

contaminated water. Zinc contamination in soils and water can hinder plant growth and aquatic life due to its toxicity at high concentrations (Alawadhi et al., 2024).

Aluminum is abundant in the environment but has no known biological role in humans. It is associated with neurotoxicity, particularly in individuals with impaired renal function, and has been linked to neurodegenerative diseases such as Alzheimer's disease (WHO & FAO, 2011). Aluminum can cross the blood-brain barrier, accumulate in neural tissues, and induce oxidative stress.

Nickel is essential in small amounts for certain enzymes but is also a common allergen causing contact dermatitis (EFSA, 2020). Excessive exposure, especially via inhalation in industrial settings, can cause respiratory problems, lung fibrosis, and even carcinogenic effects, notably nasal and lung cancers (IARC, 2012). Nickel contamination can originate from mining, alloy manufacturing and waste disposal, adversely affecting soil and water quality and harming biota.

Selenium is a trace element required for the function of antioxidant enzymes like glutathione peroxidase (Rayman, 2000). Its deficiency can impair immune function, but excess selenium causes selenosis, characterized by hair loss, nail brittleness, and neurological symptoms (El Mehdawi et al., 2018). Elevated selenium levels in water can bioaccumulate, affecting aquatic organisms.

Iron is crucial for oxygen transport and cellular respiration via hemoglobin. Iron deficiency leads to anemia, while excess iron can cause oxidative damage, liver cirrhosis, and cardiovascular disease (Huff et al., 2025). Iron overload conditions like hemochromatosis result from genetic predisposition and can be exacerbated by environmental exposure. Iron runoff from mining and industrial waste can lead to sediment pollution, affecting aquatic ecosystems' oxygen levels and biodiversity.

1.5 Aim of the study

The aim of this study was to determine the level of heavy metals for example: Hg, Pb, Cd, Se, Zn, Fe, Al etc. in turmeric and chili spices sold in the local market of Bangladesh.

1.6 Objectives of the study

The objectives of the study are to-

- Address the health risk assessment for both children and adults by assessing carcinogenic and noncarcinogenic exposures.
- Evaluate the extent of health risk through multiple indices: target hazard quotient (THQ), estimated daily intake (EDI), total target hazard quotient (TTHQ).
- Estimate heavy metal levels to compare with levels recommended by international organizations (WHO).

Chapter 2: Methodology

2.1 Sample collection

Two distinct types of samples, specifically turmeric and chili spices, were collected from various regions of the local markets in Bangladesh. For sample collection, four different areas were selected where spices are sold, with the aim of encompassing a broad range of regions. We collected 250 grams of turmeric and chili spices from each area and placed them in Ziploc bags. A total of 27 samples (10 branded and 17 non branded) were collected and divided into four categories: branded chili and turmeric and grinded (non-branded) chili and turmeric from mill to determine the concentrations of heavy metals present in spices. Furthermore, each sample was labelled before being transported to the laboratory for experimentation. The experiments were conducted at the Bangladesh Council of Scientific and Industrial Research (BCSIR).

2.2 Digestion of the sample

Acid digestion is an essential preparatory step in analyzing heavy metals across various matrices, including soils, sediments, biological tissues, and water samples. The purpose of this procedure is to break down complex organic materials and release bound metals into a solution that can be detected and quantified, typically using techniques like atomic absorption spectroscopy (AAS), inductively coupled plasma mass spectrometry (ICP-MS) or inductively coupled plasma optical emission spectrometry (ICP-OES).

Using a mortar and pestle, the samples are grinded to achieve homogenization, sieve through a mesh, and store them in a Ziplock bag. A digital analytical balance (Mettler Toledo, Model AG 204, Switzerland) with a precision of ± 0.0001 g for weighing the samples is used. For

drying, an oven (Digitheat, J. P. Selecta, Spain) is used. Additionally, a refrigerator (Hitachi, Tokyo, Japan) is used for sample digestion. To measure the volume of both the sample and the standard, micropipettes (1–10 μL and 100–1000 μL) are used.

Nitric acid (HNO_3) and hydrogen peroxide (H_2O_2) are frequently employed together in acid digestion for detecting heavy metals in samples such as spices, owing to their strong oxidative and solubilizing properties.

The acid digestion method involves treating a sample with strong acids to break down organic matter and mineral matrices, and convert metals into soluble forms. Nitric acid (HNO_3) is commonly used due to its strong oxidative properties, which effectively decompose organic compounds and oxidize metals into their respective ionic forms. Hydrogen peroxide (H_2O_2) is often included as an additional oxidizing agent to further enhance the breakdown of organic material, particularly in samples with high organic content, ensuring complete digestion.

Moreover, a typical digestion process involves placing a known weight of the sample in a digestion vessel, adding a specific volume of concentrated HNO_3 , and heating the mixture to promote digestion. The addition of H_2O_2 , often in the form of a 30% solution, is carried out either before or during digestion to facilitate the oxidation of organic matter. The process continues until the sample is fully digested, indicated by a clear or pale-colored solution, after which the digest is cooled, filtered, and diluted to a known volume for analysis.

