

## Microbial Spoilage of Some Meat Luncheon Samples Collected from Damietta City, Egypt

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### ABSTRACT

The microbiological examination of local meat luncheon samples obtained from local market of Damietta governorate was studied. These samples were belonging to two different companies in Egypt. Bacterial counts of luncheon samples were 2500 and  $310(\times 10^2 \text{ cfu/g})$ ; *Bacillus cerues* were 470 and  $30(\times 10^2 \text{ cfu/g})$ ; Staphylococci counts were 1<sup>vo</sup> and  $30(\times 10^2 \text{ cfu/g})$ ; fungi counts were 30 and  $1(\times 10^2 \text{ cfu/g})$  in company A and B, respectively. The high level of *Aeromonas* sp. in luncheon was in company A, while the lowest value was in company B. In company B, coliform bacterial count and anaerobic bacterial count were 10 and  $450(\times 10^2 \text{ cfu/g})$ , respectively, and they not detected in company A. *Listeria* sp.; *Salmonella* and *Shigella* spp. were not detected in both company. Total bacterial count, Staphylococci count and anaerobic bacterial count were higher than the permissible limit of the Egyptian Organization for Standardization and Quality (EOSQ) of Luncheon No 1114. On the other hand, *Salmonella* and *Shigella* and coliform count were in the permissible limit of the EOSQ. Fourteen microbial isolates were obtained from Luncheon samples, six isolates from company A and eight isolates from another company B. Three isolates obtained for each Nutrient agar and PDA media. Two isolates obtained for each selective agar base; Baird-Parker agar and m-*Aeromonas* selective agar media. One isolate obtained form EMB agar and cooked meat Media. No isolates obtained from *Listeria* agar base and SS agar. Among of eleven bacterial isolates, six isolates were long rods, three isolates were short rods and two isolates were coccoid shaped cells. Six isolates were Gram positive and five isolates were Gram negative. Seven isolates were non spore forming bacteria and four isolates were spore formers. All obtained isolates gave negative results in acid fast stain. Moreover, *Rhizopus oryzae*, *Penicillium aurantiogriseum* and *Cladosporium herbarum* were isolated and identified from Luncheon samples.

**Keywords:** Luncheon, Microbiological evaluation, *Penicillium aurantiogriseum* and *Cladosporium herbarum*

### INTRODUCTION

Meat luncheon generally containing finely chopped meat and fat with or without some added grains treated with spices, salt, nitrite and heat processed. Luncheon has a high spread in the world due to their high nutritional value, acceptable price, agreeable taste and easy during eating (Mahmoud *et al.*, 2016). Meat luncheon is one of the most acceptable food products, widely consumed and used for fast meats. It is considered a quick easily prepared meat meals and solve the problem of fresh and expensive meat shortage, which is not within the income reach of many families with limited income in Egypt (Shawish and Al-Humam, 2016). The Ready-to eat meat such Luncheon is considered a high risk food group, because it is often consumed without a cooking step. Luncheon is traditionally produced as industrially vacuum-packaged loaves, and afterwards is sliced and repackaged at retail stores, therefore the product may be exposed to the contamination hazard at any time (Ehrampoush *et al.*, 2017). The increasing of contamination in food industry especially meat products such luncheon by pathogenic bacteria has raised a great concern of the public (Abd-El-Malek *et al.*, 2010). So that, food poisoning outbreaks are widespread in all regions of the world. Inspection is not available in developing countries but the information indicates that the prevalence is higher than that in developed countries (Tewari *et al.*, 2015). Thus, there is a need to detect these serious food poisoning bacteria in meat products such luncheon to limit their effect. Up to 4000 deaths and 5,000,000 ill every year are caused by contaminated meat and meat products with food poisoning bacteria specially, *Staphylococcus aureus*, *Salmonella* sp., *Escherichia coli* and *Clostridium perfringens* (Hassanien, 2004). Thus, the aim of this study to make a comparison study about the contamination level of meat luncheon obtained from tow different companies, and comparing these results with the Egyptian Organization for Standardization and Quality of Luncheon.

### MATERIALS AND METHODS

#### Luncheon samples collection:

All samples were vacuum-packed luncheon meats and it collected from different local market of Damietta city. These luncheon samples produced from two different companies in Egypt A and B. Samples were aseptically transferred into sterile polyethylene bags, and transported in ice box to the laboratory of Microbiology Department, Faculty of Agriculture, University of Damietta for microbiological examination.

#### Microbiological examination of luncheon samples:

All of samples were examined microbiologically for pathogenic bacteria and fungi. Ten grams of each sample (vacuum-packed luncheon meat) were transferred into 90.0 ml sterilized water and shaken well to homogenate. One ml of suitable dilution ( $10^{-3}$ ,  $10^{-4}$  and  $10^{-5}$ ) was put into Petri dish or tubes, and then melted suitable medium was poured and mixed well then left to solidifying. Petri dishes were then incubated at suitable temperature for limited period. Obtained single developed microbial colonies of different morphologies in all used cultivation media were picked-up. Colonies were transferred into suitable media plates for sub culturing to obtain pure microbial isolates (Ronald, 2010).

