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Original Paper

Evaluation of Antifungal Analysis of Selected Food Products Obtained In the Local Markets of Peshawar KPK Pakistan

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***Abstract*— Foodborne diseases are very easily transmitted from contaminated food and food handlers. Bacteria and fungi are the major factors for fruit spoilage. Food spoilage causes losses to producers, distributors, and consumers in the form of reduced quality and quantity and higher prices. The objective of this study was to isolate and evaluate the antifungal analysis of food-borne fungal pathogens. For the evaluation of pathogenic fungi Milk, Chutney, Apple, Spinach, Salad, Guava, Fish, and Peas were taken. All the other food samples were taken in polythene zip bags. The vegetables and fruits were cut into small pieces with a sterilized blade and then plated on Sabouraud dextrose agar (SDA) aseptically and then incubated for 5 days. To find foodborne pathogenic fungi in various food items, a total of 320 were collected for evaluation. Out of the total food samples (65), 20.31% were found positive for fungal pathogens in various foodstuffs while 255 (79.68%) food samples were found negative. Among the positive food samples, Fusarium spp was more predominant 28(43%) led by Aspergillus spp 25(38.5%). In comparison, Mucor and Cladosporium were found in low frequency with percentages of 7(10.8%), 5(7.7%). For antifungal activity, a panel of selected drugs was used to assess the susceptibility of pathogens. Among the antifungals best activity was shown by Voriconazole and intermediate activity was shown by Fluconazole. While high resistance was observed in Nystatin as compared to other antifungals.**

Keywords*— *Antifungal, Pathogens, Foodstuffs, Contamination, Food spoilage

I. INTRODUCTION

[1] Foodborne diseases are very common and can easily transmit from contaminated food and food equipment. Among the foodborne pathogens, strains of Staphylococcus aureus, Bacillus cereus, Listeria monocytes, and Clostridium are very important because of their presence in a wide range of foodstuffs. [2] Weather conditions can selectively promote the growth of particular fungal species, which cause rotting and spoilage of grape berries before harvest. The fungi may also lead to produce mycotoxins and cause contamination.

[3] Food spoilage is any change in the appearance, smell, or taste of food products. Food spoilage causes losses to producers, distributors, and consumers in the form of reduced quality and quantity and higher prices. The factors that affect microbial growth in food and constantly the association that develops also determine the nature of spoilage and any health risks. The influence of each factor on growth in a food system the factors are present together and exert effects on microbial growth in combination, either favorably or adversely.[4] Bacteria are the major and important factor for fruit spoilage. They will survive in suitable temperatures with the presence of food and water, which caused changes in the appearance, color, and smell of the fruits. [5] Vegetables are frequently consumed raw without being exposed to the processes that reliably eliminate pathogens. The washing of vegetable fruits and can reduce fungal levels but cannot eliminate pathogens. Eating or drinking contaminated foods or drinks can cause food-borne disease. [6] Post-harvest fungal infection of fruits and vegetables is mainly caused by fungal pathogens that can be harmful to both humans and animals as they produce mycotoxins, post-harvest diseases in fruits and vegetables are a serious problem that results in the loss of a large percentage of crops and some fruits. Microorganisms, including bacteria and fungi, cause considerable economic losses by spoiling not only harvested fruits and vegetables but also crops in their fields. The identification of food-borne fungi

is a good step to control them. [7] The wide array of available dairy foods challenges the microbiologist, engineer, and technologist to find the best ways to prevent the entry of microorganisms includes spoilage include aerobic psychrotrophic, Gram-negative, bacteria, yeasts, molds, heterofermentative lactobacilli, and spore-forming bacteria. [8] Certain fungi such as Aspergillus, Fusarium, and Penicillium are commonly occurring filamentous fungi found in vegetables and their growth may result in the production of mycotoxins, which can cause a variety of illnesses in humans, from an allergic response to immune suppression and cancer. [9] The transmission of this disease by hand, food, and kitchen surfaces had been reported from 5 to 100%. Several reports have been indicated that food-borne diseases are a global problem. Contaminated food causes 1.5 billion cases of diarrhea in children annually, Leading To More Than 3 Million Deaths All Over The World.

