The Assessment of Contamination in Irrigation Water and Soil and Its Effect on Crops in Muzaffargarh District



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Supervisor: Dr. Khurram Yousaf

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DEDICATION

I dedicate this work to **my mother and siblings**, whose unwavering moral and financial support have been essential in the completion of this work. I also extend my deepest gratitude to my mentor **Dr. Shazia Jabeen** and my best friend, **Fatima Raheem**, whose continuous support and guidance have been invaluable throughout this academic journey. Their belief in my potential and their encouragement have been fundamental to the successful completion of this research.

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LIST OF SYMBOLS, ABBREVIATIONS AND ACRONYMS

µg/L	Micrograms per liter
mg/kg	Milligrams per kilogram
BCF	Bioconcentration Factor
BAF	Bioaccumulation Factor
BTF	Biotranslocation Factor
HQ	Hazard Quotient
CR	Carcinogenic Risk
pН	Power of Hydrogen
EC	Electrical Conductivity
EC (µS /m)	Electrical Conductivity in Micro Siemens per meter
CSF	Cancer Slope Factor
ADD	Average Daily Dose
RfD	Reference Dose
С	Concentration of arsenic in drinking water
IR	Ingestion Rate
ED	Exposure Duration
EF	Exposure Frequency
BW	Body Weight
AT	Averaging Time
WHO	World Health Organization
EPA	Environmental Protection Agency

SPSS	Statistical Package for the Social Sciences
GIS	Geographic Information System
LOD	Limit of Detection
HPLC	High-Performance Liquid Chromatography
ICP-MS	Inductively Coupled Plasma Mass Spectrometry
AAS	Atomic Absorption Spectrophotometer
CEC	Contaminants of Emerging Concern
РТМ	Potential Toxic Metals
As	Arsenic
Cd	Cadmium
Hg	Mercury

- Cu Copper
- Ni Nickel

ABSTRACT

Agriculture is a cornerstone of Pakistan's economy, causative significantly to GDP and providing livelihoods for much of the rural population. However, the sustainability of this sector is increasingly threatened by environmental contaminants, hefty metals like arsenic. Arsenic contagion in drinking water and soil poses serious risks to crop yields, food safety, and municipal health, especially in the Muzaffargarh district of Punjab, where groundwater is heavily used for irrigation. This research examines the extent of arsenic contamination in the Muzaffargarh district by analysing arsenic levels in groundwater, soil, and wheat crops, and assessing the related health risks. The study exposes that arsenic absorptions in groundwater across 18 sampled sites range from 17.3 µg/L to 294.0 µg/L, with an normal of 76.88 µg/L far exceeding the "World Health Society's" safe limit of 10 µg/L. Soil samples show arsenic levels between 18.65 mg/kg and 68.29 mg/kg, with an mean of 39.95 mg/kg, surpassing the 20 mg/kg agricultural safety threshold. In wheat plants, arsenic absorptions in roots range from 10.236 mg/kg to 34.46 mg/kg, while grains contain between 1.279 mg/kg and 18.16 mg/kg, indicating significant uptake of arsenic. The study employs bioconcentration, bioaccumulation, and biotranslocation factors to evaluate arsenic mobility within wheat plants, revealing substantial absorption by roots but limited translocation to aerial parts. Health risk assessments show alarmingly high hazard quotient values between 1672.9 and 9936.1, and carcinogenic risk values ranging from 0.7528 to 4.4713, highlighting significant public health risks for communities reliant on this polluted water for drinking and agriculture. The findings highlight the urgent need for mitigation policies, including the cultivation of arsenic-resistant crops, regular monitoring of soil and water quality, and public health interventions to reduce arsenic exposure, essential for ensuring food safety and sustaining agricultural productivity in the Muzaffargarh district.

Key Words: Arsenic contamination, Groundwater, Soil, Wheat crop, Bioaccumulation, Translocation, Muzaffargarh district, Health risk assessment, Agricultural productivity, Food safety, Environmental contaminants.

CHAPTER 1: INTRODUCTION

The farming area holds a important role in the global frugality, serving as a fundamental pillar of economic stability and growth (Svatoš 1999). Agriculture plays a pivotal role in Pakistan's economy, ranking just behind the services sector in its contribution to the national GDP. Specifically, it accounts for about 18.9% of the country's GDP. Additionally, around 63% of Pakistan's population, predominantly residing in rural areas, depend on cultivation for their occupations, either directly or indirectly. This sector is crucial for employment and economic stability in these regions, encompassing both crop production and the equally vital sub-sector of livestock (Kakar et al, 2016).

The "agricultural sector" is pivotal in enhancing food accessibility and securing food supply, thereby playing a crucial role in achieving food security (Wegren and Elvestad 2018). Although it is widely acknowledged that global food demand will rise significantly in the coming decades, there are doubts about whether global cultivation can meet this claim over increased food creation. ('Cook et al, 2011'). Considering the current trends and future projections, it is crucial to significantly boost agricultural production by an estimated 60% to 70% to adequately supply food for the anticipated global population by the year 2050 (Silva George 2018).

Heavy metals and metalloids are widely acknowledged as environmental contaminants (Maksymiec 2007). Dense metals are logically present in the environment and are essential for various biological functions. However, they can become harmful when they accumulate in living organisms (Mitra et al, 2022). Certain heavy metals, such as copper, zinc, selenium, manganese, nickel, cobalt, chromium, and molybdenum, are crucial for essential biological functions and thus contribute positively to crop productivity. (Salla, Hardaway, and Sneddon 2011) Iron such as zirconium, mercury, antimony, arsenic, and cadmium, which do not play any essential roles in metabolic processes, can substantially decrease crop productivity when their concentrations exceed optimal levels (Shahid et al, 2015). Components such as cadmium (Cd), and arsenic (As), and are considered nonessential, as they do not contribute any beneficial roles in plants, animals, or humans. Additionally, these elements have no nutritional value and are highly toxic (Khan et al, 2015).

"Heavy metal contamination" in soil and water represents a significant environmental challenge, severely impacting food safety, human health and crop production. The accumulation of these toxic rudiments in farming soils poses a threat to crop production, food

eminence, and the complete sustainability of agricultural systems. Yields can uptake heavy elements from polluted soils over their cause, with these metals subsequently translocating to various plant parts, including those consumed by humans. This process creates potential pathways for human exposure to these toxins. Consequently, heavy metals pose toxicity risks to both plant health and human well-being (Vasilachi, Stoleru, and Gavrilescu 2023).

Arsenic is widely familiar as a potent pollutant and is regarded as one of the most hazardous chemicals globally (Shankar, Shanker, and Shikha 2014). Arsenic pollution arises from human activities and both natural. Natural processes include enduring, mineral dissolution, and biochemical reactions. Human activities contributing to arsenic contamination include mining, the extreme use of arsenic-based insecticides or herbicides, and the discharge of industrial effluents. This pollution has significantly harmed both soil ecosystems and aquatic systems (Sevak and Pushkar 2024). As per the 2022 ranking by the Agency for "Toxic Substances and Disease Registry" (ATSDR), arsenic is identified as the most toxic metalloid (ATSDR 2023). "The International Agency for Research on Cancer" (IARC) classified arsenic as a Group 1 human carcinogen (Van Halem et al. 2009). Arsenic, a human carcinogen, can negatively impact human health level at little absorptions of 0.002 mg/L. (Jang, Somanna, and Kim 2016)

Arsenic pollution in groundwater is a world-wide problem (Rasool et al. 2021). The widespread digestion of arsenic polluted eating water has adversely impacted over 2 million individuals globally (Ullah et al. 2023a). Reports indicate that around 70% of groundwater and surface water resources in Pakistan are polluted by a combination of organic, biological pollutants and inorganic (Shahid et al. 2018). A significant number of individuals in the country may be consuming arsenic-contaminated drinking water, putting them at elevated risk for health complications. In certain regions of Pakistan, many shallow subsurface aquifers and tube wells comprise arsenic absorptions that greatly surpass the suggested frontier of 10 ppb (Malik et al. 2009; Shahid et al. 2018b; Shakoor et al. 2015).

Arsenic impurity in groundwater also presents a global challenge to irrigation supplies, posing substantial food security and safety concerns, especially in regions reliant on groundwater for agricultural purposes. This contamination arises chiefly from the natural dissolution of arsenic from sedimentary iron oxides under reducing conditions. Affecting over 70 countries, this issue impacts approximately 19.7% of global crop production on irrigated lands tainted with arsenic. Over time, the build-up of arsenic in these soils threatens both current and future agricultural productivity, thereby exacerbating food security challenges (Sun et al. 2024).

The substantial addition of arsenic in cultivated soils facilitates its handover through the food chain, ultimately leading to human exposure and posing severe health risks (Rehman et al. 2021). Raised levels of arsenic in soil suggestively impair fertility and soil properties, harm valuable soil microbes, and upset plant physiological processes, prominent to inhibited plant development and growth. These disruptions subsequently disturb the food chain and food web (Zhang and Yan 2021).