#### Microbiological determination of luncheon samples:

Total bacterial count of luncheon samples were done on nutrient agar (NA) cultivation medium at 30°C for 24hrs (Atlas, 2010). Detection of coliform group was done in two stages, the first step was to detect the presence of acid and gas "presumptive test" using MacConkey broth medium at 37°C for 24-48 h. where the second step is to ascertain the presence of coliform bacteria by "confirmed test" which done on Eosin Methylene Blue (EMB) agar plates at 37°C for 24-48 hours. The metallic sheen colonies as well as typical coliform colonies were recorded as positive confirmed test. Confirmed organisms (typical coliform colonies) were transferred into MacConkey broth and onto agar slants. After incubation at 37°C for 24-48 h., the production of acid and gas in the broth medium and the

presence of Gram negative short rods in smears prepared from slants were considered as a positive test (APHA, 1998). Coliform counts were done using Most Probable Number method (MPN) and calculated from standard Table (LQAD, 2008).

Staphylococci was counted in Baird-Parker agar medium (Merck, 2010) and incubated at 37°C for 72 h. Black, shiny and convex colonies of 1-5 mm in diameter with a narrow, white edge surrounded by a clear zone of 2-5mm wide considered to be positive (Merck, 2010). Opaque rings within the clear zones only appear after 48 h. of incubation. The obtained colonies surrounded by an opaque zone were monitored (APHA, 1998).

Cooked meat medium (Atlas, 2010) was used for counting and isolation of *Clostridium* spp. Dilution frequency technique was adopted to determine the count of anaerobic spore forming Clostridia. The inoculated tubes were sealed with sterile mixture of Vaseline and Paraffin oil in 1:1 ratio (Vaspar layer) and incubated at 37°C for up to 7 days. The presence of Clostridia was detected at the end of the incubation period by accumulation of gases pushing the Vaspar layer up using MPN method (AOAC, 2012). *Clostridium* spp. Count was calculated from standard Table (LQAD, 2008).

*Listeria* sp. was aerobically counted on *Listeria* agar base (Oxoid, 2006) at 35°C for 48 h. *L. monocytogenes* was grown as brown-green colored colonies with a black halo (Ronald, 2010). *Bacillus cereus* was counted on selective agar base (Atlas, 2010) at 37°C for 72 h. *B. cereus* grow as moderate-sized (5mm) colonies, which was turquoise, surrounded by a precipitate of egg yolk and turquoise (Ronald, 2010). *Aeromonas* sp. was counted on m-*Aeromonas* selective agar (Atlas, 2010) at 37°C for 72 h. The obtained colonies which showed visible yellow color were monitored as *Aeromonas* sp. (Ronald, 2010). *Salmonella* and *Shigella* were counted on modified *Salmonella Shigella* agar, (modified S S agar,) at 37 °C for 48-72 h(Atlas, 2010). The obtained colonies which were black-center colonies or pink to red colonies were monitored as *Salmonella* or *Shigella* (Ronald, 2010).

Total fungi was counted on Potato dextrose agar, PDA (Atlas, 2010) at 25°C for 5 days after the incubation period, developed fungal colonies were counted per each plate and the mean count of three plates was recorded to calculate the fungal count (APHA, 1998).

#### Isolation and identification of microorganisms.

Single developed microbial colonies of different morphologies in all used cultivation media were picked-up. Colonies were transferred into suitable media plates for sub culturing to obtain pure microbial isolates (Ronald, 2010). The obtained bacterial isolates were maintained on suitable media slants at 5°C until use. The following microbiological methods were carried out to identify the obtained bacterial isolates according to Brenner *et al.* (2005). Shape, arrangement of the cells, the Gram reaction, spore stain and acid fast stain were microscopically examined in stained preparations of 24 - 48 h. (Benson, 2001).

Identification of fungal isolates was based on the visual observation of fungal isolates grown on PDA medium. Plates were carefully identified by morphological characteristics in clouding colure of the colonies and growth on PDA, malt extract agar (MEA) and Czapek

yeast extract agar (CYA) media at 25 °C. In addition, the vegetative and reproduction strictness observed in microscope were also considered for the fungal isolates. The taxonomic keys described in Samson *et al.* (2000) was used for identification which was done at the Regional Center For Mycology and Biotechnology (RCMB), Al-Azhar university.

