Journal of Prevention, Diagnosis and Management of Human Diseases
Vol: 04, No. 02, July-Dec 2024
ISSN: 2799-1202, DOI: 10.55529/jpdmhd.42.28.34

Research Paper

Isolation and diagnosis of bacteria and fungi from some areas of tikrit and some villages

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Article Info

Article History:
Received: 17 April 2024
Revised: 24 June 2024
Accepted: 30 June 2024
Published: 18 August 2024

Keywords:
Isolation
Diagnosis
Bacteria and Villages
Fungal Identification
Air Quality Assessment

ABSTRACT

Background: Airborne microorganisms, including bacteria and fungi, contribute to environmental air pollution and may pose health risks, especially in agricultural regions.
Objective: To assess airborne bacterial and fungal contamination in selected areas of Tikrit, including Al-Alam, Al-Bu Ajil, Al-Buhyazaa, and Al-Karaat village.
Methods: A descriptive study was conducted during November and January 2021–2022 using the passive settle plate method. Petri dishes containing Potato Dextrose Agar (PDA) were exposed to air for 15 minutes across sampling sites. After incubation, microbial growth was analyzed. Bacteria were cultured on MacConkey agar and Eosin Methylene Blue (EMB) agar. Gram staining was used for bacterial identification, while Lactophenol cotton blue staining was used for fungal identification.
Results: Fungal contamination was higher than bacterial contamination, with Penicillium spp. predominating in agricultural areas. The highest colony count was observed in Al-Bu Ajil (sample G2.1), with over 96 colonies, approximately two to three times higher than other sites, likely due to favorable environmental conditions such as humidity and temperature. Most bacterial isolates were Gram-negative, mainly Klebsiella spp., which showed growth on MacConkey agar in Tikrit, Al-Alam, and Al-Karaat, indicating environmental adaptability. No bacterial growth was detected in Al-Buhyazaa (sample 3.1 H), possibly due to unfavorable conditions.
Conclusion: Airborne microbial contamination varied across study sites, with higher fungal loads in agricultural areas and widespread presence of Gram-negative bacteria. Environmental factors significantly influence microbial distribution in air.

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1. INTRODUCTION

The common environmental pollutants found in the air are chemicals, particles, or biological materials that have an impact on human and other organism health [1]. Furthermore, indoor sources of air pollution include hairspray, room deodorizers, photocopiers, paints, solvents, printers, computers, and air purifiers. Due to people's excessive time spent at home and at work, household air pollution has a negative impact on health [2]. Many microbes, the majority of which are pathogenic, proliferate in the human environment. While some bacteria were not infectious when they were small, they have the potential to cause infections when they grow to optimal levels. These bacteria are known as opportunistic bacteria [3].

2. RELATED WORK

Many of the fungi that cause allergies in humans are classified as Ascomycota, Basidiomycota, or yeast [4]. These fungi include Aspergillus, Rhizopus, Penicillium, Cladosporium, and Alternaria, and they can grow in the air and on surfaces made of natural and artificial materials. They are present throughout the year, but are most prevalent in the fall [5], [6]. Moreover, temperature, humidity, air movement, and air exchange rate are the primary environmental factors that support the development and reproduction of airborne microorganisms [7].

2.1.The Aim of this Study

The purpose of this study was to evaluate the amount of fungi and bacteria that are causing air pollution in some locations of Tikrit, including the Al-Alam region, Al-Bu Ajil, Al-Buhyazaa, and the town of Al-Karaat [8].

3. METHODOLOGY

3.1. Material and Methods

[9] The biology labs at Tikrit University in Iraq's College of Science conducted the current study. The purpose of the study was to evaluate the amount of bacteria and fungi that cause air pollution in a few different areas of Tikrit, including the Al-Alam area, Al-Bu Ajil, Al-Buhyazaa, and the village of Al-Karaat. Samples for the study were first collected in November and January of 2021–2022.

3.2. Macconkey Agar

To prepare this medium, dissolve 51.5 g of MacConkey agar in 1 liter of distilled water. Transfer the mixture into a sterilizing container. Distribute the mixture into petri dishes and store in the refrigerator at 4 oC until needed [1].