Wheat is recognized as the second most vital cereal crop globally, with production volumes comparable to those of rice. It serves as a staple food, particularly in Europe, where the average adult's daily consumption of cereals and cereal products is approximately 0.25 kilograms, predominantly comprising wheat-based items (Zhao et al. 2010). In Pakistan, wheat is cultivated by 80% of farmers, making it the second most-produced crop annually (Ali et al. 2024). The quality of wheat grains envisioned for human ingesting is influenced by a multitude of factors. These comprise ecological conditions such as temperature and rainfall, soil characteristics like organic and texture matter content, and agrarian performs involving the use of composts, insecticides, and herbicides. (Basit and Hussain 2024) However, wheat production is significantly impacted by its sensitivity to the harmfulness of possible toxic metals. These metals can harshly hinder its growth and yield (Ali et al. 2020).

Excessive and prolonged ingesting of lethal inanimate arsenic (As) over intake water and food over a dated of 5–10 years can lead to 'arsenicosis'. This term broadly refers to health issues associated with arsenic exposure, including skin cancers, skin disorders, internal cancers (affecting the bladder, kidneys, and lungs), vascular diseases in the legs and feet, potential diabetes, elevated reproductive disorders and blood pressure (Guidelines for drinking-water quality: fourth edition; 2017; Santra et al. 2013).

Muzaffargarh is a significant district in Punjab, particularly in terms of agricultural productivity (Akram et al. 2014). In Muzaffargarh, as in other parts of Punjab, wheat is a staple crop (Muzaffargarh district gazette; 2019). The agricultural area in Muzaffargarh is irrigated using both canal and groundwater sources. This study specifically focuses on wheat, which is irrigated using groundwater accessed through tubewells. Canal water is available from May to August, while the wheat sowing and harvesting season in Muzaffargarh spans from November to April. The underground water quality in Muzaffargarh district fails to meet WHO standards, particularly after the August 2010 flood, which led to substantial degradation due to stagnant floodwaters. The district comprises three distinct water zones: brackish, sweet, and

contaminated, with arsenic contamination emerging as a critical issue. As an industrial hub with around 50 industrial units, Muzaffargarh experiences daily deterioration in underground water quality (Ullah et al. 2024).

The quantification of arsenic in in Muzaffargarh district was last conducted in 2005, (Nickson et al. 2005a) and no subsequent readings have been carried out to reassess the current arsenic levels. This leaves a significant gap in understanding the present condition of groundwater arsenic contamination. Furthermore, even though soil arsenic concentrations and rice-grain arsenic levels have been investigated in detail, this area of research remains understudied in the case of wheat. This shortage is felt most in Pakistan and in particular in Muzaffargarh district where the authors of this paper found that no exploration has been directed to determine the extent of arsenic build-up in wheat. Besides, there is no study available to measure the absorption of arsenic in the soil of Muzaffargarh district. The purpose of this study will be to make some advances in filling these gaps through analysing the arsenic absorptions in portable water, measuring arsenic in the soil, and exploring the arsenic uptake in the wheat crops of Muzaffargarh district.

2.1 Objectives:

The primary aims of this study are to:

- 1. Quantify the concentrations of arsenic in irrigation water and soil within the Muzaffargarh district.
- 2. To measure the health hazards connected with arsenic exposure in drinking water in the Muzaffargarh District.
- Conduct a study on the bioaccumulation and translocation of arsenic in Triticum aestivum (wheat) with a special reference to the approval of arsenic and its mobility in the plant.

3 CHAPTER 2: LITERATURE REVIEW

3.1 Agricultural Sector in Pakistan

3.1.1 Importance and Contribution to the Economy

Pakistan appreciates a diversified structure of economy where the agriculture sector has significant contribution, it contributes 24% in the gross domestic product (GDP) and occupies 37%. 4% of employment. The predominance of agriculture in the economy underscores its importance as a key driver of employment, economic growth, and poverty discount. The agricultural sector's extensive linkages with other sectors further emphasize its critical role in the broader economic framework, as it not only supports food security but also stimulates growth in related industries (Pakistan Economic Survey 2023-24). (Islam et al. 2023) noted that the agricultural sector also makes a substantial contribution to Pakistan's foreign exchange earnings. According to (Baig and Khan 2006), this sector supplies essential raw materials to domestic agro-based industries, including sugar, vegetable processing, leather, and textiles. Consequently, the significance of agriculture to Pakistan's economic development and the welfare of its population cannot be overemphasized.

3.1.2 Employment and Livelihood in Rural Areas

Previous research in Pakistan indicates that agriculture continues to be a critical sector for employment, involving approximately 65% of the rural populace and accounting for 38% of the overall national labor force (Agricultural Statistics of Pakistan 2021-2022). (Rasheed et al. 2024) emphasizes that farming is a main source of employment, food, and income, particularly for the rural population, which forms the popular of the country's inhabitants. Pakistan is the fifth most crowded nation globally, with approximately 34.7% of its citizens living in urban areas, while the remaining population resides in rustic areas and depends severely on farming for their livelihood.

3.1.3 Crop Production and Livestock Sub-sectors

Crop production in Pakistan primarily consists of three main cropping systems: Such systems include cotton-wheat, rice-wheat, and mixed cropping systems. A brief introduction to the major cash crops of Pakistan, weed fields of which are burning with great intensity in the Thar desert these days, includes raw cotton, wheat, and rice which, in total, constitute 3%. Contributes about 6% of the GDP and gives a share of 44% to the food basket of the country (Shahzad et al. 2019). As cited in (Azam and Shafique 2017) cotton, wheat, sugarcane, rice, and maize are the main crops that form a large part of Pakistan's GDP and agricultural value chain, providing food security and export returns. The livestock sector which holds 58% of the total production is one of the most significant sections of the Bangladeshi economy. 11% of agricultural value as cess for the development of the agriculture sector. This 6 percent of GDP is critically important for rural economy and poverty reduction. Also, the poultry and fisheries industries contribute tremendously to the economy through production and employment. The sustainable management of these sectors is integrity to the smooth running of the economy and the improvement of the standard of living of people in the rural areas.

3.2 Global Food Security and Agricultural Production

3.2.1 Increasing Food Demand and Production Challenges

(Chen et al. 2024) pointed out that increasing NO₃ production alone is insufficient to solve the multifaceted issues of food security. Crop production in the world was assessed to have stood at 2.790 billion tonnes, with an annual increase of 3 percent. Concurrently, the per capita consumption of food has been on an upward trend, thereby intensifying the overall demand for food resources. In previous studies (Galanakis 2024) told us that the global population is projected to spread nearly 9.7 billions by 2052, resulting in a marked upsurge in nourishment demand. Urbanization and economic development are major contributors to heightened food consumption, especially of processed and animal-derived products, which are resource-intensive to produce. Consequently, the escalating global food demand calls for significant enhancements in the efficiency and sustainability of food production to meet future needs without further environmental degradation. (Yang et al. 2024) observe that global food production must address the dual challenge of enhancing yields while maintaining environmental challenges, including soil degradation, water scarcity, and loss of biodiversity.

(McKenzie and Williams 2015) concludes soil degradation, water shortages, and the overuse of fertilizers and pesticides have depleted vital natural resources, creating substantial obstacles for food production.

3.2.2 Importance of Boosting Agricultural Productivity

(Baig et al. 2023) noted that the Positive development in farming production is a significant factor in the advancement of the farming segment and the economic development of nations. Since the time of Smith Adam, scholars have generally agreed that the rate of agricultural output growth is slower associated to other areas of the frugality. (Banerjee et al. 2024) focused on enhancing agricultural productivity which is crucial to address the increasing global food demand driven by population growth and environmental degradation. This requires efficient resource utilization, the adoption of advanced technologies, and sustainable farming practices. Such measures ensure food security, stimulate economic growth, and alleviate poverty, particularly in rural regions. (Shah et al. 2024) also emphasizes the critical importance of increasing agricultural productivity to satisfy global food requirements. Enhanced agricultural productivity is essential for augmenting food availability, alleviating poverty, and fostering economic development, especially in developing countries. He argues that increasing productivity can counteract the negative impacts of environmental degradation, secure sustainable food supplies, and support smallholder farmers' livelihoods. This previous study also underscores that technological advancements, better resource management, and sustainable agricultural practices are vital to realizing these objectives.

3.3 Environmental Contaminants in Agriculture

3.3.1 Definition and Types of Environmental Contaminants

Environmental contaminants encompass any physical, chemical, biological, or radiological substances or materials that adversely affect air, water, soil, or living organisms. These substances frequently originate from human activities, resulting in notable deviations from the standard environmental composition. When these contaminants occur in concentrations exceeding normal levels and exert harmful effects on the environment or valuable elements within it, they are classified as pollutants (D'surney and Smith 2005).

Types of Environmental Contaminants (Khanmohammadi et al. 2020)

1. Primary and Secondary Contaminants:

- Primary contaminants refer to substances that are harmful in the form they are initially released into the environment.
- Secondary contaminants emerge from chemical reactions occurring in the environment, often originating from less harmful precursor substances.
- 2. Chemical Contaminants:
- Thick elements: Such as mercury, lead, arseni, and, zinc And copper.
- Hydrocarbons: Including polychlorinated biphenyls, bisphenol A, catechol, and paracetamol.
- Organophosphorus compounds: Examples are methyl parathion, diazinon, and chlorpyrifos.
- 3. Biological Contaminants:
- These contaminants arise from biological entities including bacteria, yeast, fungi, viruses, prions, protozoa, or their toxins and by-products.