## RESULTS AND DISCUSSION

### Microbiological evaluation of luncheon samples

All luncheon samples were in the production date. Results in Table 1 showing, the mean values of the tested luncheon samples. The highest value of the total bacterial count in luncheon was in samples of company A being 2500 ( $\times 10^2$ cfu/g), but the lowest value was in the samples of company B being 310 ( $\times 10^2$  cfu/g). These values are higher than that of the permissible limit of the Egyptian Organization for Standardization and Quality of Luncheon No. 1114 (EOSQ, 2005), which premised the total bacterial count at maximum value of 10 ( $\times 10^4$  cfu/g). So the examined samples are not accepted according to the EOSQ.

**Table 1. The mean value of microorganisms ( $\times 10^2$  cfu/g) presented in examined luncheon samples collected from two different companies**

Cultivation media	Company	
	A	B
Nutrient agar	2500	310
<i>Listeria</i> selective agar base	ND*	ND
<i>Bacillus cerues</i> selective agar base	470	30
Barid parker agar	170	30
m- <i>Aeromonas</i> selective agar base	20	10
S S agar	ND	ND
MacConkey	ND	10
Cooked meat	ND	450
PDA	30	1

ND\* = Not Detected

This high value of the total bacterial count of the meat products such as luncheon because of luncheon is a good medium for bacterial growth (Nowak and Krysiak, 2005). Obtained results were lower than that obtained by Abostate *et al.* (2006) who studied the total bacteria in meat luncheon in Cairo and Giza supermarkets in Egypt being,  $1.02 \times 10^7$  and  $1.33 \times 10^7$  cfu/g, respectively.

*Listeria* sp. (Table 1) was not detected in both luncheon samples of the two examined companies. Although authors did not detect *Listeria* sp. in the samples, but another authors detect it, so that, authors suggest to add it in the EOSQ, because *Listeria* sp. did not excised in the EOSQ of luncheon (EOSQ, 2005). Vacuum package luncheon inhibits the growth of *Listeria* sp. and delays the development of spoilage because of the slow propagation of *Listeria* sp. which tolerate the anaerobic conditions (Nowak and Krysiak, 2005). Obtained results are in disagreement with those obtained by Abd-El-Malek *et al.* (2010) who isolated *Listeria* sp. from luncheon in Assiut, Egypt. Also, these results are a disagreement with those obtained by Jamali *et al.* (2013) who isolated *Listeria* spp from luncheon in Malaysia. Moreover, Zaghoul *et al.* (2010) determined *Listeria* sp. ( $20 \times 10^2$  cfu/g) in luncheon sample in Cairo governorate, Egypt. On the other hand, obtained results are similar to that obtained by Ahmed *et al.* (2012) who did not find *Listeria* sp. in any samples of luncheon in Port-Said City, Egypt.

*B. cereus* (Table 1) was counted in luncheon and it were 470 and 30 ( $\times 10^2$ cfu/g) in samples of company A and B, respectively. Authors suggest to add *B. cereus* count in the EOSQ of luncheon (EOSQ, 2005), because of *B. cereus* considered as major foodborne pathogen during the last few decades. It cause two types of food poisoning, known as the emetic and the diarrheal types. *B. cereus* produces several virulence factors including toxins and enzymes, which are considered the most important factors such as lecithinase, hemolysin and protease (Abd-El-Tawab *et al.*, 2015). So, the examined samples are not accepted according to the EOSQ. The presence of *B. cereus* in luncheon because the heat treatment at 72° C for 15 min. (Lawley *et al.*, 2012) did not enough to kill the endospore forming bacteria. Obtained results are lower than that obtained by Abostate *et al.* (2006) who counted total bacilli in meat luncheon in Cairo and Giza, Egypt being  $1.66 \times 10^5$  and  $7.53 \times 10^4$  cfu/g, respectively.

The mean values of Staphylococci count of luncheon listed in Table 1. These values were 1<sup>vo</sup> and 30 ( $\times 10^2$  cfu/g) in samples of company A and B, respectively. These values are higher than the permissible limit of the EOSQ of luncheon (EOSQ, 2005) which was did not premise Staphylococci count in luncheon. So, these samples are not accepted according to the EOSQ. Is heated luncheon at 72 °C for 15 min to eliminate non spore forming bacteria and to prevent the acquisition of luncheon for the taste cooked by the effect of heat settlement (Lawley *et al.*, 2012), this treatment did not enough to kill Staphylococci bacteria. Obtained results are similar to that obtained by Hassanien (2004) and Shawish and Al-Humam (2016) who isolated *Staphylococcus aureus* from luncheon samples in Kalyobia governorate and Kingdom of Saudi Arabia (KSA), respectively.

*Aeromonas sp.* (Table 1) was counted in luncheon samples. The results showed that a high level of *Aeromonas sp.* in luncheon was in samples of company A ( $20 \times 10^2$  cfu/g), while the lowest value  $\cdot$  cfu/g was in company B. Authors suggest to add the count of *Aeromonas sp.* in the EOSQ of luncheon (EOSQ, 2005), because of it caused many of diseases for consumers. Similar results were obtained by Júnior *et al.* (2006) who isolated *Aeromonas sp.* from luncheon samples in São Paulo.