II. Materials And Methods

This research study was conducted in the Microbiology Research Laboratory, Abasyn University Peshawar from April 2021 to May 2022. The work focused on isolation, identification and antibiogram analysis of pathogenic fungal species from various food stuffs.

TABLE I OVERALL FREQUENCY OF FUNGAL PATHOGENS IN VARIOUS FOOD ITEMS

S.no	Food type	Aspergillus	Fusarium	Mucor	Cladosporium
1	Milk	5	6	0	0
2	Chutney	6	5	0	0
3	Apple	0	2	3	5
4	Spinach	0	5	4	0
5	Salad	4	3	0	0
6	Guava	4	3	0	0
7	Fish	6	0	0	0
8	Pea	0	4	0	0
Total		25	28	7	5

A total of 320 food samples were examined. For the evaluation of pathogenic fungi Milk, Chutney, Apple, Spinach, Salad, Guava, Fish and Pea were taken. All the others food samples were taken in polythene zip bag. The fruits were cut into small segments with a sterilized blade an d then plated on Sabouraud dextrose agar (SDA) aseptically and then incubated at 28ºC for 5 days. Culture was obtained and sub-culturing each of the different colonies that emerged onto the SDA plates and were incubated at 28ºC for 5 days. As a control, each of the healthy food samples was sterilized with 75% ethanol.

A. Description of the Study Area

A total of 320 randomly selected spoilt fruits and another 100 healthy looking fruits were examined. The fruits were cut into small segments (3 mm in diameter) with a sterilized blade, surface sterilized in 1% hypochlorite for 2 min, plated on Sabouraud dextrose agar (SDA) aseptically and then incubated at 28ºC for 5 days. A pure culture was obtained and maintained by sub-culturing each of the different colonies that emerged onto the SDA plates and were incubated at 28ºC for 5 days. As a control, each of the healthy fruits was sterilized with 75% ethanol. The fruits were cut into small segments (3 mm in diameter) with a sterile blade, placed on SDA and then incubated at 28ºC for 5 days

B. Identification Method for Fungi

Agar disc diffusion method was used for screening antifungal activities of each antibiotic. Yeast inoculums in 0.85% NaCl solution was spread on the surface of yeast extract-peptone-glycerol (YPG) agar plate. Sterile filter paper discs with 50 µg of Nystatin, 25 µg of Fluconazole, 1 µg of Voriconazole were used. Sterilized water was used as negative control.

III. Results

To find foodborne pathogenic fungi in various food items, a total of 320 were collected for evaluation. Out of the total food samples (65), 20.31% were found positive for fungal pathogens in various foodstuffs while 255 (79.68%) food samples were found negative (Table I) Among the positive food samples Fusarium spp were more predominant 28 (43%) leading by Aspergillus spp 25 (38.5%). Mucor and Cladosporium were found in low frequency with percentages of 7 (10.8%), and 5 (7.7%) (Figure 1).

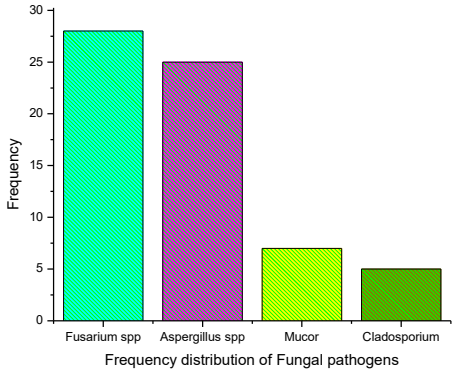

Fig 1. Frequency distribution of fungal pathogens isolated from various food items

For the evaluation of pathogenic fungi Milk, Chutney, Apple, Spinach, Salad, Guava, Fish and Pea were taken (Tab. 1,

Figure 2). The percentages of fungi were high in Milk leading by Chutney, Apple and Spinach. Intermediate frequency was found of Salad, Spinach, Guava and Fish while very low frequencies of fungal pathogen were found in Pea.

