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Research Article

## A MIXED FOOD OUTBREAK WITH SALMONELLA AND STAPHYLOCOCCUS AUREUS, ALYOTMAH VILLAGE, MARCH-2017

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#### Abstract:

**Introduction:** Food poisoning is one of the major public health problems worldwide. Salmonella and S.aureus are primarily responsible for food poisoning. The current study focuses on a food poisoning outbreak in Alyotmah Village, Saudi Arabia, in 2017, which was due to eating from the Farooj Madinati Restaurant.

*Objectives*: To assess the extent of the food poisoning outbreak and identify its source along with providing recommended measures to prevent future outbreaks.

**Design:** Epidemiological study was carried out among the affected individuals, who were interviewed with questionnaire made by the Food Safety Department of the Ministry of Health of Saudi Arabia. This was followed by the environmental survey and field inspection of the restaurant. Laboratory samples were collected including food and restaurant food preparation tools, in addition to biological samples of the four food handlers and stool samples of 11 patients.

**Results:** Epidemiological study on 69 individuals showed that they developed predominant symptoms of abdominal pain, diarrhea, and fever in less than 6 hours of incubation period. Salmonella and S.aureus were found to be present in most of the laboratory samples.

**Conclusion:** The study identified that the food handlers were the asymptomatic carriers of the organisms, resulting in the contamination of the tools used in the preparation of food.

Maintaining proper hygiene within the restaurant, along with increasing the hygiene awareness of the food handlers can go a long way in reducing such outbreaks in future.

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#### **INTRODUCTION:**

Worldwide, Salmonella is the most common organism causing food poisoning and leading to major public health problem.(1) There are more than 2600 serotype of Salmonella e.(2) One of Salmonella serotype is nontyphoidal Salmonella e (enteriditis, and typhimurium), which is responsible for a frequent food poisoning outbreak globally.(2)

The incubation period of Salmonella food poisoning ranges between 6 to 12 hours and may reach up to 72 hours.(3) Salmonella infects humans through ingestion of contaminated food or contaminated water, as well as eating undercooked meat, poultry and eggs.(4) About 4% of patient with Salmonella e enteriditis may develop chronic or carrier stage; chronic\carrier patients may spread the infection to many people; particularly those who work in food industries.(5) The symptoms of Salmonella food poisoning include nausea, vomiting, abdominal pain, fever, headache, and diarrhea; these symptoms are self-limiting which may persist for two to seven days.(6) However, patients with long and severe disease may need intravenous therapy along with an antibiotic to prevent dehydration and bacteremia.(7)

Contaminated food with Salmonella maybe influanced with insuffcient refregeration. The optimum temperature for Salmonella growth is ranging between 35 to 37°C.(4) The temperature of 70°C or higher would kill Salmonella (4), hence using high temperature while cooking will make food safe to be utilized. The optimum pH for the Salmonella e growth is ranging between 6.5 and 7.5. Salmonella e growth inhibition occurs at pH<3.8(4), therefore acidic pH may act as bactericidal.

In 2010, it was estimated that more than 93 million cases of gastroenteritis are due to Salmonella species, with 155,000 deaths that occur each year.(8) Each year in the United States, the estimated foodborne diseases were 9.4 million, about 11% were due to nontyphoidal Salmonella (NTS).(9) Additionally, more than 228 thousand were admitted to the hospital; 35% of this admission were due to NTS.(9)

According to the food safety department of Ministry of health, Saudi Arabia, the total outbreaks during 2016 were 372, and the total reported cases were 2113.(10) More than 35% of reported cases were due to Salmonella .(10) Additionally, the incident rate of the food poisoning outbreak decreased from 17.9/100,000 in 2004 to 11/100,000 in 2016. (10) Moreover, the incident rate of Salmonella food poisoning reached its maximum rate (7.81/100,000)

in 2007. After which a sharp decline to reach the minimum rate (2.34\100.000) by2016. (10)