*Salmonella* and *Shigella* spp. (Table 1) was not detected in both luncheon samples of the two examined companies. The results are in the permissible limit of the EOSQ of Luncheon (EOSQ, 2005) which did not premise *Salmonella* and *Shigella* count in Luncheon. So that, these samples are accepted according to the EOSQ. It could explain this by the heat treatment may be did not reach to 72° C for 15 min. which is enough to kill *Salmonella* and *Shigella* spp. Obtained results are similar to that obtained by Hassanien (2004) who did not found *Salmonella* and

*Shigella* in luncheon samples. On the other hand, obtained results are disagreement with that obtained by Karmi (2013) who isolated *Salmonella sp.* from luncheon Aswan, Egypt.

The mean values of coliform count of luncheon are in Table 1. The mean value was ( $10 \times 10^2$ cfu/g) in samples of company B and not detected in samples of company A. These values are in the permissible limit of the EOSQ of luncheon (EOSQ, 2005) which premise coliform count in luncheon at a maximum value of  $10 \times 10^2$  cfu/g. So, these samples are accepted according to the EOSQ. EOSQ of luncheon did not premise *E. coli*. Obtained results of company B was higher than that obtained by Hassanien (2004) who counted coliform in luncheon samples being  $7.94 \times 10^2$  cfu/g.

Anaerobic bacterial count (Table 1) in luncheon was 450 ( $\times 10^2$ cfu/g) in samples of company B, while it is not detected in samples of company A. These values are higher than the permissible limit of the EOSQ of luncheon (EOSQ, 2005) which did not premise anaerobic bacterial count. So, these samples are not accepted according to the EOSQ. The presence of anaerobic bacteria in luncheon may be because of the heat treatment (72° C for 15 min.) did not enough to kill the endospore forming bacteria. Obtained results of company B was similar to that obtained by Hassanien (2004) who isolated *C. perfringens* from luncheon samples in Kalyobia governorate, Egypt. Obtained results of company A was disagreement with that of Hassanien (2004). While obtained results in company B was higher than that obtained by Atwa and Abou-El-Roos (2011) who counted *C. perfringens* from luncheon in Menoufeya and Gharbia governorates, Egypt being 21 cfu/g.

Total fungi and yeasts (Table 1) were counted in luncheon being 30 and 1 ( $\times 10^2$ cfu/g) in samples of company A and B, respectively. The results showed a high level of contamination with total fungi in samples of company A while the lowest value was in samples of company B. Obtained results are similar to that obtained by Ismail *et al.* (2013) who isolated fungi from luncheon samples in Cairo and Giza, Egypt. Authors suggest to add the count of the total fungi and yeasts in the EOSQ of luncheon (EOSQ, 2005), because it causing many of diseases and producing mycotoxins.

**Characterization and identification of obtained microbial isolates**

Table 2 showing that, fourteen different microbial isolates were obtained from Luncheon samples and its numbers, six isolates from samples of company A and eight isolates from samples of company B. Three isolates obtained form nutrient agar medium with Nos. of 111, 116 and 112. Another three isolates obtained from PDA medium with Nos. of 3, 6 and 11.

**Table 2. Numbers and sources of obtained microbial isolates**

Examined samples	Company	Microbial isolates obtained on cultivation media of									
		Nutrient agar	<i>Listeria</i> agar base	<i>Bacillus cereus</i>	Baird parker	<i>Aeromonas</i>	SS agar	Mac Conkey	Cooked meat	PDA	Σ
Luncheon	A	111	-	117	125	123	-	-	-	3 and 6	6
	B	116 and 112	-	126	122	124	-	22	1	11	8
Σ	2	3	0	2	2	2	0	1	1	3	14

Two isolates obtained from each selective agar base; Baird-Parker agar and m-*Aeromonas* selective agar

media with Nos. of 117, 126, 125, 122, 123 and 124, respectively. One isolate obtained from EMB agar and

cooked meat media with Nos. were 1 and 22, respectively. No isolates obtained from *Listeria* agar base and SS agar media.

Table 3 showing, bacterial cell shape and results of some staining of obtained bacterial isolates. Of eleven bacterial isolates, six (Nos. 1, 111, 112, 116, 117 and 126) of them were long rods, three isolates (Nos. 22, 123 and 124) were short rods and two isolates (Nos. 122 and 125)

were coccoid. Eight isolates (Nos. 1, 111, 112, 116, 117, 122, 125 and 126) were Gram positive. Three isolates (Nos. 22, 123 and 124) were Gram negative. Six isolates (Nos. 1, 111, 112, 116, 117 and 126) were spore formers and five isolates (Nos. 22, 122, 123, 124 and 125) were non spore forming bacteria. All obtained isolates gave negative results in acid fast stain. The identification testes were done according to Brenner *et al.* (2005) as follow.

