

Ministry of Higher Education
and Scientific Research
University of Diyala
College of Education for Pure Science
Department of Chemistry



***CHEMICAL AND MICROBIOLOGICAL CONTAMINANTS ANALYSIS OF
SOIL, IRRIGATION WATER, AND AGRICULTURAL PRODUCT IN
DIFFERENT REGIONS OF IRAQ***

A Thesis

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By

Alaa Ahmed Chyad

**B.Sc. Chemistry, College of Education for Pure Science,
University of Diyala 2011 -2012**

Supervised by

Prof. Dr.

Ahmed Mahdi Saeed

Dr.

Abeer Saleh Alhendi

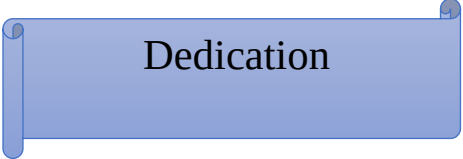
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بِسْمِ اللَّهِ الرَّحْمَنِ الرَّحِيمِ

(يَرْفَعُ اللَّهُ الَّذِينَ آمَنُوا مِنْكُمْ
وَالَّذِينَ أُوتُوا الْعِلْمَ دَرَجَاتٍ وَاللَّهُ
بِمَا تَعْمَلُونَ خَبِيرٌ)

صدق العظيم الله



Dedication

I Dedicate My Work

To My Father and My Mother God blesses them

To My Dear Brothers and Sisters

To All My Colleagues in Work and Study

To All Researchers in This Field

Acknowledgments

First of all, I would like to thank ALLAH the Most Gracious, the Most Merciful, for helping me Finishing this work and I pray that this blessing may last throughout my life. I would like to express my gratitude to my supervisor Prof. Ahmed Mahdi Saeed & Dr. Abeer Saleh Alhendi for the useful comments, remarks, and engagement through the learning process of this study. I would like to thank my beloved ones, who have supported me throughout the entire process, by keeping me harmonious and helping me putting pieces together. I will be grateful forever for their help.

Abstract

This study is divided into three parts; the steps for each part are summarized as shown below:

Part One: -

Estimated of a total number of bacteria (TB), a total number of *coliform* (TC), *E-Coli*, and molds and yeasts in 39 irrigation water samples, 75 soil samples, 75 paddy samples, and 75 rice samples of two Governorates of Iraq, Diyala and Salah al-Din was made. Three fields of each province were used to collect samples. A pour plating method was used to determine TB, TC, and molds and yeasts; while most probable number method was used to determine *E-Coli*. Iraqi Quality Standardization (IQS) and Food and Drug Administration (FDA) were considered the limits.

The results showed that all irrigation water samples have exceeded the permissible limit for TC molds and yeasts, while all soil and paddy samples were exceeded for total bacteria, total coliform, and molds and yeast. Rice samples were exceeded for only molds and yeasts. *E- Coli* in soil and rice was less than 3 cell/g, which was the detection limit of the procedure. While in water and paddy, *E- Coli* was less than the FDA limit. The molds and yeasts counts were very high, for example, molds and yeasts of paddy and rice were ranged from $(2.06- 21.5) \times 10^7$ cfu/mL, $(1.20-12.0) \times 10^6$ cfu/g respectively, while the limit was 10^2-10^4 for good and accepted quality respectively. In conclusion, rice samples were considered unsafe for human consumption, and irrigation water and soil were the sources of the contamination, therefore treatment for irrigation water should be strongly considered.

Part Two: -

Estimated of total aflatoxins, B1, B2, G1, and G2, in 75 rice samples that taken from the three fields of each province was perform. High-performance liquid

chromatography (HPLC) was used to determine total aflatoxin. Commission of the European Communities (CEC) was considered the limits, 4 µg/kg, because there is no Iraqi limit yet. The results showed that most of the rice samples contained aflatoxin higher than the permissible limit. The lowest and highest aflatoxin concentration in Diyala field were 1.19 µg/kg and 16.40 µg/kg respectively. The number of samples that exceeded the permissible limit of total aflatoxin was 66 samples out of the 75 rice samples. Rice of the two study provinces area was unsuitable in terms of its contamination of total aflatoxin therefore the produced rice was considered non-safe for human consumption. The reason for the high concentration of aflatoxin in rice samples is due to the high molds and yeasts count in irrigation water, soil, paddy, and rice. Therefore, a treatment to the contamination sources of fields has become a very necessary, and it must be resolved quickly.

Part Three: -

Determined the levels of heavy metals, Lead (Pb), Cadmium (Cd), Iron (Fe), manganese (Mn), Cobalt (Co), Magnesium (Mg), Aluminum (Al), and Copper (Cu) of 39 irrigation water samples, 75 soil samples, 75 paddy samples, and 75 rice samples in both governorates were made. Three fields of each province were used to collect samples. Atomic Absorption Spectrophotometer was used to determine heavy metals concentrations. Iraqi Quality Standardization (IQS) and World Health Organization (WHO) were considered the limits. The results showed that all irrigation water samples were exceeded the permissible limit for Pb, Cd, Fe, and Mn, while all soil samples were exceeded Mn and Mg. Paddy and rice samples were exceeded for only Pb limit. The Pb limit of paddy and rice was very high, for example, Pb concentrations of rice and paddy were ranged from 1.805-4.776 mg/kg, 0.642-3.481 mg/kg respectively, while the limit was 0.2 mg/kg. Paddy and rice samples had heavy metals concentration within the limit for all other tested heavy

metals. Cobalt and aluminum concentrations were less than the detection limit for all the tested samples. In conclusion, rice samples were considered unsafe for human consumption in term of its heavy metals content, and irrigation water was the source of the contamination. A treatment for irrigation water should be strongly considered.

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List of Symbols and Abbreviations

Symbol or Abbreviation	Definition
A. A	Ammonium Acetate
ACN	Acetonitrile
Afs	Aflatoxin
AFB1	Aflatoxin B1
AFB2	Aflatoxin B2
AFG1	Aflatoxin G1
AFG2	Aflatoxin G2
AAS	Atomic Absorption Spectroscopy
CEC	Commission of the European Communities
cfu	colony formation unit
cm	Centimeter
DON	Deoxynivalenol
EC	Eosin Methylene Blue agar
FAO	Food and Agriculture Organization
FDA	Food and Drug Administration Circular
g	Gram
HPLC	High Performance Liquid Chromatography
IARC	International Agency for Research on Cancer
IMVIC	Indole – Methyl red -Voges-Proskauer -Citrate
IQS	Iraqi Quality Standardization
Kg	Kilogram
LST	Lauryl Sulphate Tryptose broth
L	Liter
MEOH	Methanol
mg	Milligram
mL	Milliliter
µg	Micrograms
MPN	Most Probable Number
ng	Nanogram
OTA	Ochratoxin A
PH	Power of Hydrogen
S.D	Standard Deviation
TB	Total bacteria

TC	Total coliform
WHO	World Health Organization
ZEA	Zearalenone

Chapter One
Introduction
&
Literature Review

1. Introduction

Rice (*Oryza sativa* L) is one of the most important staple foods worldwide ¹, which is a staple food for more than half of the world's population. Rice is the second commonly used grain after wheat ². Rice is being cultivated on approximately 167 million hectares of area under varied climatic conditions in tropical and subtropical regions ^{3,4}, which is cultivated in 144 out of 196 countries worldwide ⁵. More than 661 million tons of rice crops were produced annually around the world ⁶, which means about 487.35 million milled rice production was in 2018 ⁷. China is the number one rice-producing country around the world followed by India, Indonesia, Bangladesh, and Vietnam ^{2,8}. Regarding to Iraq, Rice is a strategic crop, which is a staple food for the majority of the Iraqi people. The variety Amber is the most important local Iraqi rice variety, and it is characterized by high quality in terms of its flavor, aromatic character ⁹. The production quantity of rice in Iraq for 2015, 2016, and 2017 were 200, 43, and 100 thousand tons respectively ¹⁰. Rice's nutritional importance comes from the high amounts of easily digestible carbohydrates ¹¹, which provide about 20% of the world's dietary energy supply ¹². As well as its protein contains a balanced level of Lysine, essential amino acids, compared to other grain crops ¹³. Rice is considered beneficial to human health because it contains vital nutrients such as fibers, iron, calcium, and other micronutrients ¹⁴.

Food safety is described as the assurance of food that will not cause harm to the consumer when prepared and/or eaten as directed ¹⁵. Due to the importance of grains as one of the main sources of human consumption, an inspection of the safety and quality of grains is a guarantee for human health ¹⁶. Rice is one of these grains, which can be infected with biological and chemical contaminants during harvest or after harvest ¹⁷. Most rice contaminates are microorganisms, mycotoxins, heavy

metals, and pesticides^{18,19}.

Microorganisms are one of the major food degradation, toxification, and pathogenicity, affecting food quality and safety. Biological contamination of the environment includes several microorganisms such as total coliform, *E-coli*, molds and yeasts, etc.^{20,21}. Also, Enterobacteriaceae family including coliform and fecal coliform have been known to cause severe health problems in humans²².

Mycotoxins are secondary metabolites of fungi that are usually nephrotoxic or carcinogenic in nature²³. Mycotoxins can contaminate foods and cause serious health problems and significant economic losses of food resources. Grains are very susceptible to mycotoxin contamination²⁴. Among the known mycotoxins, aflatoxin group especially aflatoxin B1 (AFB1) and ochratoxin A (OTA) have been identified as potent and potential carcinogens by the International Agency for Research on Cancer (IARC) respectively²⁵. Also, aflatoxins are considered the most harmful microbial toxins to humans among other toxins such as deoxynivalenol (DON), Zearalenone (ZEA)²⁶.

Heavy metals are metals with densities greater than 5 g/cm³²⁷, and they are a carcinogenic and mutagenic compounds in nature²⁸. Some metals such as calcium, iron, magnesium, zinc are essential nutrients that become harmful and toxic when their concentrations exceed recommended standards²⁹. Other heavy metals such as cadmium, lead, aluminum, etc. accumulated in the human body causing toxic effects such as diarrhea, vomiting, dizziness, and even cancer^{23,30}.

Therefore, microbiological and chemical testing are the most used methods for determining the safety of cereal crops around the world³¹. In Iraq, the Quality Control Department, Grain Board of Iraq, routinely conducts chemical and microbiological tests on imported rice. While there is no such microbiological or chemical testing for domestic rice received by Iraqi farmers. Microorganisms,

mycotoxins, and heavy metals in paddy and rice were measured at many mills in Iraq, and it was discovered that some paddy samples were contaminated before processing ³². Therefore, the contamination might be from the fields too not just as a result of paddy and rice storage or processing steps.

1.2 Literature Review

1.2.1 Rice Production and Consumption Worldwide

Rice is one of the main crops in the world and ranks second after wheat crop in terms of the planted area and the production rate ^{33,34}. Rice cultivation requires a large amount of land and water, and it grows in alkaline, acidic, and saline soils, which means rice planting resists the desert, humid, flood, hot, dry, and cold conditions ^{3,28}. More than 163 million hectares of rice were harvested in more than 100 nations, where the global production was about 480 million metric tons of milled rice each year ³⁵. Asia produces and consumes more than 90% of the world's rice, while South Asia harvests more than 108 million hectares at a 66%. India and China are producing and consuming 50% of the world's production. According to the estimation, rice output in Asia will need to expand by 1.2–1.5% per year by 2035 to fulfill population demands ^{36,37}. The number of rice consumers around the world is approximately three billion people, and the rice consumption rate will be increased approximately 1 to 3% per year in the future ^{38,39}. The global population was estimated to increase to 9 billion by 2050. Therefore, the global area occupied by rice fields should be increased in the coming decades to meet global demand. However, rice cultivation is not possible in all parts of the world, thus international trade of rice is the only way to consume it for unproduced countries ⁴⁰.

In Iraq, rice is the most important summer crop, and the growing season starts in June/July and ends in October/November ⁴¹. The cultivation is still mainly dependent on manual labor, and the rate of seed used in planting is 120 kg/hectares ⁴². The production area of rice is mainly in central and southern Iraq with a large concentration in the Al-Najaf and Al-Qadisiya provinces due to their climate. The

climate presents the quality of the soil for cultivation and the availability of irrigation water. Other produced provinces at a lower level are Al-Muthanna, Misan, Dhi-Qar, Wasit, Diyala, and Salah al-Din ⁴¹. The best class is called locally (Anbar), which is characterized by an aromatic smell that is considered a high demand in Iraq for more than twenty years ⁴³. Indian type of rice is grown along the banks of the Tigris and Euphrates rivers, while Japanese type of rice is cultivated in small quantities in the north of Iraq where irrigation is possible and locally known as (Bazian). Despite the extremely favorable agro-climatic conditions ⁴⁴, Iraq is a country that imports cereals included rice because the domestic rice production is not enough to meet the needs of its population ⁴⁵. In Iraq, there was yearly fluctuation in the harvested area of rice and thus the production over the past twenty years. The reasons for the low productivity were the failure to adopt high-productivity varieties and the lack of use of modern technologies to serve the land and crops in addition to environmental conditions ^{46,47}. The average area of cultivated rice in Iraq was 59,046 hectares in 2019, the production quantity was 125 thousand tons. The rice production was approximately 209,612 tons in the year 2013, and the lowest production was only 18,200 thousand tons in the year 2018 ⁴⁸. The yield rate was 3.55 tons/ hectar, however, the yield was recorded with 6-8 ton/hectare for some parts that were used improved varieties. The yield might be increased all over Iraq to 6 tons/hectare by using a highly productive rice variety when applying an improved technology ^{49,50}.

1.2.2 Nutritional Importance of Rice

Rice is a nutritious grain for humans and is essential to the food security of millions in Asia, Latin America, and Africa. It is considered directly as food for humans or indirectly as animal feed ⁵¹. Studies showed that 100g of the rice crop contains about 7.1g of protein, 3.33g fat included 0.83g saturated fatty acids, 66.67g carbohydrates, and 8.3g fiber ⁵². In addition, rice contains vitamins and minerals such as calcium (Ca), magnesium (Mg), and phosphorous (P), with a small amount of copper (Cu), iron (Fe), manganese (Mn), and zinc (Zn) ^{53,54}. Rice is a huge source of human energy, about 20% of energy, and for its low price people are consuming it a lot ¹². On the other side, rice waste also contains a high value of vitamins and proteins, therefore it was used as animal feed ⁵⁵.

1.2.3 The Rice Safety

The safety and quality of cereals are of great importance because they reflect one of the most important sources of food and feeds in the world ⁵⁶, especially rice. Rice is considered one of the basic food crops for many people ². Contamination of cereals occurs from irrigation water to soil then to field crops ⁵⁷. Rice pollution may come from a variety of sources, including air, dust, dirt, water, insects, rats, birds, livestock, humans, storage and shipping containers, and handling and processing equipment ⁵⁸. The most dangerous sources of rice contamination are biological and chemical contamination such as coliform, *E- Coli*, molds and yeasts, mycotoxins, and heavy metals ⁵⁹. Hence monitoring exposure to these harmful materials protects consumers' health ⁶⁰. Consumers are at risk from bacteria and fungi if such microbes involve species that are pathogenic or toxic ⁶¹. Diseases caused by fungal pathogens, which significantly reduce productivity and quality, are among the most serious rice

cultivation issues ⁶². The reduction of microbes could greatly increase food safety and lower the incidence of foodborne illnesses worldwide ⁶³.

1.3 Microorganisms

Finding a living organism may indicate the presence of a community of organisms that could indicate the presence of disease-causing bacteria such as (*E- Coli*) ⁶⁴. The indicator organisms most commonly used are coliform bacteria and fecal coliform bacteria. Enterobacteriaceae family includes (*Coliforms*, *Escherichia spp.*, *Enterobacter spp.*, *Klebsiella spp.*, *Salmonella*, and *Citrobacter spp*) ⁶⁵. Coliform bacteria are defined as facultatively anaerobic, Gramnegative, non-spore-forming rods. Coliform bacteria generally belong to four genera of the Enterobacteriaceae: *Citrobacter*, *Enterobacter cloacae*, *Enterobacter aerogenes*, *E- Coli*, and *Klebsiella pneumonia* ⁶⁶. Some coliform bacteria are associated with the intestines, which consist of about 1% of the intestine at birth colon of warm-blooded animals therefore it is called fecal coliforms, while others are related to plant materials ⁶⁷. Coliform bacteria are considered as indicator organisms because their presence in foods indicates that circumstances are suitable for the presence of enteric pathogens and may signify insufficient sanitary conditions ⁶⁸. Total coliforms include organisms that are found normally in nature with the ability to survive and grow in water ⁶⁹. Therefore, they are not just useful as an indicator of fecal pathogens, but may be used as an indicator of the efficacy of treatment or to evaluate the cleanliness and integrity ⁶¹.

The genus *Escherichia* includes five species, and the most important of them is *Escherichia coli* (*E- Coli*). *E- Coli* was identified as the only species in the coliform group found exclusively in the intestinal tract of humans and other warm-blooded animals ⁷⁰. The presence of *E- Coli* is evidence of fecal contamination because of the ability of *E- Coli* to spread in different environments. *E- Coli* contaminates food

and causes many diseases to humans in an opportunistic manner whenever the opportunity arises ⁷¹. The potential of infection of *E- Coli* gives its importance among those species belonging to the intestinal family ^{71,72}. Their optimum temperature for growth is 37, which is the same as the temperature of the human buddies ⁷³. In many cases, *E- Coli* coexists with pathogenic organisms thus its presence may reflect the presence of enteric pathogens such as urinary tract infections and meningitis ⁷⁴. Nonpoint sources of human and animal waste, municipal wastewater discharge, domestic pets, livestock, land application of manure pasture areas, storm runoff, etc. can be a source of *E- Coli* contamination. Several environmental factors including temperature, pH, dissolved oxygen, nutrients, and the native microorganisms are affecting *E- Coli* growth ^{75,76}.

Fungi, molds and yeasts, are distributed widely in soils, water, and air ⁷⁷. Yeasts are ubiquitous groups of fungi that are associated with a wide range of ecological. Therefore, yeasts of different species can live in very diverse habitats including leaf surfaces, seawater, rivers, lakes, and on/in warm-blooded animals ⁷⁸. Molds and yeasts are distributed widely in decaying plant materials. Therefore their presence on unprocessed plant and animal foodstuffs is almost assured by harvesting, handling, distribution, and storage. Inadequate preservation of these foodstuffs can result in mycological spoilage ⁷⁹. The presence of potentially mycotoxigenic molds may indicate a public health hazard. Therefore mycological evaluation of food and food ingredients has become routine in quality assurance programs ⁸⁰.

Knowledge of the number of molds and yeasts in foods is useful when assessing the quality and extent of spoilage as well as predicting the shelf life. Cereals usually carry a variety of fungi from the field and also contain storage molds ⁸¹. Determination of the mycological quality of cereals and derived cereal products is a very important issue since these commodities are used extensively as foods and

animal feeds. Also because some of the organisms encountered in these products are capable of producing toxic secondary metabolites, mycotoxins ⁸².

1.3.1 Microorganisms in Irrigation Water

Total coliforms and fecal coliforms, especially *E- Coli*, have long been employed as indicators of water microbiological quality ⁸³. Total coliform (TC) bacteria are commonly found in the environment, and it is utilized to indicate the presence of other pathogens in water. Human feces poses the greatest threat of waterborne sickness ⁸⁴. *E- Coli*, an intestinal bacteria, is not generally harmful, and it is found in significantly higher concentrations in surface waters than other pathogens ⁸⁵. The World Health Organization (WHO) presently recognizes *E- Coli* as the best fecal indicator bacteria for detecting fecal contamination of water however, fecal coliforms are an acceptable alternative ⁸⁶. *E- Coli* is found at tolerable concentrations in all mammalian feces, although it does not multiply appreciably in the environment. The presence of pathogens caused waterborne infections is indicated by high quantities of these bacteria ⁸⁷. Typhoid fever, hepatitis, ear infections, gastroenteritis, and dysentery are among the diseases and ailments that can be found in water with high fecal coliform levels ⁸⁸. Agriculture irrigation water contamination has a significant impact on soil quality, and irrigation water is a source of numerous diseases that impair food quality and protection ⁸⁹.

The attention towards the occurrence of fungi in water has increased last decade, and they are now considered possible contaminants responsible for causing health problems in humans ⁹⁰. Fungi are commensal or environmental saprophytic microorganisms widely distributed in nature, which can cause opportunistic infections in immunocompromised individuals ⁹¹. The occurrence of fungi in water was rarely considered because attention was directed towards the occurrence of bacteria and viruses these microorganisms unlike fungi cause acute diseases ⁹².

A study was undertaken in Taiwan in 2004 to isolate microbes to detect coliforms and *E-Coli* from 640 total wells, obtained from 11 distinct locations to detect coliforms and *E-Coli*. The results show with 224 samples contain *E-Coli*, 359 samples contain Coliform, 60 samples are clean ⁹³.

1.3.2 Microorganisms in Soil

The biodiversity of microbial communities in soil has important implications for the stability and functioning of ecosystem processes. However, very little is known about the environmental factors that define the microbial role and how this influences the composition and activity of microbial communities ⁹⁴. Soil is a fascinating medium for developing microorganisms since it contains a variety of nutrients required for the digesting system of the organisms ⁹⁵. Soil is one of the richest reservoirs of microorganisms that billions of microorganisms belonging to thousands of different species are found in one gram of agricultural soil ⁹⁶. Despite microorganisms are making up less than 0.5% of the soil mass, microorganisms have a significant impact on soil properties and processes positively ⁹⁷. The soil microorganisms play an important role, nutrient cycling, and organic matter turnover in terrestrial ecosystems ⁹⁸. They may act as a nutrient source and are involved in humification processes, degradation of pollutants, and maintenance of soil structure ⁹⁹. A well-functioning soil microbial community is, therefore, a prerequisite for soil fertility and resilience to external factors ⁸⁶. The destruction of the soil microbiota due to bungle or natural contamination causes plant and animal populations above ground to decrease ¹⁰⁰. Disease-causing microorganisms are frequently found in soil and water ¹⁰¹. Soil microbes include bacteria, archaea, fungus, protozoa, and viruses ¹⁰². Fungi are one important group of microorganisms in the soil these eukaryotic creatures can be single-celled or multicellular depending on environmental conditions, and they live by decomposing and absorbing the organic

materials in which and on which they grow ¹⁰³. Molds and yeasts are single-celled fungi that can alter microbial and plant development as well as have a role in the creation of soil aggregates and the maintenance of soil structure ¹⁰⁴.

In a study conducted in 2019 in Oman on soil and well-irrigated contamination, total and fecal coliforms in well 1 were >2400 cfu ml⁻¹, and no pathogenic bacteria were found in the soil 2–3 meters distant from the well-irrigated ¹⁰⁵.

1.3.3 Microorganisms in Paddy and Rice

Field crops may be contaminated with enteric bacterial pathogens, and the sources of contamination during the pre-harvest stages are soil, feces, irrigation water, insecticides, dust, insects, manure, and wild or domestic animals ²⁰. While, during the post-harvest stages could be contaminated by contact with polluted water, human carriers, harvesting equipment, and transport containers ¹⁰⁶. Rice crops are subjected to be infected by many factors that may be biotic (pathogens) or abiotic (environmental factors) ¹⁰⁷. These diseases either attack rice plants at a growth stage or infect rice grains after harvest, and in both cases, the quantity and quality losses in rice crops have occurred ¹⁰⁸. Contamination occurs mostly before harvesting, however, the storage of rice is an important step in terms of keeping rice in a good quality and safe ¹⁰⁹. One of the main factors contributing to the spoilage of rice in storage is microbial development ¹¹⁰. Rice downgrading due to discoloration in storage presents a significant loss to growers and processors. Therefore, proper pre-harvest and post-harvest management of the rice is crucial to maintain the grain quality and safety ¹¹¹.

Foodborne illnesses caused by *E-Coli* are one of the most important gastrointestinal diseases and represent a public health risk. Therefore, the presence of *E-coli* can be used to evaluate the microbiological quality of Field crops ¹¹². Also, contamination

by molds and yeasts is still a serious concern in international food hygiene and security ¹¹³. Postharvest losses of field crops are high, such losses are mainly due to pathogenic fungi ¹¹⁴. Annually, post-harvest fungal diseases cause 10% to 30% losses in fruit, vegetables, and grains. Because cereal is generally harvested with a water content of 20 -22 percent, or 0.92 -0.95, drying is essential to prevent mold formation during storage ⁶¹. Various countries have implemented risk pre-warning mechanisms and food safety test requirements to prevent and control dangers. Because molds and yeasts produce mycotoxins in rotten food, these mycotoxins are exceedingly dangerous, mutagenic and carcinogenic when people or animals consume mold-contaminated food or feed ¹¹⁵.

Anindita and Prapti, investigated coliform in Indian rice in 2014. The presence of total coliforms was determined in 12 samples from 60 samples produced in Kolkata, West Bengal. The majority of the samples had a coliform count of 10^3 - 10^5 cfu/g. Microbial and biochemical analysis revealed the presence of enteric bacteria, particularly *E- Coli*, in both conventional and organic rice cultivars ¹¹⁶.

1.4 Mycotoxins

Mycotoxins are secondary metabolites from molds that can contaminate foods and cause serious health problems in consumers ⁷³. Grains are very susceptible to mycotoxin contamination and their metabolites have been linked to toxicological effects ranging from mild gastroenteritis to deadly cancer disease ²⁶. Over 450 different types of mycotoxins and their metabolites are known but only a few of them are dangerous to humans and animals ¹¹⁷. Mycotoxins based on the severity of the effect were classified into five major groups: 1, 2A, 2B, 4, and 5 ¹¹⁸. Group 1 is considered carcinogenic for humans such as Aflatoxins B1, B2, G1, G2 (Table 1.1). Group 2A such as ochratoxin A (OTA) it has immunosuppressive and carcinogenic properties (Table 1.1) ¹¹⁹. Group 2B, fumonisin B1, B2, were defined as likely

carcinogenic for humans it causes kidney failure. Group 4 such as zearalenone (ZEA) and group 5 such as deoxynivalenol (DON) are not classified as human carcinogenic toxins due to insufficient evidence It causes liver failure ^{120,121}. Temperatures of 22 to 29 °C and water activity of 0.90 to 0.99 are ideal for mold growth and mycotoxin production ⁶². Rice is prone to be contaminated with mycotoxin as it is usually produced in climatic environmental conditions favorable to fungal infection and growth ¹²². Rice, like other cereals, can be subject to contamination by mycotoxins, both in field and during storage posing a risk to human health and economic loss ^{122,123}.

