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Study Of Bioaccumulation Of Arsenic In Plants Near Hatti Industrial Area Raichur

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ABSTRACT

The bioaccumulation of arsenic in edible plants poses significant environmental and public health concerns, particularly in industrialized regions such as Hatti in Raichur, Karnataka. This study investigates arsenic contamination levels in various edible plants grown in the vicinity of the Hatti industrial area. Samples were collected from multiple locations around the industrial zone, and arsenic concentrations in plant tissues were analysed using Inductively Coupled Plasma Optical Emission Spectroscopy (ICP-OES). Results revealed elevated arsenic levels in plant samples, with variation observed among different plant species. Factors influencing arsenic uptake, including soil properties, plant species, and environmental conditions, were examined to elucidate bioaccumulation mechanisms.

Specifically focusing on pigeon pea plants, suspicion of arsenic presence prompted extraction using an alkaline leaching method with temperature assistance, following a modified wet digestion approach. Initial chemical tests confirmed arsenic presence, with subsequent instrumental analysis via ICP-OES yielding the following observations (mg/kg): N1t (0.098), N2t (0.094), N3t (0.097), N4t (0.098), F1t (0.096), F2t (0.048), F3t (0.089), and F4t (0.092).

Although arsenic concentrations in pigeon pea plants were below toxicity limits, ingestion of larger quantities could pose health risks. Acute oral ingestion of arsenic can result in gastrointestinal irritation, decreased blood cell production, abnormal heart rhythm, blood vessel damage, and impaired nerve function. Chronic exposure may lead to skin changes, skin cancer, and various internal cancers, along with potential impacts on diabetes, blood pressure, reproductive health, and cognitive function. This study contributes to understanding arsenic bioaccumulation in edible plants.

<u>Key words</u>: Bioaccumulation, Alkaline Leaching method, Inductively Coupled Plasma Optical Emission Spectroscopy, Arsenic Uptake, Pigeon Pea plants, Gastrointestinal Irritation, Chronic Exposure

1. INTRODUCTION

Arsenic (As) is a very poisonous heavy metal that presents a considerable risk to both the health of humans and the environment. Arsenic pollution in soil is almost often caused by human activities, such as mining, manufacturing, or agricultural methods, and it may be present in the environment for very long periods of time. When Arsenic is introduced into the soil, it immediately begins to undergo a number of biogeochemical changes, which ultimately makes it available to plants. When it comes to the bioaccumulation of Arsenic from the ground, plants are absolutely essential. They do this by absorbing Arsenic through their roots and then transporting it to various plant tissues, where it accumulates to varied degrees. The mechanisms that are involved in the absorption and transport of Arsenic inside plants are complicated and rely on the plant species, the qualities of the soil, and the circumstances of the environment. When Arsenic accumulates in plants, it can have a negative impact on the plants' ability to grow and develop, as well as their general health. In addition, Arsenic may make its way into the food chain through the ingestion of plant-based products that have been polluted with arsenic, which poses a risk to human health. Ali et al. (2019)

Pigeon pea is a perennial shrub normally cultivated as an annual crop and, in India, can be used in rotation and intercrop systems with different cereal crops.

Classification: The scientific classification of the pigeon pea (Cajanus cajan) is as follows:

Kingdom: Plantae Clade: Tracheophytes Order: Fabales Family: Fabaceae Subfamily: Faboideae Genus: Cajanus Species: C. cajan



Image 1.1 Tropical Homestead, OCTOBER 14, 2013

Moreover, pigeon pea develops a deep root system, making it drought tolerant. These traits encourage cultivation in rain-fed drylands, although the poor growth conditions (e.g., aridity, nutrient-poor soils) mean that yields remain low. Effective symbiosis may improve nitrogen (N) content in this pulse legume and, hence, seed quality and quantity. However, legume-rhizobium symbioses are sensitive to drought, and, therefore, N fixation can be inefficient (Serraj et al., 1999; Mula and Saxena, 2010; Varshney et al., 2012)

Pigeon pea plants are taken as a sample:

Pigeon pea is an important commercial pulse crop for the farmers of Karnataka, a south Indian state. Pigeon pea (*Cajanus cajan*), a leguminous crop, is widely grown in the arid and semi-arid tropical regions. About 6.8 million tons are produced globally, out of which Asia accounts for 83% while Africa and America account for 14.2% and 2.2% respectively (FAOSTAT,2017)). Pigeon pea seeds are commonly consumed as a protein-rich food in Asia, Latin America, and Africa. Pigeon pea has a typical root system which helps it to cope up with the water limiting conditions and make it a choice of crop for rain-fed cultivation in the arid as well as semi-arid tropical regions. Besides having well-adopted plant architecture, pigeon pea cannot sustain under longer dry spells frequently occurring during the cropping season (Upadhyaya et al., 2012). Similarly, water logging conditions also hamper pigeon pea is essential to avoid losses under extreme water regimes