**Table 3. Bacterial cell shape and some of staining results**

Isolates Nos.	Cell morphology and staining				
	Cell shape	Arrangement	Gram stain	Spore stain	Acid fast stain
122 and 125	coccoid	single or clusters	+	-	-
1, 111, 112, 116, 117 and 126	long rods	single	+	+	-
22, 123 and 124	short rods	single	-	-	-

One typical colony of Staphylococci was black, shiny and convex. Black color colony on Barid Parkar agar was picked up and streaked onto nutrient agar plate. After growth, the colony was golden yellow on nutrient agar. The microscopic examination proved that the cells were spherical, Gram positive, arranged in clusters, non-spore formers non-acid fast, and it was catalase positive, oxidase-negative and coagulase positive. So it can be suggested that isolate No. 122 considered as *Staphylococcus aureus*. Similar results were obtained by Cetin *et al.* (2008) who identified *S. aureus* using Baird–Parker agar supplemented with egg yolk telluride emulsion. The colonies of *S. aureus* have typical morphology and it tested for catalase and positive coagulase test. Obtained results are similar to that obtained by Hamad (2010) who identified *S. aureus* using positive coagulase test. Obtained results are also similar to that obtained by El-Hadedy and Abu-El-Nour (2012) and Can *et al.* (2013) who identified *S. aureus* by using selective isolation media (Baird-parker), and confirmed by molecular method based on 16S rRNA gene. The colonies had a typical black colored view surrounded by a light colored area.

Isolate No. 125 was Gram positive coccoid shaped cells. Typical colony was small and brown. Colony was picked up and streaked onto the same medium slant. After growth, the microscopic examination appeared the shape of spherical cells, Gram positive, arranged in single or irregular, non-spore forming and non-acid fast. Isolate No. 125 can be considered as *Micrococcus* sp. This result is in agreement with those of Osakue *et al.* (2016) who isolated *Micrococcus* sp from luncheon in Benin City, Nigeria.

After anaerobic bacterial growth on the cooked meat medium, Gram and spore stain were done to confirm the presence of bacterial spores. The bacteria were long rod and Gram positive, spore-formers which confirm the presence of *Clostridium* spp (isolate No. 1). Obtained results are similar to that obtained by Hassanien (2004), Ismail (2006) and Atwa and Abou-El-Roos (2011) who isolated *C. perfringens* from Luncheon samples collected from Kalyobia, Ismailia and Menoufia and Gharbia governorates, respectively.

Colonies which were white colored and grown on nutrient agar medium were picked up and streaked onto the same medium slant. After growth, the microscopic examination showed that, the cells were long rods, Gram positive, spore forming bacteria and non-acid fast, arranged in single, pair or chains. Isolates Nos. 111, 112 and 116 can be considered as members of the Genus

*Bacillus* sp. Similar results were obtained by Tallent *et al.* (2012) who isolated the genus *Bacillus* sp. from Luncheon samples.

Colonies which were blue colored with zone grown on *B. cereus* selective agar base medium were picked up and streaked onto the same medium slant. After growth, the microscopic examination showed that, the cells were long rods, Gram positive, spore forming bacteria and non-acid fast, arranged in single, pair or chains. Isolates Nos. 117 and 126 can be considered as members of the Genus *Bacillus*. Obtained results are similar to that obtained by Hassanien (2004), Güven *et al.* (2006) and Abd-El-Tawab *et al.* (2015) who isolated *B. cereus* from luncheon samples collected from Kalyobia Governorate, Turkey and Gharbia Governorate, respectively.

Bacteria isolated from MacConkey broth which gave acid and gas was picked up and streaked onto EMB medium. After growth, the colony was green and metallic sheen. The cells were short rods, Gram negative, non-spore formers and non-acid fast, arranged in single cells oxidase-negative. Isolate No 22 considered as *Escherichia* sp. Obtained results are similar to that obtained by Hassanien (2004), Hamad (2010), Al-Mutairi (2011) and Hassanin *et al.* (2015) who isolated *E. coli*. from luncheon samples from Kalyobia governorat, Khartoum State, Giza governorate, Cairo governorate and Assiut, Egypt, respectively.

Colonies Nos. 123 and 124 appeared on m-*Aeromonas* agar medium which gave yellow color, were picked up and streaked onto the same medium slant. After growth, the microscopic examination proved that the cells were short rods, Gram negative, non-spore formers and non-acid fast, arranged in single cells and it was catalase positive, oxidase-positive. Isolates Nos. 123 and 124 considered as *Aeromonas* sp. Similar finding was clear by Júnior *et al.* (2006) and Sarimehmetoglu (2009) who isolated and identified the genus *Aeromonas* sp. from luncheon samples collected from São Paulo and turkey, respectively.