1.4.1 Aflatoxins

The term “aflatoxin” comes from three words: (i) the “a” that represents the *Aspergillus* genus; (ii) The “fla” that represents the species *flavus*; and (iii) the “toxin” that means poison ¹²⁴. Aflatoxins (AFs) are mycotoxins formed by *Aspergillus* species such as *A. parasiticus* and *A. flavus* that are toxic to humans and animals ⁶⁰. Aflatoxigenic molds can cause a decrease in production, a loss of nutritional value, and a diminution of the market value of agricultural products ¹²⁵.

Aflatoxin B1 (AFB1), aflatoxin B2 (AFB2), aflatoxin G1 (AFG1), and aflatoxin G2 (AFG2) are the four major species based on their fluorescence under ultraviolet light (blue or green) ¹²⁶. AFB1 and AFB2 are produced by *Aspergillus flavus* and *Aspergillus parasiticus*, aflatoxin B1 is classified by the International Agency for Research on Cancer (IARC) as a class 1 toxin due to its demonstrated carcinogenicity to humans especially liver cancer, while AFG1 and AFG2 are only produced by *Aspergillus parasiticus* ¹²⁷. IARC determined that maximum residue limit (MRL) of aflatoxins for human consumption ranges from 4 to 30 µg /kg in food

¹²⁸.

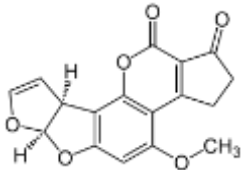
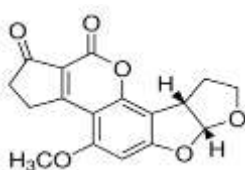
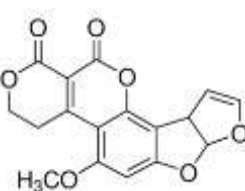
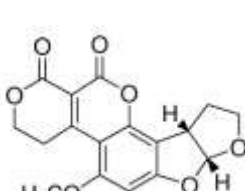
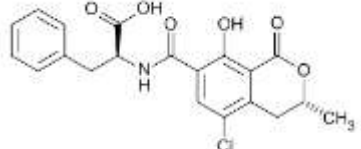
Albakeri and others, investigated the toxicity of *Aspergillus flavus* from infected rice in Iraq in 2020. The results revealed the total numbers of aflatoxins produced from aflatoxigenic isolates were 49, 47, 20, and 16 for aflatoxin B1, aflatoxin G2, aflatoxin G1, and aflatoxin B2 respectively ¹²⁹.

In 2021, a study was carried out to analyze the danger of aflatoxins in Colombian rice. Rice samples, 246 samples, were measured, and aflatoxins were found in 38.6% of the 246 samples, with levels ranging from 0.08 to 41.75 ¹³⁰.

1.4.2 Ochratoxin A

Ochratoxin A (OTA) is a fungal toxin produced by certain fungal species of the genera *Aspergillus* and *Penicillium* including *P. verrucosum*, *A. carbonarius*, *A. niger*, and *A. ochraceus* across a large range of climates in multiple foodstuffs. OTA was first detected by Van der Merwe et al. in an *Aspergillus Ochraceorosei* in 1965 ¹³¹. Toxigenic and filamentous fungi in food and feed contain the majority of OTA because of their thermostability. OTA can be found in agricultural products such as cereals, grapes, coffee, spices, cocoa, and derivatives ¹³². It has been shown to mostly affect the kidney, but it can also be teratogenic ¹¹⁹.

Table (1.1) Types of Aflatoxins

Mycotoxin	The chemical structure	Molecular formela	Descriptions
Aflatoxin B1 (AFB1)		$C_{17}H_{12}O_6$	Molecular weight: 312 g/mol Melting-point (°C) 268–269 ¹³³ .
Aflatoxin B2 (AFB2)		$C_{17}H_{14}O_6$	Molecular weight: 314.3 g/mol Melting -point (°C) 286–289 ¹³³ .
Aflatoxin G1 (AFG1)		$C_{17}H_{12}O_7$	Molecular weight: 328.3 g/mol Melting-point (°C) 244–246 ¹³³ .
Aflatoxin G2 (AFG2)		$C_{17}H_{14}O_7$	Molecular weight: 330.3 g/mol Melting-point (°C) 237–240 ¹³³ .
Ochratoxin A (OTA)		$C_{20}H_{18}ClNO_6$	Molecular weight: 403.8 Melting-point (°C) 159 ¹³³ .

1.5 Heavy Metals

Heavy metals are normal components of the Earth's surface, but their concentrations have reached toxic levels in many environments as a result of anthropogenic pollution¹³⁴. Minerals are an example of a nutrient. Minerals are required for cellular metabolism in some cases, for example calcium, phosphorous, magnesium,

sodium, potassium, iron, manganese, zinc, copper, selenium, chromium, cobalt, and iodine are all considered essential minerals ¹³⁵. These minerals are divided into two categories based on their functions in the body; which are macro minerals and micro or trace minerals ¹⁶. Macro minerals, such as calcium, magnesium, phosphorus, sodium, and potassium, are those that the body needs more than 100 mg of in a single day; while trace minerals are those that have a required amount from 0.5 to 50 mg. Trace minerals include zinc, iron, manganese, selenium, copper, and cobalt ^{136,137}. Heavy metals are highly persistent in the atmosphere because they are non-biodegradable and non-thermally degradable, which allowing them to accumulate to toxic levels quickly ¹³⁸. Arsenic (As), Cadmium (Cd), Lead (Pb), Chromium (Cr), Copper (Cu), Mercury (Hg), Nickel (Ni), and Zinc (Zn) are non-degradable toxic elements ¹³⁹. Table (1.2) explains the toxicity of heavy metals.

1.5.1 Heavy Metals in Irrigation Water and Soil

Industrial and municipal wastewaters are increasingly being used for irrigation in urban and peri-urban agriculture due to their availability ¹⁴⁰. As a result, long-term irrigation of industrial/municipal wastewater causes heavy metal buildup in soils ¹⁴¹. Also, heavy metals and metalloids can accumulate in soils due to emissions from rapidly developing industrial areas, mine tailings, high-metal waste disposal, leaded gasoline, and paints, fertilizer land application ¹⁴². Consequently, high levels of heavy metals can impair soil function cause plant toxicity and contaminate the food chain by compromising the quality and safety of food ⁶⁴. Since plants can uptake toxic compounds from soils via their roots, the soil is a source of many contaminants for plants ¹⁴³. “Soil pollution” is characterized as the presence of specific chemicals or substances in the soil that is unsuitable and/or in higher concentrations than natural causing harm to a non-target organism. Soil contamination must be controlled because food safety, human health, and long-term economic ¹⁴⁴. Heavy

metal contamination of soil is also of concern because of its effect on groundwater and surface water ¹⁴⁵.

A study was conducted in 2018 to determine heavy metal contamination of irrigation water in Bangladesh. They found that the Cd concentration was (0.001 mg/L), Cu concentration ranged from 1.43 to 6.04 mg/L, and the Pb concentrations ranged from 0.35 to 1.75 mg/L. The average Cd concentration did not exceed the FAO's permissible limit, but it was close to it, while Cu concentration was higher than the permissible limit. Only Pb concentration was below the permissible limit, 5.0 mg/L ¹⁴⁶.

A study was conducted in 2014 to determine the heavy metal concentration in the soil samples from agricultural lands of Bhaktapur district, Nepal. The ranges in the content of metals in soil samples were 13.76 - 60.56 mg/kg for Cu, 58.67-180.62 mg/kg for Fe, 4.67-67.43 mg/kg for Pb, 0.08- 1.52 mg/kg for Cd, Below detection limit (BDL) to 0.60 mg/ kg ¹³⁹.

1.5.2 Heavy metals in Rice

The increase of toxic pollutants from heavy metals in rice fields threatens food safety. Consumption of food crops contaminated with heavy metals is a major risk factor for human health and safety ¹⁴⁷. Intake of minerals and exposure to toxic levels to humans can occur through the consumption of contaminated agricultural products and field crops ¹⁴⁸. These come from irrigation water and soil and thus accumulate in rice grains and other grain crops through the food chain ^{52,149}. The concentration of heavy metals is not affected by cooking rice and soaking it only with water or soaking with water and table salt or water and citric acid. However, heavy metals can be decreased when rice is soaked with water, table salt, and citric acid for 20 minutes and cooked ¹²³. Also, the method of cooking rice in the normal

way or by using a microwave or a pressure cooker had no effect on the concentration of heavy metals in rice ¹⁵⁰.

In 2018, Anxiang Lu and others estimated heavy metals in rice in China, and they found that Cd concentrations in rice grains ranged from 0.005 to 6.21 mg/kg, with a mean of 0.564 mg/kg. The maximum permissible concentration of Cd in rice grains is 0.2 mg/kg. For Fe, Cu and Pb concentrations in rice grains, they were varied from 0.003 to 0.192, 0.001 to 0.012, and 0.001 to 0.07 mg/kg respectively ¹⁴.

Table (1.2) Toxic Effect of Heavy Metals on Humans Body

Heavy Metals	The Diseases
Lead	Headaches, nausea, vomiting, antisocial behavior; decreased hemoglobin synthesis; impaired renal function; deafness, blindness, and retardation ¹⁵¹ .
Cadmium	Toxicity to organ-like liver and kidney, hepatic, pulmonary, osteoporosis, carcinoma (primarily prostate and renal) ¹⁵² .
Iron	The accumulation of iron within the central nervous system is associated with neurodegenerative diseases, including Alzheimer's disease, Parkinson's disease, Huntington disease , amyotrophic lateral sclerosis ¹⁵³ .
Mercury	A causative agent for various disorders neurological, nephrological, immunological, cardiac, insomnia, weakness, muscle atrophy, twitching, headaches, disturbances in sensations, changes, and performance deficits on tests of cognitive function ¹⁵⁴ .

Copper	Acne, allergies, alopecia, anemia, anorexia, arthritis, autism, epilepsy or seizures, elevated cholesterol, cancer, diabetes, hormone imbalance, weakness, migraine headaches, hyperthyroidism, infections, inflammation, insomnia, multiple sclerosis, myocardial infarction, nausea, premenstrual tension, scoliosis, all skin disorders, tooth loss, urinary tract infections ¹⁵⁵ .
Chromium	GI Tract hemorrhage and blood loss; liver/kidney necrosis; skin ulcers, irritative dermatitis, and gastrointestinal carcinomas ⁷⁹ .
Nickel	Kidney failure, heart attack, cancer of the mouth, intestines, and lungs, skin issues, nausea, vomiting, hemorrhages, malaise, low blood pressure, muscle tremors, tetany, and paralysis ¹⁵⁶ .
Cobalt	Excess of this metal might increase the action of thyroid and bone marrow, which might, in turn, lead to overproduction of erythrocytes, fibrosis in lungs, and asthma ¹⁵⁷ .
Zinc	Metal fume fever signs include flu-like symptoms, stomach and intestinal upset, and/or liver dysfunction, moderate weakness, headaches, nausea, and vomiting ¹⁵⁸ .
Aluminum	Excessive aluminum levels are responsible for characteristic clinical symptoms associated with levels of intake that the organism is not able to eliminate (renal excretion mainly) ¹⁵⁹ .
Magnesium	Caused of paralytic ileus, pancreatitis, and renal failure ¹⁶⁰ .

1.6 Mycotoxins Measuring Instruments

Several sensitive analytical devices have been used including thin-layer chromatography (TLC), high-performance liquid chromatography (HPLC), gas chromatography (GC), mass spectrometry, electrochemical assay, and enzyme-

linked immunoassay (ELISA) to identify and quantify mycotoxins in food and feed samples with high accuracy and precision ^{60,161}. High-performance liquid chromatography (HPLC) is the most popular and accurate instrumental technique for detecting levels of aflatoxins (AFs) produced by fungi ^{59,124}.

1.6.1 High-Performance Liquid Chromatography (HPLC)

High-Performance Liquid Chromatography is one of the most essential analytical chemistry methods available, developed from traditional column chromatography by the Russian botanist *M.S Tswett* in 1903 ¹⁶². The premise is that a sample solution is injected into a porous material column (stationary phase), and a liquid (mobile phase) is pumped through the column at high pressure. The changes in migration rates through the column resulting in the different partitioning of the sample between the stationary and mobile phases are used to separate samples. Elution occurs at different times depending on the distinct components ^{163,164}. The most important technical types of HPLC are: -

-Normal phase HPLC

In this technique, polar static phases of silica SiO_2 or alumina Al_2O_3 are used, size of minutes ranging between 0.3-0.5 micrometers for the analytical column ¹⁶⁵. This technique is used for compounds that are less soluble in water and buffer solutions and is of little use for the separation of polycyclic aromatic hydrocarbons. The disadvantages of this technique in practical application are the lack of selectivity of the Stationary phase ^{166,167}.

- Reverse Phase HPLC

This technique is one of the most important and modern techniques used in analysis and separation than the rest of the known chromatographic techniques. In 1973,

about 20% of the separation processes used reverse-phase technology. However, recently more than 80% of the separation processes of HPLC technology use the reverse phase. In reverse phase HPLC, the stationary phase consists of a non-polar fine particles column, while the mobile phase is a polar solution. The idea of using non-polar microparticles as a stationary phase and a polar mobile phase was developed for the first time by the scientist *Boscott*, who used cellulose as a stationary phase 1967^{9,168}.

1.6.1.1 Components of a High-performance Liquid Chromatograph

1. Solvent Tank

Conical flasks, each of a capacity of 1 liter, are used to store the mobile phase. The containers are attached to a filter Poly Tetra Fluoro (PTFE) tubes whose inner diameter is about 0.2-0.5 μm to carry the mobile phase from the tank to the pump. This filter is to prevent the passage of unauthorized particles and impurities likely to be present in the mobile phase⁶⁴.

2. Pumps

It may include more than one pump. Each pump contains a piston that drives the mobile phase to the chromatographic column at a constant flow rate of 0.10-1.0 mL/min and at constant pressure, 600-6000Pa¹⁶⁹.

3. Connecting Tubes

The tubes connecting the pump to the syringe are made of stainless steel rust with (1.6mm x 0.8 mm I.D) dimensions that connect the syringe and the shaft. The other tubes connect the shaft and the connection cell with dimensions (1.6mm x 0.3 mm I.D), preferably as narrow as possible; This is to reduce packet spread and dead size¹⁷⁰.

4. Solvent Mixer

It is a steel container in which solvents are mixed for solvent injection after mixing and blending homogeneously ¹⁷¹.

5- Prototype Injection System

Standard solutions and samples are transferred to the column through an injector. The injection can be done by using micro or manual injection during the injection system or by using an automatic injector.

6. Column

The heart of the device, where the separation process takes place, is made of stainless steel and corrosion .It is found in all devices ranging in length from 5-25 cm and with an internal diameter from 3-5 milliliters ¹⁷².

7. Detectors

The detector is the component responsible for turning a physical or chemical attribute into a measurable signal corresponding to concentration or identity.

Types of Detectors

1. UV-ViS detector
2. Fluorescence detector
3. Refractive Index detector
4. Evaporative light scattering detector
5. Electrical conductivity detector
6. Electrochemical Detector
7. Mass spectrometer Detector ¹⁷³.

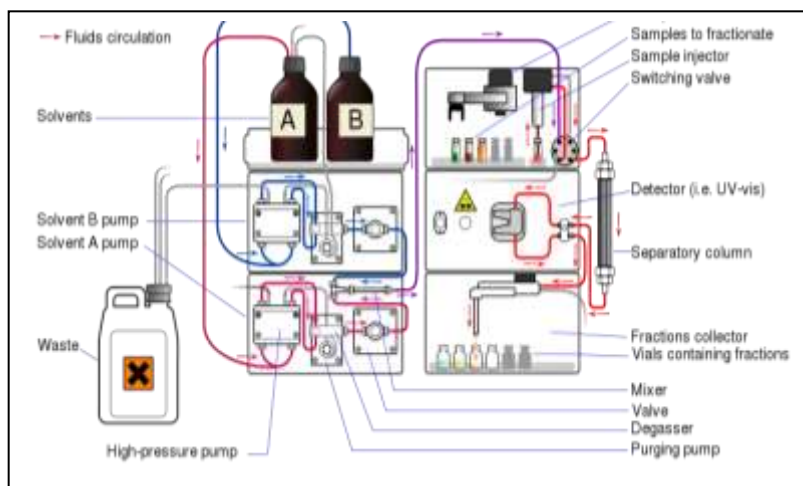


Fig. (1.1) High-Performance Liquid Chromatography Device ¹⁷⁴.

1.6.1.2 Applications of HPLC

High-performance Liquid Chromatograph technology (HPLC) is used to analyze a number of water contaminants, as well as to separate amino acids, Proteins, vitamins, nucleic acids, pesticides, sugars, and various pharmaceutical compounds. It is a widespread separation method for organic analysis, and it is now routinely used for the separations and analysis of pharmaceutical and biological materials. Also, it has been used in the fields of medicine, environmental and industrial, such as the pharmaceutical industry, oil derivatives, and organic industries. The widespread of HPLC used is due to the conditions that lead to an increase in speed, linearity, speed of passage of the sample through the column, better efficiency, and higher sensitivity compared to conventional liquid chromatography ^{175,176}.

HPLC coupled with mass spectrometry (MS) techniques has offered abundant qualitative and quantitative information from an analysis of residues in foodstuffs. HPLC-MS has demonstrated high sensitivity and selectivity, as well as a low detection limit. However, when the number of target ions increases, the detection sensitivity of this approach may fall dramatically ¹⁷⁷.

Tandem mass spectrometry hyphenated to liquid chromatography separation systems (HPLC–MS/MS) has evolved into a significant technology in clinical chemistry. Target analysis in therapeutic medication monitoring and metabolic disorders diagnosis are two of the most prominent application sectors ¹⁷⁷. HPLC–MS/MS has several key advantages, including the possibility for high analytical specificity, a wide range of applicability to tiny and big compounds, and the capacity to quickly create powerful assays with a high degree of flexibility, because of the potential dangers that these medications pose to human health, a number of approaches based on ultra-high-performance liquid chromatography-tandem mass spectrometry (UHPLC-MS/MS) have been developed for screening and quantification of various drug families ^{177,178}.

1.6.1.3 Advantages of HPLC

1. This technique is distinguished from other techniques by the possibility of analyzing several samples at the lowest cost, as well as the separation speed and accuracy ¹⁷⁹.
2. This technique has been widely adopted at present for qualitative separation, diagnosis, and quantification of mixtures of complex compounds due to its simplicity and the ease and accuracy of interpretation of the results ¹⁸⁰.
3. Used to identify compounds and liquids with high boiling points.
4. Samples can be injected at room temperature.
5. The materials to be used are liquids or solids dissolved in a liquid of varying degrees of relatively high evaporation ¹⁸¹.

1.7 Heavy Metal Measuring Instruments

For the determination of heavy metals in various sample matrices, several analytical techniques have been developed, including flame atomic absorption spectrometry (FAAS), electrothermal atomic absorption spectrometry (ETAAS), UV-VIS spectrophotometry, voltammetry, inductively coupled plasma-optical emission spectrometry (ICP-OES), and inductively coupled plasma-mass spectrometry (ICP-MS). For identifying trace elements, flame atomic absorption spectrometry (FAAS) is an effective detection technique because of its low cost, ease of service, high sample throughput, and good selectivity. Therefore, this technique has been widely used for heavy metal determination^{182,183}.

1.7.1 Atomic Absorption Spectrometry (AAS)

Atomic absorption spectroscopy is an analytical method for identifying metals and metalloids in their formation of a wide range of samples. The measuring method depends on the ability of free and neutral atoms to absorb a resonant beam when passed in its atomic vapor. The first atomic absorption was observed by *Wollaston* in 1802, but the idea of analysis and quantitative elements by flame atomic absorption technique was put into practice in 1952 by physicist Australian *Walsh* when his first research appeared in 1955. In the year 1959 the Russian physicist *vov'L* made a major breakthrough by describing the basis of non-flame atomic absorption by making a graphite furnace cell, which is tracing lower concentration than the flame atomic^{184,185}.

1.7.1.2 Atomic Absorption Spectroscopy Components

1. Light Source: The source is a Hollow cathode lamp, which gives a spectrum in the range of 190 - 800 nm. The lamp radiates an atomic spectral light source for the desired element its estimation is in the name of the element itself ¹⁸⁶.

2. Atomizer: The process of withdrawing the solution of the tested element to be quantified. The solution, about 3-4 mL/min, is transformed into a fine mist, which makes the response fast ¹⁸⁷.

3. Flame: It is required that the flame temperature by 2000 k or more, so the gas is ignited with an oxide such as air, nitrous oxide, or oxygen with nitrogen or acetylene. The mixture of air with acetylene is considered one of the best types of mixtures as it gives a suitable temperature for the estimation of about 39 samples ¹⁸⁸.

4. Light Interrupter: It controls the passage of the signal that allows the passage of rays emanating from a light source into flame at a constant speed in succession, which leads to magnification of the rays ¹⁸⁹.

5. Monochromator: It is used as a unifier to distinguish the wavelengths emitted by a light source that passes through the flame to allow only the desired wavelengths to pass through ¹⁹⁰.

6. Detector: photomultiplier tube is used to detect the intensity of the beam by converting it into an electric current whose intensity depends on the intensity of the light falling on its photovoltaic cell. It magnifies the electrical current produced by the photovoltaic cell and converts it into measurements ¹⁹¹.

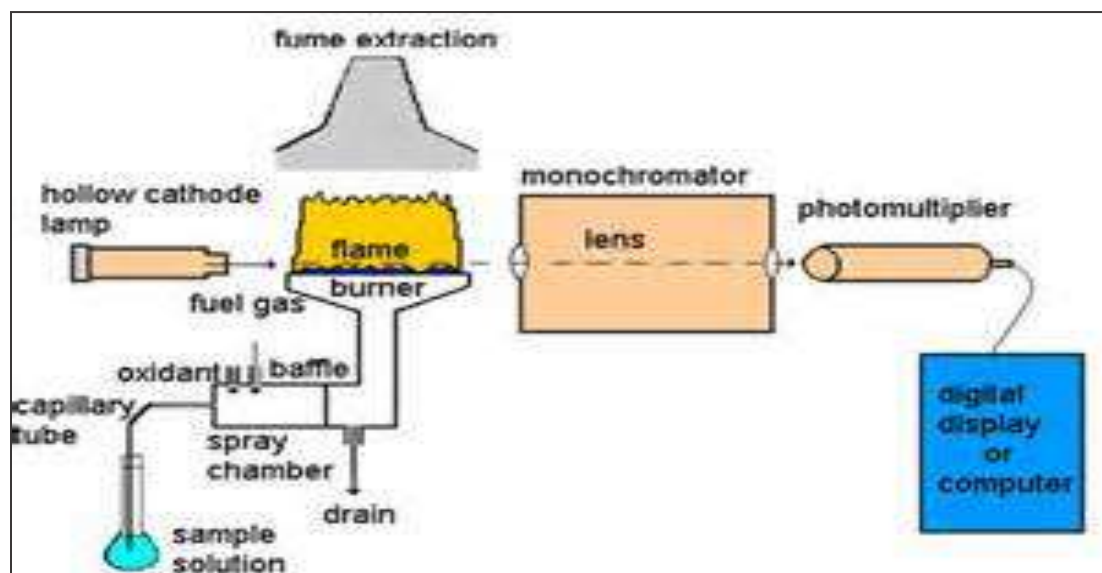


Fig. (1.2): Atomic absorption spectrometry Device ¹⁹².

The Aim and the Objectives of the Study

The overall goal of this project is to investigate the safety of the paddy and rice from fields before processing and storage, in addition to determining the source of the contamination. The specific objectives that will be met in this project are:-

1. Estimating microbial contamination in paddy, rice, soil, and irrigation water of several fields in two provinces of Iraq.
2. Estimating the total aflatoxins concentration in rice of several fields in two provinces of Iraq.
3. Estimating heavy metals contents in paddy, rice, soil, and irrigation water of several fields in two provinces of Iraq.

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Chapter Two
Materials
&
Methods

2. Materials and Methods

2.1 Biological Devices and Tools

The laboratory equipment and Devices detailed in Table (2.1) were used for procedure experiments and research studies

Table (2.1) Biological Devices and Tools used in This study

No.	The Device Name	The manufacture company	Origin
1	Incubator	Memmert	Germany
2	Refrigerator	Beko	Turkey
3	Fume hood (chamber)	Biomerix	France
4	Sensitive balance	Mittler	Switzerland
5	Distillater	Biomerix	France
6	Benzen burner	Beko	Turkey
7	Autoclave	Litrayama	Japan
8	Water bath	Memmert	Germany
9	Vortex	Leb.Teeh	Germany

No.	The Device Name	The manufacture company	Origin
10	Micro pipette	Brand	Germany
11	Beakers	Memmert	Germany
12	Scroll bottle	Memmert	Germany
13	Cylinder	Memmert	Germany
14	Conical flask	Memmert	Germany
15	Funnel	Memmert	Germany
16	Test tube	Memmert	Germany
17	Ruck	Memmert	Germany

2.2 Biological Materials

The Culture Media and Chemical Reagents are shown in Table (2.2) were used to conduct Biological Tests

Table (2.2) Biological Materials used in This study

No.	Materials name	Origin
1	Sabouraud Dextrose Agar	India
2	Nutrient Agar	Europ
3	Lauryl Tryptose Broth (lauryl Sulphate Broth)	India
4	MacCONKEY Agar	United Kingdom
5	PEPTONE WATER	United Kingdom
6	EOSIN METHYLENE BLUE AGAR (Levine)	United Kingdom
7	EC Broth	India
8	M.R.V.P. MEDIUM	United Kingdom
9	SIMMONS CITRATE AGAR	Englind
10	Tartaric acid	Englind

No.	Materials name	Origin
11	Kovacs reagent	Englind
12	Methyl red reagent	Englind
13	Voges-Proskauer reagent	Englind

2.3 Chemical Devices and Tools

The Chemical Devices and Tools shown in Table (2.3) were used to conduct Chemical Tests

Table (2.3) Chemical Devices used in This study

No.	Device name	Origin
1	high-performance liquid chromatography (HPLC)	Shimadzu, LC-20AD, Japan
2	Atomic Absorption Spectrometry (AAS)	Shimadzu U, AA-6200, Japan
3	pH meter	Mettle Toledo, Japan
4	Hot plat	ISO –LAB, Tecam
5	Sensitive balance	Mettler (Switzerland)
6	Oven	Italy
7	Ultrasonic water bath sensitive	APEL, PD-303 UV, Japan
8	Crucible	Germany
9	Volumetric Flask	Germany

2.4 Chemical Materials

The Chemicals shown in Table (2.4), the information of the manufacturers, and the purity were used, as all the materials used were of high purity.