Staphylococcal Aureus(SA) is another common organism that cause food-born disease which result from ingestion of Staph enterotoxins, produced by enterotoxigenic strains of coagulase-positive Staphylococci.(11) Staphylococci are classified to more than 50 species, according to its ability to produce coagulase. (11) SA lives as a normal flora in the skin and mucus of mammals and birds; transfer of SA to food occurs either by human carriage during food processing or by dairy animal in cases of mastitis.(11) The incubation period for SA depends on amount of enterotoxin ingested, and it ranges between half hour to 8 hours. (11) The symptoms of SA include nausea, vomiting, abdominal pain, diarrhea, dizziness, general weakness, and sometimes moderate fever.(11) The optimum temperature for the growth of SA is 37°C, and optimum temperature to produce S. Enterotoxin range between 37-45°C. Additionally, the SA grows in pH that range between 4-8, and the production of S. Enterotoxin occurs in pH ranging between 4-9.6.(11) Therefore, SA can be controlled at temperature higher than 45°C, and pH lower than 4.

#### **BACKGROUND:**

Alyotmah is a small town located 75km south west of Al Madinah Al Monowarh. The families in Alyotmah usually eat from outside during weekend, and there about four restaurants. There is one Primary Healthcare Center (PHC) along with Wadie Alfera hospital which serves as its referral hospital. According to the director of Alyotmah PHC, there were no outbreaks in the town till 3<sup>rd</sup> of March 2017: when the first case visited the PHC in Alyotmah Village, and a total number of cases were seven on the first day. However, the outbreak of food poisoning was confirmed and announced in next day (4<sup>th</sup> March) when the number of new cases reached to fifteen and announced in newspaper. After which FETP department assigned a team to visit the restaurant in the region along with patients. The Food Safety Department (FSD) of General health directorate of Madinah was involved along with FETP team.

#### **OBJECTIVES:**

- To assess the extent of this food poisoning outbreak
- To identify the source of the outbreak, and
- To provide the recommended measures to prevent future outbreaks.

#### **METHODOLOGY:**

#### A) Epidemiological:

#### Study design:

Descriptive study. As this outbreak is public, the best study design for this case is a case-control study but there are not enough controls available.

#### Study Population:

Any person who became ill after eating from the restaurant, and visited any health service in Alyotmah between 2<sup>nd</sup> –4<sup>th</sup> March- 2017.

#### Case Definition:

A case definition was set as any person who ate from Farooj Madinati Restaurant between 2<sup>nd</sup> to 3<sup>rd</sup> March, and developed symptoms between 2<sup>nd</sup> to 4<sup>th</sup> March 2017. The symptoms include either abdominal pain, diarrhea, vomiting, fever, or nausea.

#### Data Collection and Analyses:

An interviewed questionnaire, made by Food Safety Department of the Ministry of Health of Saudi Arabia, was used for data collection. This questionnaire composed of demographics, history of illness, symptoms and food items ingested (appendix). The line list of all people and their food item were listed. Data were cleaned using excell, then entered into database analyses system (EPI-Info Ver 7.2.0.1)

#### Study Variables:

The study variables included demographics (age, sex, nationality), history of illness (time of eating, and the onset of symptoms, incubation period, symptoms, food items), and the outcome.

#### A) Environmental Survey and Field Inspection:

The team visited the restaurant, which we inspected for any insects and cleanness of cooking and preparation area, the tools used in making the food, washtub used in melting the chicken, and the serving area. We also meet with the management of the restaurant.

#### A) Laboratory

We took samples from uncooked chicken, Mayo Sauce, along with swabs from tools used to make the food, and Mayo-Sauce blender machine to test them for Salmonella along with serotyping, and Staphylococci aureus. All food handlers (4 persons) were asked to visit the central laboratory in Madinah where stool samples, as well as swabs from nails, throat, and nose, were taken for Salmonella along with its serotyping, Staphylococci aureus and parasite analysis. We also collect stool samples for eleven patients for culture of Salmonella with its serotyping and Staphylococci aureus. However, we didn't take

samples for fried chicken, cooked potato, or eggs as they were not available.

