



## Microbiological Profile of Food Served and Related Surfaces in a Libyan General Hospital, Libya

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**C**OMBATING foodborne pathogens in hospitals is a growing concern. A major aspect is a safe nutrition for patients in hospitals. It is still a hot topic concerning patients, media, and politicians. In Libya, it is a rising trend to inspect food and surfaces' safety aspects to assure hospital hygiene regulations. In this study, a survey of the general microbiological quality of food and surfaces in a General Teaching Hospital was undertaken in Alshatii, Libya. Fifty food and forty surface samples were cultured on different kinds of selective culture media. Significant colonies were counted and identified via microscopic and biochemical examinations. This study reported satisfactory levels of *Escherichia coli*, *Staphylococcus aureus*, *Enterobacter* spp., *Klebsiella* spp. and *Listeria* spp. when compared with guidelines for the published microbiological quality of ready-to-eat food. No detectable levels of *Salmonella* spp. were recorded in our study. We identified 9 out of 40 surface samples with unsatisfactory levels. *Listeria* spp. were isolated from eight surface samples that indicate improper hygiene compared with data on the general microbiological quality of surfaces. Food samples from the General Hospital in Libya was of relatively accepted microbiological quality but surface hygiene needs an improved strategy to prevent cross-contamination that causes public health problems. This inspection acts as a hygiene alert to improve the management of hospital cleaning and it will have a positive impact on the public health of Libyan society.

**Keywords:** Foodborne pathogens, Food Hygiene, HACCP, Libyan hospitals, Public health.

### Introduction

Microbial food safety is an instant public health concern worldwide in both developed and developing countries, despite the achievements in medical care, food science, and technology (Frank et al., 2011; Faour-Klingbeil & Todd, 2018; Ouf et al., 2019). World Health Organization (WHO) claimed an increase of foodborne diseases in both European and emerging countries by 38.3 and 915.8 in each hundred thousand of the population, respectively (Pereira et al., 2009; Braden & Tauxe, 2013; Ouf et al., 2019). In 2019, WHO proposed that each year about 600 million people get infected with diarrheal

diseases due to eating contaminated food all over the world (WHO, 2019). It was estimated that foodborne illness cases are high in the Eastern Mediterranean sub-region, and it is expected to increase with the population growth and the limitations of valuable resources (Faour-Klingbeil & Todd, 2018). Everyone is susceptible to foodborne diseases, but the immune-compromised hospitalized patients are particularly at risk of picking food-borne illnesses and suffer more serious consequences as a result of consuming contaminated food. Foodborne diseases were reported to highly affect children and the elderly than any other age group. Outbreaks of foodborne infection in healthcare settings can result in severe

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illness, wastage of medical treatments, the spread of infection between patients and staff members (Sockett & Rodgers, 2001).

Microorganisms, their associated toxins, and chemical contaminants are the major causes of foodborne diseases (Ingelfinger, 2008; Frank et al., 2011). Some of the foodborne pathogens responsible for foodborne diseases are *Staphylococcus aureus*, *Listeria* spp., *Salmonella* spp., *Bacillus* spp. and *Escherichia coli*. These pathogens can cause foodborne diseases in hospitals as well as the community as a whole (Havelaar et al., 2015). Foodborne infection outbreaks in hospitals can result in a serious illness. Risk factors include the nature of microorganisms, the quality of the food, the physiological state of the patient, contamination rate, insufficient cooking temperatures, inadequate sanitation, contaminated equipment, cross-contamination, and poor personal hygiene. These factors can lead to foodborne illness with serious health and economic consequences. The hospital food service program is classified to be the most complex production process within the hospitality sector. A balanced healthy diet must be served for patients in hospitals as the main treatment (Wilkinson et al., 1991; Edwards & Hartwell, 2006; Bannerman et al., 2008).

In (2006) the World Health Organization (WHO), highlighted the main contributors to foodborne illnesses including foods from unsafe sources, cross-contamination, poor personal hygiene among food handlers, inadequate cooking, and improper storage of the food (WHO, 2006). In (2008), WHO reported that it is important to handle food in such a way that the microorganisms present do not have a chance to multiply and to prevent food from becoming contaminated with other microorganisms. Significant statistical evidence reported that 70% of all cases of food poisoning are induced by food service staff (Griffith, 2000). Additionally, it was reported that surfaces act as reservoirs for microbes and could be able to transmit hospital pathogens. It also could increase the danger of cross-contamination through indirect contact with the patients (Otter et al., 2013). It is highly reported that assessments of microbial quality via the implementation of hazard analysis and critical control point principles (HACCP) lead to microbiological safety in hospitals and different settings (Griffith, 2000; Angelillo et al., 2001;

Moore & Griffith, 2002).