Table (2.4) Chemical Materials used in This study

No.	Materials name	Purity	Chemical symbol	Company and origin	Molecular weight g/mol
1	Nitric acid	70%	HNO ₃	BDH- U.K	63.012
2	Hydrochloric acid	37%	HCl	BDH- U.K	36.46
3	phosphoric acid	99%	H ₃ PO ₄	BDH- U.K	98
4	Ammonium acetate	98%	C ₂ H ₇ NO ₂	BDH- U.K	77.03
5	Acetonitrile HPLC grade (ACN)	99.99 %	C ₂ H ₃ N	Belgian	41.05
6	Methanol HPLC grade (MeOH)	99.99 %	CH ₃ OH	Belgian	32.04

2.5 Preparation methods of Used Solution

1. **Peptone Water Solution:** 7.5 g peptone water was weighed and 500 ml of distilled water mix well and distribute into final containers sterilize by autoclaving at 121°C for 15 minutes.
2. **Tartaric acid Solution:** 1 g of Tartaric acid was weighed and placed into the container, then 500 ml of distilled water was added and mixed well.
3. **0.1 M Ammonium Acetate:** 7.7 g of ammonium acetate was weighed and placed in a volumetric flask, then 1000 ml of distilled water was added to it.
4. **0.2 M HNO₃:** 3.26 ml of Nitric acid was placed in a 250 ml volumetric flask, then was filled with distilled water.
5. **5.5 M HCL:** 23 ml of Hydrochloric acid was placed in a 50 ml volumetric flask, then was filled with distilled water.

2.6 Description of The Two Study Areas

Governorate of Diyala is one of the eighteen governorates of Iraq. Governorate of Diyala constitutes 4.1% of the area of Iraq, and its area is estimated to be 17,685 km². Between lines 22.44° - 56 .45° east of the Greenwich line, thus it represents the international borders of Iran from the east and is bordered by Salah al-Din Governorate and Baghdad Governorate from the west (Governorate of Diyala Agriculture Directorate).

Province of Salah Al-Din is one of the Iraqi provinces. Governorate of Salah al-Din constitutes 5.6% of the area of Iraq, and its area is estimated to be 24,363 km². It is located between 33.3° to 35.6° North latitude and 42.4° to 45° East longitude ¹⁹³.

2.7 Samples Collection

All irrigation water, soil, and paddy samples were collected from Diyala and Salah al-Din governorates during October to November 2020. Three fields of each province were used to gather the samples.

2.7.1 Irrigation Water Samples

Irrigation water samples, 39 samples, were collected from Diyala and Salah al-Din with a volume of 60 ml for each sample. The number of Diyala water samples was eight samples for each field. The first field was well water, the second was river water, and the third was pooled water, irrigation water that came from fields full of water or had water more than the required amount. Five irrigation water samples for each field from Salah al-Din where the first and third field was a pooled and the second field was a river. Placed samples into clean, dry, and sterilized containers and stored them inside an ice bath until they were transferred to a refrigerator to be analyzed later ¹⁹⁴.

2.7.2 Soil Samples

Seventy-five soil samples were collected manually using a shovel from Diyala and Salah al-Din, fifteen samples for each field of Diyala, and ten samples for each field of Salah al-Din. Samples were collected using a random selection as appeared in Fig (2.1), which is suitable for fields of any size, whether equal or unequal. Samples were collected from the soil surface to a depth of 20 cm below the surface. Soil samples were stored in plastic bags (polyethylene). Once the samples were in the laboratory, the grass, leaves, roots, and stones were removed by hand, and the soil samples were air-dried at ambient laboratory temperature. Air-dried soil samples were crushed with a pestle and mortar before passing a 2 mm mesh sieve for analysis ^{3,195}.

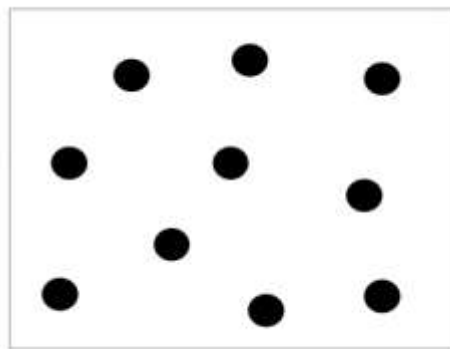


Fig. (2.1) The Random Method for Collecting Soil Samples

2.7.3 Paddy and Rice Samples

Seventy-five samples were collected from rice fields from Diyala and Salah al-Din, fifteen samples from each field of Diyala, and ten samples from each field of Salah al-Din. Samples were collected from the same soil sampling locations. They were stored in plastic bags (polyethylene) then paddy was peeled to obtain rice by using Aljaon (its machine was used for the peeling cereal in past). It was the degree of whiteness of the rice 38. Wearing gloves and sterilizing with alcohol between each sample and other ¹⁹⁶.

2.8 Microbiological Analysis

All used microbiological media were prepared as per the supplier's instructions. The media was sterilized using an autoclave at 121°C, for 15minutes. The targeted microorganisms, Total bacteria, Total coliform, and molds and yeasts were enumerated in Nutrient Agar, MacConkey Agar, and Sabarude Agar by a pour plating technique, while *E- Coli* was enumerated by following most probable number (MPN).

2.8.1 Total Bacteria Count

One ml or gm of samples was taken from irrigation water or soil and placed in 9 mL of sterile peptone water. While 10 g paddy or rice sample was taken and placed in 90 mL of sterile peptone water. The samples were carefully mixed and aseptically introduced into the dilution tubes. successive dilutions were made by mixing 1 mL of the original mixture with 9 mL of dilution water (peptone water) until 7 dilutions. Then 1 ml of the sample solution is placed in Petri dishes in a Fume hood. After that, about 15 mL of sterilized and cooled to about 45°C of Nutrient Agar was poured on Petri dishes and allowed the media to solidify. The plates were incubated inversely at 37°C for 24 hours to examine the total bacteria count ¹⁹⁷. The obtained results are shown in table (3.1) and fig. (3.1)

2.8.2 Total Coliform Count

The same series of dilutions that were made above were used to determine total coliform bacteria. One mL of sample solution was placed inside Petri dishes, and about 15 mL of sterilized and cooled to about 45°C of Violet Red Bile Lactose agar (VRBL-agar) was poured over it and mixed well before allowing it to solidify. Plates were incubated at 37°C for 24 h before counting the coliform bacteria in the samples ¹⁴³. The obtained results are shown in table (3.2), and fig. (3.2).

2.8.3 Molds and Yeasts Count

The same series of dilutions that were made above were used to determine molds and yeast count. One ml of sample solution was placed inside Petri dishes, and about 15 ml of sterilized and cooled to about 45°C of Sabouraud agar was poured over it and mixed well before allowing it to solidify. Then the plates were incubating at 26°C for 3 days to count the molds and yeasts in the sample. Molds colonies were

larger and more diffusive than yeasts colonies after incubation ¹⁹⁸. The obtained results are shown in table (3.4) and fig. (3.3).

2.8.4 *E- Coli* Count

The most probable number (MPN) method was used to count *E- Coli* for all samples. The technique for determining the MPN takes several days (up to 5 days). One ml of the three dilutions, 0.1, 0.01, and 0.001 were introduced to three tubes of lauryl sulfate tryptose (LST) broth and incubated at 37 °C for 24 h. The tubes were checked for gas production. Tubes that developed gas bubbles were assumed to be positive, and a loopful of the culture was transferred into an *E- Coli* (EC) broth tube. The EC tubes were incubated at 37°C for 24 hours before being checked for gas generation. A loopful of gas-producing EC tubes was scattered across brilliant green lactose bile broth (BGLB, a selective both comprising bile salts and bright green as inhibitors) and incubated at 37 °C for 24 hours. Finally, biochemical studies (IMVIC tests) verified typical colonies on vivid green lactose bile broth agar plates ¹⁹⁹. The obtained results are shown in table (3.3).

2.8.4.1 Indole – Methyl red -Voges-Proskauer -Citrate (IMVIC)

Tests were a set of four tests, including Indole, Methyl Red, Voges Proskauer, and Citrate Utilization, that were used to identify members of the Enterobacteriaceae family. This is done in accordance with the medium's color-changing after the indicators have been added. The experiments were carried out using the approach described by Chholgyel N et al. A positive Indole response is demonstrated by the emergence of a red ring in the Indole test, but a stable red color in MRVP broth indicates a positive MR test, indicating the presence of *Escherichia coli*. A positive V-P test is also indicated by the appearance of pink color in the VP broth. Citrate-positive organisms proliferate and turn blue during the citrate agar incubation time

¹¹⁶. Microorganisms analyses were done at University of Diyala Biology Department.

2.9 High- Performance Liquid Chromatography Method

2.9.1 Optimum condition selection for Aflatoxin estimation

This method was used for the determination of Aflatoxin in rice, where this method was implemented by injecting 10 μ m of the prepared solution using manual injection. The solution of standard Aflatoxin was injected after filtration using a microfilter (0.45 μ m) through the injection system (Reverse high-performance liquid chromatography) device in a stream of mobile phase on a column type ODS - C18 (25 cm L * 0.46 cm I.D. and 5 μ m particle size diameter) and UV detector was used. This study was accomplished by study many different types of parameters (mobile phases types, pH, flow rate, and wavelengths) to obtain the best conditions for the separation of mycotoxins in rice. The results obtained were shown in paragraph (3.2.1) as table (3.5).

2.9.2 Application on Standard Aflatoxins

10 μ m standard of aflatoxin concentrations (1 ng/ml, 3 ng/ml), respectively was injected through the injection system of HPLC device under optimum conditions (mobile phase consist of ammonium acetate 50 %, acetonitrile 30 % and methanol 20%, a wavelength at a 270 nm UV detector, flow rate of the mobile phase 1.5 ml/min, measurement time of 4 minutes and pH equal 4 using a separation column of dimensions ODS - C18 (25 cm L * 0.46 cm I.D. and 5 μ m particle size diameter). The results obtained were shown in paragraph (3.2.2) as table (3. 5), and fig (3.4), (3.5).

2.9.3: Estimation Aflatoxin in Rice by Using HPLC Device

0.1 g of milled rice was weighed and 10 mL of mobile phase solution was added to it, which consisted of (50% acetyl acetate and water, 30% acetonitrile, and 20% methanol) at pH = 4 (optimal conditions). The solution was placed in an ultrasonic device for 30 minutes, then the solution was filtered by using filter papers and filtered again by using microfilters. The mycotoxins were injected after filtration using a microfilter (0.45 μ m) through the injection system (Reverse high-performance liquid chromatography) device in a stream of mobile phase on a column type ODS - C18 (25 cm L * 0.46 cm I.D. and 5 μ m particle size diameter) under optimum conditions (mobile phase consist of ammonium acetate 50 %, acetonitrile 30 % and methanol 20%, a wavelength at a 270 nm UV detector, flow rate of the mobile phase 1.5 mL/min, measurement time of 4 minutes and pH equal 4) and UV detector was used. The results obtained were shown in paragraph (3.2.3) as table (3.5), and fig. (3.6), (3.7), (3.8), (3.9) (3.10), (3.11), (3.12), (3.13), (3.14), (3.15). Aflatoxins analyses were done at University of Diyala Department of Chemistry.

2.10 Preparation Samples for Heavy Metals Measurement

2.10.1 Preparation of Irrigation Water Samples

Irrigation water samples were flittered by using a Whatman filter paper. Then a few drops of strong nitric acid were added to samples (60 ml) to inhibit metal precipitation and biological growth, and then Atomic Absorption Spectrophotometer (AAS) was used to quantify heavy metals ²⁰⁰, and the results in tables (3.6), (3.7), (3.8), (3.9), (3.10), (3.11), and fig. (3.20), (3.21).

2.10.2 Preparation of Soil Samples

Fifteen ml of 30% HCL and 70% HNO₃ (3:1 ratio) was added to one gram of dried soil and leftover a day. The mixture was placed on a hot plate for 45 minutes in a fume hood (chamber), and then the solution was quantitatively transferred to a volumetric flask and the volume was completed to 100 ml by adding ion-free distillate water. After that, they were filtered by filter papers and heavy metals were measured by AAS device ²⁰¹, and the results in tables (3.6), (3.7), (3.8), (3.9), (3.10), (3.11), and fig. (3.20), (3.21).

2.10.3 Preparation of Paddy and Rice Samples

Ten grams of well-ground paddy or rice were placed in a crucible and burned in the oven at a temperature of 600 °C for 6 hours. Then 5 ml of HCl (5.5M) were added and placed on a hot plate, and 20 ml of HNO₃ (0.2M) was added and left for 90 min. Then they were filtered by filter papers for measuring heavy metals in the AAS device ²⁰², and the results in tables (3.6), (3.7), (3.8), (3.9), (3.10), (3.11), and fig. (3.20), (3.21).

2.11 Atomic Absorption Spectrophotometer Analysis (AAS)

The metals were measured by using an Atomic Absorption Spectrophotometer (AAs; AA-7000 Atomic Absorption Spectrophotometer, Shimadzu, Japan), which was used an air acetylene flame. The AAS measured the elements, Iron (Fe), Lead (Pb), Copper (Cu), manganese (Mn), Cadmium (Cd), Magnesium (mg), Aluminum (Al), and Cobalt (Co). The calibration curves fig. (3.17), (3.18), (3.19), for all of the metals were created individually using varied concentrations of standard solutions by running the reagent blanks, the instrument was reset to zero. The standard solutions were produced right before the analyses. The prepared samples were inhaled into the nebulizer and turned into aerosol before entering the atomizer. The

linear calibration curve for each metal was used to determine the concentrations. Heavy metals analyses were done at University of Diyala Department of Chemistry, University of Baghdad Biology Department, Ministry of Industry Ibn- Sina Company.

2.12 Statistical Analysis

One-way analysis of variance (ANOVA) was performed for statistical analysis of data. Tukey's test of means was implemented by using IBM SPSS Statistics –Version 23.0. Significant differences were considered at $\alpha = 0.05$ level.

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Chapter Three
Results
&
Discussions

3. Results and Discussions

In this part of the thesis, estimation of the microbiological analysis, total count of bacteria, total coliform, *E-Coli*, and molds and yeasts for irrigation water samples, soil samples, paddy samples, and rice samples were presented. Estimation of aflatoxin of rice samples by High-performance liquid chromatography method was shown, and estimation of heavy metals for irrigation water, soil, paddy, and rice samples by using Atomic Absorption Spectroscopy were presented.

3.1 Microorganisms

3.1.1 Total Bacteria of Irrigation water, Soil, Paddy, and Rice

Table (3.1) demonstrated total bacteria count in irrigation water, soil, paddy, and rice from Diyala and Salah al-Din fields. Total bacteria count for all irrigation water samples of the tested fields were lower than the permissible limit of the FDA. The range of TBC in irrigation water in this study from was found to be from 21.9×10^3 to 80.0×10^4 cfu/mL, and it was higher than TBC in Nigeria river ranges, 2.0×10^2 cfu/mL to 60.0×10^2 cfu/mL²⁰³ and higher than TBC in well water from America, 1×10^4 cfu/mL²⁰⁴. Total bacteria count for all soil, and paddy samples were higher than the permissible limit of the FDA in all tested fields of Diyala and Salah al-Din. The highest total bacteria count of soil in Salah al-Din was 80.0×10^5 cfu/g. Fig. (3.1) explained the number of samples that exceeded the permissible limit of total bacteria, which showed that all 75 samples of soil and paddy had total bacteria more than the permissible limit. All paddy samples were higher than the FDA limits for total bacteria. The results of this study were much higher than the results obtained by Alhendi and Mohammed (2018) for wheat grown in Iraq, whose results were less than the detection limit for IQS and FDA for all samples²⁰⁵. The reason for the rise of rice microorganisms from wheat is due to the conditions of cultivation and

harvesting of paddy in a high moisture content that helps the growth of microbes, unlike wheat. Total bacteria count of rice samples was lower than the permissible limit of the FDA. The highest number found in the two study areas was 76.0×10^3 cfu/g, and it was higher than the TBC of rice taken from several mills of Iraq, 8.8×10^2 cfu/g³². The reason could be because the paddy was stored for months before processing, which led to reduce the moisture, and that reduced the microorganisms count. Also, the study areas were different, in this study was include Diyala and Salah al-Din fields, while in their study, they included Babel, Diwania, and Alnajaf, which might be the reason behind these differences. Abdul-Nabi and Qasim (2020) mentioned that imported and local rice was collected, 48 samples, from the local markets of the city of Baghdad in Iraq, and they found that the highest percentage of the total number of bacteria were 16×10^5 cfu/mL for Pakistan rice sample²⁰⁶. Fig. (3.1) showed that there was no irrigation water or rice samples had total bacteria more than the FDA permissible limit.

Table (3.1) Total Bacteria cfu/mL, or cfu/g in irrigation water, agricultural soils, paddy, and rice.

	Diyala			Salah al-Din			Limit	
Samples	Field 1	Field 2	Field 3	Field 1	Field 2	Field 3	FDA	IQS
Water x10 ⁴	24.6±2.6 ^{ab}	20.2±5.4 ^b	22.8±4.3 ^{ab}	28.4±2.1 ^a	22.6±3.6 ^{ab}	28.3±2.5 ^a	10 ² -10 ⁶	—
Soil x10 ⁵	24.7±4.5 ^{bc*}	22.1±4.9 ^{b*}	20.1±3.8 ^{c*}	28.8±2.5 ^{a*}	23.3±6.9 ^{bc*}	23.0±5.3 ^{bc*}	10 ² -10 ⁶	—
Paddy x10 ⁵	27.5±3.1 ^{a*}	18.7±3.3 ^{b*}	21.1±3.2 ^{a*}	28.3±2.5 ^{a*}	18.0±3.6 ^{b*}	27.4±2.9 ^{a*}	10 ² -10 ⁶	—
Rice x10 ³	27.6±3.1 ^a	16.1±2.4 ^b	24.7±10.2 ^{ab}	25.8±4.8 ^{ab}	17.1±2.7 ^{ab}	26.4±4.7 ^{ab}	10 ² -10 ⁶	—

Note: Values are expressed as a mean (SD). Means with different letters within the same category are significantly different at $P < 0.05$. IQS = Iraqi Quality Standardization (2006), FDA = Food and Drug Administration Circular (2013), * overpassed FDA acceptable limit.

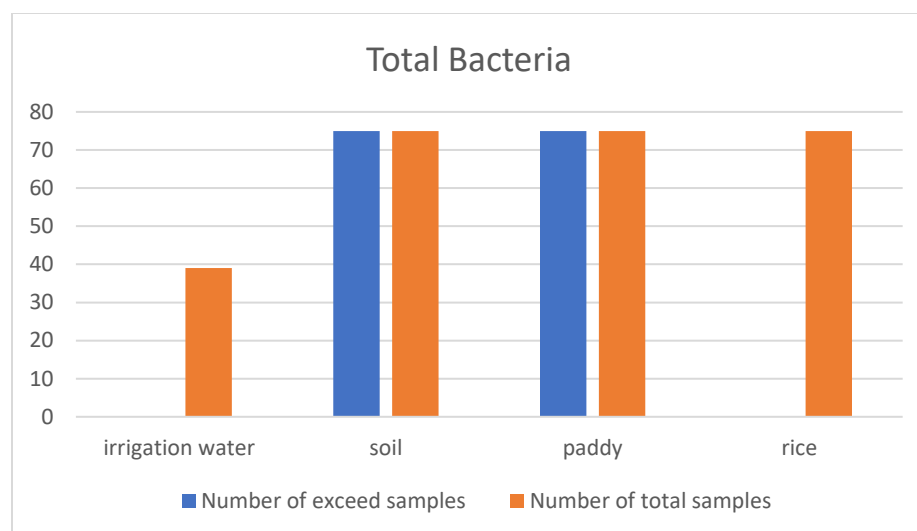


Fig. (3.1): Number of Samples Exceeded FDA Limit for The Total Bacteria Count to The Total Number of Samples.

3.1.2: Total Coliform of Irrigation Water, Soil, Paddy, and Rice

Table (3. 2) illustrated the total coliform count in irrigation water, soil, paddy, and rice samples from Diyala and Salah al-Din fields. Total coliform count in irrigation water and soil were higher than the permissible limit of FDA. The highest TC number in water was 75.0×10^3 cfu/mL from the first field of Salah al-Din, while the highest total coliform in soil was 82.0×10^3 cfu/g. The high count of microorganisms might be attributed to the rise of wild animals, rodents, flies, feces of poultry, and livestock raised by farmers in fields. As well as agricultural practices involving the use of poor microbial quality irrigation water and fertilization of crops with non - composted animal waste can also add several pathogenic microorganisms to those crops^{207 208}. The total coliform count in paddy from Diyala and Salah al-Din fields was higher than the permissible limit of FDA. However, all rice samples had TC lower than the permissible limit of the IQS and FDA in all tested fields. The highest number in the two study areas of total coliform was 1.1×10^3 cfu/g, which was higher than the results of the study conducted by AS Alhendi et al. (2020) on rice taken

from different mills of Iraq the highest number was 1.5×10^3 cfu/g³². However, the highest count of TC was lower than the reported results of Abdul -Nabi and Qasim (2020) study, 3×10^3 cfu/mL. Their study was on 48 samples of imported and local rice that collected from the local markets of the city of Baghdad in Iraq²⁰⁶. Fig (3.2) showed that irrigation water, soil, and paddy samples had total coliform more than the permissible limit, while there was no rice sample had total coliform more than the permissible limit.

Table (3.2) Total Coliform cfu/mL, or cfu/g in irrigation water, agricultural soils, paddy, and rice.

	Diyala			Salah al-Din			Limit	
Samples	Field 1	Field 2	Field 3	Field 1	Field 2	Field 3	FDA	IQS
Water x10 ³	22.0±3.4 ^{ab*}	19.4±3.9 ^{b*}	19.3±3.5 ^{b c*}	26.3±2.7 ^{a*}	14.8±2.0 ^{c*}	19.7±2.0 ^{bc*}	10 ³	—
Soil x10 ³	25.4±2.7 ^{a*}	23.8±24.5 ^{a*}	22.1±2.2 ^{a*}	28.4±3.5 ^{a*}	22.0±4.2 ^{a*}	29.9±2.7 ^{a*}	10 ³	—
Paddy x10 ⁵	27.6±3.9 ^{ab*}	19.1±2.6 ^{d*}	21.1±2.1 ^{cd*}	31.0±3.8 ^{a*}	23.8±2.7 ^{bc*}	27.2±2.7 ^{ab*}	10 ² -10 ⁴	—
Rice x10 ²	8.2±0.96 ^a	5.2±0.95 ^c	7.0±0.83 ^b	9.2±0.86 ^a	9.1±0.47 ^a	9.1±0.85 ^a	10 ² -10 ⁴	10 ² -10 ³

Note: Values are expressed as a mean (SD). Means with different letters within the same category are significantly different at $P < 0.05$. IQS = Iraqi Quality Standardization (2006), FDA = Food and Drug Administration Circular (2013), * overpassed FDA.

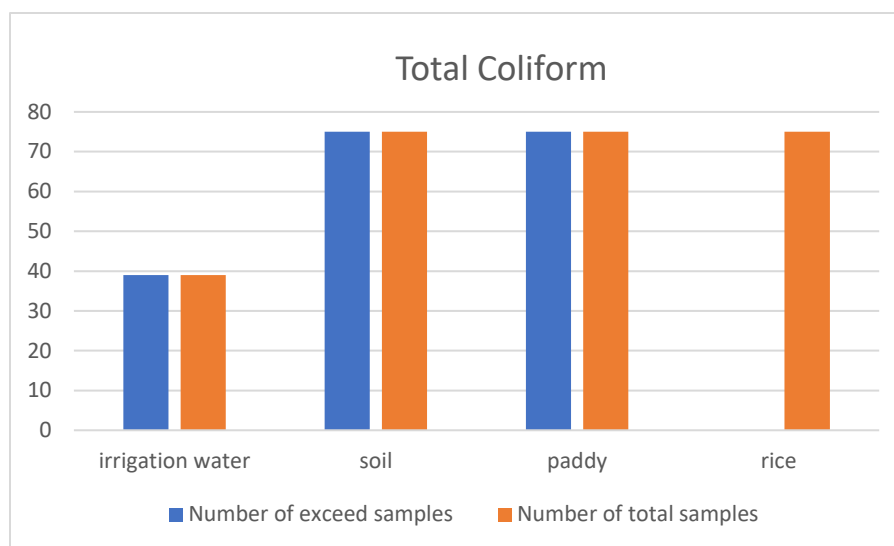


Fig. (3.2) Number of Samples Exceeded the Total Coliform Count

FDA Limit to The Total Number of Samples.

3.1.3 *E-Coli* of Irrigation Water, Soil, Paddy, and Rice

Table (3.3) demonstrated *E- Coli* in irrigation water, soil, paddy, and rice from Diyala and Salah al-Din fields. *E- Coli* in soil and rice was less than 3 cells/g, which was the detection limit of the procedure. *E- Coli* in water and paddy was less than the FDA limit. There was no sample that overpassed the FDA limit for all the tested samples. The results of irrigation water in this study were less than the irrigation water of rice fields in Korea, which ranged from 10^{-2} – 10^{-5} MPN²⁰⁷, and in another study mean of Irrigation water and mean of Paddy field were 1.69×10^{-3} , 5.99×10^{-4} respectively were higher than the results of two areas study²⁰⁹. Microbial contamination can occur during any of the steps involved in the production, harvesting, and packing. However, the steps in the production chain that have the greatest potential for pathogen contamination are soil preparation (use of organic manures) and planting and growing (use of contaminated irrigation water and animal manures^{208, 210}).

Table (3.3) *E-Coli* cell/mL or g in irrigation water, agricultural soils, paddy, and rice.

	Diyala			Salah al-Din			Limit	
Samples	Field 1	Field 2	Field 3	Field 1	Field 2	Field 3	FDA	IQS
Water	10.0±3.0 ^a	8.0±3.0 ^a	9.0±1.0 ^a	9.0±2.0 ^a	10.0±2.0 ^a	7.0±2.0 ^a	10 ² -10 ³	–
Soil	< 3	< 3	< 3	< 3	< 3	< 3	10 ² -10 ³	–
Paddy	5.0±5.0 ^a	4.0±5.0 ^a	7.0±6.0 ^a	7.0±5.0 ^a	6.0±6.0 ^a	6.0±5.0 ^a	10 ² -10 ³	–
Rice	< 3	< 3	< 3	< 3	< 3	< 3	10 ² -10 ³	–

Note: Values are expressed as a mean (SD). Means with different letters within the same category are significantly different at $P < 0.05$. IQS = Iraqi Quality Standardization (2006), FDA= Food and Drug Administration Circular (2013), * overpassed FDA acceptable limit.