Arsenic bioaccumulation in plants is a serious environmental problem that has serious consequences for ecosystems and human health. Arsenic is an element that occurs naturally in the Earth's crust in a number of different forms. Arsenic is a metal that, while necessary for some species in small doses, may be toxic if present in the environment in large enough quantities. The presence of Arsenic pollution commonly originates from two primary sources: natural geological occurrences and anthropogenic activity, including mining, industrial operations, and the utilisation of pesticides and fertilisers containing arsenic. Following its discharge into the environment, Arsenic has the ability to last for prolonged durations, hence presenting a substantial hazard to ecosystems

The process by which plants take up Arsenic from the soil and water and store it in their tissues is known as **Bioaccumulation**. Numerous elements, such as plant type, ambient circumstances, and soil characteristics, affect how much Arsenic plants can absorb. Certain plants, referred to as hyperaccumulators, have developed defence mechanisms that allow them to absorb extremely high concentrations of Arsenic without becoming harmful. Because of their special qualities, researchers are examining the use of hyperaccumulators in phytoremediation—a technique that uses plants to concentrate and remove heavy metals from polluted soils. The buildup of Arsenic in plants can have ripple effects that spread across ecosystems. When herbivores ingest plants that have been polluted with arsenic, they run the risk of being exposed to potentially dangerous quantities of arsenic, which can then travel up the food chain. Because of this bioaccumulation, both wildlife and people who ingest food that

has been polluted are at danger. (Huang et al., 2006; Karimi and Alavi, 2016). (Karn, 2015; Sadee et al., 2016; Taylor et al., 2017).

• <u>1.1 HISTORY OF ARSENIC</u>

Arsenic is a chemical element with the symbol As and its atomic number 33. It is classified as a metalloid, which means it exhibits properties of both metals and nonmetals. Arsenic is known for its toxicity and has a complex history dating back thousands of years.

- i. Atomic Number: 33
- ii. Atomic Weight: Approximately 74.92 atomic mass units (amu)

• <u>1.2 Historical Use:</u>

The longest-used form of Arsenic has been in its compounds, such as Arsenic sulphides. It was utilised for beauty and therapeutic purposes in many different civilizations. Arsenic-containing substances were utilised by the ancient Greeks and Romans in their cosmetics and medicines.

<u>1.3 Discovery and Naming:</u>

The discovery of Arsenic as a distinct element is credited to the German alchemist Albertus Magnus in the 13th century.

The name <u>"Arsenic"</u> is derived from the Greek word <u>"Arsenikon"</u> which means <u>"yellow orpiment"</u>.

One of the biggest risks to the public's health is Arsenic, a toxic heavy metal. Sources of exposure to Arsenic include the workplace or contaminated water and food. Arsenic has a lengthy history of use as a metalloid material and as a pharmaceutical. The king of poisons and poison of kings are two well-known names for it. Food, water, and the environment all contain it as a pollutant. Metalloid (As0), inorganic (As³⁺ and As⁵⁺), organic, and Arsine (AsH₃) are the different forms of Arsenic. The small intestine is the primary site of assimilation. Other exposure pathways include skin-to-skin contact and inhalation. Arsenic is metabolised to monomethylarsonic acid (MMA) and dimethylarsinic acid (DMA), the latter of which is the major form in the urinary excretion of arsenic, after distribution to several tissues and organs in the body, including the lungs, heart, kidneys, liver, muscles, and neural tissue. Arsenic poisoning, both acute and chronic, is linked to the malfunctioning of many important enzymes. Similar to other heavy metals, Arsenic can prevent the operation of enzymes that contain sulfhydryl groups. Furthermore, Arsenic binds to the lipoic acid moiety of pyruvate dehydrogenase to block it. Inactivating pyruvate dehydrogenase can stop the Krebs cycle and prevent oxidative phosphorylation. Cell damage results from a decrease in ATP generation as a result. (*Arsenic Poisoning: Causes, Symptoms & Treatment*, n.d.)

<u>1.4 Symptoms:</u>

- Immediate symptoms of Arsenic poisoning may include- abdominal pain, nausea and vomiting, diarrhoea, cough, chest pain, shortness of breath, sore throat, abnormal heart rhythm (arrhythmia), low blood pressure, pins and needles feeling in fingers and toes, red, swollen skin, garlic Odor in your breath and body tissues.
- Severe Arsenic poisoning symptoms include- skin pigmentation changes, warts and lesions, hard patches on the palms of hands and soles of feet, white lines on your nails (Chronic Arsenic poisoning)
- Hight oxidative stress, Affect the structure of function of cardiovascular system Vitamin A deficiency, Night blindness, Heart disease, Eye inflammation, Hyperkeratosis and hyperpigmentation, Blushed complexion.