Therefore, the combination of nitric acid and hydrogen peroxide provides a reliable and efficient method for digesting samples in heavy metal analysis. Proper optimization of digestion conditions—such as acid volumes, temperature, and digestion time—is essential to achieve complete oxidation and accurate quantification of heavy metals.

2.3 Heavy metal detection

For analysis, an inductively coupled plasma mass spectrometer (ICP-MS) is employed due to its high sensitivity, allowing for the detection of very low concentrations (ppq = parts per quadrillion level). ICP-MS combines a high-temperature argon plasma—typically sustained via radio-frequency induction—to atomize and ionize sample constituents, with a mass spectrometer that separates and detects ions by their mass-to-charge ratio (m/z). Moreover, samples, often introduced as an aerosol via a nebulizer, are ionized in a ~6,000–10,000 K plasma. The ions pass through an interface into a vacuum system and are focused by ion optics into a mass analyzer—commonly a quadrupole—which filters ions based on m/z before detection. ICP-MS instrument components and operating conditions are mentioned “in Table 1”

Table 1- ICP-MS Instrument components and operating conditions

Instrument component	Type / value
Nebulizer	ST PFA Microflow
Spray Chamber	Quartz cyclonic
Torch	One - piece quartz torch ,2mm i.d. Injector
Cones	Nickel sampler and skimmer Aluminium hyperskimmer
Peristaltic pump tubing	carrier/ internal standard: orange and green (0.38mm i.d.) Waste : gray/gray santoprene (1.33mm i.d)
Sample uptake rate	0.2 mL/min
Operating condition	Type/value
Rf power	1600 W
Plasma gas flow	15 L / min
Auxiliary gas flow	1.2 L /min
Nebulizer gas flow	Optimized for CeO +, C
Cell gas	Oxygen, Helium

2.4 Operating procedure of ICP-MS

The inductively coupled plasma mass spectrometry (ICP-MS) analysis was conducted using optimized instrumental parameters to ensure high precision and accuracy in elemental detection. The instrument was equipped with an ST PFA Microflow nebulizer and a quartz cyclonic spray chamber. A one-piece quartz torch with a 2 mm internal diameter injector was used for efficient sample introduction. Nickel sampler and skimmer cones, along with an aluminium hyperskimmer, were employed to enhance ion transmission. Peristaltic pump tubing of orange and green (0.38 mm i.d.) was used for carrier and internal standards, while gray/gray Santoprene tubing (1.33 mm i.d.) was used for waste. The sample uptake rate was maintained at 0.2 mL/min. The operating conditions included an RF power of 1600 W, plasma gas flow of 15 L/min, auxiliary gas flow of 1.2 L/min, and a nebulizer gas flow optimized for CeO^+ and C. Oxygen and helium were used as cell gases to minimize spectral interferences and improve detection limits.

Chapter 3: Result

3.1 Heavy metal assessment

A total of 27 samples were collected from various local markets across Bangladesh and categorized into four different sections: non-branded chili, branded chili, non-branded turmeric and branded turmeric. The concentration of heavy metals present in these samples were determined through Inductively Coupled Plasma Mass Spectrometry (ICP-MS).

Table-2 represents the overall concentration of heavy metals detected in the collected samples, while table-3 summarizes the concentration level according to the categorized sections. Depending on these analytical results, four distinct bar charts were developed to illustrate the distribution of heavy metals: non-branded chili (figure1), branded chili (figure 2), non-branded turmeric (figure 3) and branded turmeric (figure 4).

Table 2: Concentration of heavy metals in the samples

Samples	As	Se	Pb	Cd	Cr	Ni	Cu	Zn	Hg	Mn	Co	Fe	Al	Be
Non-branded CH-1	0.058	0.123	0.504	0.101	1.137	0.378	7.549	37.825	0	10.089	0.075	119.972	41.998	0.002
Non-branded CH-2	0.075	0.081	0.544	0.126	1.519	0.834	8.68	19.08	0.7	23.471	0.348	186.788	138.41	0.004
Non-branded CH-3	0.07	0.078	0.376	0.278	2.076	0.606	8.331	12.107	0.091	29.863	0.187	105.04	60.049	0.005
Non-branded CH-4	0.049	0.063	0.689	0.02	0.538	0.889	5.962	35.057	0	19.568	0.144	75.535	61.257	0.002
Non-branded CH-5	0.055	0.06	1.744	0.141	1.361	0.897	9.054	16.482	0	21.932	0.358	168.374	135.401	0.006
Non-branded CH-6	0.108	0.047	1.503	0.194	1.583	0.733	8.645	21.771	0	24.576	0.219	216.826	124.253	0.006
Non-branded CH-7	0.117	0.099	0.549	0.189	1.958	0.86	8.985	11.198	0	21.591	0.226	224.595	108.413	0.004
Non-branded CH-8	0.115	0.216	0.546	0.186	1.732	2.141	9.301	23.46	0	22.728	0.271	232.18	125.518	0.009
Non-branded CH-9	0.082	0.118	0.017	0	1.898	0.561	8.24	16.567	0.519	9.17	0.123	123.018	64.113	0.002
Branded CH 1	0.116	0.099	0.645	0.361	3.251	1.095	11.179	26.337	0.324	31.387	0.35	127.244	115.15	0.005
Branded CH 2	0.062	0.067	0.343	0.066	2.057	0.532	7.085	111.753	0.28	13.909	0.18	92.971	48.953	0.003
Branded CH 3	0.04	0.091	0.743	0.259	1.588	0.678	8.227	23.62	0.122	13.94	0.202	77.303	44.951	0.002
Branded CH 4	0.034	0.107	0.195	0.04	2.159	0.922	7.844	22.551	0.176	10.158	0.157	84.054	42.35	0.002
Branded CH 5	0.028	0.134	0.014	0.016	2.238	0.731	7.064	11.976	0.054	10.372	0.215	82.497	36.264	0.001
Non-branded TU-1	0.085	0.052	0.695	0.124	1.239	0.441	3.32	24.788	0.032	40.94	0.287	953.343	100.537	0.004
Non-branded TU-2	0.049	0.038	0.001	0.02	0.951	0.098	2.863	11.873	0.102	55.385	0.174	116.079	60.566	0.001