#### **RESULTS:**

#### 1) Epidemiological Results:

We were able to reach 69 people who developed any of the following symptoms: abdominal pain, diarrhea, vomiting, nausea, fever, and headache. As shown in table 1, age of patients ranged between 3 to 70 years, mean age is 28.86±14.75, and 49.3% of ill people are in the age range between 20 to 40 years old. Additionally, table1 shows the demographics distribution of Alyotmah outbreak, men (63.8%) are affected more than women (36.2%) and that 69.6% of affected people are Saudis. Table 2 shows the most frequent food items consumed by the patients, where Mayo-Sauce (98.6%) and fried chicken (95.7%) constitute the most eaten items. Table 3 shows the frequency of the symptoms; the predominant symptoms were abdominal pain (94.2%) and diarrhea (89.86%). Table 4 shows only 10.1% had moderate to severe symptoms requiring hospital admission and no deaths were reported. Table 5 shows the incubation period range between 4.26 hours to 40.15 hours, and mean incubation period is 16.98±8.33; table 4 shows incubation period of more than 6 hours constitutes more than 88.4%. Table 6 shows the stratification of frequent symptoms by incubation period group where 100% of patient, who developed illness in less than 6 hours, had abdominal pain, diarrhea, and fever.

Line list of patients shows that the first person ate from the restaurant at 4 PM in 2<sup>nd</sup> March, she developed the symptoms of nausea, vomiting, diarrhea, abdominal pain, and headache after 17 hours which is 9 AM 3<sup>rd</sup> of March. Line list also shows that the first person reported same symptoms after almost 5 hours of eating fried chicken with Mayo-sauce.

Figure 1 is an hourly histogram of EPI Curve which shows a common or a pointed source outbreak, with two peaks in two different days.

# 2) Results of Environmental Survey and Field Inspection

The restaurant is newly opened three weeks before the event. All the four food handlers have the fitness medical certificate. The restaurant did not obtain the certificate to open from municipality department of Alyotmah village. On inspecting the area where food was prepared, we found that all tools, mayo-machine blinder, and washtub were dirty. No insects were found at the restaurant. The ketchup, nuggets, and potato came manufactured sealed and packet. The manager of the restaurant denied that he or other food handlers were sick before the event. He explained the processing of making the mayo-sauce which

consisted of garlic, uncooked eggs, garlic powder, oil, and salted lemon. All are mixed in a mayomachine blinder. The mixture-sauce is kept in small compartments at room temperature. He also explained how they prepare the chicken, where they thawed the chicken in a washtub. Then, the chicken is dipped in the spice dish and then in the eggs and breadcrumbs. After that, the chicken is fried at a temperature above 160°C. Additionally, they cleaned the tools used in preparation along with mayomachine with the only piece of clothes along with water, without using disinfectants, detergent or soaps.

#### 3) Laboratory Results

Table 7 & 8 summarize the laboratory results for swabs of nail, throat and nose, along with stools of four food handlers. All food handler's nail swabs (100%) were positive for Staph. Aureus. Additionally, 75% of the throat swab of food handlers were positive for Staph aureus. Only 25% of the stool sample (one food handler) was positive for Salmonella, whereas all stool sample of the food handlers is positive for Ent. Histolytica.

Table 9 shows the laboratory result for stool samples of 11 patients; of 11 patients, eight patients (72.73%) were positive with Salmonella, and three patients (27.27%) were positive for Staph aureus.

Table 10 shows the laboratory results of food items and tools used in the preparations of food. We took three samples for mayo-sauce, and two samples for uncooked chicken for laboratory analysis and culture. For the mayo-sauce sample, two samples were safe for human use while one sample was positive for both Salmonella and Staph aureus. One uncooked chicken sample was positive for Salmonella while another chicken sample was positive for Salmonella serotyping were done, and result was S. enteriditis type 6704552 in all samples. Table 10 demonstrates that mayomachine was positive for Salmonella. Additionally, the tools used in preparing of food and mayo-sauce were positive for Staph Aureus.