<u>Case study</u>

Bangladesh

77 million people (1/2 population of crowded bangladesh) may have been exposed to toxic levels of Arsenic, More than 20% of deaths are caused by Arsenic Ground water is contaminated with As

United kingdom 1990 & 1991

In uk 6000 people had the Arsenic poisoning and 70 of them died The beer contained 15ppm of Arsenic, Glucose is contaminated to Arsenic Contaminated during the refining process

Japan 1995

In japan 12,130 children suffered from Arsenic poisoning and 130 of them died. Milk from the tokushima factory was contaminated to Arsenic

Symptoms: Loss of appetite, vomiting, diarrhea, boil, skin colour changed to black anemia, hypertrophy of the liver. After effect Blur red vision, hard of hearing, low score in school record, abnormal brainwave, epilepsy, headache, dizziness, cold hands and feet

Arseni cand ADHD study (Roy et al 2010)

526,6-7 years children in Terreon Mexico ,Urinary Arsenic levels were measured No significant relationship found between any measure of urinary Arsenic and parents ratings of behaviour .However, higher urinary Arsenic was associated with high scores on Cognitive Problem exams Mees' lines) persistent sore throat, constant digestive issues

As we know, any research is mainly concerned with a particular place. And these places helps us in knowing the several activities of that specific area. Raichur and Hatti are two places in the Indian state of Karnataka, each with its unique historical, cultural, and economic significance. Raichur and Hatti are two places in the Indian state of Karnataka, each with its unique historical, cultural, and economic significance economic significance.

Raichur district

Raichur is located in the eastern part of Karnataka, situated between the Krishna and Tungabhadra rivers. It serves as an important agricultural and industrial hub in the region. The region around Raichur is known for its cotton, paddy, and pulses production, thanks to the irrigational benefits from the Krishna and Tungabhadra rivers. Additionally, Raichur is known for its thermal power station and the solar photovoltaic power plant, which contribute significantly to its economic development.



1.2 image- Raichur map

<u>Hatti</u>

Hatti, also known as Hutti, is located in the Raichur district. It is primarily known for its gold mines. The history of gold mining in Hatti goes back to ancient times, with evidence suggesting that mining activities were carried out during the period of the Mauryan empire and continued through various periods of Indian history. The primary economic activity in Hatti is gold mining. The Hutti Gold Mines are among the top gold producers in India. Managed by the Hutti Gold Mines Company Limited (HGML), a government of Karnataka undertaking, these mines are the only active gold mines in India. The mining operations not only contribute significantly to the local economy but also offer employment opportunities to the local population. Both Raichur and Hatti, despite their economic and historical significance, are also known for their rich cultural heritage. This includes festivals, local cuisines, and traditional practices that reflect the diversity and richness of Karnataka's cultural landscape.

When writing about Raichur and Hatti, it's essential to delve into both the historical context and the current socio-economic conditions. This provides a comprehensive understanding of their significance in the past and their role in contemporary society.



1.3 image- Hatti outline map

REVIEW OF LITERATURE

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OBJECTIVES

- > Objective 1: To collect plants samples near the Hatti industrial area
- > **Objective 2:** To extract the Arsenic from the plant sample
- Objective 3: To find out the concentration of heavy metals from the extracted sample.

METHODOLOGY

- > Aim: To determine the bioaccumulation of Arsenic in plants near Hatti industrial, Raichur.
- Samples: Plants.
- Sampling method: Stratified sampling and Random sampling.
- > Number of sample chosen: Total 8 pigeon pea plants were collected
- Inclusion Criteria: Arsenic, is chosen because even in very small quantities these heavy metals are dangerous to human body.
- Exclusion Criteria: Zinc, Copper is not chosen for the study because Cu and Zn are nutritionally vital elements whose tissue concentrations are homeostatically controlled, making them less harmful to humans than other non-essential metals with stronger toxic effects.

Objective 1: Plant samples near the Hatti industrial area were collected using both random and stratified sampling techniques.

Sampling Methodologies:

Random Sampling: This method involved randomly selecting samples from the entire study area without any specific pattern or criteria. Random sampling ensured that each plant in the area had an equal chance of being selected for the sample. It helped to minimize bias and ensured the representation of the overall plant population near the industrial area.

Stratified Sampling: Stratified sampling involved dividing the study area into distinct subgroups or strata based on certain characteristics. Samples were then collected from each stratum in proportion to its representation in the overall population. This method ensured adequate representation of different environmental conditions or variables that might influence plant distribution and health.