Non-branded TU-3	0.072	0.031	0.33	0.105	1.278	0.518	2.94	19.946	0.021	121.361	0.168	163.385	129.881	0.01
Non-branded TU-4	0.06	0.036	0.155	0.016	0.992	0.175	2.241	5.24	0.034	24.141	0.146	135.897	113.536	0.004
Non-branded TU-5	0.041	0.05	0.102	0.039	1.084	0.181	2.749	14.157	0.052	59.41	0.182	103.77	70.876	0.002
Non-branded TU-6	0.238	0.048	1.342	0.119	1.107	0.658	2.777	6.895	0.009	28.451	0.226	336.944	277.337	0.017
Non-branded TU-7	0.161	0.041	0.466	0.125	1.168	0.697	2.295	11.78	0.24	50.607	0.165	70.14	172.528	0.013
Non-branded TU-8	0.216	0.053	0.221	0.118	0.941	0.364	3.473	27.342	0.028	54.534	0.099	207.415	109.654	0.005
Non-branded TU-9	0.244	0.046	0.151	0.027	1.111	0.303	2.049	22.48	0	19.857	0.219	364.901	347.445	0.024
Branded TU-1	0.046	0.025	0.422	0.083	0.67	0.315	2.525	25.804	0.118	62.318	0.128	172.672	119.804	0.007
Branded TU-2	0.059	0.043	0.476	0.05	1.05	0.283	2.171	4.544	0	39.179	0.133	131.527	81.132	0.004
Branded TU-3	0.037	0.055	0.695	0.072	0.826	0.389	2.401	54.64	0	60.424	0.099	107.947	71.511	0.006
Branded TU-4	0.077	0.056	0.66	0.073	1.494	0.354	2.764	31.241	0.02	47.322	0.125	82.843	100.743	0.004
Branded TU-5	0.081	0.025	1.519	0.041	1.376	0.757	0.549	13.667	0	31.054	0.205	382.239	2721.179	0.057

Table 3: Concentration of heavy metals (mean± SD) in non-branded chili, branded chili, non-branded turmeric and branded turmeric.

Heavy metals	Chili		Turmeric		Permissible Limit
	Non-branded	Branded	Non-branded	Branded	
As	0.08 ±0.03	0.06±0.04	0.12±0.08	0.06±0.01	<5 ppm
Se	0.09±0.051	0.09±0.02	0.04±0.01	0.04±0.01	1.2-2.2 ppm
Pb	0.72 ±0.55	0.38±0.30	0.38±0.41	0.75±0.44	<10 ppm
Cd	0.14±0.08	0.14±0.15	0.077±0.04	0.06±0.01	<1.5 ppm
Cr	1.53±0.4	2.25±0.60	1.09±0.12	1.08±0.35	2.14 ppm
Ni	0.88±0.51	0.79±0.21	0.38±0.21	0.41±0.19	1.2 ppm
Cu	8.31±1.02	8.27±1.69	2.74±0.48	2.08±0.88	<150 ppm
Zn	21.51±9.38	39.24±40.89	16.05±7.90	25.97±19.10	< 50 ppm
Hg	0.15±0.27	0.19±0.11	0.05±0.07	0.02±0.05	0.1 ppm
Mn	20.33±6.70	15.95±8.82	50.52±30.31	48.05±13.46	100 ppm
Co	0.22±0.09	0.22±0.07	0.18±0.05	0.13±0.03	3.50 ppm
Fe	161.36±57.56	92.81±20.05	272.43±274.98	175.45±120.24	300 ppm

Heavy metals	Chili		Turmeric		Permissible Limit
	Non-branded	Branded	Non-branded	Branded	
Al	95.49±38.09	57.53±32.53	153.59±97.24	618.87±1175.37	1200 ppm
Be	0.004±0.002	0.002±0.00	0.01±0.01	0.015±0.02	Not specific