HACCP is recognized as an effective food safety assurance system. To achieve a successful HACCP system, training and constant supervision of employees is very important for producing safe foods (Moore & Griffith, 2002; El-Wehedy et al., 2019). Although the implementation of the HACCP system on hospital food service is still voluntary in most countries, several hospitals have adopted these principles to ensure that hospital food is safe for consumption by high-risk patients (Angelillo et al., 2001; Moore & Griffith, 2002). Our study is based on past epidemiological evidence reporting global outbreaks, general trends in foodborne disease, and local pilot surveys in Libya. The lack of data is the main reason for underreporting and the consequent inaccurate estimation of the burden associated with foodborne diseases especially in hospitals. According to Faour-Klingbeil, only food poisoning cases were recorded from 2001-2004, Food and Agriculture Organization report in Libya (FAO/WHO, 2005; Faour-Klingbeil & Todd, 2018). The recent study defines the hygienic and microbiological quality of the food and surfaces in Libya's General Hospital. It also has a positive impact on the strengthening of the general concept of health services and protection in all hospitals, and hence on the public health of Libyan society and internationally.

## **Materials and Methods**

### *Study frame and sampling protocol*

In this study 50 food samples and 40 surface samples were collected from a Libyan General Hospital. The frequency of food sampling was three times a month and to guarantee the result's reliability, samples were collected aseptically in hygienic conditions by the same person, for every sample of food and surfaces. Hence, food samples were collected in sterilized bags, using a sterilized spoon. Surface sampling was performed by a moist sterile swab with a sterile isotonic liquid. Samples were taken by friction from studied surfaces of locals, equipment, and staff's hands. Then, samples were transported to the laboratory in a cooled icebox maintained at 4°C.

### *Food sampling*

Fifty food samples were collected from the kitchen of the hospital just before loading trays to their destination in the hospital wards. Foods served were rice, pasta, cuscus based on chicken

soup and salad. About 25 g of different foods were collected in sterile containers labelled with the type of sample, date, temperature, time of collection, and name of the hospital kitchen. Samples were transported immediately in an icebox to the microbiology laboratory for processing (Meldrum et al., 2009). Samples were homogenized in a stomacher in peptone water (total volume 100mL) and incubated at 37°C for 30min. The suspension was then centrifuged and 10 fold dilutions of the supernatant from 10<sup>-1</sup> to 10<sup>-4</sup> were made.

#### *Surfaces sampling*

Forty samples were taken on the same time of food sampling from the hands of food handlers as well as from the surfaces to which the food is exposed (the kitchen pans, meat cutting boards and knives, salad cutting boards and knives, boxes used for delivering the food, plates where the food is served). According to their nature, there were twenty samples of local surfaces (vegetables and fruits preparation rooms, staff's dining rooms, etc.), ten samples of materials (patients trays, milk thermos, water tanks, and glasses, etc.), and ten of the kitchen staff hands. Ethical considerations were conducted before taking samples from the chef and other assistants. The temperature of each surface sample was recorded and swabs samples from surfaces were released in 1mL of sterilized physiological water. Then, this bacterial suspension served as a mother solution to inoculate the same culture media used for microbiological analysis.

#### *Determination of the total bacterial count*

An aliquot of 1mL of each dilution was pour plated onto a nutrient agar medium (Oxoid Ltd.) The plates were incubated at 25°C for 24 to 48hrs. Plates were examined after 48hrs, each colony was counted, and the total number of colonies was multiplied by the corresponding dilution to get the total number of bacterial colonies per gram product. Values obtained were compared to guidelines of food microbiological quality and food surface hygiene standards (Gilbert et al., 2000; Agency, 2009; Osimani et al., 2014).

#### *Isolation of bacterial contaminants on selective media*

The following bacterial identification protocol was based on a modified method of the Health Protection Agency (HPA) General Standard Method F20. Samples were routinely related

to guidelines of food microbiological quality (Agency, 2009), and surface hygiene guidelines (Gandhi Joan Webster, 2013).