Methods for collecting Plant samples:

- Hatti industrial area is a gold mining area.
- Locations that were taken into consideration for sample collection were named N1, N2, N3, N4, F1, F2, F3, and F4.
- N (1,2,3,4) were plots that were in closer vicinity to the Hatti area.
- F(1,2,3,4) were the plots further away from the Hatti area.
- N1- Midnapore
- N2- Kotha
- N3- Tawag
- N4- Chikkanagnur
- F1- Veerapura
- F2- Hosagudda
- F3- Rodalabanda
- F4- Yaradoni

Butter Paper Bags: Butter paper bags were used for collecting plant samples to prevent moisture loss and maintain the integrity of the samples during transportation and storage. The breathable nature of butter paper allowed for adequate air circulation, preventing the buildup of humidity that could lead to fungal growth or decay of plant material.

Ziplock Bags: Ziplock bags provided an additional layer of protection against moisture and contamination. They were sealed to prevent any leakage or spillage of plant samples, ensuring their preservation until further analysis. Ziplock bags were particularly useful for collecting samples in areas where environmental conditions, such as humidity or rainfall, may have affected the quality of the samples.

Labelling: Each sample was labelled according to its location code, which allowed for easy identification and tracking of samples throughout the study. Location codes have included information on the names of villages.

Objective 2: To extract Arsenic from the plant samples.

Sample Preparation:

The plant samples were first dried to remove moisture, ensuring accurate measurements and preventing microbial growth during storage. Subsequently, the dried samples were ground into a fine powder to increase the surface area available for extraction and facilitate the release of Arsenic compounds present within the plant tissues.

Extraction Process:

a. Sample Weighing: A precise amount of 1 gram of the ground plant sample was measured for extraction, ensuring consistency and reproducibility in the analysis.

b. Addition of Sodium Carbonate: 0.7 grams of sodium carbonate were added to the 1 gram of plant sample to create alkaline conditions favourable for the extraction of Arsenic. Sodium carbonate acted as a reagent to promote the release of Arsenic ions from the plant matrix.

c. Addition of Distilled Water: 15 ml of distilled water were added to the mixture of plant sample and sodium carbonate. Distilled water served as a solvent, aiding in the dissolution of Arsenic compounds and facilitating their extraction from the plant material.

d. Heating: The mixture was heated to 90 degrees Celsius, optimizing the extraction process by increasing the solubility of Arsenic compounds in the alkaline solution. Heating also accelerated the chemical reactions involved in the release of Arsenic ions from the plant matrix.

e. Filtration: After heating, the mixture was filtered to separate the liquid solution containing extracted Arsenic from the solid residue. Filtration removed any undissolved plant debris or insoluble particles, ensuring the purity of the extracted solution.

> **<u>Objective 3:</u>** To find out the concentration of heavy metals from the extracted sample.

HEAVY METAL	PRELIMINARY TEST
Arsenic	Gutzeit Test:
	 1 ml of the sample was taken in a test tube. 2 pellets of pure zinc metal were put into it. 5 ml of dil. sulphuric acid was poured over the contents. The evolved gas was purified by passing over lead acetate paper (to absorb H₂S gas) and was reacted finally with mercuric chloride test paper. A yellow stain on the paper indicates the presence of Arsenic.

Protocol for the Detection of Arsenic (DFSS Toxicology Manual)

PRELIMINARY TEST (Colour Test)

Scheele's Green:

Addition of Cupric Sulphate: After heating, cupric sulphate was added to the mixture. The addition of cupric sulphate resulted in the formation of a green colour, indicating the presence of Arsenic. This

colour change served as a visual indicator of successful extraction and presence of Arsenic ions in the solution.

INSTRUMENTAL ANALYSIS

With the help of an instrument Inductively Coupled Plasma Optical Emission Spectroscopy (ICP-OES).

An analytical method known as inductively coupled plasma-optical emission spectroscopy (ICP-OES) is used to determine the atomic composition of a specific sample

Principle: ICP OES uses an inductively coupled plasma to excite the atoms in the sample. A generator supplies argon gas with enough energy for it to jump to the next physical state. With a high particle density, the plasma reaches temperatures of 5,000 to 10,000 K. The measurement solution is then injected into the plasma. It takes a fraction of a second for the sample to dry out, melt and finally to vaporize. The gas molecules, which are now also being excited, are then atomized and ionized. They emit the electromagnetic radiation that is used for the actual analysis. Transfer optics direct the radiation to another optical component that can separate out the various wavelengths. After the waves are split, a detector registers the intensity of each wavelength, which is proportional to the concentration of the respective analyte. As the analysis depends on the correlation between the light intensity and the element concentration, calibration standards come into play. These calibration standards help to derive a mathematical function which relates radiation to concentration.

Advantages of ICP-OES

- Can be used to measure almost all the elements in the periodic table.
- The technique has a wide dynamic concentration range and can measure elements at trace to high concentrations.
- Detection limits for most elements are in the range of micrograms per litre to milligrams per litre.
- Multi-elemental quantitative analysis can be carried out in a period as short as 1 min with a small amount of solution (0.5–1.0 mL).
- Technique that combines good quantitative multielement capability, wide linear dynamic ranges, good sensitivity, limited spectral and chemical interferences, low detection limits, and speed and ease of data handling and reporting with widespread (multiple-vendor) instrument availability and reasonable cost.