3.1.4 Molds and Yeasts of Irrigation Water, Soil, Paddy, and Rice

Table (3.4) demonstrated molds and yeasts count in irrigation water, soil, paddy, and rice from Diyala and Salah al-Din fields. Molds and Yeasts for all irrigation water, soil, paddy, and rice were much higher than the permissible limit of the IQS and FDA in Diyala and Salah al-Din governorates. The molds and yeasts of Salah al-Din fields were significantly higher than those of Diyala. The largest number of molds and yeasts found in paddy, irrigation water, soil, and finally in rice which were 32.0×10^7 , 17.0×10^7 , 15.0×10^6 , and 14.0×10^6 cfu/mL or g of different fields respectively. The average of molds and yeasts in irrigation water in this study is from 1.02×10^7 cfu/mL to 14.2×10^7 cfu/mL. The microbial quality of irrigation

water can be affected by the design of wells, depth to groundwater, and rainfall ²⁰⁷. The reason for the rise of microorganisms is might be because of the harvest that takes place in a high content of moisture, which helps them grow them. In addition to the rain just before harvesting leads to wide colonization of the grains, which causes fungal contamination that can be observed on the surface of the grains and under the shell ¹¹³. The highest results in the two study areas of Molds and Yeasts in rice was 10.7×10^6 cfu/g, which was higher than the results of the study conducted by AS Alhendi et al. (2020) on the rice taken from different mills of Iraq. The highest reported count of molds and yeast in their study was 8.8×10^1 cfu/g ³². Abdul-Nabil and Qasim (2020) mentioned in their study on 48 samples of imported and local Iraqi rice that the highest total number of fungi was 11×10^3 cfu/mL ²⁰⁶. One of the reasons for the rise of molds and yeasts in rice was probably because the samples were taken from fields and the moisture content of them was high, which is the main condition for molds and yeasts growth ²⁰⁶.

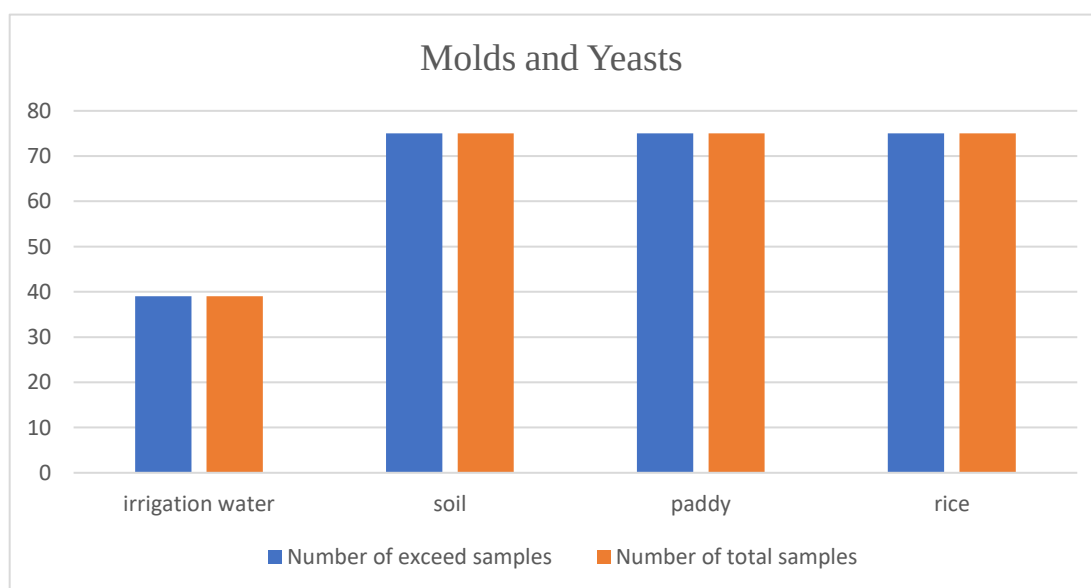


Fig. (3.3) Number of samples Exceeded the Total Molds and Yeasts
Count Limit to The Total Number of Samples

Table (3.4) Molds and yeasts cfu/mL, or cfu/g in irrigation water, agricultural soils, paddy, and rice.

	Diyala			Salah al-Din			Limit	
Samples	Field 1	Field 2	Field 3	Field 1	Field 2	Field 3	FDA	IQS
Water x10 ⁷	1.11±0.18 ^{c*}	1.02±0.20 ^{c*}	1.12±0.18 ^{c*}	14.0±2.0 ^{ab*}	14.2±1.6 ^{a*}	12.2±1.3 ^{b*}	10 ² -10 ³	—
Soil x10 ⁶	1.16±0.34 ^{b*}	1.32±0.26 ^{b*}	1.22±0.23 ^{b*}	12.1±1.79 ^{a*}	11.9±1.59 ^{a*}	11.9±1.28 [*]	10 ² -10 ³	—
Paddy x10 ⁷	2.09±0.17 ^{b*}	2.18±0.19 ^{b*}	2.06±0.15 ^{b*}	21.5±4.14 ^{a*}	21.4±1.83 ^{a*}	20.4±2.63 ^{a*}	10 ² -10 ⁴	10 ² -10 ⁴
Rice x10 ⁶	1.20±0.18 ^{c*}	1.22±0.14 ^{c*}	1.23±0.15 ^{c*}	9.60±2.46 ^{b*}	21.4±1.83 ^{a*}	12.0±1.56 [*]	10 ² -10 ⁴	10 ² -10 ⁴

Note: Values are expressed as a mean (SD). Means with different letters within the same category are significantly different at P < 0.05. IQS = Iraqi Quality Standardization (2006), FDA = Food and Drug Administration Circular (2013), * overpassed FDA

3.2 Estimation of Aflatoxin by High -Performance Liquid Chromatography

3.2.1 Selective the Optimum Condition for Aflatoxin Estimation

These tests were conducted to choose the appropriate conditions for the determination of aflatoxin according to the method of work described in section (2.9.1). Several practical tests were conducted to select the optimal conditions as shown in table (3.5), which presented a summary of the obtained results. The optimal conditions were determined based on the best peak shape and retention time. The mobile phase was 50% (Ammonium acetate + water), 30% acetonitrile, and 20% methanol. The best flow rate was 1.5 mL/min, which gave an appropriate time to separate the aflatoxin without interfering the peak with the solvent. The optimal wavelength was 270 nm and pH, 4.

Table (3.5) Types of Solutions Used as Mobile Phases

Parameters	Type parameters	Optimum value
Mobile phase	50% acetate solution + 50% methanol 50% acetate solution + 50% acetonitrile 50% acetate solution + 30% acetonitrile + 20% methanol	50% acetate solution + 30% acetonitrile + 20% methanol
pH	3,4,5	4
Flow rate mL/min.	1.0, 1.2, 1.5	1.5
Wavelength nm.	270, 254, 235	270

3.2.2 Application on Standard Aflatoxins

As in (2.8.2), 10 μ l standard of total aflatoxins, 1 ng/mL and 3 ng/mL, was injected through the injection system of HPLC device under optimum conditions mentioned in Table (3.5). The separation column of dimensions ODS - C18 (25 cm L * 0.46 cm I.D. and 5 μ m particle size) diameter were used. The obtained results as in Fig. (3.4) and (3.5) showed that the standard solution of aflatoxin gave four partial separated peaks which referred to four types of aflatoxins (G1, G2, B1, and B2). The total area of the four peaks in the chromatogram was considered for the determination of total aflatoxins for the tested samples.

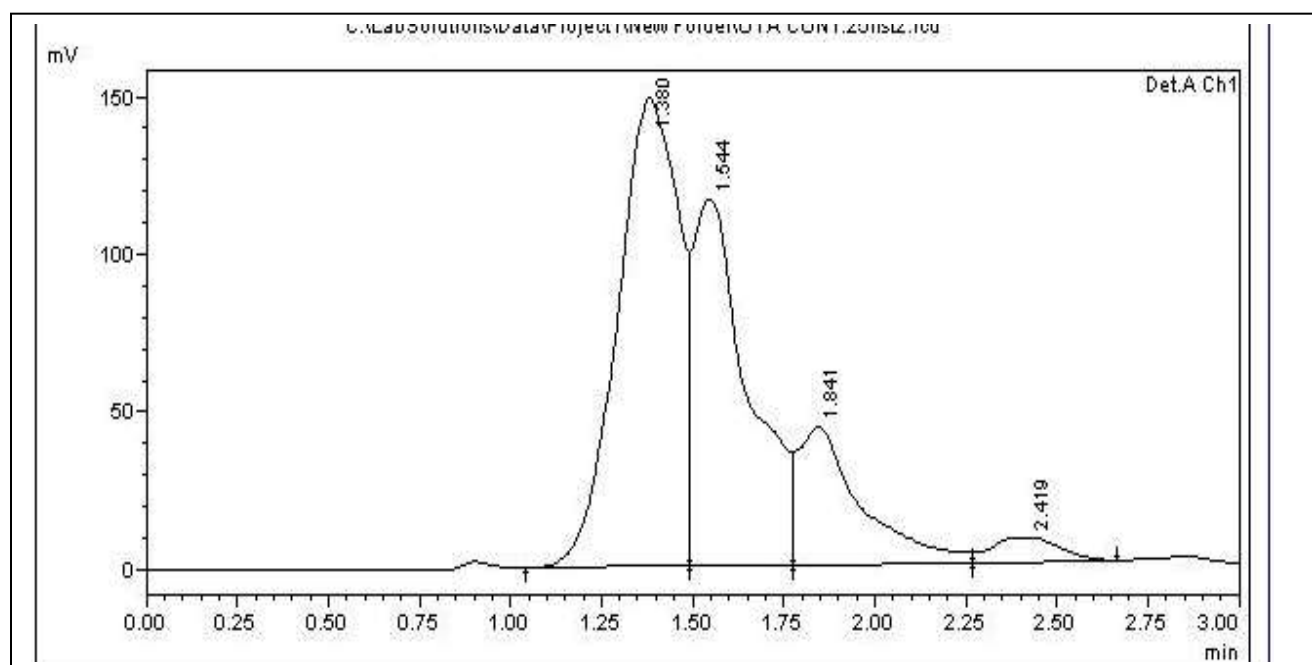


Fig. (3.4): A chromatogram of Standard Aflatoxin (1 ng/mL).

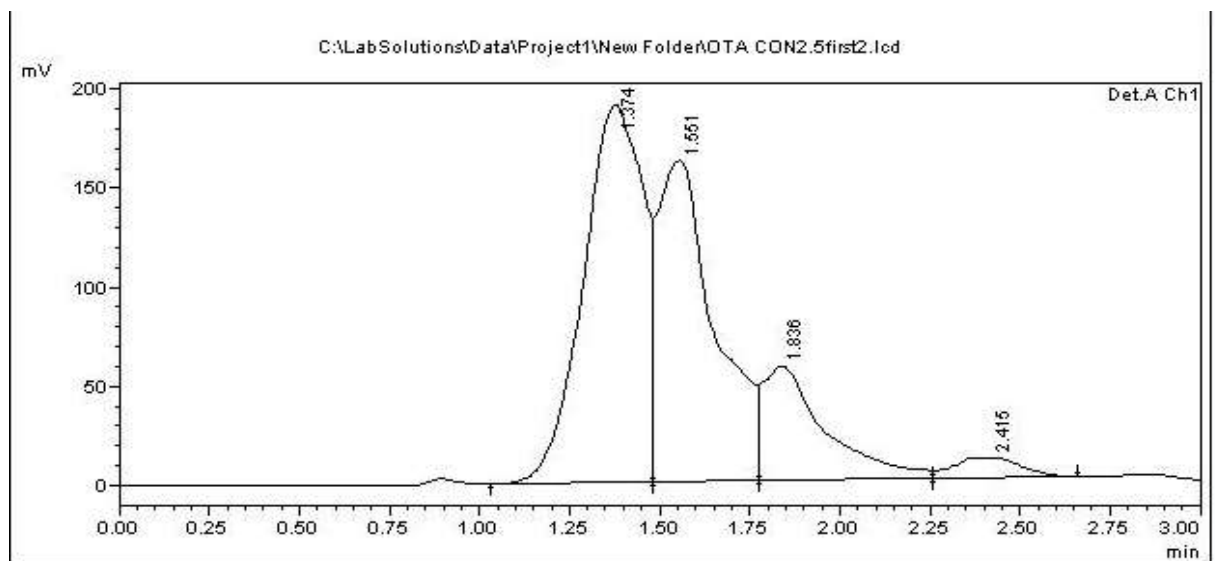


Fig. (3.5): A chromatogram of Standard Aflatoxin (3 ng/mL).

3.2.3 Estimation of Aflatoxin in Rice by Using HPLC Device

As in (2.9.3) the rice was injected after filtration using a microfilter, 0.45 μm , through the injection system of Reverse high-performance liquid chromatography device. The results obtained were shown in table (3.5) and fig. (3.6), (3.7), (3.8), (3.9), (3.10), (3.11), (3.12), (3.13), (3.14), (3.15), which revealed that all samples contained different values and different types of aflatoxins.

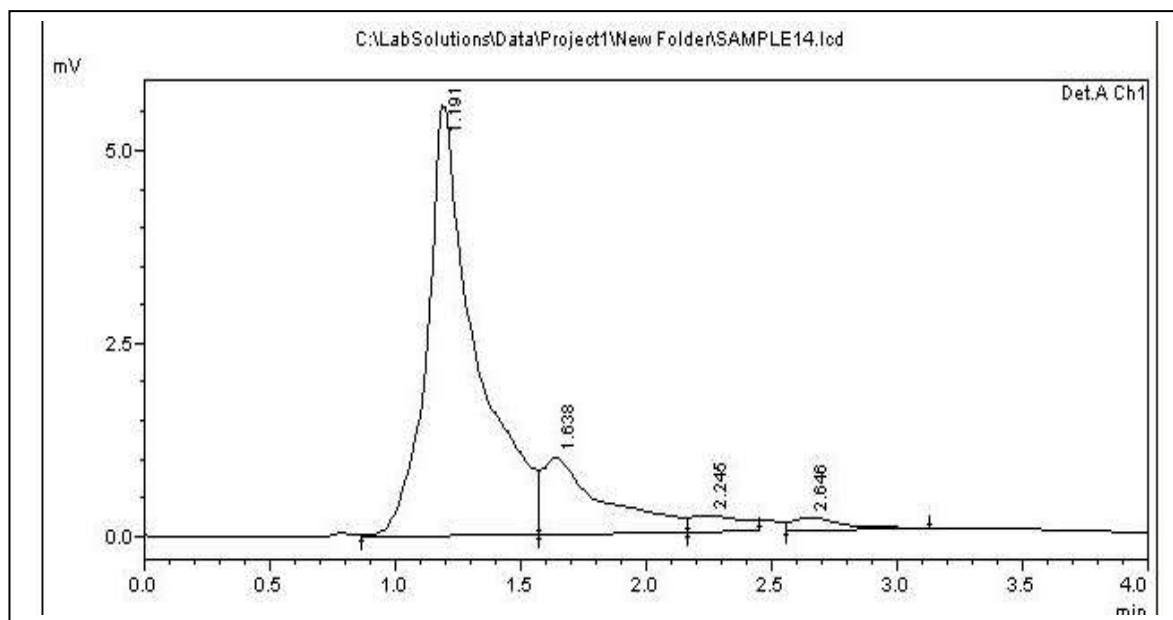


Fig. (3.6): A chromatogram of Rice Sample (14)

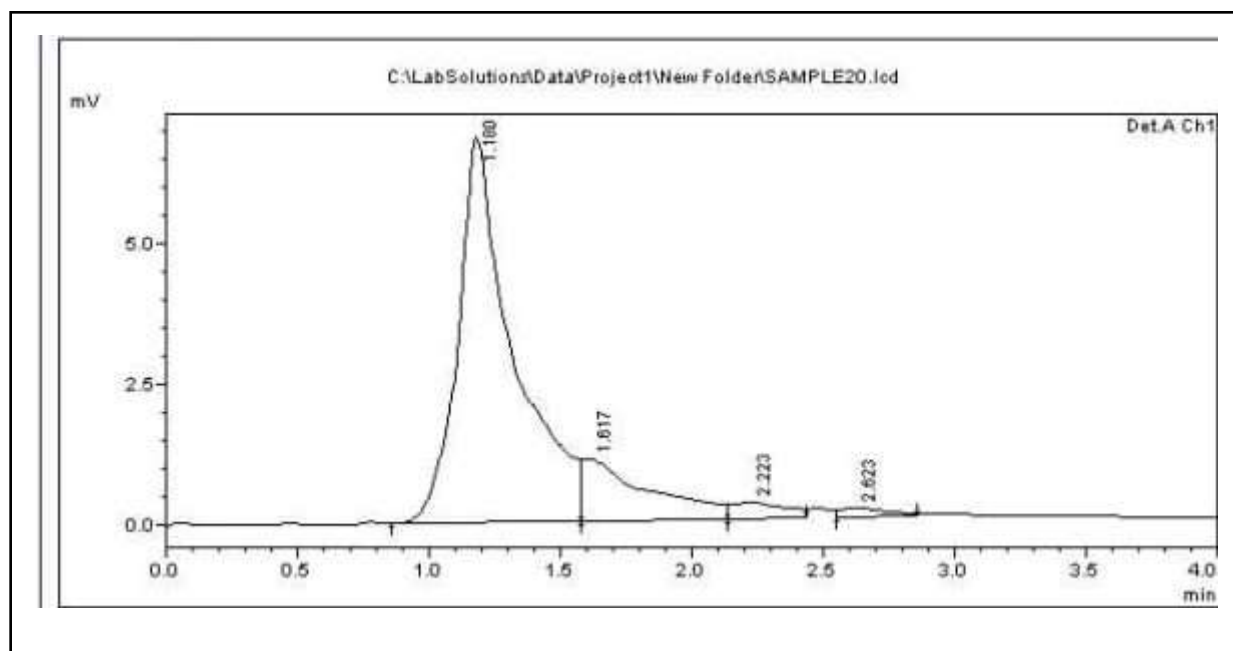


Fig. (3.7): A chromatogram of Rice Sample (20)

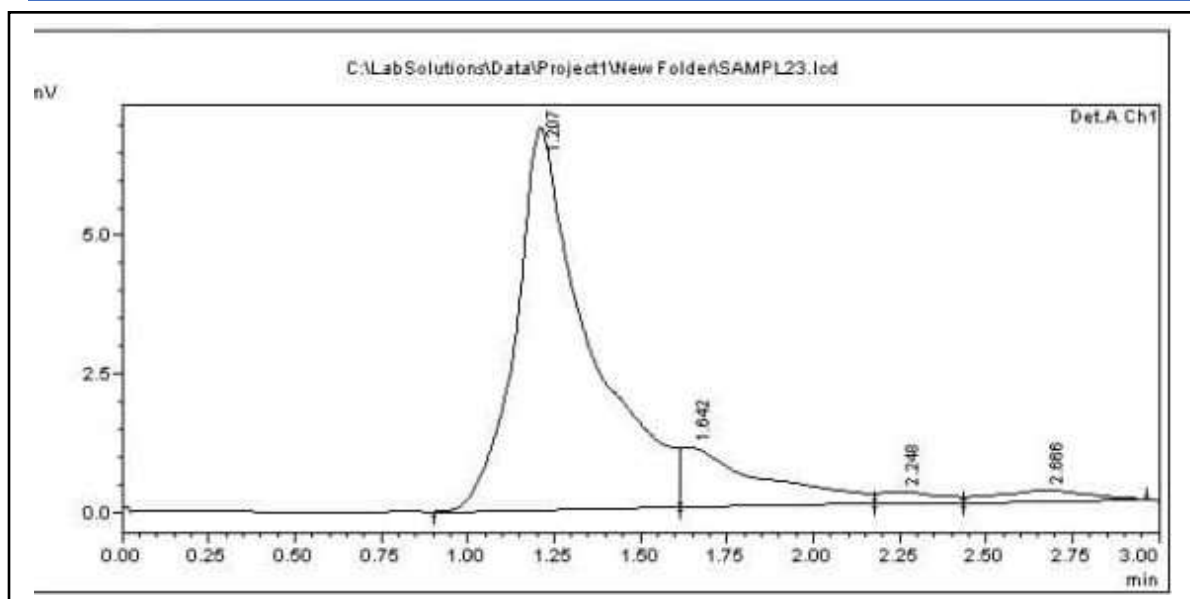


Fig. (3.8): A chromatogram of Rice Sample (23)

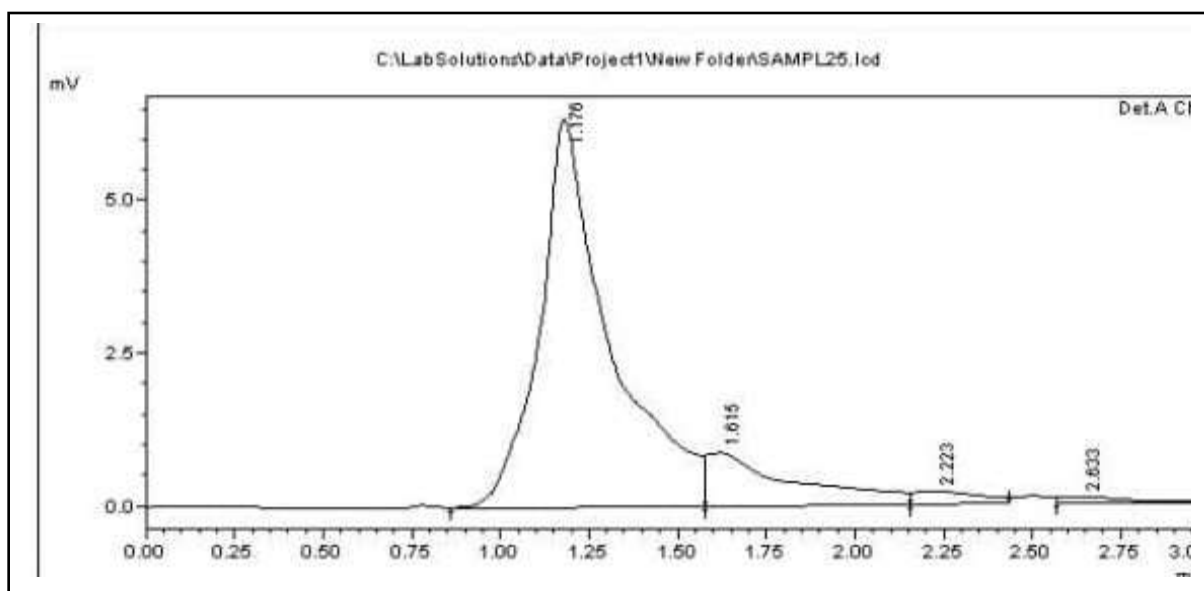


Fig. (3.9): A chromatogram of Rice Sample (25)

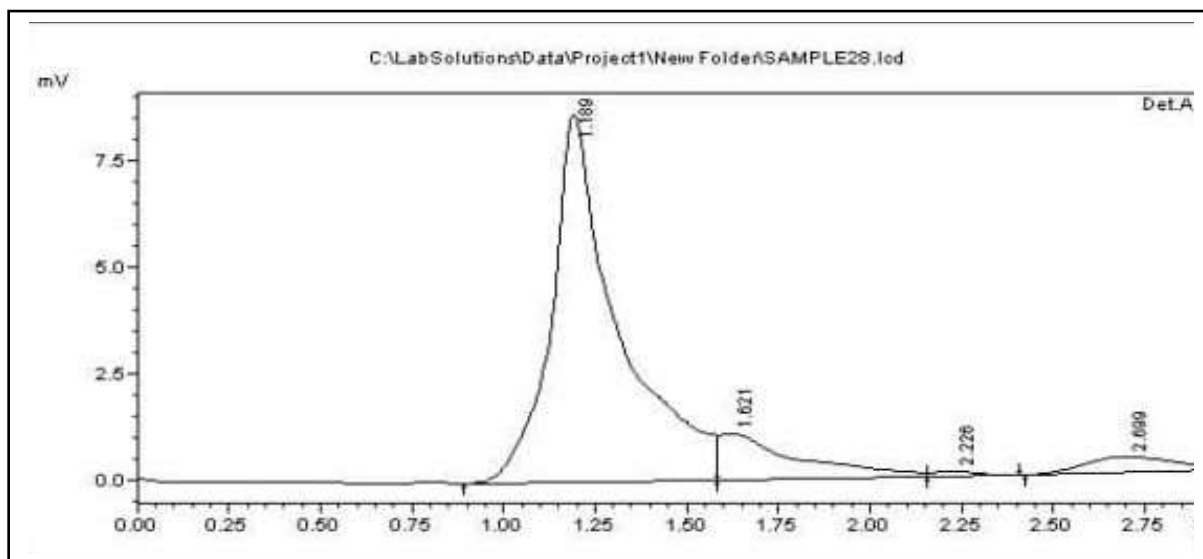


Fig (3.10) A chromatogram of Rice Sample (28)

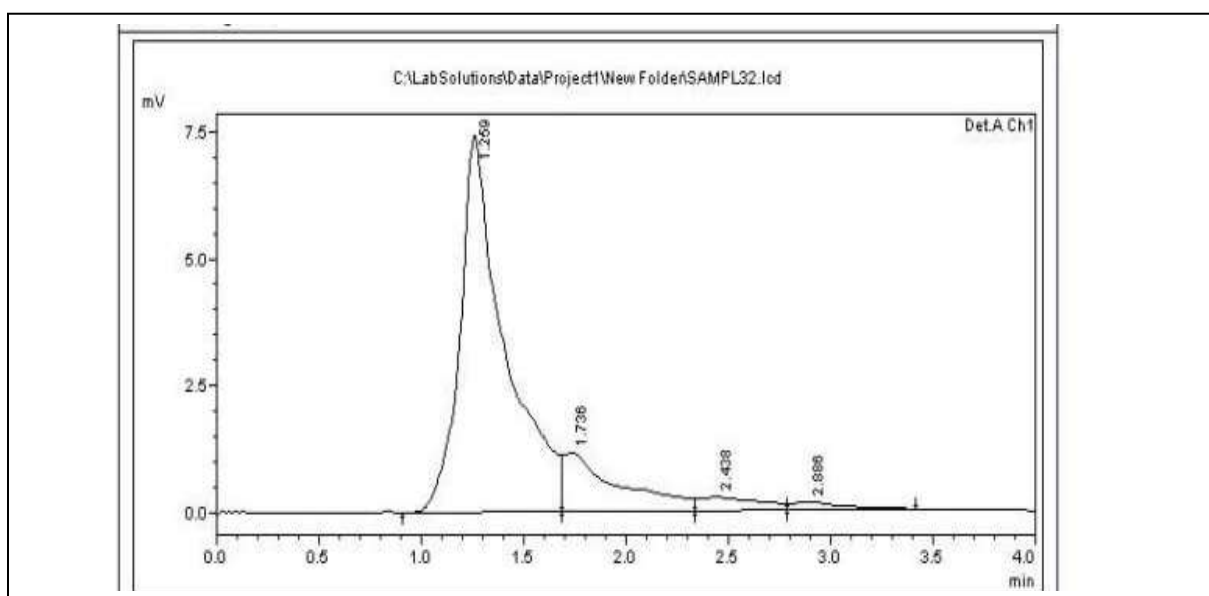


Fig. (3.11): A chromatogram of Rice Sample (32)