#### *Staphylococcus aureus*

A sample of 0.1mL of the 10<sup>-1</sup> dilution was inoculated onto a Baird Parker agar plate (Oxoid Ltd.) and spread via a sterile plastic spreader (Agency, 2009). The plate was incubated at 37°C for 24 to 48hrs. After incubation, the plate was examined for *S. aureus* colonies that appear as shiny, black, convex colonies with a zone of opacity surrounded by a clear zone.

#### *Listeria species*

A sample of 0.5mL of the 10<sup>-1</sup> dilution was inoculated and spread using a sterile plastic spreader onto a Listeria selective agar plate (OxoidLtd.) (Agency, 2009). The plates were incubated at 30°C for 48hrs.

#### *Bacillus cereus*

A sample of 1ml of the 10<sup>-1</sup> dilution was inoculated onto a *B. cereus* selective agar plate (Oxoid Ltd.) and spread using a sterile plastic spreader (Agency, 2009). The plates were incubated at 30°C for 24 to 48hrs. The plates were checked after 18 to 24hrs of incubation, and typical *Bacillus* spp. colonies were picked for further analysis. Confirmation of *B. cereus* was made with the BBL Crystal GP system (Becton Dickinson Co., Shannon, Ireland), following the manufacturer's instructions.

#### *Escherichia coli*

A sample of 1mL of the 10<sup>-1</sup> dilution was inoculated onto a desoxycholate agar plate (Oxoid Ltd.), spread via a sterile plastic spreader, and incubated at 30°C for 48hrs (Agency, 2009). The chromogenic substrate in the agar (5-Bromo-4chloro-3indolyl--D-glucuronic acid) detected the glucuronidase reaction of *E. coli*, resulting in blue colonies.

#### *Salmonella species*

A sample of 0.1mL of the buffered peptone water was inoculated into 10mL of Rappaport Vassiliadis broth and incubated at 44°C for 24hrs (Agency, 2009). The Rappaport Vassiliadis broth was then subcultured onto xylose lysine desoxycholate agar and incubated at 37°C for 24hrs. These plates were then observed for typical *Salmonella* spp. colonies.

### Coliforms

A sample of 1mL of the  $10^{-1}$  dilution was inoculated onto a violet red bile lactose agar plate (Oxoid Ltd.) (Agency, 2009). The medium contains bile salts and crystal violet which inhibit some Gram-positive bacteria, mainly staphylococci. The inoculum was spread using a sterile plastic spreader and incubated at 30°C for 48hrs. Neutral red is used as the pH indicator. Lactose-fermenting bacteria produce pink to red colonies that are generally surrounded by a reddish zone of precipitated bile. Non-lactose-fermenting bacteria result in colorless colonies (Moore & Griffith, 2002).

### Microscopic and biochemical identification of bacterial contaminants

The bacterial colonies isolated on different kinds of selective media were subjected to the purification step. Preliminary identification of isolates was performed using Gram staining, coagulase test, catalase test, indole production test, methyl red test, Voges-Proskauer test, urease test, citrate utilization test, and sugar fermentation test (glucose, lactose, rhamnose, and fructose), and oxidase test according to Bergey's Manual of Systematic Bacteriology (Holt & Williams, 1989; Holt et al., 1994; Harrigan, 1998). In the case of *L. monocytogenes*, the API Listeria system (BioMerieux, La Balme-Les-Grottes, France) was used as a confirmatory test (Setiani et al., 2015). Additionally, the identification of Gram-negative bacterial isolates was confirmed by the API 20E kit (BioMerieux, La Balme-Les-Grottes, France) following the manufacturer's instructions. An aliquot of 0.1mL of a suspension of each isolate was inoculated in each microtubule of the APIkit strip and incubated for 24- 48hrs at 37°C. After incubation, the result of each reaction was identified using the Catalogue Analytique.

### Statistical analysis

Descriptive and statistical analysis of the data was undertaken by using Microsoft Excel and Minitab Info version 16.1. Relative proportions were compared by using the Chi-Squared test ( $\chi^2$ ) and One Way Anova, Two Way Anova, T- Value tests. A probability value of less than 5% was defined as significant (Sokal, 1995).

## Results

### The temperatures of food and surface samples

The temperature of food samples, before serving (BS) and after serving (AS) was measured.