The characteristics of isolate No. 3 showed that, culture examination was as following; colonies on CYA attending 2-3 cm diameter at 25°C, radially, white, grayish, to green. Reverse colorless. Microcolonies grow on CYA at 5°C and no growth at 37 °C. Microscopic examination showed that, conidiophore diameter 2.8 µm. Rami 17.2x 3.8 Metulae 11.5x3.0µm swollen in the end. Phialides 7.0 x 2.0 µm. Conidia were elliposoidal to sub spherical 3.0x3.2 µm. Identification was mainly based on current universal

keys of Samson *et al.* (2000). From these observations, isolate No. 3 were identified as *Penicillium aurantiogriseum*. Obtained results are similar to that obtained by Brr *et al.* (2004) and Ismail *et al.* (2013) who isolated *Penicillium* sp. from luncheon samples collected from Tanta city, Egypt and Cairo and Giza, Egypt, respectively.

The characteristics of isolate No. 6 showed that, culture examination was as following; colonies on MEA at 25 °C attaining a diameter of 5.5 cm in 7 days, olivaceous green with olivaceous black reverse. Microscopic examination showed that, conidiophores 3 – 6 µm in diameter. Conidia were ellipsoidal to cylindrical with rounded ends 10.0 X 4.0 µm. Identification was mainly based on current universal keys of Samson *et al.* (2000). From these observations, isolated No. 6 was identified as *Cladosporium herbarum*. Obtained results are similar to that obtained by Brr *et al.* (2004) and Ismail *et al.* (2013) who isolated *Cladosporium* sp. from luncheon samples collected from Tanta city, and Cairo and Giza, Egypt respectively.

The characteristics of isolate No. 11 showed that, colonies were whitish becoming greyish-brown, up to 1 cm high. Microscopic examination showed that, sporangiophores was single or in tufts, brown, 16 µm wide, mostly unbranched. Sporangia was Spherical, 125 µm, brownish grey to black. Columella comprising 50-70%, spherical. Sporangiospores were grayish green-sub-spherical to ellipsoidal, 6.3X4.6 µm. Chlamyospores were single or in chain. Identification was mainly based on current universal keys of (Samson *et al.*, 2000). From these observations, isolate No. 11 was identified as *Rhizopus oryzae*. Obtained results are similar to that obtained by Brr *et al.* (2004) who isolated *Rhizopus* sp. from luncheon samples collected from Tanta city, Egypt.

## CONCLUSION

Finally, it could be concluded that, the values of total bacterial count, Staphylococci count and anaerobic bacterial count were higher than the permissible limit of the Egyptian Organization for Standardization and Quality (EOSQ) of Luncheon No 1114. On the other hand, the values were in the permissible limit of the EOSQ of luncheon in case of *Salmonella*, *Shigella* and coliform bacteria. So that, these samples are not accepted according to the EOSQ. Authors suggest to add the count of *Aeromonas*, *Listeria*, *B. cerues* and total fungi and yeasts in the EOSQ of Luncheon, because of it caused many of diseases for consumers.

## REFERENCES

Abd-El-Malek, A. M.; S. F. H. Ali; R. Hassanein; M. A. Mohamed and K. I. Elsayh (2010). Occurrence of *Listeria* species in meat, chicken products and human stools in Assiut city, Egypt with PCR use for rapid identification of *Listeria monocytogenes*. *Veterinary World*.3(8): 353-359

Abd-El-Tawab, A. A.; F. I. El-Hofy; D. F. Khater and Y. M. AL-Baaly (2015). Molecular studies on toxigenic strains of *Bacillus cereus* isolated from some meat products. *Benha Veterinary Medical J.*, 29(1):129-133.

Abostate, M. A. M.; D. A. Zahran and H. N. EL-Hifnawi (2006). Incidence of *Bacillus cereus* in some meat products and the effect of gamma radiation on its toxin(s). *International J. of Agriculture and Biology* 8(1):1-4

Ahmed, H. A.; A. A. Megahed and G. M. O. Mohammed (2012). Molecular detection of *Listeria monocytogenes* pathogenic genes as food poisoning micro-organism isolated from ready- to- eat meals in Port-Said City markets. *Assiut Vet. Med. J.* 58(132):1-17

Al-Mutairi, M. F. (2011). The incidence of Enterobacteriaceae causing food poisoning in some meat products. *Advance J. of Food Science and Technology* 3(2): 116-121

AOAC, Association of Official Analytical Chemist (2012). *Official Methods of Analysis*, 20<sup>th</sup> Ed., AOAC international, suite 500, 481 North Frederick Avenue, Gaithersburg, Maryland 20877-2417, USA

APHA, American Public Health Association (1998). *Standard Methods For the Examination of Water and Wastewater*. 20<sup>th</sup> Ed. APHA, Inc. New York

Atlas, M. R. (2010). *Handbook of Microbiological Media*. 4<sup>th</sup> Ed. CRC Taylor and Francis Group Boca Raton London New York, USA

Atwa I. E. and N. A. Abou-El-Roos (2011) Sincidence of *Clostridium perfringens* in meat products at some Egyptian governorates. *International J. Microbiological Research*. 2 (3): 196-203

Brenner, D.J., Krieg, N.R., Staley, J.T. & Garrity, G.M. (2005). *Bergey's Manual of Systematic Bacteriology*, 2nd ed., Springer-Verlag, New York, NY.