Disadvantages of ICP-OES

- Expensive
- Poor precision lack of reproducibility in results obtained for the same sample.
- Sample drift situation in which the signal is not stable and changes in position over time.
- Non-ideal detection limits means that in many cases, the detection limits obtained through the use of ICP-OES are higher than desired for the target application.

Inaccurate identification refers to situations in which the ICP-OES signal identifies a signal as corresponding to one element when it in fact belongs to a different element.

OBSERVATION AND RESULTS

Sl.No	Location	Unit	As concentration
1	N1 t Midnapore	mg/kg	0.098
2	N2 t Kotha	mg/kg	0.094
3	N3 t Tawag	mg/kg	0.097
4	N4 t Chikkanagnur	mg/kg	0.098
5	F1 t Veerapura	mg/kg	0.096
6	F2 t Hosagudda	mg/kg	0.048
7	F3 t Rodalabanda	mg/kg	0.089
8	F4 t Yaradoni	mg/kg	0.092

Results obtained for Preliminary test





Sample N1t showing Positive result



Sample N2t showing Positive result



Sample N3t showing Positive result



Sample N4t showing Positive result

Discussion

The suspected Pigeon pea plants were selected for the detection of Arsenic and extraction was done using alkaline leaching method assisted with temperature (modified form of wet digestion method). Initial preliminary chemical tests were done for suspicion of presence of Arsenic, followed by instrumental analyses using ICP-OES. On instrumental analysis these observations were made which are mentioned below;

- 1. sample N1t showed presence of 0.098 mg/kg,
- 2. sample N2t showed presence of 0.094 mg/kg,
- 3. sample N3t showed presence of 0.097 mg/kg,
- 4. sample N4t showed presence of 0.098 mg/kg
- 5. sample F1t showed presence of 0.096 mg/kg
- 6. sample F2t showed presence of 0.048 mg/kg
- 7. sample F3t showed presence of 0.089 mg/kg
- 8. sample F4t showed presence of 0.092 mg/kg

Based on the samples analysed above it can be clearly seen that the presence of Arsenic is there in these edible Pigeon pea plant. Although the amount present is less than the toxicity limit. Ingestion of 70 to 180 mg can be fatal, but initial effects may be delayed for several hours or days. Acute oral ingestion of lower doses can result in these symptoms

Irritation of stomach and intestines with symptoms such as stomach-ache, nausea, vomiting and diarrhea. Decreased production of red and white blood cells, which may cause fatigue, abnormal heart rhythm, blood-vessel damage resulting in bruising and impaired nerve function causing a "pins and needles" sensation in hands and feet. White bands, called Mees'lines, are visible in the nails.

Still it can have hazardous effect on lethal exposure at a higher consumption or for long period of time Chronic effect

Signs and possible impacts of Arsenic exposure:

Skin changes including patches of darkened skin and the appearance of small "corns" or "warts" on the palms, soles, and torso; these are often associated with changes in the blood vessels of the skin.

Skin cancer

Cancer of the liver, bladder, kidney or lungs

Possibly diabetes, high blood pressure, reproductive disorders and lower IQ scores.⁽¹⁴⁾⁽¹⁵⁾

It is clearly indicative that the plant sample taken for analysis is only close to 1 gram (whole plant was collected, dried, crushed and only one gram was taken for analysis) where it is giving results in decimals of milligram per kilo gram. But the normal consumption can have many peas involved which can cause acute to chronic effects based on the age group involved in consumption.

Comportment Model of Arsenic in the Body:

Arsenic, a toxic metalloid, exhibits various behaviours within the human body, largely influenced by its chemical forms and routes of exposure. Upon ingestion, Arsenic undergoes complex distribution and redistribution processes, facilitated by physiological mechanisms. The comportment model makes clear these dynamics, describing the journey of Arsenic from entry to eventual elimination.

Following ingestion, Arsenic primarily enters the bloodstream through the gastrointestinal tract, where it encounters absorption barriers and mechanisms. Arsenic species may undergo biotransformation within the gut, leading to the formation of different metabolites with varying toxicities and affinities for specific tissues. Once absorbed, Arsenic distributes throughout the body via the circulatory system, reaching target organs such as the liver, kidneys, lungs, and skin.

Within tissues, Arsenic undergoes further metabolism, involving methylation, reduction, and oxidation reactions, mediated by enzymatic pathways. These metabolic processes contribute to the formation of metabolites with altered chemical properties, influencing their reactivity, solubility, and excretion kinetics. Arsenic compounds may undergo sequestration within cellular compartments, such as lysosomes and mitochondria, or binding to biomolecules, including proteins and DNA, affecting cellular function and integrity.