The highest temperature was recorded on the second and third days. It was 26°C before food serving and 25°C and 24°C after food serving, respectively. While, the lowest temperature of food before serving was 21°C and after serving was 20°C on the seventh, eighth, and ninth days (Fig. 1). The temperature decreases one or two degrees between the sampling before and after food serving. No significant statistical difference was recorded between them ( $P > 0.05$ ). By measuring the temperature of surface samples, it was found that the temperature pattern differed on the five days from 18°C to 26°C (Fig. 1).

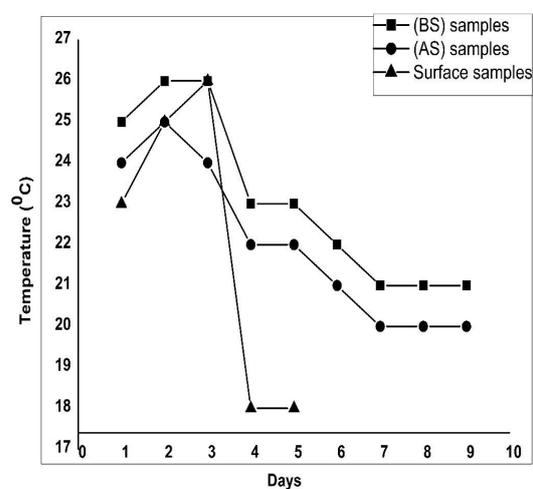


Fig. 1. The temperature pattern of food and surface samples [T Value= -1.17, P Value= 0.259].

### Detection of total bacterial count isolated from food samples

Our results showed that fifty food samples retrieved from the hospital kitchen showed variable total bacterial count. Samples number (34 and 44) recorded the highest microbial count with  $1056 \times 10^4$  CFU in 25g of food. On the other hand, the lowest microbial count was recorded in sample number (14) with  $4 \times 10^4$  CFU in 25g of food as shown in Table 1.

### Identification of key pathogens isolated from food samples

Preliminary identification of bacterial colonies was based on their isolation on selective media. Further microscopic and biochemical identification was conducted according to Bergey's Manual of Systematic Bacteriology (Table 2 and Supple. Fig. 1, 2).

TABLE 1. The total bacterial count (CFU) isolated from food samples ( $\bar{X}$ = 28.06, P value= 0.000).

Number of food samples	CFU/25g of food sample X10 <sup>4</sup>	Number of food samples	CFU /25g of food sample X10 <sup>4</sup>
1	28	26	180
2	216	27	20
3	360	28	799
4	450	29	211
5	180	30	1000
6	200	31	620
7	868	32	560
8	732	33	692
9	520	34	1056
10	72	35	496
11	580	36	40
12	368	37	976
13	856	38	536
14	5	39	976
15	769	40	536
16	680	41	976
17	70	42	540
18	33	43	752
19	21	44	1056
20	808	45	312
21	105	46	200
22	960	47	240
23	190	48	192
24	219	49	520
25	60	50	364

TABLE 2. Morphological and biochemical features of *S. aureus*, *L. monocytogenes*, and *B. cereus* isolated from food and surface samples.

Biochemical test	<i>S. aureus</i>	<i>L. monocytogenes</i>	<i>B. cereus</i>
Growth on selective media	Baird Parker agar	Listeria selective agar	<i>B. cereus</i> selective agar
Gram staining	Gram-positive cocci	Gram-positive rods	Gram-positive rods
Oxidase test	-	-	-
Urease test	+	-	-
Voges-Proskauer test	+	+	+
Indole production test	-	-	-
Methyl red test	+	+	-
Citrate utilization test	+	-	+
Catalase test	+	+	+
Sugar fermentation tests			
Glucose	+		+
Lactose	+	+	
Rhamnose	+	+	
Fructose	+		+

\* (+): Indicates positive results, (-): Indicates negative results.

Some key foodborne pathogens were recorded in food samples in our study including *L. monocytogenes*, *Enterobacter* spp., *Klebsiella* spp., *E. coli*, *S. aureus*, and *B. cereus*. Fluctuation in the number of colonies of all bacterial types was recorded, however, an observable increase was detected in (AS) food samples. Food samples before serving showed no colonies of *L. monocytogenes*. However, it was found in two (AS) food samples on the third day and the tenth day with (1) and (5) CFU per gram, respectively. Similarly, *Enterobacter* spp. wasn't recorded in (BS) food samples, while it was recorded in six (AS) food samples. Additionally, *Klebsiella* spp. was recorded in one (BS) sample only, while it was detected in six (AS) samples. Furthermore, *E. coli* and *S. aureus* were isolated from two (BS) food samples, however, they were isolated from five and three (AS) food samples, respectively. On the fifth day, only one sample contained *B. cereus* in both (BS) and (AS) food samples. The highest count of foodborne pathogens was recorded in (AS) food sample on the fourth day including *Enterobacter* spp., *Klebsiella* spp., *E. coli* with (31, 31, and 22) CFU per 25g, respectively. No colonies were detected in (BS) and (AS) food samples examined on the second day. Statistical analysis showed the presence of significant differences between the isolated numbers of foodborne pathogens isolated from (BS) and (AS) food samples as shown in Table 3.