Benson, H. J. (2001). *Microbiological Applications Lab Manual*, 8<sup>th</sup> Ed. The McGraw-Hill Companies, UK

Brr, A. H.; N. Y. Moustafa and A. M. Edris (2004). Incidence of moulds and aflatoxins in some meat products. *Benha Vet.Med.J.* 15(2): 65-75

Can, O. P.; S. Şahin; M. Erşan and F. Harun (2013). Sivas kofte and examination of microbiological quality. *Biotechnology in Animal Husbandry*. 29 (1): 133-143

Cetin, O.; E. B. Bingol and H. Akkaya (2008). The microbiological, serological and parasitological quality of cig kofte (raw meatball) and its lettuce marketed in Istanbul. *Polish J. of Environ. Stud.* 17(5):701-706

Ehrampoush, M. H.; S. M. Mazloomi; S. Zarei; F. Hemmati; M. T. Ghaneian; B. Hajimohammadi and A. Dehghan (2017). Microbiological quality of Sausage during slicing at food retail stores in Shiraz, Iran. *International J. of Nutrition Sciences*. 2(1): 20-25

El-Hadedy, D. and S. Abu-El-Nour (2012). Identification of *Staphylococcus aureus* and *Escherichia coli* isolated from Egyptian food by conventional and molecular methods. *J. Genetic Engineering and Biotechnology*. 10:129-135

EOSQ, Egyptian Organization For Standarization and Quality (2005). *Egyptian Standards for Requirements of Luncheon No:1114*

- Güven, K.; M. B. N. mutlu and Ö. Avci (2006). Incidence and characterization of *Bacillus cereus* in meat and meat products consumed in Turkey. J. of Food Safety. 26:30-40
- Hamad, N. M. H. (2010). Prevalence of *Escherichia coli*, *Salmonella* and *Staphylococcus aureus* in Processed Meat in Khartoum State. M. Sc. Thesis. Faculty of Veterinary Medicine, Khartoum Univ. Sudan.
- Hassanien, F. S. (2004). Bacterial hazards associated with consumption of some meat products. Benha Vet. Med. J. 15(2): 41-54
- Hassanin, F. S.; M. A. Hassan and S. A. Helmy (2015). Enterobacteriaceae in meat products sold by street vendors. Benha Veterinary Medical J. 29(2):74-79
- Ismail, S. A. (2006). Microbiological quality of hawawshy consumed in Ismailia, Egypt. J. of Food Safety. 26. 251-263
- Ismail, S. A.; A. A. Shehata and E. M. El-diasty (2013). Microbiological quality of some meat products in local markets with special reference to mycotoxins. Global Veterinaria. 10 (5):577-584
- Jamali, H.; L. C. Chai and K. L. Thong (2013). Detection and isolation of *Listeria* spp. and *Listeria monocytogenes* in ready-to-eat foods with various selective culture media. Food Control. 32:19-24
- Júnior, O. D. R.; L. A. Amaral; A. N. Filho and R. P. S. Iturrino (2006). Bacteria of the genus *Aeromonas* in different locations throughout the process line of beef slaughtering. Revista Portugues Ciências Veterinárias. 101:125-129
- Karmi, M. (2013). Prevalence of *Salmonella* in meat products. Global Veterinaria. 11 (5):685-688
- Lawley, R.; L. Curtis and J. Davis (2012). Chemical Hazards in Food. Food Safety Info, 163 Fox Lane, London N13 4AU.
- LQAD, Laboratory Quality Assurance Division (2008). Most Probable Number Procedure and Tables. United States Department of Agriculture Food Safety and Inspection Service, Office of Public Health Science, 950 College Station Road, Athens, GA 30605.
- Mahmoud, F. S. A.; M. E. Y. Kdous and A. M. Bayomey (2016). Evaluation of new dried blends of fast processed luncheon meat. Middle East J. of Applied Sciences. 6(1): 113-119
- Merck, (2010). Merck Microbiology Manual 12<sup>th</sup>. Darmstadt, Germany
- Nowak, A. and E. Krysiak (2005). Predominant microflora of vacuum-packed frankfurters. Polish Journal of Food and Nutrition Sciences. 14/55(1): 91-94
- Osakue, O. P.; J. O. Igene; P. A. Ebabhamiegbegbo and S. E. Evivie (2016). Proximate analysis and microbial quality of ready-to-eat (RTE) fried chicken parts. J. of Food & Industrial Microbiology. 2(1):2-8
- Oxoid (2006). "Oxoid Manual", 9<sup>th</sup> Ed. Oxoid Limited, Wade Road, Basingstoke, Hampshire, England
- Ronald, M. A. (2010). Hand Book of Microbiological Media. CRC Taylor and Francis Group Boca Raton London New York, USA
- Samson R. A; E. S. Hoekstra and J. C. Frisvad (2000). Introduction to Food and Airborne Fungi. Utrecht, Netherlands
- Sarimehmetoglu, C. K. B. (2009). Isolation and identification of motile *Aeromonas* spp. in Turkey meat. Ankara Üniv. Vet. Fak. Derg. 56:95-98
- Shawish, R. R. and N. A. Al-Humam (2016). Contamination of beef products with Staphylococcal classical enterotoxins in Egypt and Saudi Arabia. GMS Hygiene and Infection Control. 11:2196
- Tallent, S. M.; K. M. KoTewicz; E. A. Strain and R. W. Bennett (2012). Efficient isolation and identification of *Bacillus cereus* group. J. of AOAC. International. 95(2):446-451
- Tewari, A.; S. P. Singh and R. Singh (2015). Incidence and enterotoxigenic profile of *Bacillus cereus* in meat and meat products of Uttarakhand, India. J. Food Sci Technol. 52(3):1796-1801
- Zaghloul, R. A.; M. A. El-Shenawy; N. A. Neweigy; H. E. Abou-Aly; R. K. El-dairouty; W. I. El-Kholy; M. T. Fouad; J. M. Soriano; J. Mañes and L. Mico (2010). *Listeria* spp. and Enterobacteriaceae group in sandwiches of meat and meat products. British Microbiology Research J. 4(4):360-368