The redistribution of Arsenic within the body occurs through dynamic equilibria between various tissue compartments, driven by factors such as blood flow, tissue perfusion, and metabolic activity. Arsenic may undergo compartmentalization within organs, with preferential accumulation in certain tissues over others, dictated by factors like lipid solubility, protein binding, and membrane permeability. The dynamics of Arsenic redistribution contribute to its systemic toxicity and chronic health effects, as it persists within the body over extended periods, exerting cumulative impacts on cellular and organ function.

Conclusion

The bioaccumulation of Arsenic in edible plants in the Hatti industrial region in Raichur, Karnataka, is a significant environmental and public health problem. Plants in the surrounding area absorb Arsenic from the soil and water as a result of industrial operations such as mining and metallurgy, which accumulate this harmful element. As a result, edible plants grown in these places become poisoned with arsenic, posing serious health hazards to people when consumed. According to studies, persistent Arsenic exposure can cause a variety of health concerns, including skin lesions, respiratory disorders, cardiovascular ailments, and even cancer. As a result, it is critical to address this issue through comprehensive environmental management measures, including rehabilitation of polluted areas, implementation of stronger industrial laws, and monitoring of Arsenic levels in agriculture.

Limitations:

- > The analysis only considered a 1-gram subsample from a potentially larger representative sample.
- Further investigation is needed to determine the arsenic distribution within different plants parts (peas vs leaves/stems)

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Annexure

Sample collection



INSTRUMENTAL ANALYSIS



SHIVA ANALYTICALS (INDIA) PRIVATE LIMITED

Plot No. 24D(P) & 34D, KIADB Industrial Area, Hoskote, Bangalore - 562 114. Karnataka, India Tel:+91-80-2801 5333 Email:info@shivaanalyticals.com Website:www.shivaanalyticals.com CIN:U24230KA1994PTC125297 | GSTIN:29AACCS8434K1ZV

Test Report No: CERT2024003918_BB Order No: 2024003918

Test Report

Product Classification	Plant Products				
Customer Name	JAIN (DEEMED - TO - BE) UNI	JAIN (DEEMED - TO - BE) UNIVERSITY, SCHOOL OF SCIENCES, JC ROAD, BANGALORE - 560027			
Address	Reena Susan Philip, Head of the Department, Department of Forensic Science, School of Sciences, Jain (Deemed - to - be) University, J C Road, Bangalore - 560027 560027 Bangalore				
Sample Reference Name	N1t				
Sample Reference No	NA	Date of Receipt	06-03-2024		
Date of Registration	06-03-2024	Date of Start of Analysis	06-03-2024		
Date of End of Analysis	11-03-2024	Date of Issue of Report	11-03-2024		
Batch No	NA				
Discipline	Chemical				
Group	Food & Agricultural Products				
Sample Id	NA				
Invoice No., Date & Consignee Details:	NA				
PO	TRF				
PO Date:	06-03-2024				

Test Results

S.No	TEST PARAMETER	UNIT	RESULT	TEST METHOD
1	Metals			
1.1	Arsenic as As	mg/kg	0.098	SAIL/FOOD/MOA/017

	Terms And Conditions
S.No	Description
1	The results reported above pertains only to the tested sample and applicable parameters, where the Laboratory has not performed the Sampling.
2	Samples which are degradable will be disposed immediately after testing and others will be disposed after one month from the date of issue of test certificate unless otherwise specified.
3	Total liability of our laboratory is limited to the invoiced amount.
4	This report is not to be reproduced either wholly or in part and cannot be used as an evidence in the Court of Law and should not be used in any advertising media without prior written permission.
5	In case any reconfirmation of contents of this test certificate is required, please contact our office.
6	Sampling is not done by us unless specified.
7	Any discrepancy in the Test Certificate should be notified within 30 days.

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Test Report No: Order No:

CERT2024003920_BB

2024003920

Test Report

Product Classification	Plant Products			
Customer Name	JAIN (DEEMED - TO - BE) UNIVERSITY, SCHOOL OF SCIENCES, JC ROAD, BANGALORE - 560027			
Address	Reena Susan Philip, Head of the be) University, J C Road, Banga	Reena Susan Philip, Head of the Department, Department of Forensic Science, School of Sciences, Jain (Deemed - to - be) University, J C Road, Bangalore - 560027 560027 Bangalore		
Sample Reference Name	N2t			
Sample Reference No	NA	Date of Receipt	06-03-2024	
Date of Registration	06-03-2024	Date of Start of Analysis	06-03-2024	
Date of End of Analysis	11-03-2024	Date of Issue of Report	11-03-2024	
Batch No	NA			
Discipline	Chemical			
Group	Food & Agricultural Products			
Sample Id	NA			
Invoice No., Date & Consignee Details:	NA			
PO	TRF	TRF		
PO Date:	06-03-2024			

Test Results

S.No	TEST PARAMETER	UNIT	RESULT	TEST METHOD	1
1	Metals				1
1.1	Arsenic as As	ma/ka	0.094	SAIL/FOOD/MOA/017	1

Remarks .