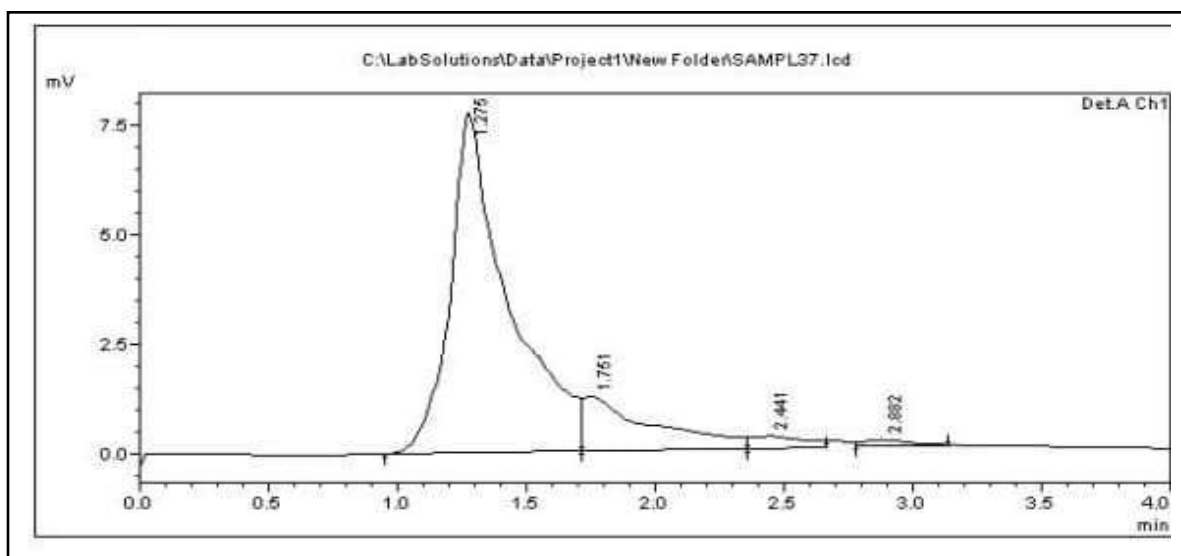


Fig. (3.12): A chromatogram of Rice Sample (37)

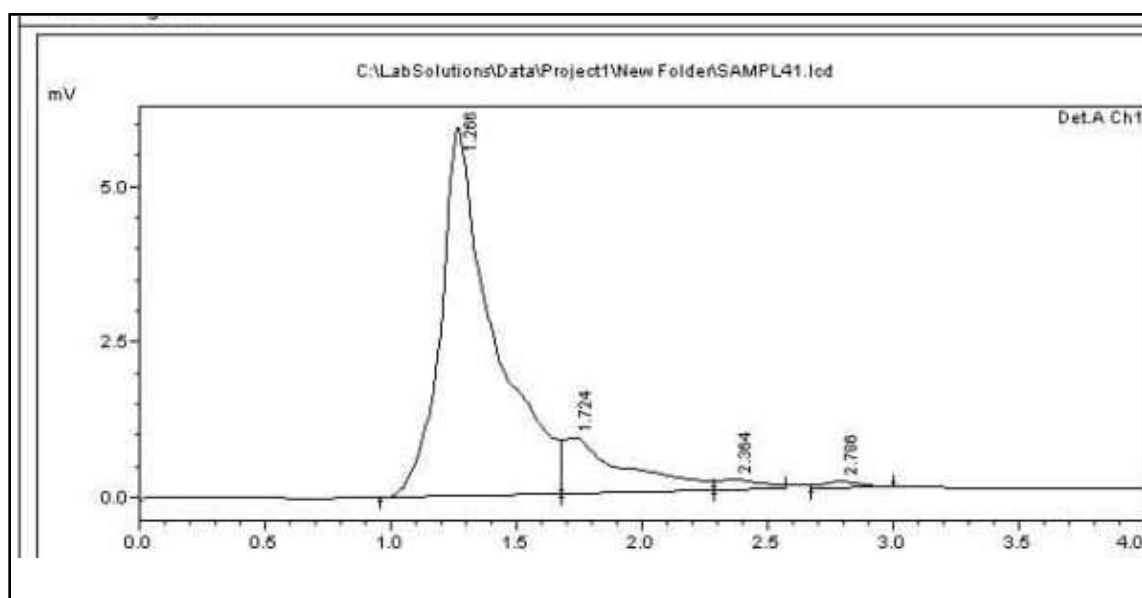


Fig. (3.13): A chromatogram of Rice Sample (41)

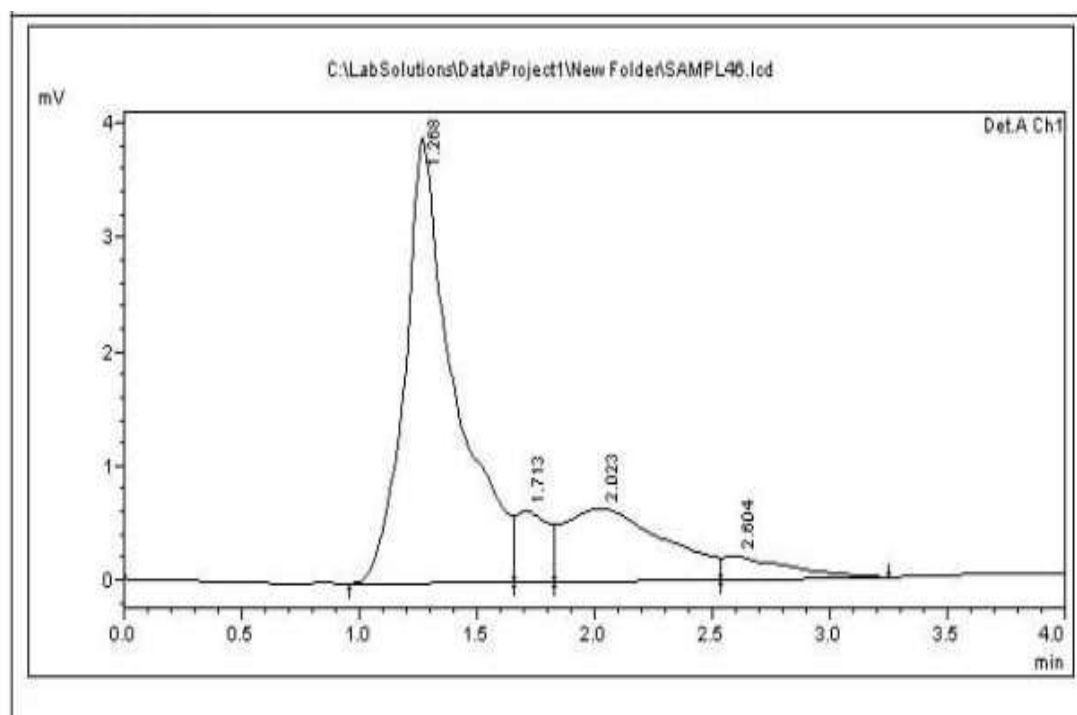


Fig. (3.14): A chromatogram of Rice Sample (46)

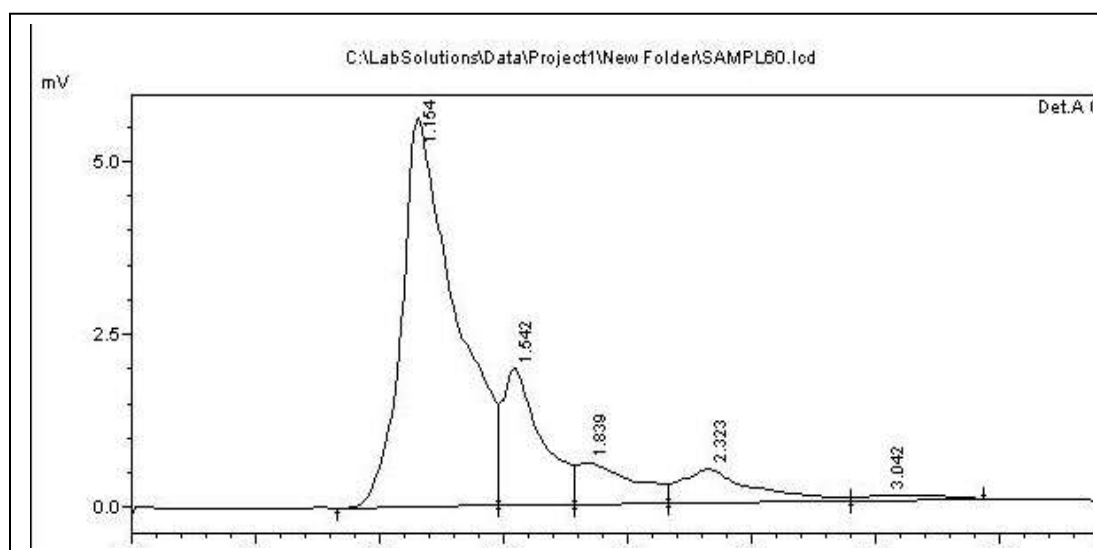


Fig. (3.15): A chromatogram of Rice Sample (60)

3.3 Aflatoxins Concentration

The results of aflatoxin for most rice samples of Diyala and Salah al-Din fields were higher than the limit of the CEC, 4µg/kg. fig. (3.10) explained the number of samples that exceeded the permissible limit of total aflatoxin, which showed that out of the 75 rice samples, 66 samples were containing total aflatoxin more than the limit. The lowest and highest sample containing aflatoxin in the Diyala field was 1.19 µg/kg and 16.40 µg/kg respectively. The average in this study was 5.0 to 6.4 µg/kg. Where these results were close to the results of the study conducted by Al-Shuhaib (2020) on 109 rice samples obtained from the middle Euphrates region in Iraq, where the results showed that 32 samples were within the limit of total aflatoxin, while 77 samples were contaminated with aflatoxin ⁵⁹. Also, the results were consistent with the results of the study conducted by AS Alhendi and others (2020) for seven different mills in three provinces of Iraq, Babel, Alnajaf, and Aldiwania. They mentioned that the range of total aflatoxins was from 1.1 to 8.8 µg/kg for rice samples ³².

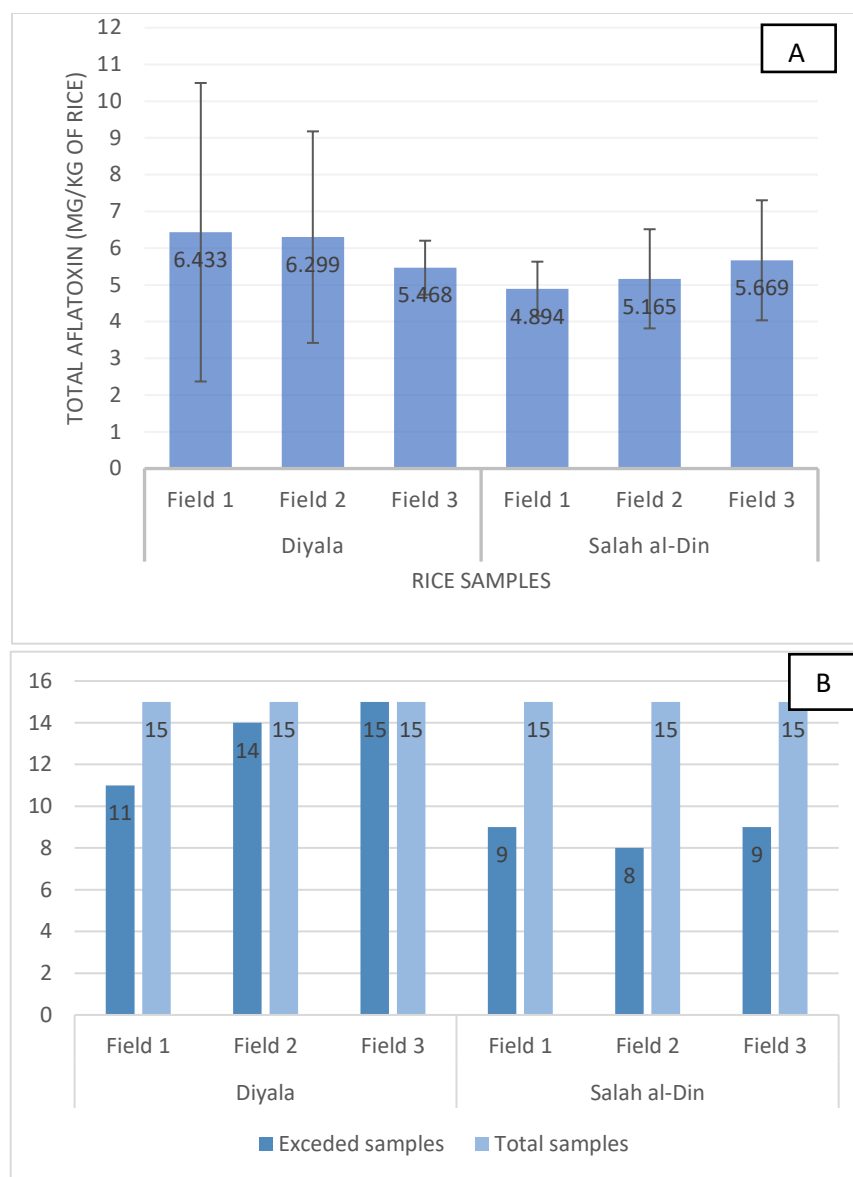


Fig. (3.16) A. Total Aflatoxin for Rice Samples of The Two study Areas (Six fields); B: Exceeded samples to Total Tested Samples of The Six Fields.

Limit was 4 μ g/kg (CEC, 2006).

3.4 Calibration Curves of Heavy Metals

Atomic Absorption Spectrophotometer Analysis (AAS) was used to measure the elements, Fe, Pb, Cu, Mn, Cd, Mg, Al, and Co. The calibration curves for all of the metals were created individually using varied concentrations of standard solutions.

The standard solutions were produced right before the analyses. The shapes of the calibration curves for the measured metals were shown in fig. (3.17), (3.18), (3.19).

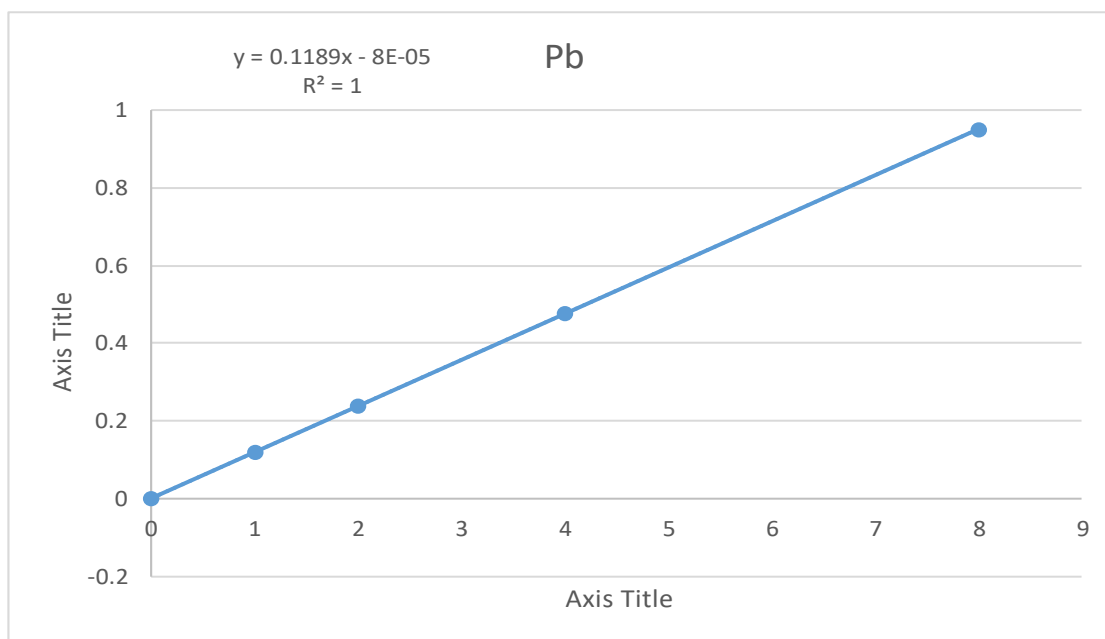


Fig. (3.17): Calibration graph for Pb using AAS.

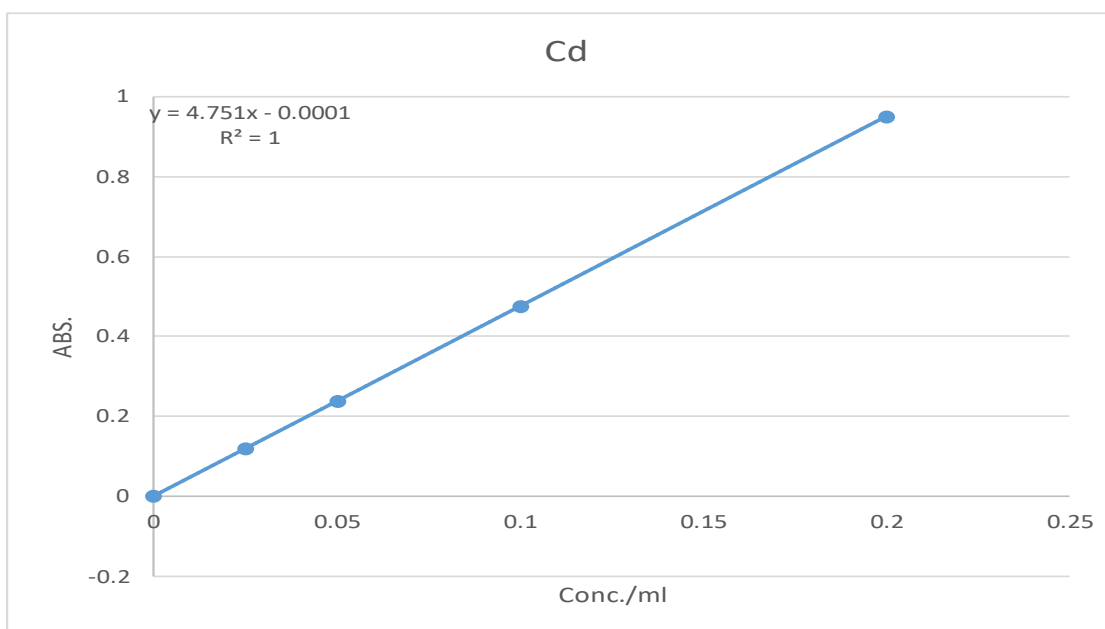


Fig. (3.18): Calibration graph for Cd using AAS.

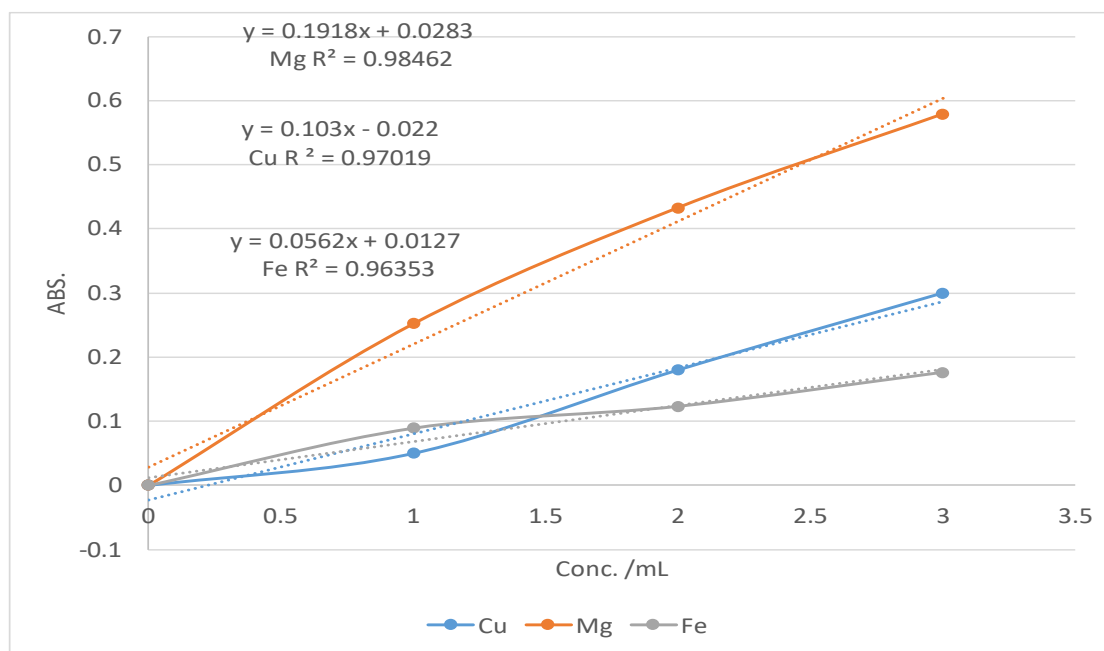


Fig. (3.19): Calibration graph for (Cu, Mg, Fe) using AAS

3.5: Estimation Heavy metals of Irrigation Water, Soil, Paddy and Rice.

3.5.1. Lead Concentration

Table (3.6), and fig (3.20) demonstrated Pb metal concentrations in irrigation water, soil, paddy, and rice from Diyala and Salah al-Din fields. The concentration of Pb for all irrigation water samples in Diyala and Salah al-Din governorates was much higher than the permissible limit of the IQS and WHO. The lowest concentration of Pb was found in field 1 of Diyala, which the source of the irrigation water was well, while the highest concentration of lead was found in the pooled source of irrigation water. The average concentration of Pb in Diyala fields was 62, 97, and 133 times higher than the permissible limit for the first, second, and third fields respectively, and it was higher than by 104, 111, and 133 times for the first, second, and third fields of Salah al-Din fields respectively. There were significant differences between Pb concentrations of irrigation water among fields. The high concentration of heavy metals mostly was more in pooled water followed by river water and lastly in the

well water as expected. Compared with the literature, Pb concentration in the Tigris River was 0.05 mg/L²¹¹, which was greater than the limit given by WHO, 0.01 mg/L. However, in this study, Pb values range was 0.286-1,801 mg/kg, which was much higher than Pb concentration of the Tigris River.

The concentration of Pb for all soil samples in Diyala and Salah al-Din governorates was less than the permissible limits of the WHO. Also, there were no significant differences between Pb concentrations of soils among fields. The lead concentration for this study ranged from 30.15-33.63 mg/kg, which was higher than the concentration of lead in the soil of Kurdistan, Iraq. In their study, they found that Pb concentration was 14.67 mg/kg for soil depth 20 cm, while it was 6.5 mg/kg for the surface layer of the soil²¹².

The concentration of Pb in paddy for all samples was much higher than the permissible limit of the WHO. Pb concentration in Diyala was 9, 10, and 10 times higher than the permissible limit for the first, second, and third fields, respectively, and it was higher by 10, 10, and 11 times for the first, second, and third fields of Salah al-Din governorate respectively. There was no significant difference between paddy concentration among the fields. Pb concentrations in the two study areas was ranged from 0.642 to 3.481 mg/kg, which was similar to the results obtained in the Malaysia, 2.06 mg/kg²¹³.

Also, all the concentration of Pb in rice was higher than the permissible limit of the IQS and WHO. Pb concentration was approximately 15 times higher than the permissible limits. There were no significant differences between Pb concentrations of rice among fields. All rice samples were exceeded the IQS and WHO limit for Pb, while all other detected metals were within the limit. Pb concentration of the produced rice from Diyala and Salah al-Din was ranged from 1.805-4.776 mg/kg, which was much higher than Pb concentration of rice produced from Alnajaf and

Aldiwaniaa provinces, Iraq that was ranged from 0.093 to 0.339 mg/kg ³². Pb concentration of rice in Al-Mishkhab, in Iraq was 0.778 mg/kg ²¹⁴. Pb concentration of wheat planted in several region of Iraq ranged from 0.017 to 0.191 mg/kg ¹⁰, which was significantly lower than the concentration of Pb in rice. The reason for the high concentration of lead in rice might be because of its high concentration in irrigation water, which is transferred from water to plant because the planting of rice needs soaking in water for a while. Internationally, Pb concentrations in Bengal rice were 0.42-6.39 mg/kg and that exceeded 100 times the WHO limit, which is posing a significant threat to human health ²¹⁵.

Table (3.6) Concentration of Pb mg/L, or mg/kg in irrigation water, agricultural soils, paddy, and rice.

	Diyala			Salah al-Din			Limit	
Samples	Field 1	Field 2	Field 3	Field 1	Field 2	Field 3	WHO/ FAO	IQS
Water	0.626±0.329 ^{b*}	0.971±0.489 ^{ab*}	1.322±0.401 ^{a*}	1.047±0.342 ^{ab}	1.111±0.359 ^{a*}	1.332±0.402 ^{a*}	0.01	0.01
Soil	32.301±1.116 ^a	31.966±1.115 ^a	31.431±0.789 ^a	31.431±0.789 ^a	31.643±1.157 ^a	31.188±3.033 ^a	85	—
Paddy	1.810±0.871 ^{a*}	2.059 ± 0.662 ^{a*}	1.970±0.750 ^{a*}	2.123 ±0.568 ^{a*}	2.257±0.556 ^{a*}	2.464±0.613 ^{a*}	0.2	—
Rice	3.071±0.815 ^{a*}	3.168±0.795 ^{a*}	3.273±0.687 ^{a*}	3.183±0.800 ^{a*}	3.586±0.829 ^{a*}	3.358±0.771 ^{a*}	0.2	0.2

Note: Values are expressed as a mean (SD). Means with different letters within the same category are significantly different at $P < 0.05$. WHO/ FAO =World Health Organization, Food and Agriculture Organization (2011). IQS = Iraqi Quality Standardization (2009). *overpassed WHO/IQS acceptable limit.