## الفساد الميكروبي لعينات لانشون اللحم مأخوذة من مدينة دمياط - مصر

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تم إجراء الفحص الميكروبيولوجي لعينات لانشون مأخوذة من الأسواق المحلية بمحافظة دمياط. العينات كانت تتبع شركتين محليتين بجمهورية مصر العربية. كان العد البكتيري الكلي ٢٥٠٠ و ٣١٠ × ١٠ وحدة مكونة للمستعمرة لكل جرام (cfu/g). بينما كان عدد الباسيلس سيريس ٤٧٠ و ٣٠ × ١٠ (cfu/g) في حين كانت أعداد الاستافيلوكوكس 170 و ٣٠ × ١٠ (cfu/g). أما أعداد الفطريات كانت ٣٠ و ١ × ١٠ (cfu/g) للشركتين أ، ب على الترتيب. وكانت أعداد الايرومونات في عينات الشركة أ أعلى من أعدادها في عينات الشركة ب. بينما كانت أعداد بكتيريا القولون والبكتيريا اللاهوائية في عينات الشركة ب ١٠ × ١٠ و ٤٥٠. على الترتيب ولم يلاحظ تواجدهما في عينات الشركة أ. وكانت العينات خالية من اللستيريا والسالمونيلا والشيجيلا في كلتا الشركتين. كانت أعداد البكتيريا الكلية والاستافيلوكوكس والبكتيريا اللاهوائية أعلى من الحدود المسموح بها من قبل الهيئة المصرية للمواصفات والجودة رقم ١١٤ والخاصة بالانشون. على الجانب الآخر كانت أعداد السالمونيلا والشيجيلا وبكتيريا القولون مطابقة للمواصفات. تم عزل وتعريف ١٤ عزلة ميكروبية من عينات اللانشون، ٦ عزلات من الشركة أ و ٨ عزلات من الشركة ب. تم عزل ٣ عزلات من على بيئة الأجار المغذى و ٣ عزلات أخرى من على بيئة أجار البطاطس والدكستروز. وعزلتين من على بيئات الأجار القاعدية الاختيارية و Pared parker أجار والايرومونات الاختيارية. وعزلة واحدة من على بيئتي الايوسين ميثيلين بلو وبيئة اللحم المطبوخ. ولم يتم عزل أي عزلات من على بيئات اللستيريا أجار والسالمونيلا شيجيلا أجار. من بين الـ ١١ عزلة بكتيرية تم تحديد ٦ عزلات خلاياها عصوى طويل و ٣ خلاياها عصوى قصير وعزلتين كرويتين. ٨ موجبة لصيغة لجرام و ٣ سالبة لصيغة لجرام. ٦ متجرثة و ٥ غير متجرثة. بينما كانت كل العزلات غير صامدة للأحماض. هذا بالإضافة إلى عزل وتعريف ٣ فطريات وكانت الريزوبس أوريزا و البيينسيليم أورانتيجريزيم و الكلاوسوبيوم هراريوم. وعموماً تعتبر العينات كلها غير مقبولة طبقاً للمواصفات التي أقرتها الهيئة