#### Identification of microbial count isolated from

**TABLE 3. The CFU numbers of bacterial species isolated from food samples before and after serving in the Libyan hospital.**

Key pathogens	<i>L. monocytogenes</i>		<i>Enterobacter</i> spp.		<i>Klebsiella</i> spp.		<i>E. coli</i>		<i>S. aureus</i>		<i>B. cereus</i>	
	(BS)	(AS)	(BS)	(AS)	(BS)	(AS)	(BS)	(AS)	(BS)	(AS)	(BS)	(AS)
No. of food samples on daily basis												
1	0	0	0	3	0	1	0	3	0	0	0	0
2	0	0	0	0	0	0	0	0	0	0	0	0
3	0	1	0	2	0	0	0	3	0	8	0	0
4	0	0	0	31	0	31	0	22	0	1	0	0
5	0	0	0	0	0	0	0	0	0	0	8	8
6	0	0	0	1	0	1	0	0	0	11	0	0
7	0	0	0	3	0	0	15	0	0	0	0	0
8	0	0	0	5	4	5	0	5	0	0	0	0
9	0	0	0	0	0	2	19	15	1	0	0	0
10	0	5	0	0	0	9	0	0	6	0	0	0

\*(BS): Before serving food samples, (AS): After serving food samples, (N.A): Not applicable, No. of colonies (CFU) was recorded in (25g) of all food samples.

#### surface samples

Our results presented the total bacterial count in forty surface samples isolated from internal and external rooms of the hospital kitchen. The number of foodborne pathogens was variable in surface samples. The highest microbial count was recorded in surface samples isolated from egg pan and tomato fruit bowls with 101 and 135 CFU in 100cm<sup>2</sup> respectively, followed by vegetables and meat cutting boards which showed a high number of microbial types with 90 CFU in 100cm<sup>2</sup>. Alternatively, no bacterial colonies were isolated from the oven surface and dishwashing sponge samples. Generally, the total number of bacterial colonies retrieved on enrichment microbial cultures ranged from 0 -135 CFU in 100cm<sup>2</sup> as shown in Table 4.

#### Identification of key foodborne pathogens isolated from surface samples

In our study, no *E. coli* colonies were recorded in all surface samples, while some coliforms were recorded including *Enterobacter* spp. and *Klebsiella* spp. *Enterobacter* spp. were detected in the outer washing basin with (2) CFU per 100cm<sup>2</sup>. Alternatively, *Klebsiella* spp. were recorded in 8 different surface samples. Egg pan recorded the uppermost *Klebsiella* spp. colony count with (70) CFU per 100cm<sup>2</sup>. On the other hand, surfaces such as meat cutting board, internal sink basin, a tray of cooking tools, and tap handle showed the lowest number with 1-2 CFU per 100cm<sup>2</sup>, as shown in Table 5.

**TABLE 4. The number of bacterial colonies isolated from the internal and external surfaces of the hospital kitchen.**

Surface sample types (100cm <sup>2</sup> )	Number of colonies (CFU)
Inner marble	14
Cutting knife	26
External marble	61
Hand of the chef's assistant	16
Vegetables cutting board	60
Cooking gas surface	15
Chicken frying pan	19
Internal sink basin	10
Tap handle	36
Food dish	43
Handle of the carriage	45
Food collection vessels	2
Food scoop	60
External marble No. 2	30
Meat cutting board	90
External sink surface	62
Hand of the chef	54
Egg pan	101
Tomato fruit bowls	135
Wall	53
Oven surface	ND
Dishwashing sponge	ND
Pot of cooking water	1
Chicken tweezers	3
External wash-faucet handle	60
Refrigerator handle	5
Food scoop No. (2)	26
Cooking vessels	2
Water cup	12
Plastic spoon	24
Collection bowl	45
Teapot nozzle	21
Knife sharpener	6
Tray of cooking tools	5
Chicken slicing knife	19
Refinery	4
Oven handle	10
Plastic plates	1
Outer washing basin	22
Potato slicing knife	14