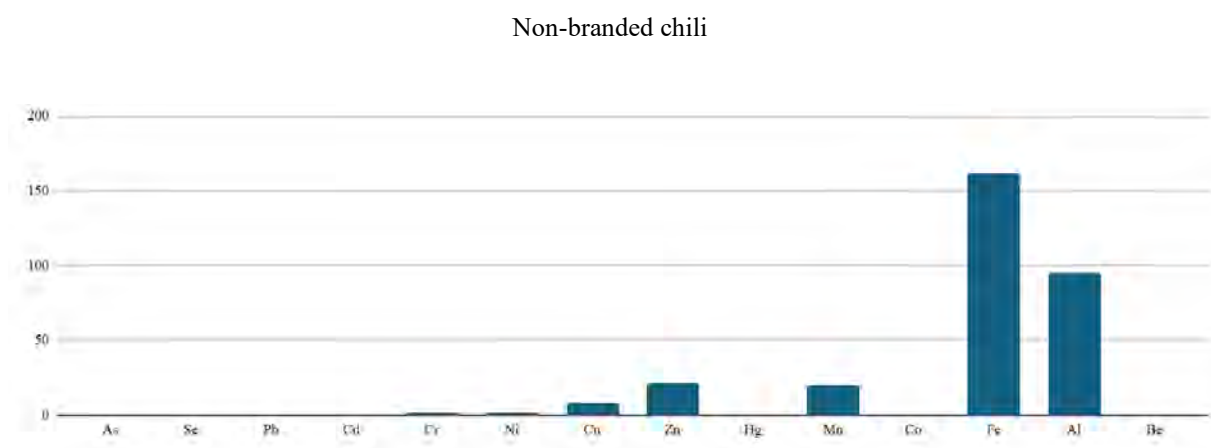


Figure 1-Concentration of heavy metals in non- branded chili

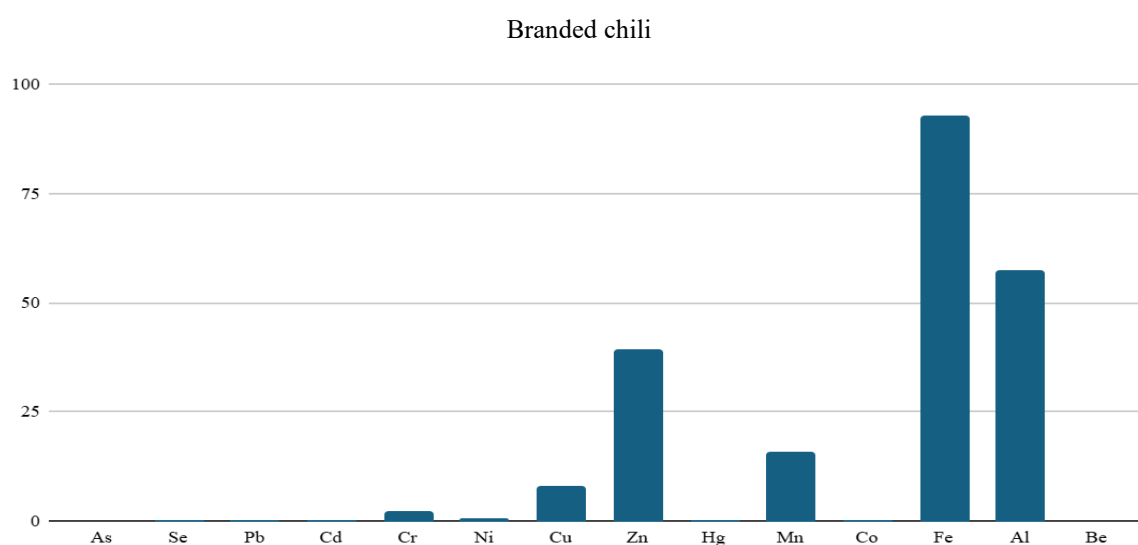


Figure 2- Concentration of heavy metals in branded chili

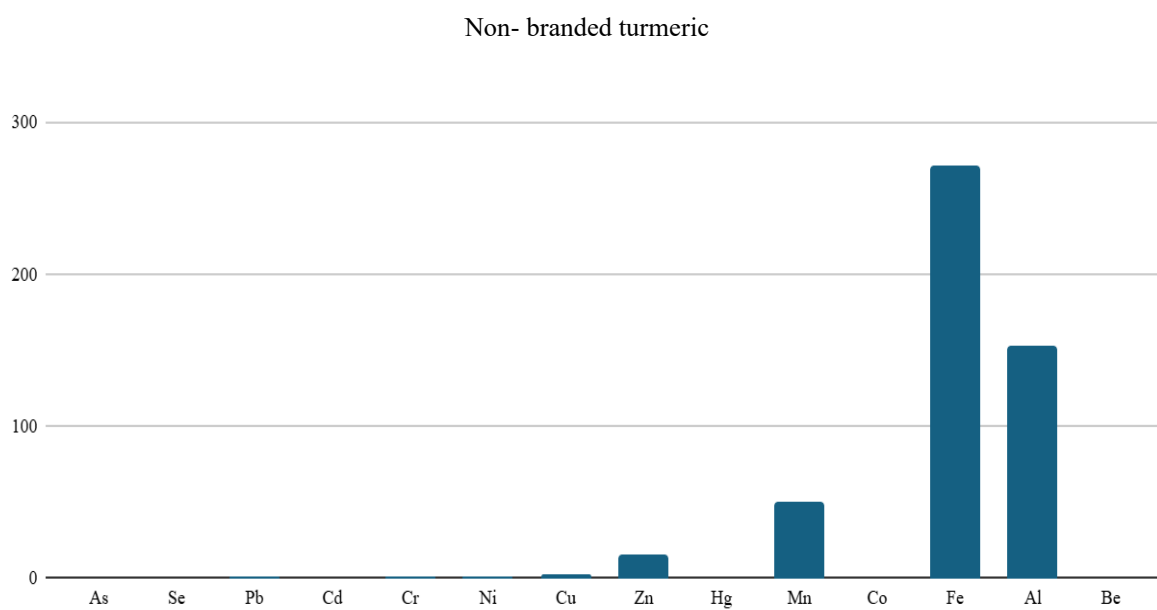


Figure 3- Concentration of heavy metals in non-branded turmeric

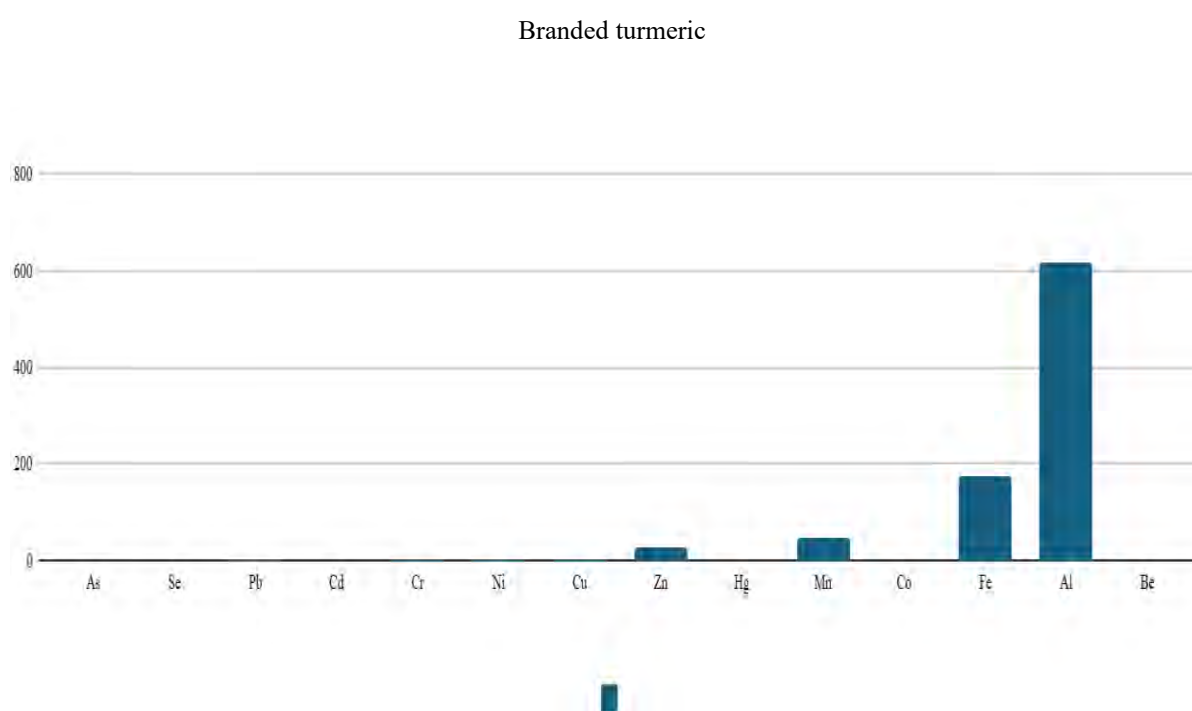


Figure 4- Concentration of heavy metals in branded turmeric

This study compared the concentrations of the trace elements in chili powder and turmeric samples with assesses reported in the literature for similar spices. Table 3 illustrates that several values documented in the literature were either higher or lower compared to those identified in the current investigation. Copper was detected in all analyzed spices and the concentrations of copper in these spices are presented in Table 3. The concentration of Cu in the spice samples ranged from 2.08 to 8.31 ppm. One-way ANOVA was utilized to assess copper concentrations among the samples, revealing significant differences in copper levels across the twenty-seven samples from two distinct groups. The maximum concentration of Cu was detected in the non-branded Chili (8.31 ppm), while the lowest concentration was detected in the branded turmeric samples (2.08 ppm). The results for all spices are within the permissible copper limits set by the WHO (20-150 ppm) for spices. In comparison to the studies shown in Table 3, the amounts of Cu in chili powder exceeded in comparison to the documented for turmeric in our findings. The concentrations of Cu in both branded and non-branded chili powder indicate that none of the results of investigation are consistent with these findings. On the contrary, for turmeric samples, the amounts of Cu were lower than those documented in both branded and non-branded chili. Overall, all examined spices exhibit minimal copper levels and pose no risk to human consumption.