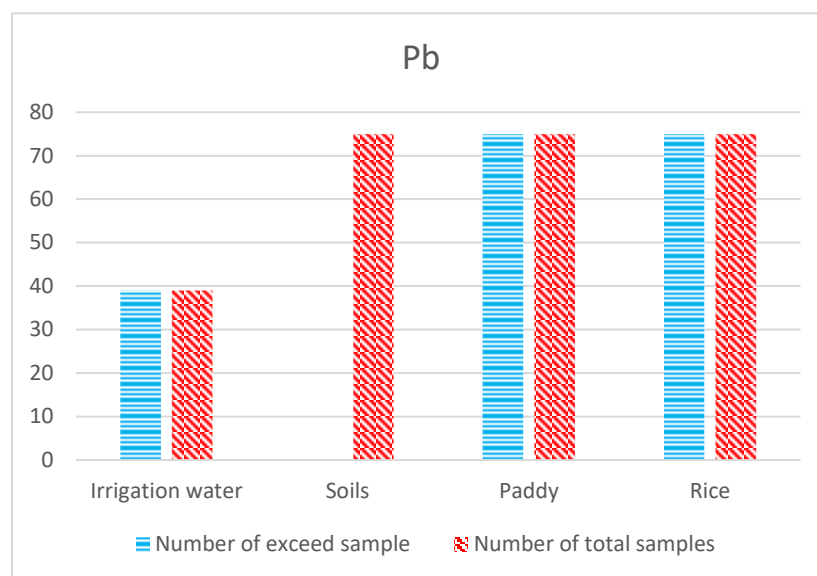


Fig. (3.20) Number of Samples Exceeded the Heavy Metals Limit to The Total Number of Samples: Pb

3.5.2 Cadmium Concentration

Table (3.7) and fig (3.21) demonstrated Cd metal concentrations in irrigation water, soil, paddy, and rice from Diyala and Salah al-Din fields. The concentration of Cd for all irrigation water samples in Diyala and Salah al-Din governorates was higher than the permissible limit of IQS and WHO. There was no significant difference between Cd concentrations of different fields. The range of Cd of irrigation water samples of this study was 0.0021-0.0096 mg/L, which was lower than the Cd concentration of a river in Cameron, 0.07- 2.83 mg/L ²¹⁶, and irrigation water of Nigeria, 0.037 mg/L ²¹⁷.

The concentration of Cd for all soil, paddy, and rice samples in Diyala and Salah al-Din governorates were less than the permissible limits of the IQS and WHO. There were no significant differences among the fields in soil and rice but there was a significant difference in paddy concentration of different fields.

The Cd concentration of rice soil in Mishkhab Area, in Iraq was ranged from 0.21-0.33 mg/kg²¹⁴, which was lower than the Cd concentration of the two study areas of this study, 0.254-1.932 mg/kg. The concentration of cadmium of soil in Mogla Bazar in Sylhet, Bangladesh was 0.05 mg/kg, which was less than the permissible limit of the WHO and less than the Cd concentration of this study²¹⁸. The concentrations of Cd in fields of Diyala and Salah al-Din were 0.009-0.092 mg/kg were less than those in Malaysia were 0.13 and 0.74 mg/kg¹³⁵. Cd concentration of rice produced at Najaf and Diwaniaa provinces, Iraq was 0.055 – 0.066 mg/kg³², which was more than the concentration of the current study, 0.014-0.095 mg/kg. Almayahi and Aljarrah mentioned that Cd concentration of Anber rice of Nasiriyah, Iraq was 0.0468 mg/kg¹⁵⁰. Internationally, Jaswant et al. (2011) reported that Cd of Indian rice was 0.014 mg/ kg²¹⁹.

Table (3.7) concentration of Cd mg/L, or mg/kg in irrigation water, agricultural soils, paddy, and rice.

	Diyala			Salah al-Din			Limit	
Samples	Field 1	Field 2	Field 3	Field 1	Field 2	Field 3	WHO/ FAO	IQS
Water	0.006±0.002 ^{a*}	0.007±0.002 ^{a*}	0.005±0.002 ^{a*}	0.007±0.002 ^{a*}	0.007±0.002 ^{a*}	0.005±0.002 ^{a*}	0.003	0.003
Soil	1.128 ±0.492 ^a	1.089 ± 0.520 ^a	1.172±0.483 ^a	1.614±0.286 ^a	1.270 ±0.372 ^a	1.276±0.453 ^a	3	—
Paddy	0.054±0.030 ^{ab}	0.072 ±0.014 ^a	0.055±0.022 ^{ab}	0.055±0.028 ^{ab}	0.057±0.022 ^{ab}	0.042 ±0.019 ^b	0.4	—
Rice	0.030±0.015 ^a	0.065±0.025 ^a	0.062±0.025 ^a	0.054 ±0.033 ^a	0.050±0.013 ^a	0.065±0.028 ^a	0.4	0.4

Note: Values are expressed as a mean (SD). Means with different letters within the same category are significantly different at $P < 0.05$. WHO/ FAO =World Health Organization, Food and Agriculture Organization (2011). IQS = Iraqi Quality Standardization (2009). *overpassed WHO/IQS acceptable limit.

3.5.3 Copper Concentration

Table (3.8) demonstrated Cu metal concentrations in irrigation water, soil, paddy, and rice from Diyala and Salah al-Din fields. The concentration of Cu for all irrigation water, soil, paddy, and rice samples in Diyala and Salah al-Din governorates was less than the permissible limits of the IQS and WHO. There were significant differences in irrigation water, soil, and rice fields, but there was no significant difference for paddy samples of different fields.

Cu concentration of Greenbelt Area North of AL-Najaf Al-Ashraf City in Iraq was 0.0079 – 0.0133 mg/L ²²⁰, which was lower than the range of the current study, 0.003-0.903 mg/L. Cu concentration of a river in Delhi region (India) was 0.64 mg/L ²⁸. Copper concentrations of all soil samples ranged 1.92-8.28 mg/kg, which were less than those ranged 13.76 - 60.56 mg/kg of rice soil in Nepal. The soil contamination with Cu could be due to wastewater irrigation, chicken manure, and pesticides in the farmland ¹³⁹. The Cu concentration of soils in Bangladesh was below the permissible limits recommended by Indian Standard Awashthi and European Union ²²¹. The concentrations Cu in the fields of Diyala and Salah al-Din were 0.064-0.962 mg/kg were less than those in Malaysia were 0.13 and 0.74 mg/k ¹³⁵. Copper concentrations of rice agreed with the Kormoker et al. (2020); Kelle et al. (2021) studies that they mentioned the Cu concentrations in Tangail district of Bangladesh and Nigerian rice samples were within the limit of WHO ^{212,219}.

Table (3.8) concentration of Cu mg/L, or mg/kg in irrigation water, agricultural soils, paddy, and rice.

	Diyala			Salah al-Din			Limit	
Samples	Field 1	Field 2	Field 3	Field 1	Field 2	Field 3	WHO/ FAO	IQS
Water	0.050±0.054 ^c	0.079±0.060 ^{bc}	0.275±0.222 ^b	0.452±0.271 ^{ab}	0.414±0.332 ^{ab}	0.665±0.327 ^a	2	1
Soil	5.507±1.235 ^{ab}	3.993 ± 1.918 ^c	5.738±1.55 ^{ab}	5.33±1.038 ^{abc}	6.343±1.152 ^{ab}	5.542±0.866 ^{abc}	140	—
Paddy	0.318 ±0.260 ^a	0.447 ±0.236 ^a	0.532±0.188 ^a	0.429 ±0.215 ^a	0.548 ±0.260 ^a	0.522 ±0.271 ^a	20	—
Rice	0.089±0.038 ^b	2.082±0.0886 ^a	0.048±0.024 ^b	0.048±0.023 ^b	0.050±0.023 ^b	0.042±0.021 ^b	10	—

Note: Values are expressed as a mean (SD). Means with different letters within the same category are significantly different at $P < 0.05$. WHO/ FAO =World Health Organization, Food and Agriculture Organization (2011). IQS = Iraqi Quality Standardization (2009). *overpassed WHO/IQS acceptable limit.

3.5.4 Iron Concentration

Table (3.8) and fig (3.21) demonstrated Fe metal concentrations in irrigation water, soil, paddy, and rice from Diyala and Salah al-Din fields. The concentration of Fe for all irrigation water samples in Diyala and Salah al-Din governorates was higher than the permissible limit of IQS and WHO. There was no significant difference between Fe concentrations of different fields. The concentration of Fe for all soil, paddy, and rice samples in Diyala and Salah al-Din governorates was less than the permissible limits of the IQS and WHO. There were significant differences among the fields. Gokalp and Mohammed (2019) reported that Fe concentration of the Heshkaro stream of Duhok province, Iraq was ranged 0.0032 - 2.20 mg/L, which was higher than the observed range of this study 0.415-0.831 mg/L of Fe. Also, Fe concentration of southwest Baghdad, wells -Yusufiyah in Iraq was 0.1-0.63 mg/L²²³. Iron concentrations of all soil samples ranged from 30.2-40.32 mg/kg, which were within the range of a peri-urban area of Lahore district India, 8.89- 35.03 mg/kg²²⁴. The concentrations of Fe in paddy were 6.062-14.37 mg/kg lower than the limit and less than the concentrations of the study conducted in India and united states²²⁵.

Table (3.9) concentration of Fe mg/L, or mg/kg in irrigation water, agricultural soils, paddy, and rice.

	Diyala			Salah al-Din			Limit	
Samples	Field 1	Field 2	Field 3	Field 1	Field 2	Field 3	WHO/FAO	IQS
Water	0.533±0.053 ^{a*}	0.452±0.138 ^{a*}	0.542±0.201 ^{a*}	0.547±0.184 ^{a*}	0.555±0.111 ^{a*}	0.678±0.159 ^{a*}	0.3	0.3
Soil	32.592±1.172 ^{ab}	34.388± 2.419 ^a	31.696 ± 1.1 ^b	32.522 1.01 ^{ab}	30.510±3.898 ^b	32.456±1.079 ^{ab}	150	—
Paddy	9.381±2.437 ^{ab}	9.082±1.903 ^b	10.231±1.691 ^{ab}	10.708±1.672 ^b	11.501±1.287 ^a	11.312±1.277 ^a	—	—
Rice	2.393±1.275 ^b	8.00±4.472 ^a	2.634±0.742 ^b	2.593±0.685 ^b	2.586±0.888 ^b	2.649±0.877 ^b	15	—

Note: Values are expressed as a mean (SD). Means with different letters within the same category are significantly different at $P < 0.05$. WHO/ FAO =World Health Organization, Food and Agriculture Organization (2011). IQS = Iraqi Quality Standardization (2009). * overpassed WHO/IQS acceptable limit.

3.5.5 Manganese Concentration

Table (3.10) and fig (3.21) demonstrated Mn metal concentrations in irrigation water, soil, paddy, and rice from Diyala and Salah al-Din fields. The concentration of Mn for all irrigation water samples in Diyala and Salah al-Din governorates was much higher than the permissible limits of the IQS and WHO. Manganese concentration of irrigation water of all fields was 10 times or more than the limit, and there was no significant difference among the fields. The concentration of Mn for all soil samples in Diyala and Salah al-Din governorates was higher than the permissible limits of the WHO. There were no significant differences between Mn concentrations among fields. The concentration of Mn for all paddy and rice samples was less than the permissible limits of the IQS and WHO.

The observed Mn concentration of Heshkaro stream of Duhok province, Iraq was 0.011-0.253 mg/L ²²⁶, which was lower than the Mn concentration, 0.315-1.811 mg/L observed in this study this result was placed within the range of the Deepmala et al. (2017) study 12.5 -53.9 mg/kg that was done in the Indian paddy soil ²²⁷. Also, a similar range was observed in Pakistan, 9.12- 30.08 mg/kg ²²⁸. Manganese concentration was 6.062-14.37 mg/kg lower than the limit and less than the concentrations of the study conducted in India and united states ²²⁵. Manganese concentration in rice within the limits of WHO and the results agreed with the ^{215,222}.

Table (3.10) concentration of Mn mg/L, or mg/kg in irrigation water, agricultural soils, paddy, and Rice

Samples	Diyala			Salah al-Din			Limit	
	Field 1	Field 2	Field 3	Field 1	Field 2	Field 3	WHO/FAO	IQS
Water	0.967±0.170 ^{a*}	0.997±0.340 ^{a*}	1.284±0.547 ^{a*}	1.111±0.414 ^{a*}	1.325±0.426 ^{a*}	1.277±0.368 ^{a*}	0.1	0.1
Soil	29.557±3.906 ^{a*}	31.447±1.149 ^a	30.513±1.834 ^{a*}	30.839±1.455 [*]	1.325±0.426 ^a	31.342±1.924 ^{a*}	26	—
Paddy	2.820 ±0.366 ^a	2.966 ±0.388 ^a	2.769 ±0.516 ^a	2.769 ±0.516 ^a	3.026 ±0.475 ^a	3.095 ±0.570 ^a	—	—
Rice	2.009±0.696 ^b	0.058±0.024 ^c	2.831±0.731 ^a	2.742±0.752 ^{ab}	2.490±0.881 ^{ab}	2.677±0.747 ^{ab}	300-500	—

Note: Values are expressed as a mean (SD). Means with different letters within the same category are significantly different at $P < 0.05$. WHO/ FAO =World Health Organization, Food and Agriculture Organization (2011). IQS = Iraqi Quality Standardization (2009). * overpassed WHO/IQS acceptable limit.

3.5.6 Magnesium Concentration

Table (3.11) and fig (3.21) demonstrated Mg metal concentrations in irrigation water, soil, paddy, and rice from Diyala and Salah al-Din fields. The concentration of Mg for all irrigation water, paddy and rice samples in Diyala and Salah al-Din governorates was much less than the permissible limits of the WHO. There was no significant difference between Mg concentrations in irrigation water and paddy samples but a significant difference in rice. The concentration of Mg for all soil samples was higher than the permissible limits of WHO. There was no significant difference between Mg concentrations of different fields.

Concentrations of Mg of irrigation water samples ranges 0.452-11.87 mg/L were less than those range 2–680 mg/L in India ²²⁹. The reason for the high concentration of heavy metals in irrigation water is probably due to the disposal of waste from the stations in this area and the disposal of some household waste. The concentration of Mg in the upper layers of the soil at a depth of 0-20 was higher than its concentration in the layers of soil at a depth of (20-40) ²³⁰. Mg concentrations of paddy fields in 0–20 cm and 20–40 cm soil layers were averaged 282 and 243 mg/kg respectively, while Mg concentrations range of the two study areas was 383.08-448.32 mg/kg, which were more than the above- mentioned study. The concentration Mg 12.435-15.890 mg/kg lower than the limit and less than the concentrations of the study conducted in India and united states ²²⁵.

The concentration of Co and Al of all samples, irrigation water, soils, paddy and rice, was less than the detection limit, therefor the data has not been stated in tables

Interestingly, the heavy metal content of Irrigation water was high whereas their concentration in soil was low. The insolubility of metals because of the high pH of the soil in addition to soil worms and redox potential of soil might reduce minerals.

Also, heavy metals were absorbed by the roots of the plant, where the highest percentage was in the root, then the stem, leaves, and finally the grain, and this was might be the reason for its decrease in rice.

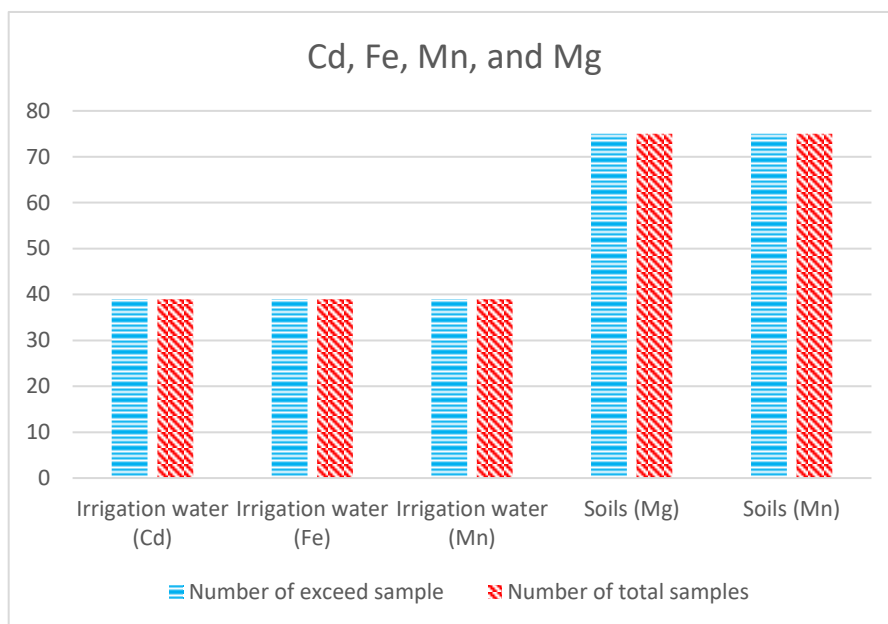


Fig. (3.21) Number of Samples Exceeded the Heavy Metals Limit to the Total Number of Samples: Cd, Fe, Mn, and Mg

Table (3.11) concentration of Mg mg/L, or mg/kg in irrigation water, agricultural soils, paddy, and rice.

Samples	Diyala			Salah al-Din			Limit	
	Field 1	Field 2	Field 3	Field 1	Field 2	Field 3	WHO /FAO	IQS
Water	11.007±0.611 ^a	11.001±0.522 ^a	11.187±0.545 ^a	10.993±0.668 ^a	11.109±0.640 ^a	11.087±0.526 ^a	50	—
Soil	399.590±17.71 ^{a*}	392.589±5.921 ^{a*}	394.661±7.419 ^{a*}	396.862±11.54 ^{a*}	395.141±5.163 ^{a*}	394.028±8.08 ^{a*}	255	—
Paddy	13.940 ±1.221 ^a	13.854 ±1.268 ^a	14.126±1.344 ^a	13.838 ±1.389 ^a	13.838 ±1.226 ^a	13.906±1.124 ^a	—	—
Rice	13.838±1.225 ^a	2.585±0.603 ^b	14.077±1.196 ^a	13.562±1.009 ^a	13.838 ±1.226 ^a	13.985±1.141 ^a	143.50	—

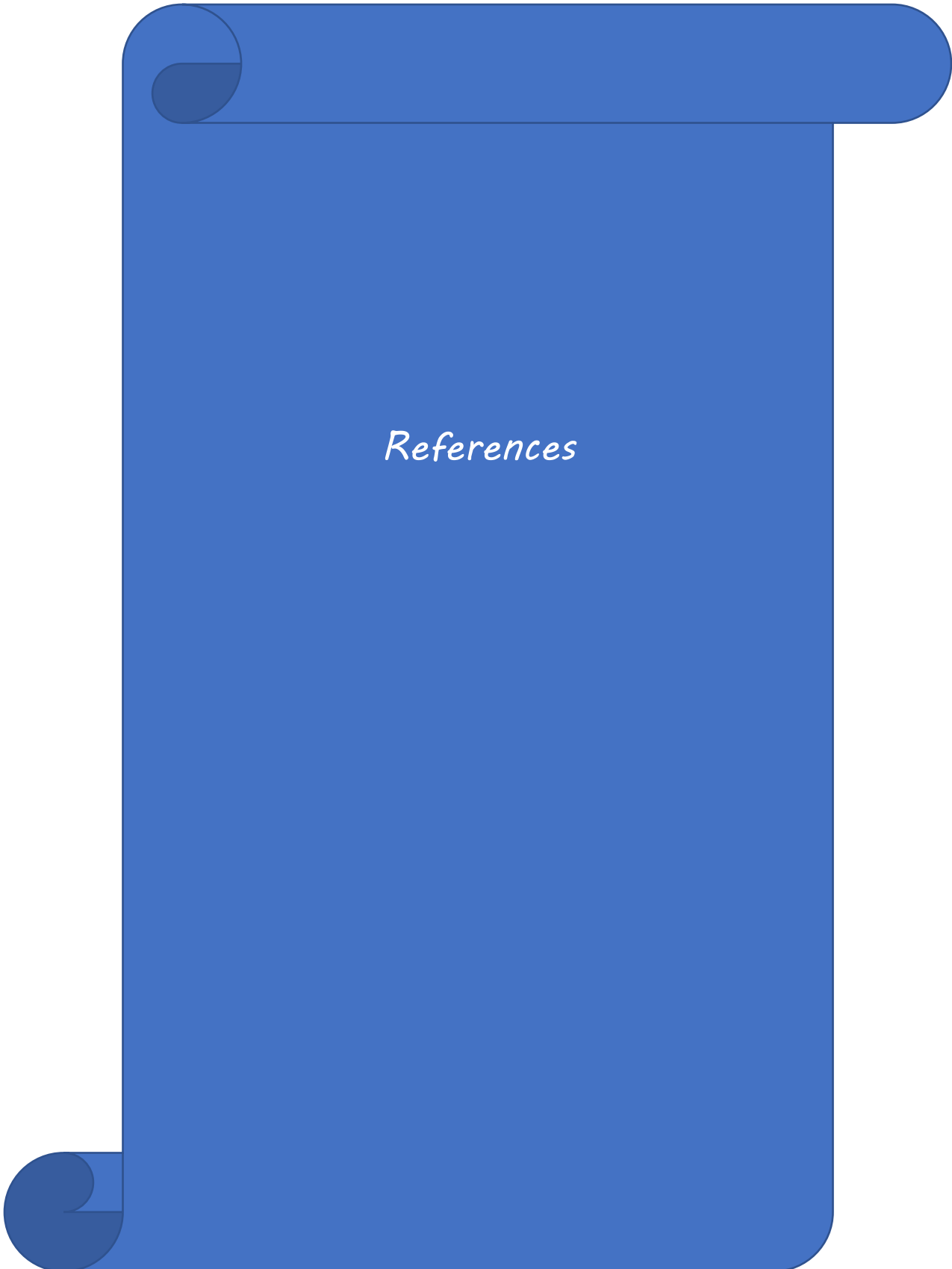
Note: Values are expressed as a mean (SD). Means with different letters within the same category are significantly different at $P < 0.05$. WHO/ FAO =World Health Organization, Food and Agriculture Organization (2011). IQS = Iraqi Quality Standardization (2009). * overpassed WHO/IQS acceptable limit

Conclusions

1. The study showed that the irrigation water is not suitable for irrigation due to the high numbers of coliform, molds and yeasts and the high concentrations of lead, cadmium, iron, and manganese.
2. Agricultural soil is not suitable for agriculture because of the high microorganisms, total bacteria, total coliform, molds and yeasts, and magnesium.
3. Paddy is not fit for human consumption due to high total bacteria count, total coliform, and molds and yeasts, and high lead concentration.
4. Rice is not fit for human consumption due to high total coliform, and molds and yeasts, and high lead concentration.

Recommendations

1. The most important recommendation to this work is to follow the source of the irrigation water contamination and find a way to reduce the contaminants.
2. Make a similar survey on paddy fields of the middle and south of Iraq, which is the main region of planting paddy.
3. Completion of other future research for the estimation of other mycotoxins such as aflatoxin B1, ochratoxin A, etc. in paddy and rice.
4. Application of the advanced method using high-performance liquid chromatography with fluorescence detectors, which is a more accurate method for mycotoxin determination.
5. Measuring other heavy metals, especially mercury, which is considered one of the main heavy metals conducted routinely worldwide.