\*ND: Not detected

*Listeria* spp. colonies were isolated from surface samples of the internal and external rooms of the hospital's kitchen with a percentage (25%) of all surface samples. *Listeria* spp. total count varied in different samples from 1- 90 CFU per 100cm<sup>2</sup>. Tomato fruit bowls recorded the topmost *Listeria* spp. colony count with 90 CFU per 100cm<sup>2</sup>. Conversely, vegetable cutting board and potato slicing knife surfaces displayed the lowermost number with 1 CFU per 100cm<sup>2</sup>, as shown in Table 6. The guidance for understanding the safety measures of microbial species isolated from ready-to-eat foods and surfaces is shown in Tables 7, 8 (Agency, 2009; Gandhi Joan Webster, 2013).

### Discussion

Diseases caused by foodborne pathogens create a massive public health problem, prevention of food contamination by these pathogens at all phases is an important goal for all officials involved in food safety ensurance (Jahid & Ha, 2012; Braden & Tauxe, 2013). The microbiological analysis outcomes are attributed to the guidelines released in 2009 by the health protection agency (Agency, 2009). Those guidelines established legislation to judge the relative quality of the sampled food for local authorities and public health bodies. Those rules identify types including satisfactory, acceptable, and, unsatisfactory. An acceptable category is used as an index showing suitable limits of microbiological quality. Unsatisfactory category means that further sampling of the infected sites is required, and if the result is unacceptable it indicates that crucial action is needed (Agency, 2009).

In our study, it was found that the temperature of the food samples varied considerably between the kitchen and the area of the patient with insignificant statistical differences ( $P > 0.05$ ). We noticed that lower temperature of food in the kitchen and after food serving can be a reason for food contamination. On the other hand, the temperature decreases one or two degrees between the sampling before and after food serving because the food delivery had been late to patients. We expect that higher temperatures in summer will induce microbial growth and enhance the replication cycles of most foodborne pathogens (Semenza et al., 2012).

**TABLE 5. The prevalence of coliforms colonies isolated from surface samples of the internal and external rooms of the hospital's kitchen.**

Surface sample types (100 cm <sup>2</sup> )	Number of colonies of coliforms(CFU)		
	<i>E. coli</i>	<i>Klebsiella</i> spp.	<i>Enterobacter</i> spp.
Internal sink basin	0	4	0
Tap handle	0	1	0
Meat cutting board	0	2	0
Internal sink basin	0	2	0
Egg pan	0	70	0
Tomato fruit bags	0	3	0
Tray of cooking tools	0	2	0
Oven handle	0	7	0
Outer washing basin	0	0	2

**TABLE 6. The prevalence of *Listeria* spp. colonies isolated from surface samples of the internal and external rooms of the hospital's kitchen.**

Surface sample types (100cm <sup>2</sup> )	The number of colonies of <i>Listeria</i> spp. (CFU)
External marble No. 1	3
Cutting knife	12
Vegetable cutting board	1
Cooking gas surface	6
Chicken frying pan	10
Tomato fruit bowls	90
Knife sharpener	5
Chicken slicing knife	2
Outer washing basin	3
Potato slicing knife	1

**TABLE 7. Guidance on the understanding of findings in ready-to-eat foods for hygiene species in general.**

Condition	Colony-forming unit (CFU)/g		
	Satisfactory	Unacceptable risk	Unsatisfactory
<i>Salmonella</i> spp.	N.D in 25g	N/A	Detected in 25g
<i>Listeria monocytogenes</i> in other ready-to-eat food	< 10	10 - < 100	> 100
<i>Staphylococcus aureus</i>	< 20	20 - <10 <sup>4</sup>	> 10 <sup>4</sup>
<i>Bacillus cereus</i>	< 10 <sup>3</sup>	10 <sup>3</sup> - < 10 <sup>5</sup>	> 10 <sup>5</sup>
<i>Escherichia coli</i>	< 20	20- < 10 <sup>2</sup>	> 10 <sup>2</sup>
<i>Enterobacteriaceae</i>	< 10 <sup>2</sup>	10 <sup>2</sup> - < 10 <sup>4</sup>	> 10 <sup>4</sup>

ND\* : Not detected, N/A: Not applicable.