BLQ-	Below Limit of Quantification	
	Terms And Conditions	
S.No	Description	_
1	The results reported above pertains only to the tested sample and applicable parameters, where the Laboratory has not performed the Sampling.	_
2	Samples which are degradable will be disposed immediately after testing and others will be disposed after one month from the date of issue of test certificate unless otherwise specified.	_
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5	In case any reconfirmation of contents of this test certificate is required, please contact our office.	
6	Sampling is not done by us upless specified	_

Any discrepancy in the Test Certificate should be notified within 30 days. 7

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Test Report No: Order No: CERT2024003921_BB

2024003921

Test Report

Product Classification	Plant Products			
Customer Name	JAIN (DEEMED - TO - BE) UNIVERSITY, SCHOOL OF SCIENCES, JC ROAD, BANGALORE - 560027			
Address	Reena Susan Philip, Head of the Department, Department of Forensic Science, School of Sciences, Jain (Deemed - to - be) University, J C Road, Bangalore - 560027 560027 Bangalore			
Sample Reference Name	N3t			
Sample Reference No	NA	Date of Receipt	06-03-2024	
Date of Registration	06-03-2024	Date of Start of Analysis	06-03-2024	
Date of End of Analysis	11-03-2024	Date of Issue of Report	11-03-2024	
Batch No	NA			
Discipline	Chemical			
Group	Food & Agricultural Products			
Sample Id	NA			
Invoice No., Date & Consignee Details:	NA			
PO	TRF			
PO Date:	06-03-2024			

		Test Resu	llts	
S.No	TEST PARAMETER	UNIT	RESULT	TEST METHOD
1	Metais			
	Areanic as As	malka	0.007	CAIL ICOOD MICA IN 17

Remark BLQ-	is : Below Limit of Quantification
	Terms And Conditions
S.No	Description
1	The results reported above pertains only to the tested sample and applicable parameters, where the Laboratory has not performed the Sampling.
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-feam Leader-Food Rajanna TC

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Test Report No: Order No: CERT2024003922_BB

2024003922

Test Report

Product Classification	Plant Products		
Customer Name	JAIN (DEEMED - TO - BE) UNIVERSITY, SCHOOL OF SCIENCES, JC ROAD, BANGALORE - 560027		
Address	Reena Susan Philip, Head of the Department, Department of Forensic Science, School of Sciences, Jain (Deemed - to - be) University, J C Road, Bangalore - 560027 560027 Bangalore		
Sample Reference Name	N4t		
Sample Reference No	NA	Date of Receipt	06-03-2024
Date of Registration	06-03-2024	Date of Start of Analysis	06-03-2024
Date of End of Analysis	11-03-2024	Date of Issue of Report	11-03-2024
Batch No	NA		
Discipline	Chemical		
Group	Food & Agricultural Products		
Sample Id	NA		
Invoice No., Date & Consignee Details:	NA		
PO	TRF		
PO Date:	06-03-2024		

	Test Results					
S.No	TEST PARAMETER	UNIT	RESULT	TEST METHOD		
1	Metals					
	Areanic as As	malka	0.008	CALL/COOD/HOL/017		

Remark - BLQ-	s : Below Limit of Quantification
	Terms And Conditions
S.No	Description
1	The results reported above pertains only to the tested sample and applicable parameters, where the Laboratory has not performed the Sampling.
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Authorised Signatory

Team Leader-Food Rajanna TC

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Test Report No: Order No: CERT2024003923_BB

2024003923

Test Report

Product Classification	Plant Products				
Customer Name	JAIN (DEEMED - TO - BE) UNIVERSITY, SCHOOL OF SCIENCES, JC ROAD, BANGALORE - 560027				
Address	Reena Susan Philip, Head of the Department, Department of Forensic Science, School of Sciences, Jain (Deemed - to - be) University, J C Road, Bangalore - 560027 560027 Bangalore				
Sample Reference Name	F11				
Sample Reference No	NA	NA Date of Receipt 06-03-2024			
Date of Registration	06-03-2024	Date of Start of Analysis	06-03-2024		
Date of End of Analysis	11-03-2024	Date of Issue of Report	11-03-2024		
Batch No	NA				
Discipline	Chemical				
Group	Food & Agricultural Products				
Sample Id	NA				
Invoice No., Date & Consignee Details:	NA				
PO	TRF				
PO Date:	06-03-2024				

S.No	TEST PARAMETER	UNIT	RESULT	TEST METHOD		
1	Metals					
1.1	Arsenic as As	mg/kg	0.096	SAIL/FOOD/MOA/017		