#### **DISCUSSION:**

A total number of 69 cases of gastroenteritis were reported within two days (from March 3<sup>rd</sup> to 4<sup>th</sup>, 2017). Clinico-epidemiological features of the outbreak provide valuable information about the causative organism. Diarrhea with or without Abdominal pain would be seen in food poisoning due to Salmonella, Clostridium perfringens, Shigella, Staph aureus, and E. coli. The C. perfringens food poisoning is associated with the abdominal pain, and

diarrhea that usually appear within 8 to 16 hours, but fever is not common.(3,12) In this outbreak, the incubation period is 2.5 to 35 hours, and fever occurred in more than 50% of the cases. Hence, this outbreak is not due to C. perfringens.

Shigella can be carried among food handlers.(13) 25 to 50% of shigella cases are associated with abdominal pain and bloody diarrhea and the incubation period is 12 to seven days.(7,13) Therefore, we excluded shigellosis. Outbreak caused by E. coli are usually characterized by afebrile or low-grade fever along with abdominal pain and bloody diarrhea.(7,12,13) Additionally, 50% of patients may experience nausea and vomiting. (7,12,13) However, the incubation period of E. coli is 2-5 days.(7,13) Therefore, we also excluded the E. coli.

The clinical feature of Staph aureus food poisoning is nausea, vomiting, abdominal pain, fever, and diarrhea. These symptoms usually appear between one hour and six hours (some reported 30 minutes to 8 hours).(11) As we mentioned earlier 100% of patient in this outbreak with incubation period less than 6 hours developed fever, abdominal pain, and diarrhea as well as 75% of patient with incubation period less than 6 hours developed nuasea. This clinical feature is ideal with Salmonella however the incubation period for Salmonella is 6 to 36 hours. Morver, the symptoms which developed in patients who had incubation period include abdominal pain, non bloody dirreha, fever, vomiting, nuasea, and headach which are identical with Salmonella . As the incubation period and clinical symptoms of this outbreak are similar with both Staph aureus and Salmonella, we cannot exclude them. Additionally, laboratory results found both organisms in the patients, food handlers, myao-sauce, and mayo blinder.

Epi curve were pointed with two peaks, this could be because of the source of infection was not controlled nor removed. Figure 2 is a three-hourly histogram of EPI Curve that shows doubling of number of patients in next day, this could be due to more of people ate from restaurants during weekend. Therefore, early identification of the outbreak and notification would have reduced the number of affected people by closing the restaurant.

Salmonella can be transmitted through eggs, which is a main ingrediant of mayo-sauce. Mayo-Sauce also consists of vinger or lemon which is an ideal for Salmonella growth along with imporper storing of the mayo-sauce in room temperature.(14) Mayo-sauce has caused many food poisoning especially during

the summer months, where the high temperature enhance the bacterial growth.(14) As mentioned earlier, a temprature of 37°C is considered as the optimum for Salmonella multiplication, so storing eggs in refrigratore would minimize the hazard that can be caused by contaminated eggs. In Saudi Arabia, many restruants use the mayo-sauce with chicken shawrma sandwich and with fried chicken. These resturants made mayo-sauce had been resposible for many food poisning outbreaks.(14)

One of the known risk factors for food poisoning is food handlers.(15) Although food handler denied the recent history of diarrhea, vomiting, or fever before & after this outbreak, isolation of Salmonella and Staph aureus from the food handlers implicate their role as an asymptomatic carrier.

The availability of laboratory facilities as well as serotyping are an important strength for this study. We tried to identify people who ate and did not get sick by calling the ill people, asking them if any of their family or neighbours ate and did not get sick. We reached to only 7 people, making case-control study difficult. So, one of the limitation of this study is the type as it was difficult to make a case-control study.