This study found that the zinc content in the analyzed spice samples ranged from 16.05 to 39.24 ppm. The lowest zinc concentration was observed in non-branded turmeric (16.05 ppm). The zinc concentrations in the analyzed spices were compared to the FAO/WHO threshold for spices (50 ppm). The zinc concentration in red pepper was higher than that in turmeric.

This analysis showed lead concentrations of 0.71 ± 0.54 in non-branded red powder, 0.38 ± 0.30 in branded red powder, 0.38 ± 0.41 in non-branded turmeric, and 0.75 ± 0.44 in branded turmeric. The lead concentrations in both chili powder and turmeric were within the acceptable

limit according to the WHO standard of 10 ppm.

The amounts of chromium (Cr) measured in non-branded chili powder, branded chili powder, non-branded turmeric, and brand turmeric samples, finding levels of 1.53 ± 0.47 ppm, 2.25 ± 0.60 ppm, 1.09 ± 0.12 ppm, and 1.08 ± 0.35 ppm, respectively. The maximum value of 2.25 ± 0.60 ppm was identified in branded chili powder, while the minimum value of 1.08 ± 0.35 ppm was recorded in branded turmeric samples. All analyzed spice samples had chromium levels below the WHO acceptable limit ($0.78\text{--}1.48$ mg kg⁻¹ or ppm), except for branded chili powder, which was marginally above the typical chromium level found in spices. The concentration of chromium in chili powder was higher than that in turmeric samples. Therefore, in conclusion, the chromium levels in all spice samples obtained from the local shop fall within the acceptable range, making them suitable for human consumption.

The findings revealed that the concentration of nickel in the analyzed samples ranged from 0.378 to 2.141ppm. The highest concentrations were found in non-branded chili powder samples, while the lowest were observed in non-branded turmeric samples. Statistically important differences were noted in the mean nickel concentrations of branded chili powder, non-branded turmeric, and branded turmeric samples. The nickel concentrations identified in this study were below the globally acceptable range of 2–7 ppm. Therefore, these spices are considered to pose no threat to human health.

The iron concentrations found in non-branded chili powder, branded chili powder, non-branded turmeric, and branded turmeric were 161.37 ± 57.56 , 92.81 ± 20.06 , 272.43 ± 274.98 , and 175.46 ± 120.24 mg kg⁻¹, respectively. The results indicated significant differences ($p < 0.05$) at the 95% confidence level for iron in red pepper and turmeric. Among the spices analyzed, non-branded turmeric had the highest iron content, while the branded chili had the lowest concentration. The iron levels detected in this study were below the World Health

Organization's permissible limit of 300 mg kg⁻¹.

This study measured the manganese concentrations in non-branded chili powder, branded chili powder, non-branded turmeric, and branded turmeric, yielding values of 20.33 ± 6.70 mg kg⁻¹, 15.95 ± 8.82 mg kg⁻¹, 50.52 ± 30.32 mg kg⁻¹, and 48.06 ± 13.46 mg kg⁻¹, respectively. The results indicated significant differences ($p < 0.05$) at the 95% confidence level for manganese levels in red chili powder and turmeric. All measured values were below the WHO acceptable threshold for manganese (100 mg kg⁻¹). Notably, non-brand turmeric displayed the highest manganese concentration at 50.52 ± 30.32 mg kg⁻¹.

The concentrations of cadmium found in non-branded chili powder, branded chili powder, non-branded turmeric powder, and branded turmeric were 0.14 ± 0.0882, 0.14 ± 0.1527, 0.07 ± 0.4942, and 0.06 ± 0.0177 mg kg⁻¹, respectively. Statistical analysis of cadmium levels indicated a significant difference ($p < 0.05$) between the mean cadmium concentrations in chili and turmeric samples. The amounts of cadmium detected in this study were lower than those of any other heavy metal analyzed across all spice samples. The acceptable limit for cadmium in spices set by the World Health Organization is 0.200 mg kg⁻¹.

The amounts of selenium (Se) measured in non-branded chili powder, branded chili powder, non-branded turmeric, and branded turmeric samples, finding levels of 0.09 ± 0.05 ppm, 0.09 ± 0.02 ppm, 0.04 ± 0.01 ppm and 0.04 ± 0.01 ppm, respectively. The maximum value of 0.09 ± 0.02 ppm was identified in branded chili powder, while the minimum value of 0.04 ± 0.01 ppm was recorded in branded turmeric samples. All analyzed spice samples had selenium levels below the acceptable limit of chili (0.4-2.2 mg kg⁻¹ or ppm) and turmeric (0.3-1.23 mg kg⁻¹ or ppm). (World Health Organization, 2011)

3.2 Health risk assessment

There are four steps for health risk assessment such as, hazard identification, exposure assessment, dose-response assessment, and risk characterization (Agency, 2021; Rezvani Ghalhari et al., 2021). It is assumed that humans are in direct contact with heavy metal explosions. The non-carcinogenic health risk of heavy metals due to direct ingestion of spices was calculated using Eq. (1) and (2):

$$THQ = EDI / RfD \quad (1)$$

$$EDI_{\text{ingestion}} = C \times DI \times EF \times ED / BW \times AT \quad (2)$$

$$TTHQ = \sum_n THQ \quad (3)$$

(Cabral Pinto et al., 2020; Rezaeiashad et al., 2022; Tajdar-oranj et al., 2018).