3.3.2 Heavy Metals and Metalloids as Contaminants

(Saidon et al. 2024) emphasizes that heavy metals, characterized by densities exceeding 5 g/cm³ and atomic numbers above 20, occur naturally. They originate from physical and chemical weathering, volcanic activities, and notably, anthropogenic sources such as industrial waste and urban runoff. These metals are swiftly dispersed among biotic components, presenting substantial environmental and health hazards due to their inherent harmfulness and long-term persistence (Angon et al. 2024) notes that heavy metal contamination represents a significant concern. Heavy metals and metalloids act as contaminants in farming soils, possessing the possible to adversely affect crop health and productivity when present at elevated concentrations. In the previous research. (Almotairy 2024) also suggests that Metalloid contamination represents a substantial global challenge, threatening food security, sustainable agriculture, and public health. Such contaminants interfere with photosynthesis, nutrient absorption, and oxidative stress regulation in plants, ultimately diminishing crop resilience and productivity.

3.4 Heavy Metal Contagion in Soil and Water

3.4.1 Causes and Pathways of Pollution

(Espíndola et al. 2024) reported that contaminants of emerging concern (CECs) predominantly infiltrate aquatic environments via wastewater effluents, agricultural runoff, and industrial

discharges. Key sources include pesticides, pharmaceuticals, and industrial chemicals. These pollutants are transferred from wastewater treatment facilities to surface waters and groundwater, where their persistence and bio accumulative nature present important hazards to both aquatic ecosystems and human health. (Jayakumar et al. 2021) focused on investigating the heavy metal contamination in soils which arises from both normal procedures, such as surviving and activity, and human events, including agricultural practices (use of fertilizers, pesticides, and biosolids) and industrial processes (mining, smelting, and wastewater irrigation). These contaminants spread through various pathways, including uptake by plants, water movement, atmospheric deposition, soil erosion, and bioaccumulation, thereby posing important hazards to human health and ecosystems. (Zheng et al. 2023) also emphasizes that soil pollution by heavy metals can result from both natural and human-induced factors, including geological processes, forest fires, irrigation, agricultural fertilization, practices, and industrial actions. As noted in the previous studies of (Jayakumar et al. 2021) hefty elements are hard to eradicate from soil and water since they are inorganic pollutants that are highly resistant and likely to accumulate in the body. All the above characteristics are a great menace to food protection and human health.



Figure 2.1: Sources and Pathways of Arsenic Contamination in the Environment (Rai et al. 2019)

3.4.2 Impact on Crop Productivity and Food Safety

According to the research done by ("Zhang et al. 2015") Heavy metals in the soil reduce crop yield significantly. Some pollutants which include cadmium (Cd), mercury (Hg), copper (Cu), and nickel (Ni) are engaged up by plant origins and reduce crop production and quality. Also, the heavy metals may be deposited in the soil, and therefore they are taken up by crops and then disbursed by people. In a study by (Qi et al. 2024), the author noticed the accretion of heavy metals in the soil which is quite dangerous in as much as it postures a risk to food safety since the metals can be taken up by crops and transferred to food products. (Guo et al. 2023) observed that the incidence of several illnesses linked to the ingesting of food products dirtied with heavy metals is on the rise, and includes cancers of the digestive system, cognitive impairments, and nutritional deficiencies. This paper titled, (Afonne and Ifediba 2020) focuses on the effect of heavy metal effluence from agricultural and industrial wastes on crop yield and food quality. He raises concerns about the effects of lethal elements in earths and plants that

are dangerous to hominoid well-being and poses danger to food security and safety and calls for measures to prevent such health risks and ensure quality and nutritional value of food.

3.5 Arsenic Contamination

3.5.1 Arsenic as a Global Environmental Challenge

(Bundschuh et al. 2022) examined that arsenic contamination is a critical global concern, affecting more than 200 million individuals across at least 105 nations. The predominant pathways for human exposure encompass the digestion of contaminated drinking water, ingesting of diet tainted with arsenic, and direct contact with arsenic-laden soil and water. (Genchi et al. 2022) noted that arsenic ranks as the 20th most plentiful metalloid in the Soil's outside and is widely acknowledged for its detrimental impact on human fitness. It disrupts numerous "cellular processes and impairs the functioning of various organs within the human body. (Bhat et al. 2024) reported that arsenic contamination affects water and soil in numerous countries, with groundwater being particularly impacted in areas where it is extensively used for irrigation, drinking, and food preparation. (Huang et al. 2024) observes that arsenic (As) is prevalent in various environmental substrates, primarily in compound forms such as inorganic arsenite (AsIII) and arsenate (AsV). Contamination of groundwater and drinking water with arsenic is a global issue, with significant concern also arising from arsenic accumulation in grains, particularly rice, which mainly contains the carcinogenic inorganic arsenic (iAs), and seafood, which typically has higher levels of organic arsenic (oAs). Grains and seafood, both essential components of the global diet, pose increasing health risks due to arsenic contamination.

3.5.2 Natural and Anthropogenic Sources of Arsenic

(Rajendran et al. 2024) examined that anthropogenic sources emit between 52,000 and 112,000 tons of arsenic annually. (Bundschuh et al. 2011.; Patel et al. 2023) defined that Human exposure to arsenic occurs through various pathways derived from both natural and humanmade sources. Geogenically, the Ground's shell is a major natural reservoir of arsenic, with regular concentrations of approximately 5 mg kg⁻¹, present in over 200 reserves, notably arsenopyrite. The natural sources include volcanic activities, the hydrothermal or geothermal activities, and weathering of minerals containing arsenic and sea water. Human activities including mining, metal producing and sweltering of fossil fuels are known sources of air, water and soil arsenic pollution. Furthermore, arsenic is used in agriculture to create arsenic-based pesticides, wood treatments, industrial processes, and waste disposal procedures that distribute arsenic throughout the environment. The natural and anthropogenic bases subsidise to the distribution of arsenic in the atmosphere, thereby requiring effective environmental and public health measures.

3.5.3 Health Risks Associated with Arsenic Exposure

According to the study conducted by (Rahaman et al. 2021), arsenic has been found to have many negative impacts on the health of the human body, such as skin diseases, neurological complications, and cancers. Long term intake of high concentration mineral arsenic over food and drinking water over a period of 5-10 years may "lead to arsenicosis", other skin ailments and malignancies of the skin and other internal organs including bladder, kidney and lung cancers. According to (kaur et al. 2024) chronic intake of low arsenic concentrations in drinking water results in skin manifestations including skin hyperpigmentation, increased keratinization, and small skin tumors. It has been associated with neurological problems such as peripheral neuropathy, cognitive impairments and developmental problems in infants. Acute arsenic poisoning, often from accidental ingestion or occupational hazards, is a significant risk with high-level exposure. (Mitra, Chatterjee, and Gupta 2020) reported that numerous experimental studies have demonstrated that chronic arsenic exposure contributes to the development of cancers in many organs, counting the skin, liver, urinary bladder, lungs and in humans.



Figure 2.2: Health Risks Associated with Arsenic Exposure (Dilpazeer et al. 2023)

3.6 Arsenic in Groundwater and Soil

3.6.1 Mechanisms of Arsenic Contamination in Groundwater

(Ali et al. 2019a) examined arsenic contamination in groundwater primarily arises through two key processes: the oxidative dissolution of arsenic-containing sulfide minerals in oxidizing environments and the reductive ending of iron oxides in anoxic conditions. Additionally, the mobilization of arsenic is influenced by pH-dependent desorption and adsorption processes, as well as microbial activity that alters arsenic speciation and availability. (Wang et al. 2023) reported that the primary mechanism of arsenic contamination in groundwater involves elevated temperatures that improve the reductive closure of arsenic-bearing iron and manganese oxides and promote the weathering of arsenic-containing silicates, important to the discharge of arsenic into the groundwater.

3.6.2 Impact on Irrigation and Agricultural Productivity

According to (Zhang et al. 2021), arsenic contamination affects nutrient uptake and essential metabolic processes in plants hence reducing crop yields and agricultural output. Measures such as good management practices and release of crops with genetic resistance are considered important in reducing these adverse effects. (Okorogbona et al. 2018) revealed that high levels of arsenic in irrigation water affect crop yield negatively. The occurrence of arsenic in irrigation water and soil has a negative influence on crop growth, yield, and quality thereby reducing the agricultural productivity. Additionally, it is a severe health concern since arsenic tends to accumulate in the edible parts of the crops. (Mishra et al. 2021) also explains that arsenic is also reported to affect the availability of water, which is used for irrigation and drinking, hence, agriculture output. This contamination put arsenic species in the environment, which is dangerous to plants, animals, and human beings.

3.6.3 Case Studies from Various Regions

The case study 20 Years of Arsenicosis Patients and Arsenic Contamination in a Community of Bangladesh investigates the long-term effects of arsenic-laden groundwater in Samta village. Over two decades, surveys conducted in 1997, 2002, 2008, and 2017 documented significant contamination in shallow tubewells and a considerable incidence of arsenicosis among villagers. Mitigation strategies, including the introduction of deep tubewells and pond sand filters, were implemented; however, arsenic exposure persisted as a critical health concern. The study underscores the partial recovery of some affected individuals due to improved water

sources and enhanced nutrition, while also emphasizing the ongoing difficulties in eliminating the contamination issue. (Akhtar Ahmad, Faruquee, and Haque Khan 2020)

The case study Human Health Risk Assessment and zoning of Arsenic and Nitrate Pollution in Groundwater of Farming Areas of the Twenty-Two Village with Geostatistics (Case Study: Chahardoli Plain of Qorveh, Kurdistan Province, Iran)" investigates the 'contamination of groundwater by arsenic' and nitrate in 22 villages within the Chahardoli Plain, Kurdistan Province, Iran. Utilizing geostatistical methods, the research assesses the spatial dispersal of these pollutants and their connected health risks. Results indicate that 73% of nitrate and 59% of arsenic samples surpass the World Health Organization's safety standards, representing a significant health risk, particularly for children. The study points to the need for appropriate measures to be taken and follow up to be made in order to protect the public's health (Solgi and Jalili 2021).