#### **CONCLUSION:**

Environmental and laboratory results pointed that the primary source of this outbreak is one of food handler who was preparing the mayo-sauce. The food handlers were infected with both Staph aureus and Salmonella but were asymptomatically; they infected the tools and the blender machine used in the preparation of mayo-sauce. Contaminated mayo-sauce along with its storing and using vinegar in its preparation promote the growth of both Salmonella and Staph. aureus. However, we cannot exclude the cooked chicken, but unfortunately, we did not find a cooked chicken sample for laboratory analysis as it was not available.

#### **RECOMMENDATIONS:**

Applying hygiene measures is the basic in prevention of food poisoning. Proper thawing of a frozen chicken is a good preventive measure that would reduce the number of outbreaks. Also, the ideal use of disinfectant to clean the tools used in processing the food. Prohibiting the preparation of mayo-sauce in the restaurant especially in summer along with using the sauce which is prepared in manufacture, sealed, and having an expire date will reduce the number of outbreaks. Adding vinegar to prepare the mayo sauce will make it a perfect media for Salmonella. Therefore, this should be incriminated.

Increasing awareness of food handlers by educating them about the proper way of washing their hands in warm running water along with soaps. Additionally, the employed food handlers who are involved in food preparation should wear gloves and facemask, and should scrub his/her nails with soap and a brush. Food handlers should be examined periodically for organism responsible for food poisoning. This examination should be carried out in governmental labs or hospitals. Increasing the awareness of public about avoiding the use of mayo-sauce during summer might lead to reduce the number of cases.

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#### Conflict of interest

There is no conflict of interest.

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Table 1: Demographics Distribution of Alyotmah Food Poisoning Outbreak, March-2017

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			No	%
	Sex	Male	44	63.8
		Female	25	36.2
cs	Age	<20	21	30.4
ihdī		20-40	34	49.3
Jgra		>40	14	20.3
Demographics	Nationality	Saudi	48	69.6
Õ	Nationality	Non-Saudi	21	30.4
		Rang	e	Mean & SD
		From	To	Weall & SD
	Age	3	70	$28.86 \pm 14.75$

Table 2: Frequency of Food Items of Alyotmah Food Poisoning Outbreak, March-2017

		No	%
	Mayonnaise	68	98.6
ms ms	Fried Chicken	66	95.7
Items	Potato	26	37.7
Food	Ketchup	17	24.6
F0	Nuggets	6	8.7
	Shrimps	2	2.9
	Burger	1	1.4

Table 3:Frequency of Symptoms of Alyotmah Food Poisoning Outbreak, March-2017

	, ,	No	%
w	Abdominal Pain	65	94.2
symptoms	Diarrhea	62	89.9
npt	Vomiting	47	68.1
Syl	Nausea	45	65.2
	Fever	36	52.2
	Headache	35	50.7

Table 4: Patients Outcomes during Alyotmah Food Poisoning Outbreak, March-2017

		No	%
	Treat in OPC	62	89.9
Outcome	Admission	7	10.1
	Death	0	0

Table 5: Incubation Period and IP Groups of Alyotmah Food Poisoning Outbreak, March-2017

			No of cases	%		
	6 Hours or less	-	8	11.6		
	More than 6 Hours	61	88.4			
<b>Incubation Period</b>	Range					
		From	to	Mean & SD		
	IP	4.26	40.15	16.98±8.33		

Table 6: Stratification of Symptoms according to Incubation Group, Alyotmah Food Poisoninh Outbreak, March-2017

		Less than 6 Hours				More than 6 Hours				
	Yes		No			Yes		No		
	No of Case	%	No of cases	%	total	No of Case	%	No of cases	%	total
Abdominal Pain	8	100	0	0	8	57	93.4	4	6.6	61
Diarrhea	8	100	0	0	8	54	88.5	7	11.5	61
Vomiting	4	50	4	50	8	43	70.5	18	29.5	61
Nausea	6	75	2	25	8	39	63.9	22	36.1	61
Fever	8	100	0	0	8	28	45.9	33	54.1	61
Headache	. 5	62.5	3	37.5	8	30	49.2	31	50.1	61

