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# A SURVEY STUDY TO ISOLATE SOME PATHOGENIC BACTERIA FOR COOKED RICE AT BAGHDAD CITY

## N. A. Handhal\* and A. S. Ahmaed

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Department of Food Sciences, College of Agricultural Engineering Sciences, University of Baghdad, Iraq.

\* **Corresponding author:** N.A. Handhal, Department of Food Sciences, College of Agricultural Engineering Sciences, University of Baghdad, Iraq. Email: <u>nn9369942@gmail.com</u>

Abstract: The current study was designed to assess the microbial safety status of rice dishes offered for sale at various local and branded restaurants. Purposely, 30 samples of rice dishes were collected from 16 local and 14 branded restaurants in Baghdad city for two seasons, summer and winter. All the collected samples were subjected to micro-biological examination to determine the prevalence and comparative enumeration of the total count, Bacillus cereus, coliform, and forming spores in rice. Results pertaining to the enumeration of the total count, Bacillus cereus, coliform, and spores-forming bacteria exhibited the highest count Logarithmic: for summer collected from branded restaurants F.S (5.81), and local restaurants coliform (7.82). And winter from collected from branded restaurants T.C (7.81), and local restaurants coliform (4.44). The highest count observed Bacillus cereus in branded restaurants, and coliform in local restaurants. Further, the results of the current study revealed that 42% of rice samples collected from local restaurants and 58% from branded restaurants have unsatisfactory safety status for B. cereus. 38% of rice samples collected from local restaurants and 62% from branded restaurants have unsatisfactory safety status for forming spores. Similarly, for coliform 63 and 42% of samples were found unsatisfactory from local and branded restaurants, respectively.

Keywords: Bacillus Cereus, Coliform, Branded Restaurants, Local Restaurants.

#### 1. Introduction

The Height incidences of foodborne illnesses have led to an increased concern regarding food handling practices and the assessment of food safety measures taken during preparation of food items. Approximately, 76 million foodborne illnesses are recorded in the United States annually. This leads to an

expected rate of 325,000 hospitalizations along with 5000 annual deaths in the United States[1]. Such outbreaks mostly result from improper food handling and poor manufacturing practices. Malpractices are adopted at various stages of food supply chain such as preparation, processing, transportation, display, and serving [2]. Furthermore, outbreaks can have detrimental effects on country's national economy, either directly or indirectly, world Health Organization (WHO) has stated that foodborne outbreaks and illnesses have seriously affected the health and economic aspects of developed and developing countries and regarded as a constant threat [3].

With increasing population, the affinity for dine out places has been increased among all age groups and the food consumption in restaurants has seen a great shift in recent years [4].

According to the WHO, each year 600 million people around the world, or 1 out of 10, become ill after consuming contaminated food. Among all these people, 420,000 die, including 125,000 children under 5 years of age, due to the vulnerability of this population to develop a diarrheal syndrome, about 43% of FBDs occur in these patients [5]. Among the microorganisms causing Disease are bacteria that have different virulence factors that give them the ability to cause a disease among these factors, we can find toxins that can be produced in food or once the pathogen has colonized the digestive tract. About 70 % of Disease result from food contaminated with a microorganism [3]. It is to be noted that main sources of contamination in restaurants branded or and local restaurants [5], [6]. Safety of food is highly dependent on the way food is handled. In addition, it is stated that restaurants are more likely to be the cause for foodborne outbreaks as they cater to a larger number of consumers and, as a result, the chances of errors in processing food are wide-ranging [7]. People involved in the food handling to be aware of food safety measures and proper food handling practices [1].

Restaurant foods can contribute as one of the best breeding places of spoilage and pathogenic microbial strains, if not subjected to proper preparation and temperature conditions. The top five risk factors identified in the FDA Code 2004, responsible for foodborne illnesses that could result in diseases, hospitalizations and deaths, include: Food from unsafe sources, improper cooking, unclean utensils, inadequate holding temperature, and poor hygiene of the staff. FDA has highlighted these risk factors repeatedly in the FDA report on the occurrence of foodborne illness risk factors in selected institutional foodservice, restaurant, and retail food store facility types [8]. Presently, in Baghdad, gastroenteritis, and are accounted as major foodborne illness and restaurant foods are usually found contaminated by potential pathogenic microorganisms. Such organisms are Salmonella typhimurium, Campylobacter jejuni, Staphylococcus, Listeria monocytogenes, Bacillus cereus, Escherichia coli, and Enterobacteriaceae. These are considered as a major threat related to food [9]. Previous studies have focused on the microbial examination of different food items being offered for sale in different regions of the world. However, no research has been reported on the microbial safety of foods being offered at various restaurants of Baghdad.