This case study describes an "urban community garden in south-eastern San Diego" as part of the case Mitigation and Monitoring and of Lethal Arsenic Accumulation and Dense Metals in Food Yields. During the four-year period of the research, the arsenic and heavy metal and content was assessed in different crops, which pointed to a high level of pollution in some cases. To this end, the researchers used raised beds with clean soil, and this removed any form of detectable contamination. This study therefore stresses the need for regular tracking and implementation of proper soil management measures to guarantee the safety of food produced in urban gardens. (Cooper et al. 2020)

3.7 Impact of Heavy Metals on Development and Crop Growth.

3.7.1 Mechanisms of Metal Uptake by Plants

In their study, (Dalvi and Bhalerao., 2013) identified that plants uptake and deposition of heavy metals occur through several different mechanisms. These include root exudates to mobilize the metal, specific root pathways for uptake, translocation from origins to shoots and sequestration in vacuoles or cell walls. (Ghuge et al. 2023) explained that the uptake of HMs in vegetation happens over the absorption of metal ions at the root level using specific plasma membrane transporters like ZIP and ABC transporters. These ions are then transported through the apoplastic and symplastic pathways into the root cells and may be stored in the vacuoles or transported to the sprout via the xylem and phloem muscles.

3.7.2 Bioaccumulation and Translocation in Crops

In its study published in 2024, (Goni et al. 2024) His research shows very high concentration of metals including iron, manganese, nickel, copper, lead, cadmium and arsenic in the roots as well as the comestible parts of plants in Bangladesh especially those irrigated with contaminated water. Rendering to (Bhattacharya et al. 2021), the arsenic content in the edible parts of plants is determined by the arsenic absorption in the earth in which the shrubberies are grown, and the flower's capacity to take up and transport the element to the edible plant parts. There is significant variation among plant species in terms of their arsenic uptake and tolerance levels. (Tudi et al. 2021a) examined that the bioaccumulation and translocation of trace rudiments in crops and soils pose significant risks not only to the location and food safety but also to human and animal health. These processes can adversely affect the reproductive, immune, and nervous systems through their integration into the food chain.

3.8 Wheat Production and Metal Toxicity

3.8.1 Importance of Wheat as a Staple Crop

(Shewry and Hey 2015) explored that wheat stands as the primary staple crop in temperate regions and its demand is rising in nations experiencing urbanization and industrialization. Beyond serving as a significant source of energy and starch wheat contributes considerable amounts of various important or health-beneficial components, containing vitamins (particularly B vitamins), dietary fiber, phytochemicals, protein, and. (Grote et al. 2021) also reported that wheat serves as the staple crop for around 35% of the world-wide populace. Over two-thirds of the world's wheat creation is utilized for human consumption, while around one-fifth is allocated for livestock feed. (Ehsan Elahi et al. 2024) also focused that in Pakistan, key staple crops include wheat, rice, maize, and sugarcane, with wheat and rice being the most prominent. (Noor Ahmed Memon; 2017) states that in Pakistan, wheat accounts for 60% of the average individual's daily diet. (Iqbal et al. 2024) observed that wheat, in Pakistan, is a major source of nutrients, widely consumed and cultivated across the country. It typically contains 1.5-2% fat, 60-80% protein, 2-3% mineral matter, and 2-2.5% glucose, though these values may vary by region and variety.

3.8.2 Metals Affecting Wheat Quality and Yield

(Lan et al. 2024) reported that crops can absorb and accrue dense metals from the earth, leading to potential health risks for both humans and animals. This issue is particularly alarming for staple crops like wheat, which are spent in large quantities and may contain high levels of these toxic substances. (Shukla et al. 2023) observed crops and vegetables grown in arsenic-contaminated regions exhibit altered growth and development, decreased yields, and increased arsenic accumulation. Although wheat, typically cultivated with less irrigation, primarily accumulates arsenate (As(V)), it is a growing carcinogenic risk. Approximately two in 10,000 individuals face carcinogenic risks from consuming arsenic-contaminated wheat, highlighting its significant contribution to human arsenic exposure even at lower accumulation levels.



Figure 2.3: "Effects of Heavy Metals on Wheat Crop" (Hussain et al. 2023)

3.9 Overview of Muzaffargarh District

3.9.1 Muzaffargarh's Agricultural Landscape

Muzaffargarh, situated in Punjab, Pakistan, is distinguished by its fertile soil and favourable climatic conditions, which support the cultivation of major crops including wheat, cotton, sugarcane, and a variety of fruits and vegetables. The district predominantly depends on canalbased irrigation systems, primarily facilitated by the Taunsa and Panjnad barrages. Nevertheless, the region contends with critical water quality problems such as arsenic contamination, salinity, and waterlogging. Groundwater arsenic pollution, driven by excessive use of tube wells, the presence of natural arsenic-bearing minerals, and various anthropogenic activities, remains problematic even during periods when canal water supply is unavailable. This situation is a chronic threat to crop production and human well-being.

The literature review points to the fact that there are gaps in the presently available literature on arsenic contamination in the Muzaffargarh district. Interestingly, the last survey on the groundwater arsenic concentration was done in 2005 which shows that there is a major data gap. In addition, although many researches directed on the accumulation of pollution in rice, little work has been done on the effects of arsenic on wheat specifically in Muzaffargarh. Furthermore, there is no data available about the absorption of arsenic in the soil of this area, which is important for the evaluation of the risk of uptake by plants and subsequent special effects on human suitability. The effect of arsenic on wheat growth and production has not been well-researched, which hampers the formulation of proper remediation measures. This study aims at addressing these research gaps through re-evaluation and estimation of the current absorption of arsenic in groundwater and soil together with the assessment of bio-uptake and mobility of arsenic in wheat crops. Thus, the results of this study provide a comprehensive risk assessment that can help agricultural and public health policies to develop strategies for combating arsenic contamination and ensuring food safety and local people's living.

CHAPTER 03: METHODOLOGY

4.1 Study Area

The floodplains of Pakistan are formed by the deposition of eroded sediments and soil brought down from the mountainous regions. Such sediments include various trace metals and nutrients and may alter the chemistry of the groundwater and surface water systems in the area. The mineralogical and geochemical properties of the Indus flood plain including the areas of Punjab and Sindh are closely associated with the effects of these mountains. Muzaffargarh District is "located in the southern region of Punjab Province", Pakistan, and has a whole space of 8,249 km with the geographical coordinates of the district being among "28°57' to 30°46' north liberty and 70°31' to 71°47' east longitude (Podgorski et al. 2017). It is located between the two major rivers, the Chenab Stream in the east" and the Indus "River in the west". This geography encompasses some of the Thal Desert and the forested areas bordering the Indus River.



Figure 3.1: (A) Study area maps: google imagery view (B) Sampling site distribution in Muzaffargarh district with managerial boundaries

The research was conducted in three tehsils within the district: These are Muzaffargarh Tehsil, which is in the center, Jatoi Tehsil in the southwest, and Alipur Tehsil in the southeast. Muzaffargarh has a semi-arid climate with hot and dry conditions prevailing in the district, the

hottest months being May to September. In the district, the cool breeze starts to prevail from mid-August and the climate remains moderate. Also, Muzaffargarh is much influenced by the monsoon rains that occur from July to September with an average precipitation of between 11mm to 45mm.

4.2 Sampling Strategy

The sampling strategy for this study involves the collection of three types of samples: Soil, groundwater, and wheat plants were the sources of the isolates used in this study. Each of the sample types will have 18 samples in total, to cover all the possibilities. The samples will be collected through a stratified random sampling method so that the data can be collected systematically and efficiently across the different strata of the study area. This method helps to make sure that every sub-group of the population is equally represented in the sample, thus making the results of the study more credible and accurate.

4.3 Samples Collection

4.3.1 Groundwater Sampling

For groundwater analysis, a total of 18 testers were calm from various tubewells at 18 different locations within the three tehsils, each at a depth of 100 feet or more. One-litre Pyrex glass bottles were used in this experiment and the carafes were first washed and prepared with purified water earlier sampling. Before the sampling, the water from the tubewells was allowed to flow for at least ten minutes. Field constraints, electrical conductivity (EC), containing temperature, and pH, were dignified on-site using a Hanna HI9829 multi-parameter analyzer (Ullah et al. 2023a). To preserve the water samples, 2 ml of "nitric acid" was added. The tasters were then deposited at 4°C in dry, dark conditions until they were sent for analysis.