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Appendices

Appendices

Appendices NO. 1

Microbiological analysis of irrigation water from Diyala first field

Samples	Total Bacteria count			Total Coliform			Molds and Yeasts	<i>E- Coli</i>
1	21900	120000	650000	2670	11300	52000	13000000	9.2
2	30000	125000	680000	2750	12000	63000	11000000	9.2
3	29500	123000	530000	2600	10400	50000	8000000	15
4	27700	107000	540000	2650	10000	56000	9000000	6.2
5	27000	100000	500000	2500	8000	38000	12000000	7.4
6	27500	103000	600000	2630	9700	46000	11000000	9.2
7	28300	119000	570000	2770	12400	65000	13000000	15
8	28100	113000	700000	2680	10600	52000	12000000	9.2

Appendices NO. 2

Microbiological analysis of irrigation water from Diyala second field

Samples	Total Bacteria count			Total Coliform			Molds and Yeasts	<i>E- Coli</i>
1	26300	92000	580000	2440	8600	66000	13000000	6.2
2	27500	104000	630000	2480	8500	56000	11000000	11
3	27300	101000	610000	2300	6500	46000	9000000	9.2
4	25700	75000	330000	2280	6300	44000	12000000	3.6
5	26500	91000	570000	2250	6000	42000	8000000	6.2
6	26900	99000	600000	2320	7000	50000	11000000	7.4
7	25000	70000	300000	2380	7500	46000	7000000	9.4
8	25300	71000	310000	2150	5500	31000	11000000	11

Appendices NO. 3

Microbiological analysis of irrigation water from Diyala third field

Samples	Total Bacteria count			Total Coliform			Molds and Yeasts	<i>E- Coli</i>
1	27100	100000	600000	2480	8500	56000	12000000	11
2	28500	120000	660000	2300	6500	44000	9000000	9.2
3	27600	103000	610000	2280	6300	42000	13000000	9.4
4	26500	95000	550000	2250	6000	50000	12000000	7.4
5	28100	118000	650000	2320	7000	56000	11000000	9.2
6	27300	103000	620000	2380	7500	30000	8000000	6.2
7	26000	83000	450000	2150	5000	66000	13000000	9.4
8	25700	75000	330000	2440	8600	46000	12000000	9.2

Appendices NO. 4

Microbiological analysis of soil from Diyala first field

Samples	Total Bacteria count			Total Coliform			Molds and Yeasts	<i>E- Coli</i>
1	196000	1000000	4400000	2250	1500	55000	1200000	<3.0
2	200000	1080000	4600000	2080	14200	64000	1500000	<3.0
3	298000	1840000	7500000	1950	1600	61000	1300000	<3.0
4	158000	950000	6300000	2150	14400	66000	1200000	<3.0
5	240000	1250000	5000000	2100	14500	65000	1700000	<3.0
6	280000	1630000	6000000	2200	16800	68000	1100000	<3.0
7	291000	1760000	7100000	2000	16400	63000	500000	<3.0
8	296000	1820000	3700000	1770	13500	57000	800000	<3.0
9	250000	1300000	5300000	1730	13200	56000	1000000	<3.0
10	290000	17540000	7000000	1650	13000	54000	600000	<3.0
11	225000	1100000	5000000	1780	15800	60000	1500000	<3.0

12	250000	1310000	4500000	1960	15300	62000	1300000	<3.0
13	200000	1050000	4600000	2230	15000	69000	900000	<3.0
14	256000	1340000	5500000	2000	16200	61000	1500000	<3.0
15	261000	1420000	5700000	1800	13300	52000	1300000	<3.0

Appendices NO. 5

Microbiological analysis of soil from Diyala second field

Samples	Total Bacteria count			Total Coliform			Molds and Yeasts	<i>E- Coli</i>
1	200000	1250000	4000000	1350	10500	30000	1200000	<3.0
2	213000	1340000	4400000	1650	12100	3500	1700000	<3.0
3	210000	1280000	4200000	1540	12000	40000	1800000	<3.0
4	220000	1350000	5000000	1680	11500	320000	1100000	<3.0
5	177000	1050000	3300000	1760	12200	41000	1300000	<3.0
6	193000	1150000	4100000	1700	14800	48000	1500000	<3.0
7	230000	1450000	5600000	1730	13000	44000	1800000	<3.0
8	233000	1980000	5800000	1720	14200	47000	1100000	<3.0
9	243000	1650000	6800000	1690	13600	48000	900000	<3.0
10	252000	1680000	7200000	1380	12500	42000	1200000	<3.0
11	235000	1500000	5500000	1440	10000	34000	1300000	<3.0

12	180000	1100000	3500000	1630	10600	38000	1400000	<3.0
13	205000	1270000	4100000	1690	13500	46000	1100000	<3.0
14	223000	1380000	5100000	1450	13400	43000	1200000	<3.0
15	244000	1600000	6600000	1670	10800	3600021	1300000	<3.0

Appendices NO. 6

Microbiological analysis of Soil from Diyala third field

Samples	Total Bacteria			Total Coliform			Molds and Yeasts	<i>E.-Coli</i>
1	210000	1350000	3300000	1460	12000	40000	1100000	<3.0
2	240000	1530000	4800000	1480	12200	43000	900000	<3.0
3	215000	1400000	3500000	1560	12800	46000	1600000	<3.0
4	295000	1800000	6100000	1590	13100	47000	1300000	<3.0
5	196000	1250000	3000000	1900	15300	57000	1200000	<3.0
6	188000	1200000	5500000	1880	15100	55000	1400000	<3.0
7	220000	1440000	3800000	1950	15800	60000	1500000	<3.0
8	225000	1460000	4100000	1800	14800	45000	1300000	<3.0
9	200000	1300000	3000000	1620	13600	48000	1100000	<3.0
10	235000	1500000	4500000	1750	14600	43000	800000	<3.0
11	185000	1150000	5100000	1710	14200	51000	900000	<3.0

12	286000	1770000	5300000	1900	15000	54000	1200000	<3.0
13	243000	1780000	5400000	1880	16600	46000	1300000	<3.0
14	223000	1550000	3200000	1650	15400	51000	1500000	<3.0
15	218000	1630000	4500000	1480	16100	54000	1200000	<3.0

Appendices NO. 7

Microbiological analysis of paddy from Diyala first field

Samples	Total Bacteria count			Total Coliform			Molds and Yeasts	<i>E- Coli</i>
1	300000	1800000	7500000	235000	1600000	7600000	20000000	6.2
2	283000	1580000	6800000	218000	1520000	7400000	22000000	<3.0
3	295000	1750000	7400000	205000	1700000	8000000	18000000	9.2
4	286000	1690000	7000000	220000	1480000	6600000	23000000	15
5	293000	1700000	7200000	215000	1460000	6500000	20000000	9.2
6	287000	1630000	6900000	222000	1700000	7000000	19000000	<3.0
7	285000	1600000	6800000	180000	1440000	5700000	22000000	3
8	277000	1530000	5600000	175000	1300000	5300000	24000000	3.6
9	275000	1510000	5400000	168000	1290000	5100000	19000000	<3.0
10	280000	1550000	6000000	185000	1480000	5900000	21000000	3
11	276000	1520000	5100000	200000	1610000	7700000	20000000	11

12	272000	1460000	5000000	225000	1730000	7100000	23000000	9.4
13	289000	1650000	6800000	212000	1650000	7800000	21000000	<3.0
14	281000	1550000	6200000	185000	1460000	5600000	22000000	6.2
15	278000	1450000	5800000	173000	1310000	5200000	20000000	3.6

Appendices NO. 8

Microbiological analysis of paddy from Diyala second field

Samples	Total Bacteria count			Total Coliform			Molds and Yeasts	<i>E-Coli</i>
1	225000	1230000	3200000	145000	1100000	3500000	23000000	<3.0
2	220000	1200000	3100000	171000	1240000	4000000	22000000	6.2
3	264000	1460000	5600000	170000	1200000	4400000	20000000	3.6
4	253000	1300000	6400000	160000	1200000	3600000	2500000	<3.0
5	262000	1424444	5000000	172000	1260000	4500000	23000000	<3.0
6	260000	1400000	5500000	180000	1520000	5200000	21000000	6.1
7	257000	1380000	4800000	175000	1350000	5000000	25000000	<3.0
8	246000	1340000	3800000	177000	1460000	5500000	22000000	<3.0
9	242000	1320000	3600000	176000	1360000	5100000	19000000	16
10	233000	1340000	3200000	173000	1280000	4700000	21000000	<3.0
11	236000	1360000	3400000	146000	1150000	3700000	19000000	11

12	251000	1320000	4400000	150000	1180000	3900000	21000000	<3.0
13	248000	1350000	4200000	155000	1200000	3400000	20000000	9.4
14	227000	1250000	3200000	168000	1220000	3800000	23000000	3
15	215000	1120000	3000000	170000	1250000	4200000	24000000	6.1

Appendices NO. 9

Microbiological analysis of paddy from Diyala third field

Samples	Total Bacteria count			Total Coliform			Molds and Yeasts	<i>E- Coli</i>
1	250000	1150000	3200000	152000	1150000	4200000	18000000	7.4
2	265000	1400000	5100000	177000	1300000	4800000	20000000	6.2
3	266000	1420000	5300000	165000	1250000	4100000	21000000	9.4
4	257000	1260000	4000000	182000	1540000	5600000	22000000	<3.0
5	263000	1380000	4800000	156000	1250000	4000000	23000000	6.2
6	273000	1520000	6000000	172000	1230000	3800000	20000000	15
7	271000	1500000	5800000	167000	1380000	4200000	18000000	11
8	262000	1310000	4600000	180000	1450000	6000000	22000000	<3.0
9	255000	1230000	3800000	172000	12180000	4300000	20000000	9.2
10	268000	1460000	5500000	168000	1240000	3900000	21000000	16
11	256000	1240000	530000	186000	1480000	6500000	23000000	<3.0

12	269000	1480000	4600000	177000	1460000	5500000	20000000	11
13	264000	1430000	4300000	176000	1470000	5300000	21000000	9.2
14	257000	1370000	4200000	164000	1350000	5400000	19000000	<3.0
15	264000	1420000	5100000	163000	1410000	4600000	21000000	<3.0

Appendices NO. 10

Microbiological analysis and total aflatoxin of rice from Diyala first field

Samples	Total Bacteria count			Total Coliform	Molds and Yeasts	<i>E- Coli</i>	Aflatoxin
1	2500	17000	63000	950	1100000	<3.0	8.67
2	2630	18500	75000	980	1200000	<3.0	11.09
3	2100	14400	68000	1000	1500000	<3.0	16.4
4	2000	16360	57000	880	900000	<3.0	8.14
5	2250	15300	63000	760	1000000	<3.0	1.19
6	1950	16200	58000	810	1300000	<3.0	9.13
7	1870	14300	60000	890	1400000	<3.0	2.26
8	1750	14000	65000	850	1100000	<3.0	7.78
9	1700	13000	46000	830	1200000	<3.0	6.93
10	1880	14200	62000	750	900000	<3.0	6.83
11	2000	16600	64000	790	1300000	<3.0	2.45

12	2130	14600	67000	780	1100000	<3.0	1.79
13	2900	22500	73000	700	1500000	<3.0	5.05
14	2750	19500	68000	730	1200000	<3.0	4.24
15	2810	2000	70000	710	1300000	<3.0	4.54

Appendices NO. 11

Microbiological analysis and total aflatoxin of rice from Diyala second field

Samples	Total Bacteria count			Total Coliform	Molds and Yeasts	<i>E- Coli</i>	Aflatoxin
1	1160	9000	31000	640	1300000	<3.0	10.7
2	1200	9600	32000	600	1100000	<3.0	5.57
3	1280	10100	36000	640	1500000	<3.0	6.22
4	1310	10800	36000	540	1300000	<3.0	5.88
5	1380	11800	38000	550	1200000	<3.0	5.4
6	1400	12000	40000	530	1000000	<3.0	5.11
7	1470	12200	50000	570	1200000	<3.0	7.55
8	1450	12100	48000	390	1300000	<3.0	5.51
9	1410	12000	41000	370	1400000	<3.0	4.26
10	1320	11000	34000	580	1100000	<3.0	4.62
11	1190	9500	32000	610	1300000	<3.0	3.72

12	1270	9800	35000	460	1200000	<3.0	14.8
13	1350	11200	36000	510	1000000	<3.0	5.75
14	1050	7700	30000	460	1100000	<3.0	5.06
15	1100	9000	31000	350	1300000	<3.0	4.34

Appendices NO. 12

Microbiological analysis and total aflatoxin of rice from Diyala third field

Samples	Total Bacteria count			Total Coliform	Molds and Yeasts	<i>E- Coli</i>	Aflatoxin
1	1250	9500	36000	830	1200000	<3.0	7.12
2	1150	9000	30000	780	1400000	<3.0	6.22
3	1850	14000	60000	640	1300000	<3.0	5.49
4	1760	13900	60000	700	1500000	<3.0	5.65
5	1940	16000	70000	680	1100000	<3.0	5.86
6	2110	14600	66000	770	1300000	<3.0	4.93
7	2000	14200	63000	730	1200000	<3.0	6.54
8	2260	15400	64000	620	1500000	<3.0	5.16
9	1830	13500	48000	660	1300000	<3.0	5.17
10	1710	13000	55000	690	1200000	<3.0	5.28
11	1850	12000	38000	800	900000	<3.0	4.6

12	1670	125000	45000	820	1100000	<3.0	4.5
13	1650	12200	38000	630	1200000	<3.0	5.4
14	1550	12800	45000	560	1100000	<3.0	4.6
15	1750	13200	84000	610	1200000	<3.0	5.43

Appendices NO. 13

Microbiological analysis of irrigation water from Salah al-Din first field

Samples	Total Bacteria count			Total Coliform			Molds and Yeasts	<i>E-Coli</i>
1	30000	135000	770000	2760	12500	60000	150000000	6.2
2	28000	113000	630000	2900	14200	62000	130000000	9.2
3	29000	129000	660000	2680	11600	55000	170000000	11
4	30000	132000	730000	2820	13300	75000	120000000	7.4
5	28600	121000	700000	2930	13500	64000	130000000	11

Appendices NO. 14

Microbiological analysis of irrigation water from Salah al-Din second field

Samples	Total Bacteria count			Total Coliform			Molds and Yeasts	<i>E- Coli</i>
1	27000	100000	640000	2440	7000	45000	160000000	9.4
2	26500	96000	600000	2350	6500	34000	150000000	9.2
3	25700	80000	450000	2400	6800	35000	150000000	11
4	26000	83000	460000	2280	5300	33000	130000000	7.4
5	27200	102000	650000	2310	6000	31000	120000000	11

Appendices NO. 15

Microbiological analysis of irrigation water from Salah al-Din third field

Samples	Total Bacteria count			Total Coliform			Molds and Yeasts	<i>E- Coli</i>
1	30000	145000	800000	2700	12500	48000	130000000	3.6
2	28500	125000	630000	2730	13000	50000	140000000	6.2
3	29300	132000	680000	2660	12000	45000	110000000	9.2
4	28700	128000	660000	2550	9300	39000	120000000	9.4
5	29100	130000	670000	2610	10000	43000	110000000	6.2

Appendices NO. 16

Microbiological analysis of soil from Salah al-Din first field

Samples	Total Bacteria count			Total Coliform			Molds and Yeasts	<i>E- Coli</i>
1	293000	1030000	7600000	2150	15200	70000	14000000	<3.0
2	300000	1250000	8000000	2350	17000	82000	12000000	<3.0
3	260000	1030000	7100000	2250	16400	67000	13000000	<3.0
4	295000	1200000	7700000	2210	17500	77000	12000000	<3.0
5	289000	1150000	7500000	2000	12200	55000	11000000	<3.0
6	270000	1100000	7300000	1700	15500	64000	9000000	<3.0
7	257000	1000000	7000000	1470	14000	60000	13000000	<3.0
8	235000	880000	5700000	1850	17000	78000	15000000	<3.0
9	287000	1180000	7600000	2000	14200	61000	12000000	<3.0
10	263000	1080000	7300000	1800	15200	63000	10000000	<3.0

Appendices NO. 17

Microbiological analysis of soil from Salah al-Din second field

Samples	Total Bacteria count			Total Coliform			Molds and Yeasts	<i>E- Coli</i>
1	255000	1450000	7500000	1700	11000	42000	11000000	<3.0
2	153000	1000000	6600000	1150	9500	31000	13000000	<3.0
3	225000	1020000	6700000	1700	13000	50000	11000000	<3.0
4	262000	1470000	8100000	15000	11200	41000	13000000	<3.0
5	135000	640000	3500000	13000	10000	35000	15000000	<3.0
6	293000	860000	5200000	14400	11400	39000	12000000	<3.0
7	180000	730000	4000000	16600	14100	56000	12000000	<3.0
8	144000	850000	3700000	16800	13400	43000	9000000	<3.0
9	230000	1040000	8000000	17500	12500	46000	11000000	<3.0
10	289000	830000	4600000	18500	13500	36000	12000000	<3.0

Appendices NO. 18

Microbiological analysis of Soil from Salah al-Din third field

Samples	Total Bacteria count			Total Coliform			Molds and Yeasts	<i>E- Coli</i>
1	250000	1400000	7000000	2280	14600	68000	9000000	<3.0
2	150000	950000	6300000	1770	12100	56000	11000000	<3.0
3	220000	980000	6300000	2150	17600	75000	13000000	<3.0
4	253000	1370000	6800000	2100	16000	72000	13000000	<3.0
5	135000	640000	3500000	1570	16200	74000	12000000	<3.0
6	185000	740000	4100000	2220	17300	79000	13000000	<3.0
7	145000	860000	3800000	2400	16700	73000	11000000	<3.0
8	155000	1000000	6500000	1960	15600	70000	12000000	<3.0
9	140000	900000	6000000	2300	18000	77000	13000000	<3.0
10	225000	1080000	6800000	2110	16200	71000	12000000	<3.0

Appendices NO. 19

Microbiological analysis of paddy from Salah al- Din first field

Samples	Total Bacteria count			Total Coliform			Molds and Yeasts	<i>E- Coli</i>
1	289000	1650000	7000000	245000	1700000	8600000	220000000	<3.0
2	285000	1610000	6900000	228000	1620000	8400000	210000000	7.4
3	278000	1550000	5700000	215000	1800000	9000000	240000000	11
4	277000	1530000	5500000	230000	1580000	7600000	180000000	9.4
5	283000	1560000	6300000	225000	1560000	7500000	210000000	6.2
6	280000	1560000	6000000	232000	1800000	8000000	190000000	16
7	285000	1600000	6800000	190000	1450000	6700000	180000000	3
8	296000	1760000	7500000	185000	1400000	6300000	320000000	6.1
9	290000	1730000	7200000	178000	1399999	6100000	210000000	<3.0
10	288000	1640000	7100000	195000	1580000	6900000	190000000	7.4

Appendices NO. 20

Microbiological analysis of paddy from Salah al- Din second field

Samples	Total Bacteria count			Total Coliform			Molds and Yeasts	<i>E- Coli</i>
1	266000	1500000	4600000	155000	1200000	4500000	220000000	3.6
2	230000	1240000	6000000	181000	1340000	5000000	210000000	9.2
3	226000	1380000	3300000	180000	1350000	5400000	180000000	<3.0
4	255000	1300000	4800000	170000	1300000	4600000	240000000	16
5	240000	1340000	4200000	182000	1360000	5500000	230000000	<3.0
6	230000	1310000	3100000	190000	1620000	6200000	210000000	9.2
7	232000	1230000	4000000	185000	1450000	6000000	190000000	<3.0
8	225000	1150000	3000000	187000	1560000	6500000	220000000	7.4
9	220000	1000000	3000000	186000	1460000	6100000	210000000	<3.0
10	200000	1250000	3000000	183000	1380000	5700000	230000000	9.2

Appendices NO. 21

Microbiological analysis and Total aflatoxin of paddy from Salah al- Din third field

Samples	Total Bacteria count			Total Coliform			Molds and Yeasts	<i>E- Coli</i>
1	293000	1700000	7500000	160000	1380000	5400000	210000000	7.4
2	276000	1540000	5600000	162000	1500000	6000000	170000000	3
3	290000	1690000	7300000	170000	1420000	5500000	150000000	<3.0
4	284000	1570000	6400000	174000	1450000	6000000	220000000	16
5	277000	1530000	5400000	205000	1670000	7100000	230000000	11
6	283000	1570000	6100000	202000	1650000	6900000	210000000	<3.0
7	290000	1740000	7300000	210000	1720000	7400000	190000000	6.1
8	275000	1500000	5600000	194000	1620000	6800000	220000000	<3.0
9	278000	1600000	6000000	176000	1500000	6200000	210000000	9.4
10	282000	1580000	6200000	190000	1600000	6800000	230000000	6.2

Appendices NO. 22

Microbiological analysis and Total aflatoxin of rice from Salah al- Din first field

Samples	Total Bacteria count			Total Coliform	Mold and Yeast	<i>E- Coli</i>	Aflatoxin
1	1300	10500	38000	850	9000000	<3.0	5.3
2	1380	12000	40000	920	12000000	<3.0	7.66
3	1420	12600	46000	890	11000000	<3.0	4.84
4	1500	13000	53000	860	12000000	<3.0	4.49
5	1360	11500	38000	930	11000000	<3.0	3.92
6	1280	9600	35000	900	10000000	<3.0	5.88
7	1380	11800	40000	940	8000000	<3.0	2.76
8	1150	7800	30000	870	9000000	<3.0	5.59
9	1280	10100	36000	960	13000000	<3.0	6.37
10	1180	10300	34000	1000	12000000	<3.0	4.84

Appendices NO. 23

Microbiological analysis and Total aflatoxin of rice from Salah al- Din second field

Samples	Total Bacteria count			Total Coliform	Molds and Yeasts	<i>E- Coli</i>	Aflatoxin
1	3000	20000	75000	1100	9000000	<3.0	4.54
2	2850	19500	70000	950	11000000	<3.0	4.99
3	2770	19000	69000	870	7000000	<3.0	5.39
4	2650	18000	68000	910	9000000	<3.0	3.73
5	2500	16000	60000	860	11000000	<3.0	4.57
6	1950	14400	56000	890	5000000	<3.0	4.34
7	1880	13000	54000	1030	13000000	<3.0	4.59
8	1660	13800	40000	830	8000000	<3.0	5.04
9	1750	12500	53000	960	12000000	<3.0	5.28
10	1600	22500	39000	850	11000000	<3.0	6.47

Appendices NO. 24

Microbiological analysis and Total aflatoxin of rice from Salah al- Din third field

Samples	Total Bacteria count			Total Coliform	Molds and Yeasts	<i>E- Coli</i>	Aflatoxin
1	2000	16300	60000	1080	12000000	<3.0	4.78
2	1910	16000	58000	930	11000000	<3.0	5.75
3	1850	15400	55000	850	9000000	<3.0	6.52
4	1670	14500	51000	880	11000000	<3.0	6.53
5	1630	13900	48000	1000	13000000	<3.0	7.98
6	1700	15100	45000	950	14000000	<3.0	7.65
7	2550	18500	62000	840	11000000	<3.0	5.79
8	2660	19000	68000	930	13000000	<3.0	2.94
9	2700	20000	78000	810	14000000	<3.0	5.31
10	2780	21000	76000	830	12000000	<3.0	3.45

Appendices NO. 25

Heavy metals of irrigation water from Diyala first field

Samples	Fe	Cu	Pb	Cd	Mn	Mg	Co + Al
1	0.234	0.501	0.987	0.0065	1.246	11.84	Not Detected
2	0.834	0.087	1.273	0.0073	0.315	11.45	Not Detected
3	0.325	0.095	0.885	0.0082	0.819	10.735	Not Detected
4	0.436	0.028	1.415	0.0053	1.765	10.651	Not Detected
5	0.665	0.126	1.691	0.0021	1.811	11.62	Not Detected
6	0.528	0.322	0.793	0.0035	0.951	10.652	Not Detected
7	0.611	0.621	1.731	0.0064	1.73	11.82	Not Detected
8	0.701	0.416	1.801	0.0046	1.634	10.721	Not Detected

Appendices NO. 26

Heavy metals of Irrigation water from Diyala second field

Samples	Fe	Cu	Pb	Cd	Mn	Mg	Co + Al
1	0.513	0.126	0.261	0.0083	1.425	10.761	Not Detected
2	0.441	0.009	1.412	0.0076	0.616	10.54	Not Detected
3	0.562	0.112	0.623	0.0035	0.706	11.54	Not Detected
4	0.262	0.008	0.731	0.0042	1.065	10.651	Not Detected
5	0.325	0.121	1.622	0.0069	1.353	11.76	Not Detected
6	0.654	0.006	1.344	0.0071	1.064	10.54	Not Detected
7	0.534	0.101	0.556	0.0096	0.534	11.561	Not Detected
8	0.323	0.145	1.217	0.0053	1.212	10.653	Not Detected

Appendices NO. 27

Heavy metals of irrigation water from Diyala third field

Samples	Fe	Cu	Pb	Cd	Mn	Mg	Co +Al
1	0.234	0.501	0.987	0.0065	1.246	11.84	Not Detected
2	0.834	0.087	1.273	0.0073	0.315	11.45	Not Detected
3	0.325	0.095	0.885	0.0082	0.819	10.735	Not Detected
4	0.436	0.028	1.415	0.0053	1.765	10.651	Not Detected
5	0.665	0.126	1.691	0.0021	1.811	11.62	Not Detected
6	0.528	0.322	0.793	0.0035	0.951	10.652	Not Detected
7	0.611	0.621	1.731	0.0064	1.73	11.82	Not Detected
8	0.701	0.416	1.801	0.0046	1.634	10.721	Not Detected

Appendices NO. 28

Heavy metals of soil from Diyala first field

Samples	Fe	Cu	Pb	Cd	Mn	Mg	Co +Al
1	33.66	5.1	33.54	1.362	32.88	391.08	Not Detected
2	33.66	5.2	30 81	1.518	23.64	383.4	Not Detected
3	34.14	8.28	33.26	0.474	23.94	448.32	Not Detected
4	31.74	6.72	31.26	0.561	28.1	421.2	Not Detected
5	33.66	7.08	33.02	1.321	37.86	398.46	Not Detected
6	33.72	5.58	32.41	0.442	31.32	391.47	Not Detected
7	31.5	3.58	33.05	1.342	31.62	391.26	Not Detected
8	34.08	4.32	31.23	1.462	23.7	403.26	Not Detected
9	32.11	4.8	32.51	1.591	25.91	387.45	Not Detected
10	31.68	4.32	33.62	1.341	30.2	395.65	Not Detected
11	32.23	4.86	32.63	1.511	30.54	421.65	Not Detected

12	33.16	5.56	31.73	0.372	30.64	385.11	Not Detected
13	31.44	6.73	30.56	0.495	31.59	395.72	Not Detected
14	30.6	4.82	33.81	1.521	31.25	391.53	Not Detected
15	31.5	5.65	30.71	1.611	30.17	388.29	Not Detected

Appendices NO. 29

Heavy metals of soil from Diyala second field

Samples	Fe	Cu	Pb	Cd	Mn	Mg	Co +Al
1	34.14	5.28	30.45	1.571	30.8	392.04	Not Detected
2	33.18	3.54	32.61	1.417	31.4	385.32	Not Detected
3	33.18	1.92	33.54	1.326	32.4	388.14	Not Detected
4	33.66	6.72	31.32	0.535	32.14	390.11	Not Detected
5	34.62	1.92	32.51	0.254	33.43	392.06	Not Detected
6	34.26	4.5	31.41	1.443	30.26	401.15	Not Detected
7	40.32	4.98	33.32	0.511	29.69	404.1	Not Detected
8	32.46	6.9	32.46	1.622	31.5	399.17	Not Detected
9	37.2	7.26	32.52	0.611	30.41	386.45	Not Detected
10	34.86	3.18	33.42	1.231	33.32	389.04	Not Detected
11	35.62	1.99	30.15	1.613	31.64	388.24	Not Detected

12	32.25	4.58	31.36	0.582	32.54	401.08	Not Detected
13	37.11	2.35	30.41	0.469	31.61	388.16	Not Detected
14	30.81	1.95	32.55	1.635	30.29	391.43	Not Detected
15	32.15	2.83	31.46	1.513	30.35	392.34	Not Detected

Appendices NO. 30

Heavy metals of soil from Diyala third field

Samples	Fe	Cu	Pb	Cd	Mn	Mg	Co +Al
1	30.53	5.28	30.52	1.318	28.62	399.02	Not Detected
2	32.12	6.91	31.61	1.521	30.53	400.63	Not Detected
3	30.14	4.76	33.53	0.631	26.24	395.24	Not Detected
4	33.62	3.25	32.22	0.713	31.83	387.55	Not Detected
5	31.52	3.76	30.43	1.495	27.41	386.43	Not Detected
6	30.91	3.53	30.34	0.312	32.13	385.23	Not Detected
7	32.72	4.81	33.46	1.425	31.62	391.65	Not Detected
8	32.63	3.76	32.51	0.931	32.62	387.21	Not Detected
9	33.41	3.53	31.33	1.516	32.54	390.54	Not Detected
10	31.62	8.41	30.42	1.916	30.43	390.54	Not Detected
11	30.53	6.61	33.42	0.422	29.81	390.42	Not Detected

12	30.57	7.35	30.21	1.301	30.76	392.45	Not Detected
13	31.83	7.52	31.51	1.601	31.11	411.12	Not Detected
14	30.56	7.25	31.15	0.917	31.19	404.34	Not Detected
15	32.46	5.87	31.16	1.561	30.85	398.76	Not Detected