3.3. Nutrinet Agar

To prepare this media, dissolve 28 grams in one liter of distilled water, put it in a sterilizing container, pour it into sterile Petri dishes, and store it in the refrigerator until needed [1].

3.4. Eosin Methylene Blue

As directed by the manufacturer, 35.96 g of this medium are dissolved in one liter of distilled water to prepare it. It is then transferred into a sterilizing container, distributed among plates, and kept in the refrigerator until needed [10].

3.5. Potato Dextrose Agar

This medium is made by dissolving 39 grams of it in one liter of distilled water, then putting it in an antibiotic to sterilize it and an antibiotic such as an anti-tetracycline with the medium. After that, the dishes are distributed, the incubator is used to make sure there is no contamination, and the media is refrigerated until needed [1].

3.6. Sterilization

All culture media is sterilized by autoclave at a temperature of 121°C \ 15 bar for 15 minutes [11].

3.7. Sample Collection

In order to determine the kind and quantity of pollutants present in the atmosphere of the study areas, Nutrient Agar, Potato Agar, and Potato Dextrose Agar media were distributed throughout the villages of Al-Karaat, Tikrit, Al-Alam, Al-Bu Ajil, and Al-Buhyazaa. The petri dish was then closed and placed in the incubator after being left open for 15 minutes [12]. The number of colonies that grew on the aforementioned dishes was calculated by counting them and recording the growth [13].

3.8. Samples Culture

After the isolates from the study areas were gathered, they were cultivated on eosin methylene blue agar, MacConkeys agar, and incubated at 37 ºC for a full day. Following this, the first growth observations were recorded and used for diagnosis [14].

3.9. Cultural Characteristics

On Nutrient Agar, MacConkeys Agar, Eosin Methylene Blue, and Potato Dextrose Agar, the features of the developing colonies were observed, including their size, shape, color, and capacity for differential growth as well as their ability to ferment. Shape, color, size, ability to grow on one medium without needing another, and fermentation were among the attributes noted [15].

3.10. Microscopic Characteristics

The bacteria underwent gram stain staining, followed by a microscope examination to document their features, including size, type (positive or negative), and how their cells were arranged in a chain or doubled up [16]. Meanwhile, the fungi were dyed blue with lactophenol to observe their external structures, including thalus and clipboard spores, and how their cells were arranged in single or doubled up groups (Alhayfat).

4. RESULTS AND DISCUSSION

4.1. Total Number of Colonies

Table 1. Represents the Total Bacteria Numbers in Different Areas

Albu Hayaza	Albu Ajil	Krayat	Alam	Tikrit	Bacterial Types	Date
41	55	55	47	30	Klebsiella, Lactococcus, Enterococcus, Brucella	7/11
42	45	45	48	29	Klebsiella, Lactococcus, Enterococcus, Brucella	22/11
40	33	33	37	34	Klebsiella, Lactococcus, Enterococcus, Brucella	5/12
41	32	32	36	31	Klebsiella, Lactococcus, Enterococcus, Brucella	19/12
45	36	36	42	32	Klebsiella, Lactococcus, Enterococcus, Brucella	10/1

47	39	39	44	33	Klebsiella, Lactococcus, Enterococcus, Brucella	22/1

4.2. Total Number of Fungi

Table 1 displays the findings of the current study. It shows that a variety of bacteria, including lactobacilli and klebsiella, grew most in November in Albu Ajil. This was attributed to the favorable environmental conditions, which included a temperature range of 12 to 43 C° and humidity fluctuations [17]. These species also increased in the spring and autumn because of the presence of virulent factors, such as toxins, capsules, and antibiotic resistance, which aid in their survival, spread, and infection [7].

The results showed that the months with the lowest growth ratio were December and November, when the temperature dropped due to the cold weather. Nonetheless, certain anaerobic bacteria were able to withstand inappropriate environmental conditions [8]. Because the formation of lactic acid lowers pH, which acts as an inhibiting substance, these bacteria are also important in the production of natural preservatives and foods [9].

However, the presence of farm animals in these villages is what caused the bacteria to spread throughout the Al-Bu Ajil and Al-Buhayaz area. They were spread through contaminated soil, milk products, and direct human contact [10]. Some of them, like Enterococcus, can grow in both rich and poor environments with oxygen because they can withstand high temperatures between 10 and 45 degrees Celsius, a pH range of 4.6 to 9.9, and high sodium chloride concentrations [3].