Table 4: Characteristics of variables included for chronic daily intake estimation.

Variable	Definition	Unit	Value	Reference
C	Concentration of heavy metals	mg/kg	Presented in Table 2	
DI	Ingestion rate	g/person/day	2	(Taghizadeh et al., 2023)
EF	Exposure frequency	days/year	365	(Taghizadeh et al., 2023)
ED	Exposure duration	year	70	(Taghizadeh et al., 2023)
BW	Body weight	kg	70	(Taghizadeh et al., 2023)

AT	Average time	exposure days	25550	(Taghizadeh et al., 2023)
RfD	Oral reference dose	mg/kg/day	Cr = 0.003, Pb= 0.004, Co= 0.0003, Zn = 0.3, Cd = 0.001, Cu= 0.04, Al = 1, Mn=0.14 As = 0.0003 Se = 0.005 Ni = 0.02 Hg =0.0003 Fe =0.7 Be = 0.002	Agency

Table 5: EDI and TTHQ VALUE of non-branded & branded chili powder and non-branded & branded turmeric powder

Heavy metals	Chili				Turmeric			
	Unpacked		Packed		Unpacked		Packed	
	EDI	THQ	EDI	THQ	EDI	THQ	EDI	THQ
As	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
Se	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
Pb	0.02	5.00	0.00	0.00	0.01	2.50	0.02	5.00
Cd	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
Cr	0.04	13.33	0.06	20.00	0.03	10.00	0.03	10.00
Ni	0.02	1.00	0.02	1.00	0.01	0.50	0.01	0.50
Cu	0.02	0.50	0.23	5.75	0.07	1.75	0.05	1.25

Zn	0.61	2.03	1.12	3.73	0.45	1.5	0.74	2.46
Hg	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
Mn	0.58	4.14	0.45	3.21	1.44	10.28	1.37	9.78
Co	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
Fe	4.61	6.58	2.65	3.78	7.78	11.11	5.01	7.15
Al	2.72	2.72	1.64	1.64	4.38	4.38	17.68	17.68
Be	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
TTHQ	35.30		39.11		42.02		53.82	

The EDI value for As, Se, Pb, Cd and Be in non-branded chili were calculated to be less than 0.01 and Cr, Ni, Cu, Zn, Hg, Mn, Co, Fe, Al in non-branded chili were calculated to be, 0.04, 0.02, 0.02, 0.61, 0.04, 0.58, 0.00, 4.61, 2.72, respectively. All the finding values are below 1 except iron and aluminum. In case of branded chili the EDI value for As, Se, Pb, Cd and Be were calculated to be less than 0.01 and Cr, Ni, Cu, Zn, Hg, Mn, Co, Fe, Al were calculated to be 0.06, 0.02, 0.23, 1.12, 0.05, 0.45, 0.01, 2.65, 1.64 respectively. In branded chili most of the values are below 1 except zinc, iron and aluminum.

In case of non-branded turmeric, the EDI value for As, Se, Pb, Cd, Hg and Be were calculated to be less than 0.01 and Cr, Ni, Cu, Zn, Hg, Mn, Co, Fe, Al in non-branded turmeric were calculated to be 0.03, 0.01, 0.07, 0.45, 0.00, 1.43, 7.78, 4.38 respectively. All the values for heavy metal are below 1 except iron, aluminum and manganese. For branded turmeric the EDI value for As, Se, Cd, Hg, Co and Be were calculated less than 0.01 and Pb, Cr, Ni, Cu, Zn, Mn, Fe, Al, were calculated to be 0.02, 0.03, 0.011, 0.05, 0.74, 0.00, 1.37, 5.01, 17.68 respectively. Here also the iron, manganese and aluminum give higher value than 1.

Chapter 4: Discussion

The concentration of all the heavy metals found in all the samples were within the acceptable range. But it has been clearly seen that the concentration of Fe, Al, Mn and Zn are quite higher than all other elements and it is not only for non-branded samples but also for branded samples. It occurs due to heavy explosion of this metal to the environment and for not using proper waste metal disposal in those industries. Though they have higher concentration, they are within the threshold limit set by WHO. Besides, their TTHQ value is very high which indicates that they are producing a heavy negative effect on the consumed population. So, it is said that though they are not producing any carcinogenic effects, they are producing non carcinogenic effects and it will create many difficulties in the long run for the consumed population.

For example, the concentration of iron reported in Ethiopia is comparable to that in the present study. The highest concentration of iron in this study is detected at 272.43 ppm which is much higher than that found in Ethiopia (Adugna et al., 2024). Excessive iron intake may lead to iron poisoning, with initial symptoms including abdominal pain, nausea, and vomiting, that can progress to more serious conditions such as cirrhosis, heart failure, and diabetes (Adugna et al., 2020). From the data analysis it is clearly seen that the iron concentration in all the samples is quite high. It occurs due to heavy iron exposure in the environment, more specifically, the cultivated field.