Study was planned to assess the previously unreported safety status of commonly consumed food items (rice dishes) offered for sale at local and branded restaurants. During the study, Prevalence of total count, forming spores, B. cereus and E. coli (in cooked rice dishes), was calculated followed by determination of comparative enumeration and safety status.

#### 2. Materials and Methods

Sampling plan: Total 30restaurants (16 local and 14 branded) were selected for obtaining randomized and representative samples. The intention behind sampling plan was to cover various restaurants located all around the city of Baghdad.

Sample preparation: Rice samples were collected in sterilized polyethylene zip lock bags from various restaurants according to the sampling plan under controlled conditions in different time intervals and transported to the laboratory as soon as possible according to the method suggested by American Public Health Association [10]. For the identification and enumeration of pathogenic bacteria. Samples were kept in ice box during transportation to the laboratory. About 11 g of sample was blended in sterilizer peptone water at 99 ml.

Microbiological examination: The specific medium agar and the supplements were purchased from Oxoid, United Kingdom and prepared according to the manufacturer's directions. Nutrient Agar to calculate the total count and forming spore. Selective media Bacillus cereusagar base, with the addition of Polymyxin-B selective with addition egg yolkafter sterilization selective media. EMB agar were prepared for the isolation of coliform bacteria. The rice samples were examined for the total count and forming spore. B. cereus and coliform, by adopting spread plate method described by [11], [12] with slight modifications.

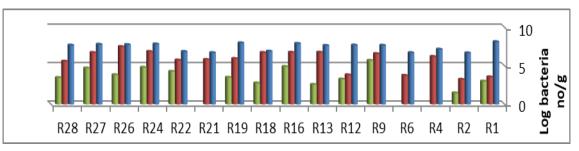
Incubation and Colony count: The petri dish was placed in an incubator at 37 C for 24–36 hr. Number of colonies were counted to know the final results afterwards.

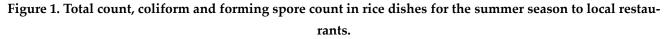
#### 3. Results and Discussion

Prevalence and enumeration of the total count, coliform and forming spore count in rice dishes for the summer season to local restaurants and branded restaurants.

As it is shown in Figure (1) and Figure (2) it is evident from the findings that among samples collected from local restaurant, highest count of  $98 \times 107$ cfu/g was noted whereas the lowest count recorded was  $71 \times 105$  cfu/g. On the other hand, the results of branded restaurants depicted the highest count of  $99 \times 107$  cfu/g while the lowest count observed was  $40 \times 105$  cfu/g. Conclusively, almost all the rice dishes procured from either local or branded restaurants of city of Baghdad have total count in range of 105-107cfu/g this results of the study matched with [13]. The preparation and storage practices of rice samples should be improved cooked in restaurants if the pollution exceeds the highest rate of the total for the possibility of satisfactory dangerous types. Local restaurants and branded restaurants rice samples was significantly high Under humid storage conditions for the summer season, the grains may deteriorate rapidly, resulting in qualitative and quantitative losses and this deterioration is accelerated [14]. Coliform count from samples collected from local restaurant, highest count of  $93 \times 107$ cfu/g was noted whereas the lowest count of  $149 \times 106$  cfu/g while the lowest count observed was  $62 \times 102$  cfu/g. Conclusively, almost all the rice dishes procured from either local or branded restaurants of city of Bagh-dat have coliform count in range of 102-107 cfu/g. Coliforms, can influence food safety and preservation because these organisms are an indicator of fecal contamination and can carry water borne pathogens [15].

Rice, may become contaminated during growth, harvesting and other agricultural operations such as processing and handling the coliforms, endospore-forming bacteria the most rice microflora. [13]. Bacteria forming spore in rice samples highest count of 73 × 104cfu/g was noted whereas the lowest count recorded was 47× 101 cfu/g. On the other hand, the results of branded restaurants depicted the highest count of 82 × 105 cfu/g while the lowest count observed was70 × 103 cfu/g. Conclusively, almost all the rice dishes from either local or branded restaurants of city of Baghdad have in range of 101–105 cfu/g.