Figure 3.2: (A) Ground water collection (B) Water preservation with nitric acid (C) Stored water

4.3.2 Soil Sampling

For soil sampling, a total of 18 samples were composed using a stratified random sampling method. These soil models were taken from the same areas where water tasters were collected, specifically from the soils irrigated by the same tubewells. Surface soil models (0–15 cm depth) were obtained from each site using a locally-made carbon steel spatula. The testers were then placed in "polyethylene zipper" bags for storage until they were prepared for analysis.



Figure 3.3: Soil Samples

4.3.3 Crop Sampling

For crop sampling, a total of 18 wheat plants, with six replicates each, were together from the same sites where soil samples were taken. The plants were placed in newspapers and stored until they were prepared for analysis.



Figure 3.4: Wheat Samples Collection
4.4 Samples Preparation

4.4.1 Soil Samples Preparation

For soil preparation, all specimen were conveyed to the test centre where they were air-dried. After drying, the tasters were sieved using a 2mm sieve, homogenized, and then stored in sealed bags. (Tudi et al. 2021b)



Figure 3.5: (A) Drying of soil samples (B) Grinding of soil samples (C) Sieving of soil samples for the digestion process

4.4.2 Wheat Samples Preparation

For the wheat samples, the roots, and grains were first separated and dressed with tap water to eliminate soil elements, tracked by cleaning with water. The cleaned portions were then cut into small parts and withered at 65°C in an air-dry oven for 48 hours until they achieved a constant weight. After drying, the samples were pulverized to a fine precipitate by using a crusher and mortar and grinder and then sieved through a 0. 2 mm sieve. The crushed models were finally kept in sealed bags to be used for other analyses. (Tudi et al. 2021b)





Character 3.6: Step by step preparation of wheat root samples





Shape 3.7: Step by step preparation of grain samples

4.5 Samples Digestion and Analysis

4.5.1 Soil Digestion

A 0.5 g dried soil taster was precisely evaluated and put into a "Teflon microwave vessel" and processed using a microwave with a 3:1 'mixture of focused hydrochloric acid and nitric acid'. The resulting suspension was "filtered through whatman filter paper and the filtrate" was then transferred to a plastic tube and the volume made up to 50ml with de-ionised water. (Khan et al. 2019)

4.5.2 Wheat Grains and Roots Digestion

All the dried samples, root and grain, is weighed to the nearest 0. It was mixed with concentrated HCl to give a total weight of 05 g and put into a Vessel. Each sample is then absorbed with 5 mL of intense 'nitric acid and 1 mL of "hydrogen peroxide" and left to stand for 24 hours. The testers are then heated at 150°C on an electric heating plate for digestion of the samples. The digested solutions are then allowed to cool to normal temperature before being watery to 25 mL with deionised water, 'stunned for ten minutes and then stored at 4°C'. (Tudi et al. 2021b)





Figure 3.8: Digestion Process of soil and wheat samples

4.5.3 Samples Analysis

Quantitative analysis of total arsenic (As) in wheat and soil crop tasters was done using an atomic immersion spectrophotometer. ("AAS, Elmer, Perkin Model 700, USA).

4.6 Calculation of Human Health Risk

To measure the possible effects of 'arsenic-contaminated' water on human health some assessment parameters have been calculated. These parameters include the carcinogenic risk (CR) average daily dose (ADD), and Azard quotient (HQ),

4.6.1 Average Daily Dose Evaluation of Arsenic (ADD)

The risks of ingesting arsenic-contaminated water were assessed using a model developed by the US EPA (Rehman et al. 2020; Shahid, Niazi, et al. 2018; US-EPA 2005). The ADD of arsenic due to ingestion of arsenic polluted water fluid is estimated through the following formula:

$$ADD = \frac{C \times IR \times ED \times EF}{BW \times AT}$$
------ (eq.3.1)

In the above equation, the symbol is used as; C for the absorption of arsenic in the drinking water in mg/L, IR for the water ingesting rate which is assumed to be 2L/day, ED for the exposure of 67 years for the assessment with the literature, EF for the contact frequency per year which is 365 days per year, BW for the body weight which is assumed to be 72kg, and AT for the usual epoch which is 24,455 days. (Rehman et al. 2020; Shahid, Khalid, et al. 2018c)

4.6.2 Hazard Quotient Calculation (HQ)

Health hazards of arsenic in groundwater over consumption of intake water that does not cause cancer can be expressed in terms of hazard quotient (HQ). The hazard measure for arsenic in groundwater in Muzaffargarh area was computed as follows (Epa and Factors Program 2011.; Rasool et al. 2017) (US-EPA 2005)

$$HQ = \frac{ADD}{RFD} - (eq.3.2)$$

Here, the term 'ADD' refers to the normal daily dose of arsenic, while 'RfD' represents the oral position dose for arsenic (0.0003mg/kg/day or 0.3μ g/kg/day). Risk assessment can be categorized as follows: if the HQ is less than one, then the risk is considered to be low or 'safe'; in contrast, if the HQ is greater than one, then the risk is perceived to be a 'potential health concern'. (Rehman et al. 2020)

4.6.3 'Calculation of Cancer Risk (CR) Assessment'

Moreover non-carcinogenic dangers arsenic is also considered to have oncogenic hazards (CR) to human health. The cancer risk was estimated using the formula below. (US-EPA 2005)

$$CR = ADD \times CSF$$
-----(eq.3.3)

In this equation, CR stands for the carcinogenic risk, while the cancer slope factor for arsenic is represented by 'CSF' and is equal to 1. 5 (mg/kg/day).

4.7 Assessment of 'Arsenic Mobility and Bioaccumulation' in Wheat Plants

4.7.1 Bioconcentration factor

Bioconcentration factor (BCF) is an essential parameter that helps in determining the uptake and build-up of pollutants in plants grown on contaminated soils. It is established by dividing the absorption of pollutant in the plant roots by the absorption of the same pollutant in the soil matrix and as depicted in the following equivalence. (Karimyan et al. 2020; Rezapour et al. 2019; Wang, Ji, and Zhu 2017)

$$BCF = \frac{C_{Root}}{C_{Soil}} - \dots - (eq.3.4)$$

Here the subscripts 'BCF' stands for bioconcentration factor, C_{Root} represents the absorption of arsenic in the roots of the wheat and C_{Soil} stands for the meditation of arsenic in the soil. This guide is useful in understanding the extent of Pollutant mobility from the soil to the plant and the capacity of the plant to uptake and store the Pollutant. High BCF values suggest that a given plant has a higher potential to uptake impurities from the soil and perhaps pass them on to higher trophic levels. (Karimyan et al. 2020)

4.7.2 Bioaccumulation factor

The bioaccumulation factor (BAF) is another parameter that can be used to characterise the transfer and concentration of a contaminant from the soil into the aerial parts of a plant. It is designed by dividing the concentration of the pollutant in the grass, scrap or the various parts of the plant by the absorption of the pollutant in the corresponding soil samples. This index is also useful for estimating the prospects of phytoremediation. The BAF of arsenic in wheat plants was estimated using the following formula. (Liu et al. 2019)

$$BAF Straw or Grain = \frac{C_{Straw or Grain}}{C_{Soil}} - \dots - (eq.3.5)$$

For example, $BAF_{Straw or Grain}$ shows the bioaccumulation factor of arsenic in wheat straw or grain correspondingly. Furthermore, $C_{Straw or Grain}$ is the concentration of arsenic in straw or "grain samples", and C_{Soil} is the concentration of arsenic in the related soils. BAF values better than 1 indicate a higher concentration or more mobility of arsenic into different parts

of the wheat plant. (Karimyan et al. 2020)

4.7.3 Biotranslocation factor

The "biotranslocation factor" (BTF) describes the capacity of a pollutant to move from the plant's roots to its airborne shares or grains. This index is well-defined as the ratio of the contaminant concentration in various parts of the plant to its concentration in the plant's roots. In this study, the bio translocation of arsenic in wheat plants was deliberate as follows. ("Liu et al. 2019; Rezapour et al. 2019")

$$BTF_{Straw or Grain} = \frac{C_{Straw or Grain}}{C_{Root}} - \dots - (eq.3.6)$$

Here, $C_{Straw or Grain}$ denotes the concentration of arsenic in the wheat grain, or straw correspondingly; C_{Root} has already been defined as the absorption of arsenic in the wheat root. (Karimyan et al. 2020)

4.8 Quality Control

All chemicals used in this experiment were of analytical grade. Solutions were prepared using deionized water provided by the Department of Agricultural Sciences & Technology at the National University of Sciences & Technology, Pakistan. For quality assurance, all samples were analyzed in triplicate. Additionally, blanks and standards were included after every batch of 10 samples to ensure accuracy and reliability.



Figure 3.9: Experimental layout

CHAPTER 04: RESULTS

5.1 Groundwater Quality of Study Area.

The groundwater quality in the study area reveals significant concerns, particularly regarding the presence of contaminants that exceed safe limits for agricultural and drinking purposes. Analysis of the groundwater samples shows elevated levels of certain pollutants, including heavy metals and other environmental contaminants, which pose a risk to both crop production and public health. These findings underscore the need for urgent intervention, including improved monitoring and management practices to mitigate the adverse effects on agriculture and the local population. The results highlight the critical importance of addressing groundwater quality issues to ensure the sustainability of agricultural productivity and the safety of water resources in the region.

comparison with who recommended mints.								
Parameters	Units	Min	Max	Average	SD	WHO	%	%
						recommended	within	out
							limit	of
								limit
EC	µS/cm	393	1350	868.28	222.705	1000	66.67	33.33
pН	_	7.05	7.78	7.338	0.1783	7.0-8.5	100	0
Arsenic	ug/I	17.30	294	76.880	71.357	10	0	100
(4.10)	~~ L							

 Table 4.1: Statistical summary of analyzed physicochemical parameters and their comparison with WHO recommended limits.