Appendices NO. 31

Heavy metals of paddy from Diyala first field

Samples	Fe	Cu	Pb	Cd	Mn	Mg	Co +Al
1	6.062	0.33	0.642	0.058	2.646	12.702	Not Detected
2	7.698	0.118	3.642	0.009	2.938	12.59	Not Detected
3	14.37	0.128	2.014	0.012	3.000	14.874	Not Detected
4	11.48	0.108	1.711	0.007	2.854	15.138	Not Detected
5	9.738	0.098	1.54	0.063	2.763	15.452	Not Detected
6	6.828	0.064	1.33	0.083	2.543	14.435	Not Detected
7	11.262	0.116	2.07	0.072	3.100	12.763	Not Detected
8	11.44	0.476	1.42	0.054	3.11	12.653	Not Detected
9	9.83	0.126	1.16	0.009	2.621	13.651	Not Detected
10	11.02	0.102	0.71	0.061	2.462	15.231	Not Detected
11	6.534	0.411	2.83	0.063	2.512	12.453	Not Detected

12	8.611	0.599	2.11	0.066	2.496	15.89	Not Detected
13	7.503	0.53	0.895	0.072	3.333	12.72	Not Detected
14	6.895	0.75	1.95	0.085	3.417	14.00	Not Detected
15	11.45	0.813	3.12	0.094	2.279	14.543	Not Detected

Appendices NO. 32

Heavy metals of paddy from Diyala second field

Samples	Fe	Cu	Pb	Cd	Mn	Mg	Co +Al
1	6.036	0.127	3.011	0.0562	2.534	15.563	Not Detected
2	7.532	0.116	1.25	0.083	3.51	12.564	Not Detected
3	8.091	0.089	3.014	0.075	2.736	12.56	Not Detected
4	9.531	0.513	2.632	0.091	2.571	15.65	Not Detected
5	11.251	0.316	2.517	0.045	3.21	13.982	Not Detected
6	8.915	0.712	1.492	0.067	2.661	12.564	Not Detected
7	11.43	0.128	1.576	0.083	2.591	13.821	Not Detected
8	7.932	0.367	2.331	0.061	3.21	12.753	Not Detected
9	9.563	0.713	1.576	0.075	3.11	12.432	Not Detected
10	12.41	0.531	1.601	0.082	2.517	14.734	Not Detected
11	7.651	0.552	1.525	0.054	2.816	15.45	Not Detected

12	8.705	0.701	2.411	0.076	2.913	12.981	Not Detected
13	6.415	0.673	1.662	0.058	3.511	15.32	Not Detected
14	9.315	0.613	3.011	0.082	3.681	14.672	Not Detected
15	11.45	0.55	1.343	0.091	2.911	12.761	Not Detected

Appendices NO. 33

Heavy metals of paddy from Diyala third field

Samples	Fe	Cu	Pb	Cd	Mn	Mg	Co +Al
1	11.04	0.113	1.553	0.056	2.415	12.871	Not Detected
2	11.07	0.296	3.431	0.091	3.11	12.652	Not Detected
3	11.97	0.312	2.242	0.048	2.163	13.832	Not Detected
4	10.71	0.413	1.326	0.037	2.221	15.761	Not Detected
5	9.53	0.415	2.114	0.081	2.672	15.67	Not Detected
6	13.61	0.591	3.675	0.022	3.131	12.413	Not Detected
7	11.53	0.631	1.501	0.063	3.14	14.62	Not Detected
8	9.315	0.611	1.46	0.014	2.353	14.62	Not Detected
9	8.761	0.592	2.158	0.076	2.353	15.91	Not Detected
10	7.513	0.583	1.312	0.065	2.493	12.76	Not Detected
11	7.669	0.613	1.115	0.085	2.592	15.76	Not Detected

12	8.653	0.815	2.221	0.046	2.496	13.93	Not Detected
13	9.715	0.765	2.272	0.039	3.849	12.96	Not Detected
14	11.55	0.572	1.509	0.048	3.739	14.652	Not Detected
15	10.83	0.665	1.661	0.053	2.615	15.432	Not Detected

Appendices NO. 34

Heavy metals of rice from Diyala first field

Samples	Fe	Cu	Pb	Cd	Mn	Mg	Co +Al
1	0.668	0.0668	2.531	0.014	3.262	12.042	Not Detected
2	3.648	0.093	4.146	0.017	2.402	13.99	Not Detected
3	3.212	0.064	2.406	0.062	2.12	14.622	Not Detected
4	2.532	0.092	1.805	0.057	2.278	12.782	Not Detected
5	0.022	0.122	2.624	0.048	1.936	15.713	Not Detected
6	0.031	0.073	2.575	0.035	1.426	13.781	Not Detected
7	1.454	0.064	3.466	0.026	0.292	12.57	Not Detected
8	3.306	0.085	4.331	0.032	2.433	15.715	Not Detected
9	3.662	0.112	3.182	0.012	2.526	12.762	Not Detected
10	3.662	0.01	4.234	0.016	2.115	13.71	Not Detected
11	3.302	0.092	4.151	0.025	2.395	12.71	Not Detected

12	2.561	0.065	2.396	0.032	1.416	13.712	Not Detected
13	2.417	0.176	2.534	0.016	1.512	12.784	Not Detected
14	3.105	0.095	3.169	0.027	2.496	15.725	Not Detected
15	2,306	0.129	2.511	0.035	1.519	14.72	Not Detected

Appendices NO. 35

Heavy metals of rice from Diyala second field

Samples	Fe	Cu	Pb	Cd	Mn	Mg	Co +Al
1	0.654	0.081	2.961	0.072	1.491	13.99	Not Detected
2	1.321	0.047	4.776	0.066	3.271	12.042	Not Detected
3	3.479	0.063	2.495	0.053	2.65	13.781	Not Detected
4	2.316	0.016	3.253	0.044	2.715	15.715	Not Detected
5	0.721	0.057	3.574	0.031	3.324	12.762	Not Detected
6	1.321	0.095	2.349	0.025	1.595	13.71	Not Detected
7	1.321	0.028	3.212	0.071	2.343	12.71	Not Detected
8	1.543	0.032	2.635	0.093	2.221	13.712	Not Detected
9	2.320	0.093	4.183	0.029	3.191	12.784	Not Detected
10	2.696	0.085	4.109	0.032	2.446	15.725	Not Detected
11	2.711	0.064	3.225	0.045	3.125	14.72	Not Detected

12	1.965	0.073	3.518	0.09	3.332	14.72	Not Detected
13	3.125	0.069	2.292	0.076	1.918	15.715	Not Detected
14	3.112	0.095	1.815	0.081	2.611	15.725	Not Detected
15	1.415	0.083	3.125	0.035	2.535	13.99	Not Detected

Appendices NO. 36

Heavy metals of rice from Diyala third field

Samples	Fe	Cu	Pb	Cd	Mn	Mg	Co +Al
1	1.824	0.047	4.213	0.075	2.703	12.834	Not Detected
2	3.233	0.0152	3.325	0.067	1.834	12.67	Not Detected
3	2.541	0.027	2.943	0.056	2.196	13.67	Not Detected
4	2.321	0.017	3.456	0.045	3.643	15.76	Not Detected
5	3.643	0.051	2.392	0.049	4.527	13.782	Not Detected
6	2.157	0.082	3.873	0.065	3.291	12.871	Not Detected
7	3.719	0.093	3.271	0.037	3.643	15.746	Not Detected
8	3.232	0.044	2.163	0.082	2.647	14.853	Not Detected
9	3.674	0.032	4.217	0.073	2.152	15.432	Not Detected
10	2.356	0.030	3.615	0.045	3.165	13.72	Not Detected
11	1.544	0.077	2.543	0.094	2.67	12.76	Not Detected

12	1.696	0.035	3.515	0.075	3.156	14.851	Not Detected
13	2.315	0.046	4.216	0.083	2.225	13.72	Not Detected
14	2.135	0.073	2.636	0.066	2.296	12.76	Not Detected
15	3.116	0.055	2.711	0.091	2.316	15.631	Not Detected

Appendices NO. 37

Heavy metals of irrigation water from Salah al -Din first field

Samples	Fe	Cu	Pb	Cd	Mn	Mg	Co +Al
1	0.501	0.315	0.905	0.0093	1.226	11.81	Not Detected
2	0.811	0.065	1.462	0.0064	1.335	10.452	Not Detected
3	0.651	0.766	1.345	0.0075	0.613	10.56	Not Detected
4	0.376	0.601	0.867	0.0056	1.616	11.63	Not Detected
5	0.395	0.511	0.654	0.0082	0.766	10.5120.	Not Detected

Appendices NO. 38

Heavy metals of irrigation water from Salah al-Din second field

Samples	Fe	Cu	Pb	Cd	Mn	Mg	Co + Al
1	0.416	0.469	1.561	0.0091	1.711	10.531	Not Detected
2	0.511	0.605	0.868	0.0064	1.583	10.563	Not Detected
3	0.603	0.073	1.432	0.0065	0.915	11.81	Not Detected
4	0.533	0.086	0.916	0.0067	0.813	10.86	Not Detected
5	0.713	0.835	0.776	0.0048	1.605	11.782	Not Detected

Appendices NO. 39

Heavy metals of irrigation water from Salah al- Din third field

Samples	Fe	Cu	Pb	Cd	Mn	Mg	Co + Al
1	0.664	0.091	1.501	0.0037	1.373	11.87	Not Detected
2	0.415	0.903	1.643	0.0045	0.935	11.34	Not Detected
3	0.713	0.779	0.866	0.0054	0.85	10.561	Not Detected
4	0.766	0.825	0.935	0.0072	1.556	10.731	Not Detected
5	0.831	0.726	1.715	0.0029	1.672	10.931	Not Detected

Appendices NO. 40

Heavy metals of soil from Salah al-Din first field

Samples	Fe	Cu	Pb	Cd	Mn	Mg	Co +Al
1	31.54	5.91	31.41	1.75	29.19	391.56	Not Detected
2	30.62	4.62	30.53	1.354	30.45	398.54	Not Detected
3	32.56	6.55	32.61	0.943	31.14	421.65	Not Detected
4	33.47	6.72	30.54	1.521	28.17	399.45	Not Detected
5	33.21	3.83	31.23	1.672	31.7	395.45	Not Detected
6	32.45	4.41	30.51	1.731	30.62	389.45	Not Detected
7	33.61	4.67	31.32	1.84	32.36	411.21	Not Detected
8	31.53	4.45	32.15	1.932	33.21	387.56	Not Detected
9	32.70	6.13	31.49	1.752	31.12	385.32	Not Detected
10	33.53	6.03	32.52	1.643	30.43	388.43	Not Detected

Appendices NO. 41

Heavy metals of soil from Salah al-Din second field

1	33.42	5.61	30.15	1.632	30.16	399.45	Not Detected
2	33.56	6.53	31.32	1.541	32.35	396.43	Not Detected
3	23.63	6.32	33.54	0.911	31.41	403.11	Not Detected
4	30.20	7.45	30.92	0.756	30.51	399.45	Not Detected
5	33.71	5.24	32.73	1.543	33.16	395.45	Not Detected
6	31.54	8.67	32.61	0.815	32.27	387.56	Not Detected
7	30.61	6.56	32.55	1.311	30.21	397.32	Not Detected
8	23.43	4.48	30.43	0.951	31.41	393.65	Not Detected
9	33.54	6.32	31.62	1.713	30.37	387.34	Not Detected
10	31.46	6.25	30.56	1.531	32.35	391.65	Not Detected

Appendices NO. 42

Heavy metals of soil from Salah al-Din third field

Samples	Fe	Cu	Pb	Cd	Mn	Mg	Co +Al
1	30.50	5.31	30.16	1.315	30.18	411.54	Not Detected
2	33.24	6.22	30.27	0.691	29.25	399.65	Not Detected
3	31.26	4.14	31.65	1.412	33.36	389.65	Not Detected
4	32.45	6.53	23.34	1.531	27.41	388.98	Not Detected
5	31.63	5.67	33.46	0.681	32.55	401.54	Not Detected
6	32.45	5.43	33.52	0.593	31.46	387.34	Not Detected
7	33.64	4.23	33.63	1.412	31.53	386.76	Not Detected
8	32.25	6.52	32.44	1.613	32,51	395.72	Not Detected
9	33.63	5.16	31.26	1.711	33.61	391.65	Not Detected
10	33.51	6.21	32.15	1.801	31.56	387.45	Not Detected

Appendices NO. 43

Heavy metals of paddy from Salah al-Din first field

Samples	Fe	Cu	Pb	Cd	Mn	Mg	Co +Al
1	10.63	0.116	2.341	0.023	3.19	12.435	Not Detected
2	9.511	0.216	1.571	0.035	2.367	12.890	Not Detected
3	11.11	0.171	1.336	0.076	2.518	12.541	Not Detected
4	12.06	0.531	3.117	0.046	2.665	13.653	Not Detected
5	11.73	0.422	1.711	0.087	3.143	15.721	Not Detected
6	11.81	0.533	2.35	0.091	3.200	12.541	Not Detected
7	13.53	0.335	2.471	0.056	2.421	14.521	Not Detected
8	9.81	0.661	1.534	0.037	2.296	15.54	Not Detected
9	7.96	0.759	2.623	0.018	3.317	15.65	Not Detected
10	8.93	0.544	2.175	0.085	2.351	12.89	Not Detected

Appendices NO. 44

Heavy metals of paddy from Salah al-Din second field

Samples	Fe	Cu	Pb	Cd	Mn	Mg	Co +Al
1	11.54	0.915	1.910	0.035	2.517	13.831	Not Detected
2	9.61	0.123	2.134	0.046	3.281	14.710	Not Detected
3	13.75	0.734	3.256	0.078	3.176	12.567	Not Detected
4	11.37	0.221	1.320	0.056	3.453	14.672	Not Detected
5	12.28	0.395	2.31	0.073	2.321	15.430	Not Detected
6	12.35	0.643	2.432	0.056	2.756	15.760	Not Detected
7	11.81	0.552	2.621	0.017	3.543	13.562	Not Detected
8	10.95	0.457	2.554	0.092	3.231	12.672	Not Detected
9	11.93	0.561	2.463	0.045	2.432	15.642	Not Detected
10	9.416	0.879	1.572	0.063	3.546	15.650	Not Detected

Appendices NO. 45

Heavy metals of paddy from Salah al-Din third field

Samples	Fe	Cu	Pb	Cd	Mn	Mg	Co +Al
1	10.150	0.613	1.821	0.051	2.120	13.721	Not Detected
2	9.419	0.122	2.331	0.062	3.691	12.982	Not Detected
3	11.250	0.731	2.242	0.043	3.351	15.541	Not Detected
4	13.180	0.246	3.153	0.031	3.295	14.650	Not Detected
5	12.570	0.492	1.824	0.047	2.317	12.532	Not Detected
6	11.370	0.384	2.716	0.076	3.593	13.731	Not Detected
7	12.460	0.343	2.695	0.026	3.216	12.870	Not Detected
8	11.560	0.962	2.787	0.023	2.465	14.621	Not Detected
9	9.530	0.457	3.481	0.025	3.432	12.870	Not Detected
10	11.630	0.873	1.591	0.046	3.465	15.541	Not Detected

Appendices NO. 46

Heavy metals of rice from Salah al-Din first field

Samples	Fe	Cu	Pb	Cd	Mn	Mg	Co +Al
1	1.941	0.065	4.434	0.091	1.467	12.672	Not Detected
2	2.132	0.043	2.689	0.054	2.348	13.630	Not Detected
3	3.432	0.021	2.767	0.012	2.235	13.245	Not Detected
4	3.574	0.046	3.546	0.032	3.606	14.631	Not Detected
5	2.365	0.054	4.645	0.037	3.515	15.724	Not Detected
6	3.416	0.016	3.145	0.028	2.121	13.760	Not Detected
7	2.257	0.035	2.136	0.095	2.372	12.672	Not Detected
8	2.740	0.037	2.921	0.017	3.561	12.872	Not Detected
9	1.563	0.094	2.717	0.094	3.462	13.870	Not Detected
10	2.521	0.067	2.833	0.083	2.729	12.543	Not Detected

Appendices NO. 47

Heavy metals of rice from Salah al-Din second field

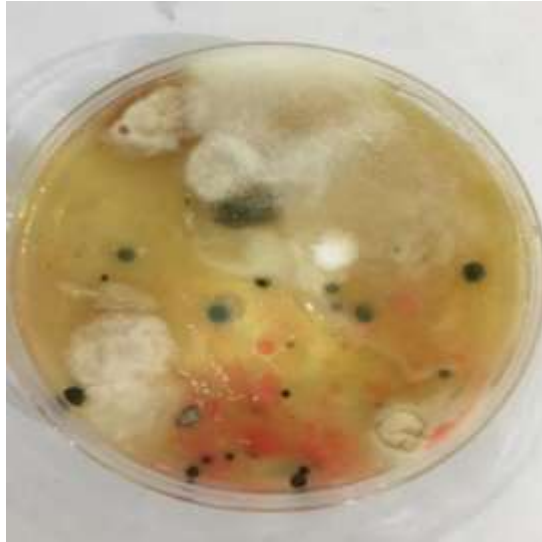
Samples	Fe	Cu	Pb	Cd	Mn	Mg	Co +Al
1	1.396	0.076	2.193	0.093	3.864	13.760	Not Detected
2	2.232	0.071	4.282	0.075	2.693	14.752	Not Detected
3	3.546	0.029	3.570	0.064	2.564	12.746	Not Detected
4	3.461	0.038	3.641	0.082	1.970	12.672	Not Detected
5	2.472	0.056	2.432	0.026	3.373	15.123	Not Detected
6	2.735	0.034	4.323	0.093	2.345	13.620	Not Detected
7	3.643	0.027	4.214	0.018	1.241	12.825	Not Detected
8	2.175	0.013	3.165	0.076	2.561	13.430	Not Detected
9	1.286	0.054	2.856	0.035	2.826	15.450	Not Detected
10	3.345	0.026	2.903	0.089	3.325	15.561	Not Detected

Appendices NO. 48

Heavy metals of rice from Salah al-Din third field

Samples	Fe	Cu	Pb	Cd	Mn	Mg	Co +Al
1	1.236	0.045	2.430	0.065	3.121	14.723	Not Detected
2	2.141	0.086	4.524	0.0541	2.734	12.640	Not Detected
3	2.465	0.046	4.493	0.043	1.456	15.340	Not Detected
4	3.932	0.053	3.521	0.037	1.271	13.630	Not Detected
5	1.914	0.053	3.473	0.042	3.653	12.741	Not Detected
6	3.676	0.0091	2.364	0.041	2.813	12.760	Not Detected
7	2.546	0.036	2.921	0.032	2.491	13.730	Not Detected
8	3.652	0.079	3.713	0.071	2.265	15.540	Not Detected
9	1.941	0.028	4.654	0.065	3.657	14.650	Not Detected
10	2.356	0.061	3.762	0.043	1.436	12.560	Not Detected

Appendix no. 49 Some pictures for molds and yeasts growth on Sabarude Agar media.



Mold and Yeast in Irrigation Water



Mold and Yeast in Soil



Mold and Yeast in Paddy

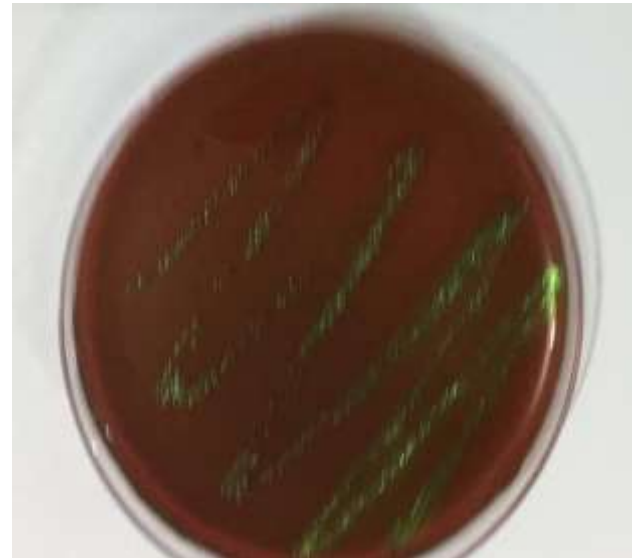


Mold and Yeast in Rice

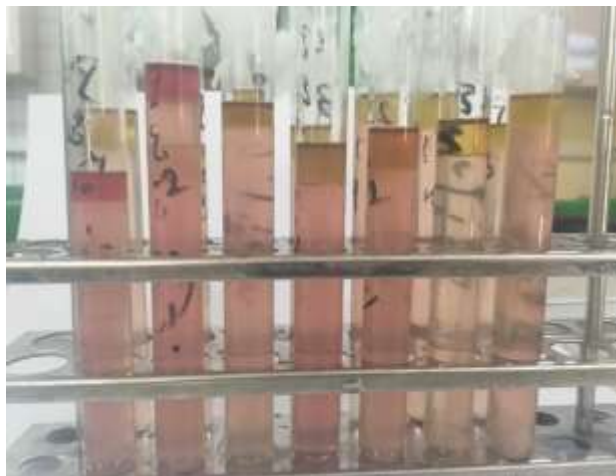
Appendix No. 50 IMVIC Test picture for E- Coli Detection.



Non-metallic color (Negative for E- Coli)



Metallic color (Positive for E- Coli)



الخلاصة

تنقسم هذه الدراسة إلى ثلاثة أجزاء ؛ يتم تلخيص خطوات كل جزء كما هو موضح أدناه:

الجزء الأول:-

أظهرت النتائج أن جميع عينات مياه الري قد تم تجاوز الحد المسموح به لكل من بكتيريا القولون والخمائر والاعفان ، بينما تم تجاوز جميع عينات التربة والأرز للبكتيريا الكلية بكتيريا القولون والخمائر والاعفان . تم تجاوز عينات الأرز للخمائر والاعفان فقط. وجد ان عدد بكتيريا *E-coli* في التربة والشلب أقل من 3 / جم ، وهو الحد الأدنى للكشف ، بينما كان في الماء والأرز أقل من حد منظمة الغذاء والدواء الامريكية. كانت أعداد الخمائر و الاعفان عالية جدًا ، على سبيل المثال ، تراوحت اعداد الخمائر والاعفان بالشلب والأرز من 2.06 $10^7 \times 21.5$ - $10^7 \times 12.0$ وحدة تكوين مستعمرة لكل غرام و $10^7 \times 1.2$ - $10^7 \times 12.0$ وحدة تكوين مستعمرة لكل غرام على التوالي ، بينما كان الحد 10^4 - 10^2 . في الختام ، اعتبرت عينات الأرز غير آمنة للاستهلاك البشري ، وكانت مياه الري والتربة هي مصادر التلوث ، لذلك هناك حاجة ملحة لمعالجة مياه الري.

الجزء الثاني:-

تم تقدير الأفلاتوكسين في 75 عينة أرز مسحوبة في ثلاثة حقول من كلا المحافظتين. تم استخدام الكروماتوجرافيا السائلة عالية الأداء (HPLC) لتحديد الأفلاتوكسين. اعتبرت الحدود الاوربية 4 ميكروغرام / كغ مرجع للمقارنة وذلك لأنه لا يوجد حد عراقي حتى الآن. احتوت معظم عينات الأرز على الأفلاتوكسين أعلى من الحد المسموح به ، حيث كانت أدنى وأعلى عينة محتوية على الأفلاتوكسين في حقل ديالى 1.19 ميكروغرام / كغ ، 16.40 ميكروغرام / كغ على التوالي. بلغ عدد العينات التي تجاوزت الحد المسموح به لإجمالي الأفلاتوكسين 66 عينة من أصل 75 عينة أرز. كان الأرز في منطقتي الدراسة غير مناسب من حيث تلوثه بالأفلاتوكسين الكلي لذلك يعتبر الأرز المنتج غير آمن للاستهلاك البشري. يعود سبب التركيز العالي للأفلاتوكسين في عينات الأرز إلى وجود عدد خمائر واعفان عالية في مياه الري والتربة والأرز. لذلك ، أصبحت معالجة مياه الري قضية ضرورية للغاية ويجب حلها بسرعة.

الجزء الثالث:-

تحديد مستويات المعادن الثقيلة مثل الرصاص (Pb) والكاديوم (Cd) والحديد (Fe) والمنغنيز (Mn) والكوبالت (Co) والمغنيسيوم (Mg) والألمنيوم (Al) والنحاس (Cu) في 39 عينات مياه و 75 عينة تربة و 75 عينة شلب و 75 عينة أرز لعينات مسحوبة من كلا المحافظتين. تم استخدام مقياس الامتصاص الذري

لتحديد تركيزات المعادن الثقيلة. تم اعتبار معايير الجودة العراقية (IQS) ومنظمة الصحة العالمية (WHO) كحدود للمقارنة. أظهرت النتائج أن جميع عينات مياه الري قد تجاوزت الحد المسموح به للرصاص ، الكاديوم ، الحديد ، والمنغنيز ، بينما تجاوزت جميع عينات التربة للمنغنيز والمغنيسيوم. تجاوزت عينات الشلب والأرز لحد الرصاص فقط. كان تركيز الرصاص مرتفعاً جداً ، على سبيل المثال ، تراوحت تراكيز الرصاص للشلب والأرز من 1.805-4.776 مجم / كجم ، 0.642-3.481 مجم / كجم على التوالي ، بينما ان الحد 0.2 مجم / كجم. احتوت عينات الشلب والأرز على تراكيز معادن ثقيلة ضمن الحدود المسموح بها لجميع المعادن الثقيلة الأخرى المختبرة. كانت تراكيز الكوبالت والألمنيوم أقل من حد الكشف لجميع العينات المختبرة. في الختام ، اعتبرت عينات الأرز غير آمنة للاستهلاك البشري من حيث محتواها من المعادن الثقيلة ، وكانت مياه الري مصدر التلوث لذلك ينبغي معالجتها.



وزارة التعليم العالي والبحث العلمي
جامعة ديالى
كلية التربية للعلوم الصرفة
قسم الكيمياء

دراسة تحليلية كيميائية وبإلوجية لملوثات نماذج التربة ومياه الري والمحاصيل الزراعية لمناطق مختلفة من العراق

رسالة

مقدمة الى مجلس كلية التربية للعلوم الصرفة، جامعة ديالى
وهي جزء من متطلبات نيل درجة الماجستير في علوم الكيمياء

من قبل الطالبة

الاء احمد جواد

بكالوريوس علوم كيمياء - كلية التربية للعلوم الصرفة - جامعة ديالى
٢٠١١-٢٠١٢

بإشراف

الدكتورة

عبير صالح الهندي

الاستاذ الدكتور

احمد مهدي سعيد

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