Test Results

Remark BLQ-	is : Below Limit of Quantification
	Terms And Conditions
S.No	Description
1	The results reported above pertains only to the tested sample and applicable parameters, where the Laboratory has not performed the Sampling.
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R ST-C ader-Food TC	Autherized Signatory (Emp. 10, 559)

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Test Report No: Order No: CERT2024003925_BB

2024003925

Test Report

Product Classification	Plant Products		
Customer Name	JAIN (DEEMED - TO - BE) UNIVERSITY, SCHOOL OF SCIENCES, JC ROAD, BANGALORE - 560027		
Address	Reena Susan Philip, Head of the Department, Department of Forensic Science, School of Sciences, Jain (Deemed - to - be) University, J C Road, Bangalore - 560027 560027 Bangalore		
Sample Reference Name	F21		
Sample Reference No	NA	Date of Receipt	06-03-2024
Date of Registration	06-03-2024	Date of Start of Analysis	06-03-2024
Date of End of Analysis	11-03-2024	Date of Issue of Report	11-03-2024
Batch No	NA		
Discipline	Chemical		
Group	Food & Agricultural Products		
Sample Id	NA		
Invoice No., Date & Consignee Details:	NA		
PO	TRF		
PO Date:	06-03-2024		

S.No	TEST PARAMETER	UNIT	RESULT	TEST METHOD
1	Metais			14
1.1	Arsenic as As	mg/kg	0.048	SAIL/FOOD/MOA/017

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Remark	is : Below Limit of Quantification
	Terms And Conditions
S.No	Description
1	The results reported above pertains only to the tasted sample and applicable parameters, where the Laboratory has not performed the Sampling
2	Samples which are degradable will be disposed immediately after testing and others will be disposed after one month from the date of issue of test certificate unless otherwise specified.
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Test Report No: Order No: CERT2024003926_BB

2024003926

Test Report

	Plant Products			
JAIN (DEEMED - TO - BE) UNIVERSITY, SCHOOL OF SCIENCES, JC ROAD, BANGALORE - 560027				
Reena Susan Philip, Head of the Department, Department of Forensic Science, School of Sciences, Jain (Deemed - to - be) University, J C Road, Bangalore - 560027 560027 Bangalore				
F3t				
NA	Date of Receipt	06-03-2024		
06-03-2024	Date of Start of Analysis	06-03-2024		
11-03-2024	Date of Issue of Report	11-03-2024		
NA		Annual and a second second		
Chemical				
Food & Agricultural Products				
NA				
NA				
TRF				
06-03-2024				
	JAIN (DEEMED - TO - BE) UNI Reena Susan Philip, Head of th be) University, J C Road, Bang: F3t NA 06-03-2024 11-03-2024 NA Chemical Food & Agricultural Products NA NA TRF 06-03-2024	JAIN (DEEMED - TO - BE) UNIVERSITY, SCHOOL OF SCIENCES, JC Reena Susan Philip, Head of the Department, Department of Forensic Si be) University, J C Road, Bangalore - 560027 560027 580027 Bangalore F3t NA Date of Receipt 06-03-2024 Date of Start of Analysis 11-03-2024 Date of Issue of Report NA Chemical Food & Agricultural Products NA NA TRF 06-03-2024		

	Test Nestits				
S.No	TEST PARAMETER	UNIT	RESULT	TEST METHOD	
1	Metals				
1.1	Arsenic as As	mg/kg	0.089	SAIL/FOOD/MOA/017	

Remark BLQ-	is : Below Limit of Quantification
	Terms And Conditions
S.No	Description
1	The results reported above pertains only to the tested sample and applicable parameters, where the Laboratory has not performed the Sampling.
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Test Report No: Order No: CERT2024003927_BB 2024003927

Test Report

Product Classification	Piant Products			
Customer Name	JAIN (DEEMED - TO - BE) UNIVERSITY, SCHOOL OF SCIENCES, JC ROAD, BANGALORE - 560027			
Address	Reena Susan Philip, Head of the Department, Department of Forensic Science, School of Sciences, Jain (Deemed - to - be) University, J C Road, Bangalore - 560027 560027 Bangalore			
Sample Reference Name	F41			
Sample Reference No	NA	Date of Receipt	06-03-2024	
Date of Registration	06-03-2024	Date of Start of Analysis	06-03-2024	
Date of End of Analysis	11-03-2024	Date of Issue of Report	11-03-2024	
Batch No	NA			
Discipline	Chemical			
Group	Food & Agricultural Products			
Sample Id	NA			
Invoice No., Date & Consignee Details:	NA			
PO	TRF			
PO Date:	06-03-2024			

	1001100010			
S.No	TEST PARAMETER	UNIT	RESULT	TEST METHOD
1	Metals			
1.1	Arsenic as As	mg/kg	0.092	SAIL/FOOD/MOA/017

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Remark BLQ-	is : Below Limit of Quantification
	Terms And Conditions
S.No	Description
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