The analyzed results of various physicochemical parameters in the groundwater tasters from the study area are summarized as follows. 'Electrical conduction' (EC) figure ranges from 393 to 1350 μ S/cm, with a regular of 868.28 μ S/cm. While 66.67% of the samples fall within the WHO recommended limit of 1000 μ S/cm, 33.33% exceed this threshold, indicating moderate enrichment of salts. The pH values of the groundwater samples range from 7.05 to 7.78, with an average of 7.3383, and all samples remain within the WHO permissible range of 7.0 to 8.5, signifying alkaline conditions. The arsenic concentrations range from 17.30 to 294.00 μ g/L, with a normal of 76.8805 μ g/L. Notably, 100% of the models surpass the WHO recommended limit of 10 μ g/L, indicating a significant occurrence of arsenic in the groundwater.

	Arsenic in Water	pH of water	Temperature	Electrical Conductivity
Arsenic in Water	1			
pH of water	0.160	1		
Temperature	0.466	0.058	1	
Electrical Conductivity	-0.025	-0.544*	-0.355	1

 Table 4.2: Pearson Correlation Coefficients Among Arsenic Concentration in Water, pH, Temperature, and Electrical Conductivity (EC)

N=18 samples, * Significant at 5%

The Pearson correlation analysis indicates that there is a weak positive connexion among arsenic concentration in water and pH (r = 0.160), as well as a moderate positive connection among arsenic absorption and temperature (r = 0.466). The correlation between arsenic concentration and electrical conductivity (EC) is both weak and negative (r = -0.025). Notably, a statistically significant negative correlation is observed between pH and EC (r = -0.544, p = 0.020), implying that higher pH levels are associated with lower electrical conductivity in the water samples. The relationships between temperature and both pH (r = 0.058) and EC (r = -0.335) are weak and statistically insignificant, reflecting minimal interdependence among these parameters in the context of the analyzed data.

The "correlation coefficient matrix" between various analysed "groundwater parameters" in the Muzaffargarh area is presented in Table 4.2. To further illustrate the relationships among these parameters, a heatmap is provided in Figure 4.1. The heatmap visually represents the direction and strength of the correlations between the groundwater parameters.



Figure 4.1: Correlation Matrix Heatmap

5.2 Arsenic Contamination in Muzaffargarh District

"Arsenic absorptions in portable water in the Muzaffargarh area exhibit a wide range, from 17.3 to 294.0 μ g/L, with an average concentration of 76.88 μ g/L. This indicates significant variability in arsenic levels across different sampling points. All analyzed samples exceed the WHO suggested intake water limit of 10 μ g/L'. Figure 4.2 (a) depicts the frequency of arsenic detection in groundwater samples, showing that arsenic levels in all samples are above the WHO safe limit. Figure 4.2 (b) provides a detailed view of arsenic concentrations at individual sampling points. It is evident that even the lowest measured concentration (17.3 μ g/L) significantly exceeds the permissible level. Notably, samples 6 and 8 exhibit exceptionally high arsenic absorptions, reaching up to 294.0 μ g/L, which is nearly 30 times the WHO recommended limit.



Figure 4.2: (A) & (B): Arsenic concentration in groundwater samples from the study area relative to the maximum allowable limit.

5.3 Spatial Distribution of Arsenic

The altitudinal circulation of arsenic concentration in the Muzaffargarh district, divided into Muzaffargarh, Jatoi, and Alipur tehsils, is depicted in the provided map. Alipur tehsil, located in the southern part of the district, exhibits the lowest arsenic levels, with concentrations below 23 μ g/L, represented in green. Adjacent areas in Jatoi tehsil show slightly higher arsenic levels

ranging from 23 to 40 μ g/L, depicted in light green. The central parts of the district, including portions of Muzaffargarh and Jatoi tehsils, display arsenic absorptions of 40 to 50 μ g/L, marked in yellow. Areas with arsenic concentrations between 50 and 70 μ g/L, shown in light orange, are also concentrated in the central regions. Higher levels of 70 to 80 μ g/L, represented by darker orange, are found in specific patches within Jatoi and Muzaffargarh tehsils. Areas surrounding the town of Muzaffargarh exhibit significantly elevated arsenic levels of 80 to 100 μ g/L, shown in red. The most extreme absorptions, reaching from 100 to 110 μ g/L and μ g/L, depicted in dark red and the darkest red respectively, are predominantly located in the central part of Muzaffargarh tehsil.



Figure 4.3: Spatial Distribution of Arsenic Concentrations in Muzaffargarh District

5.4 Health Risk Assessment

The health risk valuation conducted for groundwater in the Muzaffargarh district reveals significant concerns regarding arsenic contamination. The analysis determined that arsenic concentrations in the samples ranged from 0.0495 mg/L to 0.0844 mg/L, which is notably high. The "average daily dose" (ADD) values, intended using the US EPA model, varied among 0.501875 and 0.855722 mg/L, suggesting that residents are exposed to considerable levels of arsenic through groundwater consumption. The hazard quotient (HQ), a measure of the likelihood of non-carcinogenic effects, was determined to be between 1672. 917 to 2852. 407.

Sample ID	As (mg/L)	ADD	HQ	CR
1	0.079	0.803	2676.667	1.204
2	0.060	0.610	2034.537	0.915
3	0.067	0.680	2267.731	1.020
4	0.049	0.501	1672.917	0.752
5	0.084	0.855	2852.407	1.283
6	0.214	2.175	7252.685	3.263
7	0.125	1.268	4227.917	1.902
8	0.294	2.980	9936.111	4.471
9	0.075	0.765	2551.620	1.148
10	0.038	0.394	1316.366	0.592
11	0.077	0.783	2612.454	1.175
12	0.017	0.175	584.675	0.263
13	0.026	0.266	888.842	0.399
14	0.037	0.381	1270.741	0.571
15	0.023	0.239	797.592	0.358
16	0.051	0.523	1743.889	0.784
17	0.038	0.393	1311.296	0.590
18	0.022	0.231	770.555	0.346

 Table 4.3: "Average Daily Dose (mg/kg/day), Hazard Quotient, and Cancer Risk Assessment

 Values for Groundwater in the Study Area"

*ADD: Average Daily Dose, *HQ: Hazard Quotient, *CR: Cancer Risk Assessment

A staggering 73. 3% of the samples also had a very high Hazard quotient values above the safe limit of 1 in the areas showing high risk of possible health impacts to the large population depending on this water source. In addition, the carcinogenic risk (CR) values which estimate the lifetime probability of developing cancer because of arsenic exposure were determined to lie between 0. 752813 to 4. 47125. These CR values are extremely high, sixteen of them are above the generally accepted 5% risk level usually established by health departments indicating very high risk of cancer due to long-term consumption of water from the impacted aquifer. Such findings presume a dire importance of having appropriate and timely actions taken towards decreasing the prevalence of arsenic in the Muzaffargarh district's groundwater.

5.5 Descriptive Analysis of Soil Properties and Arsenic Content in Wheat Samples

The descriptive statistics reveal that arsenic absorptions in soil range between 18.650 and 68.290 mg/kg, with an average of 39.94956 mg/kg, while arsenic levels in wheat grains vary from 1.279 to 18.160 mg/kg, averaging 5.23728 mg/kg, and in wheat roots from 10.236 to 34.460 mg/kg, with an average of 20.39572 mg/kg

Descriptive Statistics							
Variables	Ν	Minimum	Maximum	Mean	Std. Deviation		
As Soil	18	18.650	68.290	39.94956	13.417506		
As Grains	18	1.279	18.160	5.23728	4.360900		
As Roots	18	10.236	34.460	20.39572	7.569580		
EC (1:1)	18	0.07	2.72	0.8089	0.74886		
pH (1:1)	18	7.79	8.97	8.2144	0.23781		
OM %	18	0.550	2.050	1.24167	0.315897		
N-NO ₃ - mg/kg	18	1.03	6.90	2.5006	1.59008		
P mg/kg	18	1.91	4.77	3.6011	0.85303		
K mg/kg	18	24	334	146.11	97.933		
Valid N (listwise)	18						

Table 4.4: "Descriptive Statistics of Arsenic Content and Soil Characteristics in Wheat Samples"

The soil samples show an average electrical conductivity of 80,890 μ S/m, a pH of 8.2144, an organic matter content of 1.24167%, nitrate nitrogen levels averaging 2.5006 mg/kg, phosphorus content averaging 3.6011 mg/kg, and potassium content with a broad range and an average of 146.11 mg/kg.