**TABLE 8. Guidance for understanding the count of microbial species isolated from surfaces of internal and external rooms of the hospital's kitchen.**

Type	Criteria	Borderline
	Total number of microbes	100/ 50cm <sup>2</sup>
Surface samples	Coliforms	10/ 50cm <sup>2</sup>
	<i>Listeria</i> spp.	0

It has been stated that the optimum temperature for keeping hot foods is likely 55°C in combination with a maximum holding period, which can be sufficient to prevent significant foodborne pathogens (ESR & Olsen, 2011).

From the results of our study, it was found that the total bacterial count of all food samples is acceptable when compared to guidelines of food microbiological quality and food surface hygiene standards. As all recorded numbers were found within the permissible limits between  $10^3$ - $10^5$  per gram as shown in Table 7 (CFS, 2014). Additionally, it showed that the numbers of foodborne pathogens recovered from (AS) food samples were higher than that of (BS) samples. There were statistically significant differences between the microbial load isolated from (BS) and (AS) food samples ( $P < 0.05$ ), and this indicates an increase in food contamination in patient's area than in the food preparation area. However, all foodborne pathogens in food samples were at the accepted level when compared to guidelines of food microbiological quality and food surfaces hygiene standards (CFS, 2014). We suggested that this may be due to contamination of food with airborne bacteria or bacteria transferred from patients by coughing, sneezing, or mouth infection, or it could be transmitted via workers and nurses. Similar reports mentioned that untrained employers with poor hand hygiene and inadequate glove use by the food handlers could lead to serious food poisoning (Ayçiçek et al., 2004). In addition to the low temperature of the food served as previously mentioned can be a probable cause of food contamination (ESR & Olsen, 2011).

Microbiological analysis indicated the presence of *S. aureus* in five food samples. This may be due to the contamination by food staff workers in the kitchen as normal flora on the skin, nose, and throat (Williams, 1963). Some reports declared that several outbreaks of foodborne *S. aureus* disease can be due to colonized personnel as the source of food contamination (Ferreira et al., 2014; Nasrolahei et al., 2017). In 2017, an Iranian study showed an annual checkup of nasal and fingernail content swaps of food handlers were found to harbor *S. aureus* (Nasrolahei et al., 2017). However, the recorded numbers of *S. aureus* were found within the permissible limits  $< 20$  CFU per gram (Agency, 2009). The results of the microbial analysis of food samples showed

the presence of *E. coli*, *L. monocytogenes*, *Enterobacter* spp., and *Klebsiella* spp. within the permissible limits of ready-to-eat foods (CFS, 2014). We suggest that these species enter the kitchen on raw meat, poultry, and other foodstuffs, then subsequently contaminate both raw and cooked foods after cooking (Cooke et al., 1970; Adeyanju & Ishola, 2014). Additionally, it may be due to the lack of personal hygiene of the kitchen staff as *E. coli* bacteria naturally occur in the lower part of the digestive system of human and warm-blooded animals (Shooter et al., 1970). During our study, we did not find *Salmonella* spp. bacteria in food and surface samples which is a good sign as its permissible limits are 0/g (Agency, 2009).

In this study, we identified two unaccepted surface samples (tomato fruit bowls and egg pan) due to the increase of the total bacterial count than permissible hygiene limits reported in Table 8 (Gandhi Joan Webster, 2013). Egg pan recorded 70 CFU/ 50cm<sup>2</sup> of *Klebsiella* spp., which exceeds the permissible limits of surface hygiene. The isolation of *Klebsiella* spp. from food processing environments has been reported. Consequently, it can be relocated from surfaces to food and can cause adverse effects on food quality (Møretro & Langsrud, 2017). Regrettably, we found that *Listeria* spp. in 8 surface samples, with the highest colony count in tomato fruits, bowls with (90 colony / 100cm<sup>2</sup>), and thus we propose that this is a very dangerous indicator of pollution on the food surfaces because the satisfactory sanitization limit is 0 CFU/ 50cm<sup>2</sup> (Gandhi Joan Webster, 2013). We expect that salads may be consequently contaminated causing serious illness of the patients (Shooter et al., 1970).

It would seem appropriate to infer from the findings of this food and surface survey at the General Hospital in Libya that food samples were generally of good microbiological quality. All of the food samples taken are sufficient, however, nine surface samples still contain pathogenic organisms at unacceptable levels and may pose potential hazards to consumers. This study highlights the reduction of *Listeria* spp. in food-related surfaces to prevent cross-contamination with high-risk foods consumed without heat treatment. Monitoring of food safety is an important task that must continue to reduce the current *Listeria* spp. occurrence in surface samples served in the general hospital,

which could adversely affect consumer health. This survey was a snapshot of a few days, so we recommended periodic sampling and testing of the food environments in the hospital to maintain microbiological quality and identify any possible patterns of development.