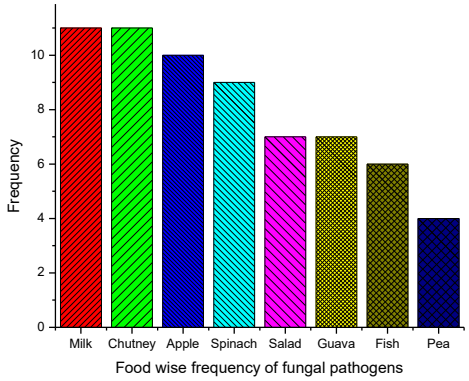

Fig 2. Food wise frequency of fungal pathogens in various food items

For antifungal activity a panel of selected drugs were used for the susceptibility of pathogens. Among the antifungal best activity were shows by Voriconazole and intermidiate activity were shows by Fluconazole. While high resistance were observed in Nystatin as compare to other antifungals (Figure 3)

IV. Discussion

Foodborne diseases are very easily transmitted from contaminated food and food handlers. Bacteria and fungi are the major factors for fruit spoilage. Food spoilage causes losses to producers, distributors, and consumers in the form of reduced quality and quantity and higher prices [1, 2]. Therefore the objective of this study was to isolate and evaluate the antifungal analysis of food-borne fungal pathogens. For the evaluation of pathogenic fungi Milk, Chutney, Apple, Spinach, Salad, Guava, Fish, and Pea were taken. The same work was conducted by [23] in Nigeria A total of 100 samples were taken results show that. the frequency of Aspergillus was found 38% high in pineapple, oranges, pawpaw, and tomatoes leading by Fusarium with the frequency of occurrence of 31% in fruits while Penicillium and Rhizopus found in low-frequency rate of 4% each food item respectively.

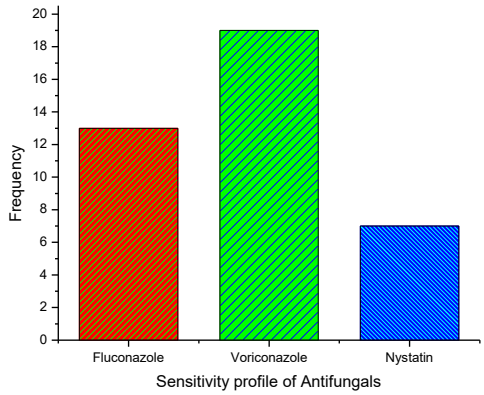

Fig 3. Sensitivity profile of commonly used Antifungals

Among the positive food samples, Fusarium spp were more predominant 28 (43%) leading by Aspergillus spp 25 (38.5%). While Mucor and Cladosporium were found in the low frequency with the percentage of 7 (10.8%), 5 (7.7%). The same research was also performed by [16, 23, 24] the most common fungal species were Aspergillus found in all fruits during storage of fruits. Other fungal pathogens Alternaria, Cladosporium, Fusarium, Mucor, Penicillium, and Rhizopus were common in fruits.

For antifungal activity, a panel of selected drugs was used for the susceptibility of pathogens. Among the antifungal best activity was shown by Voriconazole and intermediate activity was shown by Fluconazole. While high resistance was observed in Nystatin as compared to other antifungals. The same work was also conducted by [1] the isolates of the fungal pathogen were Aspergillus nigar in a high-frequency rate of (50%) led by Candida krusei the frequency rate of (19%) the low frequency of Fusarium exosporium (6%) and Mucor (25%). Antifungal analysis of fungal isolates had shown differential sensitivities toward Voriconazole. While Fluconazole and Nastatin have a high resistant rate.

V. Conclusions

Four Fungal species were successfully isolated in eight types of food samples and identified as Fusarium spp were more predominant 28(43%) led by Aspergillus spp 25(38.5%). While Mucor and Cladosporium were found in the low frequency with the percentages of 7 (10.8%), and 5 (7.7%). Further research work is required to explore more about the effect of these fungal species associated with food spoilage. Improved preservation methods, and handling, and recommended to enhance the quality of food for eating.

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