Prevalence and enumeration of the B. cereusin rice dishes for the summer season to local restaurants and branded restaurants As it is shown in Figure (3) and Figure (4) the highest B. cereus count among samples collected from local restaurants was  $16 \times 104$  cfu/g, whereas  $27 \times 103$  cfu/g was the lowest count recorded. On the other hand, among the branded restaurants, B. cereus endospores are resistant to heat, radiation, disinfectants, and desiccation, the highest count of  $32 \times 104$  cfu/g was observed while the lowest count detected was  $93 \times 103$  cfu/g. Conclusively, all the rice dishes procured from either local or branded restaurants of Baghdad reported B. cereus count in range of 103-104 with an exception of 105.B. cereus is known to be a causative organism for a

wide range of opportunistic infections, both in immunocompromised and in immune competent patients, causing two distinct foodborne illness syndromes, namely diarrhea and emesis. Estimates of the infective dose of B. cereus in food poisoning vary from103 to 1010 CFU/g<sup>-1</sup> of food [16].

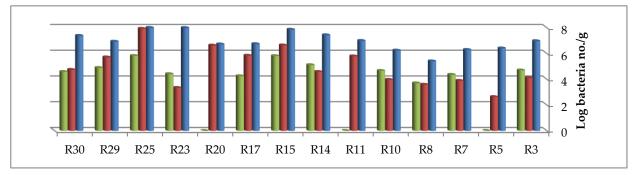


Figure 2. The total count, coliform and forming spore count in rice dishes for the summer season to branded restaurants.

Up to 104 CFUg<sup>-1</sup> in a typical serving of food is considered the upper limit to "acceptable" levels of contamination [17]. The current findings indicate that the rice dishes sold in branded restaurants are more contaminated with B. cereus as compared to the local restaurants. This may be explained by the unhygienic conditions during manufacturing, exposure during transport facility, and improper storage conditions. These results follow the same trend as reported by [18], [19].

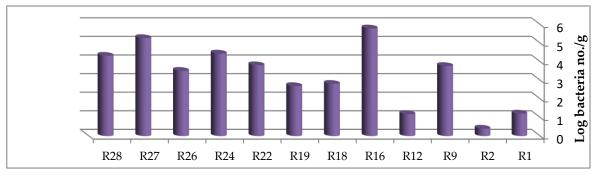


Figure 3. B. cereus in rice dishes for the summer season to local restaurants.

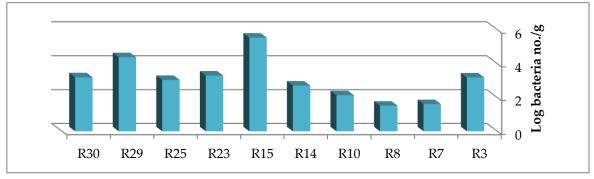


Figure 4. B. cereusin rice dishes for the summer season to branded restaurants.

Prevalence and enumeration of the total count, coliform count and forming spore count in rice dishes for the winter season to branded restaurants and local restaurants As shown in the figure (5) and (6) results vary bacterial growth for the winter season samples collected from local restaurants, highest count of  $48 \times 105$  cfu/g was noted whereas the lowest count recorded was  $31 \times 101$  cfu/g. On the other hand, the results of branded restaurants the highest count of  $91 \times 104$  cfu/g while the lowest count observed was  $14 \times 105$  cfu/g. total count in range of 101–105 cfu/g coliform count from samples collected from local restaurant, highest count of  $44 \times 104$ cfu/g was noted whereas the lowest count recorded was  $25 \times 101$  cfu/g. On the other hand, the results of branded restaurants depicted the highest count of 72 × 103 cfu/g while the lowest count observed was9 × 101 cfu/g. city of Baghdad have coliform count in range of 101–103 cfu/g. Low occurrence of contaminants on branded rice samples could be because the branded rice samples are treated properly [20]. Bacteria forming highest count of 47 × 103 cfu/g was noted whereas the lowest count recorded was 39 × 101 cfu/g. On the other hand, the results of branded restaurants depicted the highest count of 58 × 103 cfu/g while the lowest count observed was 69 × 101 cfu/g. range of 101–105 cfu/g. The microbiological quality of cooked rice in this study the presence of spores was Less than that found in the PHLS survey of cooked rice from Chinese and Indian establishments in England and Wales in 1976 [21]. The study found spores in rice samples from Indian than from Chinese establishments were of unacceptable microbiological quality in 1976 probably reflects poorer hygiene practices in these premises during handling and storage of the rice following cooking. Contamination may spores from rice raw [22].