5.6 Correlation Analysis of Soil Properties and Arsenic Content in Wheat Samples

Correlations									
	N-NO3-								
	As Soil	As Grains	As Roots	EC (1:1)	pH (1:1)	OM %	mg/kg	P mg/kg	K mg/kg
As Soil	1	-0.181	-0.124	0.021	-0.274	-0.038	-0.164	-0.419	-0.054
As Grains	-0.181	1	0.163	0.271	-0.152	0.001	0.152	.576*	-0.325
As Roots	-0.124	0.163	1	-0.360	0.210	-0.215	-0.248	0.166	-0.279
EC (1:1)	0.021	0.271	-0.360	1	491*	0.294	0.413	0.057	0.105
pH (1:1)	-0.274	-0.152	0.210	491*	1	-0.247	0.054	-0.088	-0.037
OM %	-0.038	0.001	-0.215	0.294	-0.247	1	.569*	-0.136	0.098
N-NO3- mg/kg	-0.164	0.152	-0.248	0.413	0.054	.569*	1	-0.008	0.225
P mg/kg	-0.419	.576*	0.166	0.057	-0.088	-0.136	-0.008	1	-0.367
K mg/kg	-0.054	-0.325	-0.279	0.105	-0.037	0.098	0.225	-0.367	1

Table 4.5: Pearson Correlation Coefficients Among Soil Properties and Arsenic Content in Wheat Samples

N=18 samples, * Significant at 5%

The correlation analysis indicates that electrical conductivity (EC) has a significant negative correlation with soil pH (-0.491, p = 0.039), demonstrating that as EC increases, the pH value tends to decrease, leading to more acidic soil conditions. Additionally, organic matter percentage (OM %) is significantly positively correlated with nitrate nitrogen (N-NO₃-) content (0.569, p = 0.014), suggesting that higher organic matter levels are associated with increased nitrate content in the soil. Other correlations in the dataset, including those between arsenic content in soil, grains, and roots, as well as with phosphorus and potassium levels, are generally weak and not statistically significant. For instance, arsenic in soil shows weak negative correlations with arsenic content in grains (-0.181, p = 0.473) and roots (-0.124, p = 0.625),

while arsenic in grains exhibits weak positive correlations with arsenic in roots (0.163, p = 0.518) and EC (0.271, p = 0.277), none of which reach statistical significance. Similarly, phosphorus and potassium levels demonstrate weak correlations with other variables, with no significant relationships observed.

The heatmap presents the Pearson correlation coefficients among various soil properties and arsenic content in wheat samples, using color intensity to convey the strength and direction of the relationships. A strong negative correlation is depicted between electrical conductivity (EC) and pH (-0.491), as indicated by the dark blue shading, which suggests that an increase in EC is associated with a decrease in pH, resulting in more acidic soil conditions. Additionally, a moderate positive correlation is observed between organic matter percentage (OM %) and nitrate nitrogen (N-NO₃-) content (0.569), highlighted by a lighter red shade, indicating that higher ranks of organic matter are related with increased nitrate nitrogen content in the soil. The rest of the correlations are relatively weak, as shown by the lighter colors, indicating that there are minimal linear relationships between variables such as arsenic content in soil, grains, and roots, and other soil properties like phosphorus and potassium.



Figure 4.4: "Heatmap of Pearson Correlation Coefficients Among Soil Properties and Arsenic Content in Wheat Samples"

5.7 Arsenic Distribution and Mobility in Wheat Plants Based on BCF, BAF, and BTF Values

The calculated Bioaccumulation Factor (BAF,) Bio translocation Factor" (BTF)and Bio concentration Factor (BCF) values for arsenic in wheat plants reveal distinct patterns of arsenic distribution across different plant parts. The actual BCF values demonstrate that samples, including "MP S1" and "BK1 S2" have high ability of accumulating arsenic in the roots related to arsenic in the soil with the BCF higher than 1, pointing to the high efficiency of arsenic uptake by the roots. Nevertheless, most of the samples exhibit lower BCF values of less than 1, suggesting that the roots contain comparatively less arsenic than found in the soil conditions. These include the BAF values which estimate the ability of the plant to transfer arsenic from the earth to the aerial portions of the herbal; most of the samples have less than 1, indicating

low translocation of arsenic from the soil to the straw and the grains. Namely, the sample "BD S1" depicts a slightly higher BAF value suggesting a higher degree of As transport to the aerial parts but does not rise to the level of the concentration in the soil. The BTF values provided additional information on the mobility of arsenic from the roots to the grains, and the result indicated that all samples had a BTF value of < 1 indicating low translocations of arsenic from the roots to the grains. However, some samples include "BD S1" and "BD S2" that have lower BTF values and express relatively higher tendency to mobilize arsenic from root to ounce but the content of pollution in grains is still lower than that seen in the roots.

Sample ID	BCF	BAF	BTF
BK2 S1	0.884	0.465	0.526
BK2 S2	0.191	0.071	0.370
BK2 S3	0.287	0.018	0.065
MP S1	1.096	0.297	0.270
MP S2	0.717	0.079	0.111
MP 83	0.571	0.033	0.058
BK1 S1	0.524	0.103	0.197
BK1 S2	1.073	0.199	0.185
BK1 S3	0.355	0.219	0.616
MS S1	0.272	0.036	0.132
MS S2	0.219	0.142	0.649
MS 83	0.661	0.116	0.176
BD S1	0.395	0.307	0.778
BD S2	0.450	0.273	0.606
BP	0.351	0.082	0.233
BGM	0.789	0.101	0.128
BAW	0.667	0.037	0.056
BT	0.750	0.058	0.077

Table 4.6: Calculated Bioconcentration Factor (BCF), Biotranslocation Factor (BTF and Bioaccumulation Factor (BAF), for Arsenic in Wheat Plant Samples

*BCF: Bioconcentration Factor, *BAF: Bioaccumulation Factor, *BTF: Biotranslocation Factor

CHAPTER NO 05: DISCUSSION

The outcomes of this learning specify that there is a very "high concentration of arsenic" in the groundwater of the Muzaffargarh district. The overall arithmetic mean level of arsenic in the groundwater samples was 76. 88 μ g/L, with a range from 17. The ranges for lead concentration in the water sample were from 3 μ g/L to up to 294 μ g/L to way above the WHO Guideline for safe drinking water set at 10 μ g/L. Other studies showed in different backwaters of Pakistan including the ones by (Ali et al. 2019b) and (Shahid, Niazi, et al. 2018) have found similar levels of pollution especially in areas with comparable geological and industrial characteristics. For instance, the groundwater arsenic levels in the Punjab region have been reported to be above 50 μ g/L with some parts having levels of up to 200 μ g/L (Nickson et al. 2005b). This means that contamination observed in Muzaffargarh is not an isolated problem but a part of a regional phenomenon, which has both natural and manmade causes.

The pH level of the groundwater samples varies from 7. 05 to 7. It was 78, with an average of 7. This is 34 which is well within the WHO recommended level of 6. 5 to 8. 5. This is in line with other studies for instance the work done by (Ullah et al. 2023b) who established that the groundwater in Punjab has neutral to slightly alkaline pH which can allow for the mobilization of arsenic. The "electrical conductivity" (EC) values varied from 393 to 1350 μ S/cm with an average of 868 μ S/cm. 28 μ S/cm. About 33. Of the samples 33% was found to be moderately to highly saline as 33% of the samples had a conductivity level above the WHO recommended 1000 μ S/cm. These EC values are alike to those recorded by (Malik et al. 2009b) who observed that groundwater in the Indus Basin is often salty because of dissolution of salts from the nearby sediments.

The result of the correlation analysis of the arsenic absorption in groundwater with the physicochemical parameters presented a weak positive association with temperature and a weak negative association with pH. The positive correlation with temperature may indicate that temperature theatres a role in the solubility and mobility of arsenic in groundwater, since increased temperature can increase the dissolution of arsenic-containing minerals (Xing et al. 2023). This comment is in consonance with the study by (Nickson et al. 2005b) who established that climate warming could enhance the mobility of arsenic in the groundwater system. This is in harmony with the fact that arsenic is more doable in faintly acidic to unbiased conditions of the pH scale. This is in agreement with the well-known fact that arsenic is more

doable in faintly acidic to impartial conditions which are characterized by low pH. This relationship has been well documented in studies of the chemistry of the groundwater. For example, (Smedley and Kinniburgh., 2002) noted that low pH levels in groundwater can enhance the mobility of arsenic by transforming it into more soluble species especially where arsenic is naturally present.EC had a moderate positive relationship with arsenic and therefore high salinity levels could be associated with the elevated levels of arsenic. This could be attributed to the dissolved salts which facilitate the desorption of arsenic from particle soiled to groundwater. The same observation has been made in the analysis of groundwater pollution in Pakistan. For example, (Baig et al. 2009) explained that salinity and high EC levels in the Sindh region are linked to great arsenic levels in water because these conditions enable the ion of arsenic from sediments into water thus posing a contamination threat.