Table 2. Represents the Total Numbers of Fungi

Albu Hayaza	Albu Ajil	Krayat	Alam	Tikrit	Fungus Strain	Date
43	47	11	34	9	EXpansum Penicillium, Aspergillus Nijer, Rhizopus, Arthrographis	7/11
45	49	12	33	10	EXpansum Penicillium, Aspergillus Nijer, Rhizopus, Arthrographis	22/11
40	96	14	28	8	EXpansum Penicillium, Aspergillus Nijer, Rhizopus, Arthrographis	5/12
41	94	12	26	10	EXpansum Penicillium, Aspergillus Nijer, Rhizopus, Arthrographis	19/12
42	82	13	30	11	EXpansum Penicillium, Aspergillus Nijer, Rhizopus, Arthrographis	10/1
43	85	14	31	12	EXpansum Penicillium, Aspergillus Nijer, Rhizopus, Arthrographis	22/1

[11], [12] The results of the current study were displayed in Table 2, where it was found that the predominant fungus strain is related to the genera Penicillium and Rhizobus. Arthrographis is responsible for blue mold disease, which is the most common fruit disease that affects apples.

It also infects a variety of hosts, including pears, tomatoes, corn, and rice, with Albu Ajil and Al-Buhayaz having the highest ratio of growth due to the area's agricultural nature and humidity availability [13]. These fungi are abundant in agricultural areas because some of them can harm citrus fruits [14].

Because Rhizopus fungi are heterotrophic fungi, they feed on human and plant parasites and can be found in a variety of organic materials, including fruits and vegetables [8]. Arthrographis is a type of fungus that grows in wood, soil, marine sediments, and agricultural environments. Due to the abundance of animals and air pollution from dust and high humidity, it was the cause of infectious animal disease. The areas with the largest percentage of fungal growth were Albu Ajil and Al-Buhyazaa, and the areas with the lowest percentage were Tikrit, Alam, and Al-Karaat [18].

5. CONCLUSION

This could be because the environmental conditions at the time were insufficient for bacterial growth.

Acknowledgments

The authors have no specific acknowledgments to make for this research.

Funding Information

This research received no specific grant from any funding agency in the public, commercial, or not-for-profit sectors.

Author Contributions Statement

Name of Author	C	M	So	Va	Fo	I	R	D	O	E	Vi	Su	P	Fu
Marwa M. Mahdi	✓	✓	✓	✓		✓		✓	✓	✓	✓	✓	✓	
Sarab Dalaf Khalaf	✓		✓		✓		✓		✓		✓		✓	✓
Youns R. Abdulaah		✓		✓		✓		✓		✓		✓	✓	
Teba Anwar Ahmed	✓			✓	✓		✓		✓		✓			✓

C : **C**onceptualization
M : **M**ethodology
So : **So**ftware
Va : **Va**lidation
Fo : **Fo**rmal analysis
I : **I**nvestigation
R : **R**esources
D : **D**ata Curation
O : Writing - **O**riginal Draft
E : Writing - Review & **E**diting
Vi : **Vi**sualization
Su : **Su**pervision
P : **P**roject administration
Fu : **Fu**nding acquisition

Conflict of Interest Statement

The authors declare that there are no conflicts of interest regarding the publication of this paper.

Informed Consent

All participants were informed about the purpose of the study, and their voluntary consent was obtained prior to data collection.

Ethical Approval

The study was conducted in compliance with the ethical principles outlined in the Declaration of Helsinki and approved by the relevant institutional authorities.

Data Availability

The data that support the findings of this study are available from the corresponding author upon reasonable request.

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How to Cite: Marwa M. Mahdi, Sarab Dalaf Khalaf, Youns R. Abdulaah, Teba Anwar Ahmed. (2024). Isolation and diagnosis of bacteria and fungi from some areas of tikrit and some villages. Journal of Prevention, Diagnosis and Management of Human Diseases (JPDMHD), 4(2), 28-34. https://doi.org/10.55529/jpdmhd.44.28.34

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