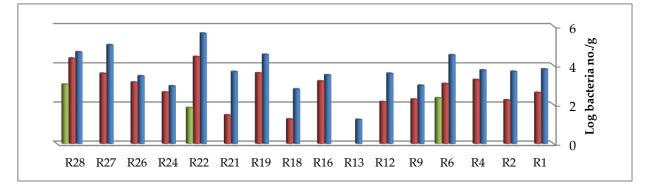
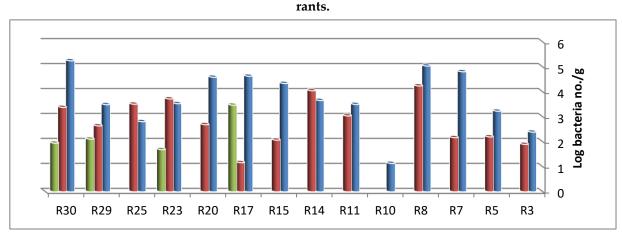
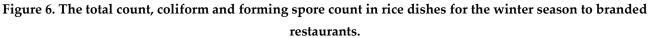


Figure 5. Total count, coliform and forming spore count in rice dishes for the winter season to local restau-





Prevalence and enumeration of the B. cereusin rice dishes for the winter season to local restaurants and branded restaurants As it is shown in Figure (7) and Figure (8) the highest B. cereus count among samples collected from local restaurants was 22× 101 cfu/g, whereas 4 × 101 cfu/g was the lowest count recorded, among the branded restaurants, the highest count of 39 × 101 cfu/g was observed while the lowest count detected was 3 × 101 cfu/g. from either local or branded restaurants of Baghdad reported B. cereus count in 101. The possible reason behind this practice could be the microbial proliferation which occurs when the rice dishes are stored overnight, waiting for its final cooking many hours later. Conversely, another study by [23]. B. cereus produce toxins, which are reported to cause gastrointestinal diseases [24]. Control measures should include proper storage practices to avoid cross contamination could reduce the number of B. cereus in the final product [17]. B. cereus grows and produces emetic toxins in a relatively short time on cooked rice and other starchy foods stored at room temperature [14].

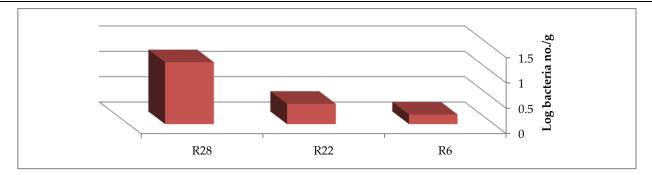


Figure 7. B. cereus in rice dishes for the winter season to local restaurants.

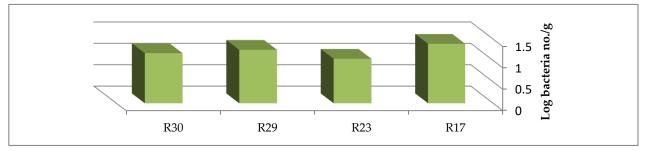


Figure 8. B. cereus in rice dishes for the winter season to branded restaurants.

## 4. Conclusion

In conclusion, the largest numbers of Bacillus cereus, coliform, and spore-forming bacteria were found in the total count. Logarithmic: collected for the summer from nationally recognized restaurants F.S. (5.81) and neighborhood eateries coliform (7.82). Winter data was gathered from nationally recognized restaurants T.C. (7.81) and coliform (4.44) as well as regional eateries. Most Bacillus cereus was found in branded restaurants and coliform in neighborhood eateries. The current study's findings also showed that 42% of rice samples taken from neighborhood restaurants and 58% from chain restaurants had unsatisfactory safety status for B. cereus. 38% of the rice samples gathered from neighborhood restaurants and 62% from restaurants with a name brand have an unacceptable safety status for spore formation. Similarly, to this, 63 and 42% of samples from local and branded restaurants, respectively, for coliform, were determined to be unsatisfactory.

# Supplementary Materials:

No Supplementary Materials.

## Author Contributions:

N. A. Handhal and A. S. Ahmaed; methodology, writing—original draft preparation, A. S. Ahmaed; writing—review and editing, N. A. Handhal; paraphrasing. All authors have read and agreed to the published version of the manuscript.

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The study was conducted in accordance with the protocol authorized by the University of Baghdad, Ethics Committee, Iraq.

## **Informed Consent Statement:**

No Informed Consent Statement.

## Data Availability Statement:

No Data Availability Statement.

## **Conflicts of Interest:**

The authors declare no conflict of interest.

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