The "concentration of arsenic" in soils of Muzaffargarh district was found to be in the range of 18. 65 mg/kg to 68. The mean was 39 mg/kg, and the range was from 29 mg/kg. 95 mg/kg. These levels are much higher than the permissible limit of 20 mg/kg for agricultural soils as suggested by the "Food and Agriculture Organization" (FAO, 2004). A similar observation has been made in the previous studies conducted in the Sindh and the Punjab region of Pakistan. As an example, (Farooqi et al. 2009) detected the occurrence of arsenic in agrarian soils of Sindh and Punjab located in the Pakistan with a concentration of 14mg/kg to 72mg/kg especially in areas where water used for irrigation is contaminated with arsenic. These results show that the soils of the Muzaffargarh district are also affected by arsenic contamination; this is troublesome for agricultural production and the security of the food source. Some of the physicochemical properties like pH and electrical conductivity of the soil were also determined. Soil samples collected from Muzaffargarh district in this study had a pH of 7. 79 to 8. It was found that the total participants were 97 with an average of 8. It registered a slightly alkaline pH of 21. This is in covenant with the results observed by (Akram et al. 2014) where it was revealed that 74% of the soils in the Muzaffargarh district had a pH above 8. pH of 5 showed that the water had a high alkaline nature which has an impact on the soil health as well as crop yields. The pH of the soil can also increase the solubility of arsenic especially in forms of arsenate and this increases the uptake of the element by plants. The electrical conductivity (EC) of soil samples in this work varied between 7000 μ S /m to 2. The salinity was 7200000 μ S /m with an average of 81000 μ S /m. The values are in conformity with the work done by (Akram et al. 2014) where they established that 94% of the soils in the Muzaffargarh district were non-saline having EC values of less than 400000 µS /m. However,

those with higher EC values especially in some tehsils were associated with the problem of water logging and the use of low-quality waters for irrigation, which may lead to soil salinity and may aenhsnace mobility of arsenic. The concentrations of arsenic in wheat grains analyzed in this study were between 1.279 mg/kg to 18. 16 mg/kg, over the Codex Alimentarius Commission's allowable concentration of 0. A maximum limit of 2 mg/kg for inorganic arsenic in rice grains, is often applied to wheat as well (Codex Alimentarius Commission, 1995). This implies that arsenic exposure is likely to occur through the consumption of wheat products. Similar findings of high As concentrations in crops grown in contaminated soils have been also presented by other authors including (Zhao et al. 2009) and (Bhattacharya et al. 2021) in areas where groundwater is used for irrigation. Soil arsenic concentrations showed a strong association with some possessions of the soil in the current study. Poor negative relationship between as absorption in soil and soil pH was observed meaning that low pH increases As solubility and availability in soil and thus uptake by plants. This finding is in agreement with the work done by (Khan et al. 2015b) and (Zhao et al. 2009) who noted that acidic conditions in the soil raises the toxicity of arsenic with regard to crops.

There was also a weak positive correlation between electrical conductivity in soil and arsenic concentration. This implies that soils with elevated salinity may be at a higher risk of being contaminated with arsenic, which may be attributed to the interaction between arsenic and other anions in the soil solution, which would promote increased approval of arsenic by plants (Tudi et al. 2021b). The same is echoed by the study by (Jayakumar et al. 2021) who reported similar effects in the contaminated soils of different agricultural zones.

The health risk assessment done in this study shows the greatness of the health impact of arsenic in the water supply in the Muzaffargarh district. The HQ values, which reflect the risks of other chronic health effects, were also worrisomely high; some of the samples had HQ values that were over 2000 times the safe levels. This suggests that there is high probability of occurrence of health impacts among the residents who depend on groundwater for their water needs (US-EPA 2005). Comparable HQ values have been found in other arsenic-endemic areas, as described by (Rahaman et al. 2021), and HQ principles are usually numerous instructions of greatness developed than permissible levels.

Furthermore, the Carcinogenic Risk (CR) values which estimates the probability of cancer occurrence due to ingestion of arsenic, were also found to be expressively higher than the permissible limits, designating very high carcinogenic risk from long-term consumption of

contaminated water. These findings are in agreement with other health risk assessments done in areas with high arsenic contamination such as those by (Jang et al. 2016). The findings suggest that there is a grim necessity for intervention strategies like offering safe water sources and arsenic removal technologies for the health of the affected people.

The bioaccumulation and biotranslocation factors estimated in this study are therefore invaluable in the understanding of the transport of As in wheat plants. The bio concentration factor (BCF) values can be used to determine the extent of arsenic uptake by the roots of the wheat samples; samples from "MP S1" and "BK1 S2" had BCF values greater than 1. These values are close to the ones presented by (Tudi et al. 2021b) where they noted BCF values between 0. 5 to 1. It was also detected in wheat plants grown in polluted soils in this learning at the concentration of 2.

All the bioaccumulation factor (BAF) values were less than one suggesting that the motion of arsenic from the soil to the shoot part of the plant was low. The Biotranslocation factor (BTF) values also agree with this, since in none of the samples the BTF values are higher than 1, meaning that the movement of arsenic from the root to the grain is not a significant one. Nevertheless, some samples like BD S1 and BD S2 had relatively high BTF which implies that there may be a variation in the varieties of rice plants in as far as arsenic uptake is concerned.

The consequences of the study also presented that the concentration of As in wheat roots is with the absorption of As in the soil suggesting that wheat roots are efficient in absorbing As from contaminated soils. However, the relationship between arsenic in the soil and arsenic in the wheat grains was not strong, which signified that there was little movement of arsenic from the roots to the grains. This trend is in agreement with the BTF values obtained which showed that arsenic was more translocated to root than to shoot of the plant.

The correlation analysis also indicated that the soil pH was inversely related to the arsenic concentration in the wheat roots, and this is in agreement with the argument that lowering of pH raises the bioavailability of arsenic for plant uptake (Bhattacharya et al. 2021). The poor relationship between soil arsenic and grains arsenic concentration implies that other factors including plant physiology and varietal characteristics may greatly influence the uptake and accretion of arsenic in edible parts.

The research of this thesis has revealed several limitations and suggestions for further improvement, which aim at both the mentioned gaps and the crucial issues of food safety and public health. A major concern in this study was the issue of funding, which affected the extent of data gathered, and the kinds of analysis that could be done. Thus, the sample size was relatively small, and this may limit the transferability of the findings. Further, the study was limited to assess the arsenic contamination of wheat and water in Muzaffargarh district without exploring other factors like the type of soil, climatic differences, or the method of irrigation. Standard analytical methods could also have had the effect of not being sensitive enough to detect or quantify low levels of contaminants – which is a drawback of the learning. To overcome these limitations and to strengthen the impact on food safety and public health, it is suggested the following recommendations: It would be beneficial to expand the research area to other areas with similar environment so that comparisons could be made. Larger sample size and including other crops apart from wheat might give a better sympathetic on the accumulation of arsenic in the food chain. In addition, the use of analytical techniques and technologies like remote sensing and machine learning may enhance the detection and analysis of contaminants.

As food safety is a major concern, there is the need to tighten the monitoring and control of groundwater used for irrigation particularly in regions that are endemic to arsenic contamination. It is important to set up routine testing of water and soil to check the level of arsenic to avoid the toxicity level. Further, research should also be directed towards the creation of arsenic tolerance crop varieties and assessment of farming practices which could help reduce arsenic accumulation in plants through adjusting the acidity of the soil and using safe water for irrigation.

To this end, there is the need to mobilize the community on the effects of consuming food and water containing arsenic. Information and propaganda, along with provision of other sources of safe water supply can greatly help in the prevention of adverse health effects. Subsequent research should focus on the chronic effects of arsenic consumption through food and its effects on susceptible groups, as well as the long-term reaction to consistent intake. Foremost, more emphasis should be placed on the identification and optimization of efficient, economical, and feasible arsenic removal technologies for local application in order to enhance food safety and public health protection. Addressing temporal variations in contamination levels through long-term studies would yield more reliable data, supporting the formulation of more effective mitigation strategies.

CHAPTER NO 06: CONCLUSION

This study conducts an in-depth examination of arsenic contamination in the Muzaffargarh district, revealing its significant impact on agricultural productivity and public health. The analysis shows that "arsenic concentrations in groundwater" are alarmingly high, ranging from 17.3 µg/L to 294.0 µg/L, well above the WHO's suggested limit of 10 µg/L. Similarly, soil arsenic levels range from 18.65 mg/kg to 68.29 mg/kg, exceeding the safety threshold of 20 mg/kg for agricultural soils. In wheat crops, arsenic levels in roots are between 10.236 mg/kg and 34.46 mg/kg, and in grains, they range from 1.279 mg/kg to 18.16 mg/kg. These findings, coupled with high bioconcentration, bioaccumulation, and biotranslocation factors, underline the serious risk of arsenic entering the food chain, thus endangering food safety. The health risk assessment underscores critically high hazard quotient (HQ) values, between 1672.9 and 9936.1, and carcinogenic risk (CR) values ranging from 0.7528 to 4.4713, suggesting significant potential for both non-carcinogenic and carcinogenic effects, including an increased risk of cancer. To counter these risks, it is necessary to carry out frequent assessment of water and soil condition, create varieties of crops that can withstand high levels of arsenic and enhance practices that minimize the uptake of arsenic. Prevention measures should aim at educating people on the dangers of arsenic exposure and ensuring availability of safe water sources. Also, it is crucial to apply and develop low-cost technologies for arsenic treatment to prevent the further spread of contamination and to ensure the safety of food products and people's health. The study has some limitation; it lacks funding to conduct the research across various regions and has restricted the data collection to a particular area, therefore recommending future studies to consider enlarging the data gathering area, increasing the sample size and using more complex statistical methods. To effectively solve the issue of arsenic pollution in the Muzaffargarh district, it is crucial to employ a multifaceted strategy that involves scientific studies, public health programs, and policy changes aimed at encouraging sustainable agriculture and enhancing the well-being of residents.

CHAPTER NO 07:

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