Notably, non-branded turmeric displayed the highest manganese concentration at 50.52 ± 30.32 mg kg⁻¹, which aligns with the reported range of manganese (184–401 mg kg⁻¹) found in Ethiopia (Adugna et al., 2020). This elevated concentration may be attributed to the presence of heavy metals in the relatively acidic soils of the agricultural land.

At higher doses, intake of Al can also cause many diseases. Such as, it can cross the blood brain barrier and cause oxidative stress. Aluminum leaching from acid soils and industrial effluents can lead to soil and water acidification, affecting plant and aquatic organism health (Khan et

al., 2010). The highest concentration of aluminum found in this study is 618.87 ppm which is lower than that highest concentration of aluminum 1222 ppm found in the spices in Tehran, Iran (Dariyan et al., 2025).

In addition, higher concentrations of zinc in the human body can be harmful due to their interference with copper metabolism. As a result, copper deficiency can lead to anemia, low white blood cell count and neurological problems such as weakness, numbness and coordination problems. The Zn concentrations in this study (16.05 to 39.2 mg/kg) are appreciable in comparison with another study in Tehran, Iran (Dariyan et al., 2025). However, when these results are compared with previous studies Zn levels with results are higher from Oromia regional state, Ethiopia (Adugna et al., 2024).

The highest concentration of manganese 50.52 ppm is found in non-branded turmeric which is much higher than the previous study of Mn found in Ethiopia (Adugna et al., 2024). But it is quite lower than the study found in Iran (Dariyan et al., 2025). Furthermore, higher amounts of manganese cause muscle spasm, hearing problems, mood changes and insomnia. Besides, in severe cases it can hamper brain development, behavioral and learning problems, especially in children and infants.

Moreover, it is clearly seen that almost all the samples for human consumption are above 1 which indicates that they are facing chronic health impact. Besides, the assumed population is in great danger. Therefore, it is also indicating that the consumed population are at great risk for heavy metal contamination and suffering metal causing diseases (Oladeji et al., 2023).

To prevent heavy metal contamination the sources of contamination should be identified and take necessary steps to control the contamination. For example, proper ETP should be established and treat the waste water before releasing it. Lined landfills and safe disposal

methods should be used for e-waste, batteries and medical waste. In addition, industries should develop technologies that use less or no use of heavy metals e.g. alternative to lead, mercury cadmium in production (Singh, P.et al.,2022). Strict laws and monitoring should be established for less emission from factories, mining and smelting. National and international limits should be followed (WHO, 2023). Moreover, organic matter should be used in the cultivated fields to reduce metal bioavailability. In addition, artificially bright colored powder should be avoided and store spices in air tight glass and food grade containers (Shrivastava, K.et al.,2020). Low metal fertilizer and certified pesticides are used to produce a low metal containing product. Moreover, sealed packaging should be maintained throughout the transportation process (FAO, 2018). Besides, spices should be stored in a dry and dust free environment (Rani et al.,2020). Therefore, further research on spice samples based on their cultivation is recommended to determine the hotspots that are responsible for iron, aluminum, manganese and zinc.

4.1 Limitations of the Study

As the study provided valuable information about heavy metal contamination in spices, several limitations should be acknowledged. Firstly, the sample was limited to only 27 and can't cover all areas of Bangladesh. Secondly, the samples were analyzed using only 14 heavy metals, excluding microbiological health indicators, which may also pose significant health risks. Moreover, the study didn't assess how heavy metals transferred or ingested in spices, which could give more comprehensive information of ecological risks.

4.2 Future research plan

Future research should focus on expanding the assessment of heavy metal contamination across a wide range of spice types and geographical regions to better understand regional variations and sources of contamination. Further studies may also explore the impact of agricultural

practices, processing and storage conditions on metal accumulation.

Chapter 5: Conclusion

To be concluded, the purpose of this study is to investigate the concentration and health risk assessment of essential metals (Fe, Zn, Cu, Cr, Mn and Ni) and non-essential metal (Cd and Pb) metals and the heavy metals (As, Hg, Se, Hg, Co) in some spices collected from local markets in Bangladesh. The samples are collected from different regions of Bangladesh and categorized into four categories such as, non-branded chili, branded chili, non-branded turmeric and branded turmeric. In this work, the ICP-MS method was used to determine the concentration of heavy metals from the samples. The carcinogenic effects are determined through the exceeding of the concentration and non-carcinogenic effects are determined through the EDI and TTHQ value. In this study, the concentration of iron, aluminum, zinc and manganese are higher than other elements and it is not only for the non-branded category but also for the branded category. However, their limits are within the range. But the alarming findings of this study is that all the categories of the samples are producing heavy non carcinogenic effects. The concentration of the essential and non-essential elements and the heavy metals are within the range but they create potential health risk because the TTHQ of the samples are quite higher than 1 and create a negative impact on the consumed population. Lastly, special attention should be given to the continuous monitoring of heavy metals, particularly non-essential metals because they are not easily excreted from the body and accumulate over time and pose a threat to consumers.

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