We recommend the implementation of the HACCP approach in the food policy of the General Libyan Hospital. The HACCP methodology is based on the use of multifunctional schemes involving the use of sanitizers and the latest techniques for disinfection to decrease the prevalence of *Listeria* spp. and *Klebsiella* spp. Many reports shed light on the application of HACCP strategies (Moore & Griffith, 2002; El-Wehedy et al., 2019). Kitchen staff in hospitals is considered a possible source of foodborne outbreaks and could introduce pathogens into foods during each step from cooking to serving (Maguire et al., 2000). We also endorse efficient personal hygiene to decrease the the colonization of *S. aureus* on staff hands by washing hands properly and wearing appropriate clothing such as hand gloves, head covers, and nose masks. Additionally, staff members should be employed after sufficient hygiene control training and they must submit with all hygiene measures to prevent food contamination. To minimize the risk of food-borne pathogens and the consequent food poisoning of customers, skilled food handlers, and food regulators must carry out regular checks on the packaging, storage, and final preparatory food stages. Additionally, cooking at high temperatures of 100°C helps killing pathogens before consumption and shortens the duration of serving (Braden & Tauxe, 2013).

### **Conclusion**

Avoidance of foodborne infection in healthcare settings is crucial. Prohibition of many foodborne outbreaks in healthcare sites could be done by following proper HACCP principles and good hygienic practice. So, the implementation of HACCP rules and similar food safety management plans in food preparation sites will provide a good hygiene practice. Appropriate supervision and food hygiene training for all staff is a legal obligation. Additionally, food safety policy should include consultants in communicable disease control, control infection officers, and catering management. Following the previously mentioned aspects will decrease the possibility

of foodborne diseases in Libyan hospitals and protect high-risk patients.

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### الكشف الميكروبيولوجي للأغذية المقدمة والأسطح المتصلة في مستشفى ليبيا العام ، ليبيا

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تعتبر مكافحة مسببات الأمراض المنقولة بالغذاء في المستشفيات مصدر قلق متزايد. جانب رئيسي هو التغذية الآمنة للمرضى في المستشفيات والذي لا يزال موضوعاً ساخناً يتعلق بالمرضى ووسائل الإعلام والسياسيين في ليبيا، هناك اتجاه متزايد لفحص جوانب سلامة الأغذية والأسطح لضمان لوائح النظافة في المستشفيات. هذه الدراسة تشمل إجراء مسح للجودة الميكروبيولوجية العامة للأغذية والأسطح في مستشفى تعليمي عام في الشاطئ ، ليبيا. حيث تم فحص خمسين عينة طعام وأربعين عينة سطحية على أنواع مختلفة من وسط غذائي انتقائي لعزل البكتيريا. تم إحصاء وتحديد المستعمرات الهامة عن طريق الفحوصات الميكروسكوبية والكيميائية الحيوية. أبلغنا عن مستويات مرضية من الإشريكية القولونية، المكورات العنقودية الذهبية، البكتيريا المعوية، كليبسيلا وليستريا عند مقارنتها بالإرشادات الخاصة بالجودة الميكروبيولوجية المنشورة للأغذية الجاهزة للأكل. لا توجد مستويات يمكن اكتشافها من بكتيريا السالمونيلا تم تسجيلها في دراستنا. لسوء الحظ، حددنا 9 من 40 عينة سطحية بمستويات غير مرضية. الليستيريا مونوسيتوجيناس تم عزلها من ثماني عينات سطحية تشير إلى سوء النظافة عند مقارنتها ببيانات الجودة الميكروبيولوجية العامة للأسطح. كانت عينات الطعام المأخوذة من المستشفى العام في ليبيا ذات جودة ميكروبيولوجية مقبولة نسبياً، لكن النظافة السطحية تحتاج إلى استراتيجية محسنة لمنع انتقال التلوث، وبالتالي التسبب في مشاكل صحية عامة. هذا التفتيش بمثابة تنبيه صحي يحسن إدارة تنظيف المستشفيات وسيكون له تأثير إيجابي على الصحة العامة للمجتمع الليبي.