Table 7: Lab results of Food Handlers, Alyotmah FPO March-2017

		Staph. Aue	erus		Stool Analysis
	Nails Swab	Nasal Swab	Throat Sawb	Salmonella	Parasites
Food Handler A	+	+	+	+	Ent. Histloytica
Food Handler B	+	-	-	-	Ent. Histloytica
Food Handler C	+	-	+	-	Ent. Histloytica+ Gardina Lam
Food Handler D	+	+	+	-	Ent. Histloytica

*Table 8: Lab results of Food Handlers, Alyotmah FPO March-2017(n=4)* 

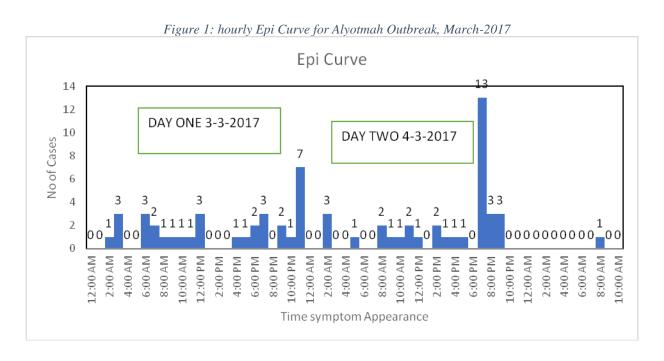
		Nasal	Throat	Nails	Stool
Stonh	Positive	50%	75%	100%	
Staph	Negative	50%	25%	0%	
Calmanalla	Positive				25%
Salmonella	Negative				75%

Table 9: Lab results of Patients Stool Analysis, Alyotmah FPO March-2017(n=11)

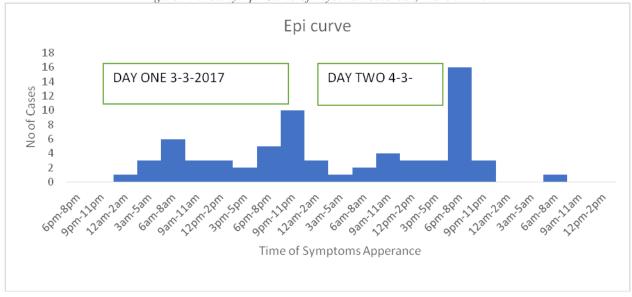
		Number	%	
Staph	Positive	3		27.27%
Stupii	Negative	. 8		72.73%
Salmonella	Positive	8		72.73%
Samonena	Negative	3		27.27%

Table 10: Lab Results of Food Item\Tools, Alyotmah FPO March-2017

Food items\tool	Staph. Aureus	Salmonella
Mayo-Sauce 1	-	-
Mayo-Sauce 2	-	-
Mayo-Sauce 3	+	+
Uncooked chicken sample 1	+	-
Uncooked chicken sample 2	+	+
Swabs from Mayo-Machine	-	+
Swabs from Tools	+	-







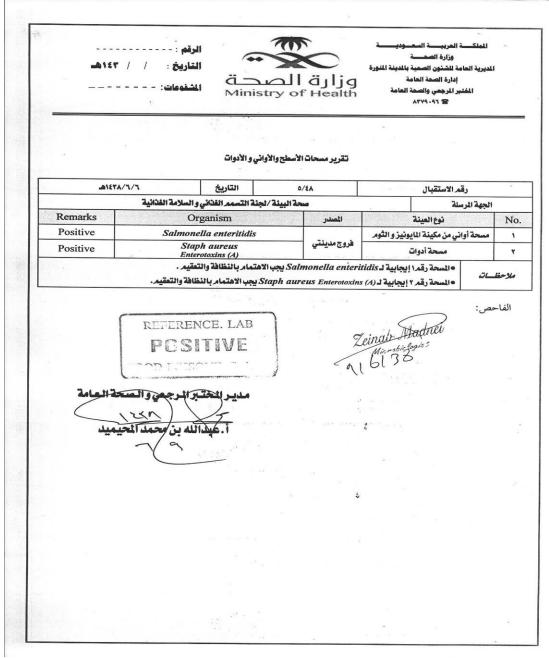
Appendix:

Appendix 1: Questionnaires Form

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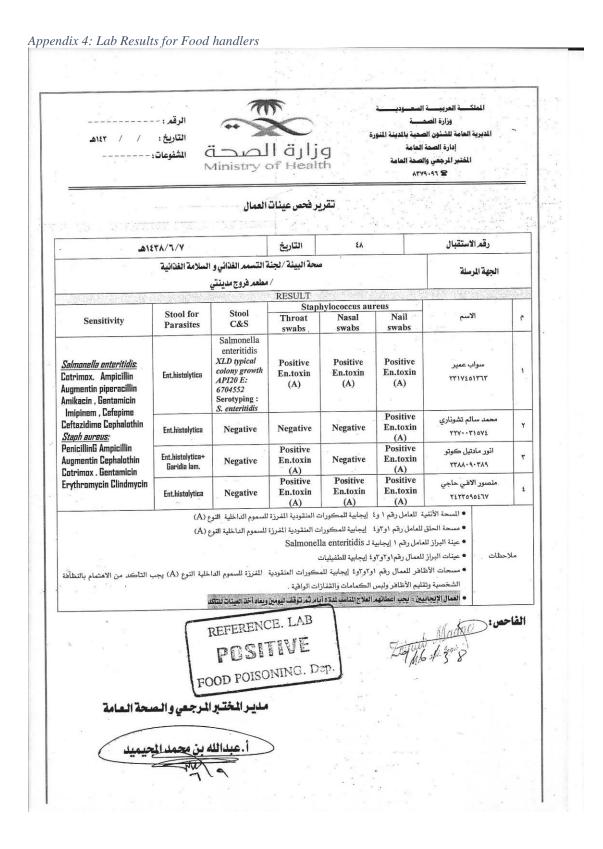
Page 17119 www.iajps.com

Appendix 2: Lab Results for Swabs of tools



Appendix 3: Lab Results for Food samples





Appendix 5: Lab Results for Stool of Patients الملكية العربيسة الس الرقم : - - - - - - -وزارة الصح التاريخ : الذيرية المامة للشنون الصحية بالدينة وزارة الصد ادارة الصحة العامة Ministry of Health الفتبر الرجعي والصحة العامة ATY4-47 8 تقرير عينات المرضى -A12TA/7/7 التاريخ رقم الاستقبال سحة البيئة / لجنة التسمم الفذائي والسلامة الفذائية الجهة الرسلة Results Culture media & Sensitivity Organism Sample Name Salmonella enteritidis: Stool سلطانة حماد Cotrimox. Ampicillin 1 Salmonella enteritidis Augmentin piperacillin XLD typical colony growth Stool إبراهيم ER12 ۲ Amikacin , Gentamicin API20 E: 6704552 Stool عانشة حميد lmipinem , Cefepime ۳ Serotyping: S. enteritidis Ceftazidime Only in samples number Stool طلال ER10 ٤ Cephalothin 1,2,3,4,5,6,10,11 Stool حصة حماد بخيت ٥ Stool ER8 ٦ Staph aureus Enterotoxins Staph aureus: (A) : Stool أمل 802-38 ٧ PenicillinG Ampicillin Mnitol S.A. growth - DNAse Augmentin Stool Positive -Coagulase Positive -غزيل عويد فهد ٨ Cephalothin Cotrimox RPLA Enterotoxins (A) Stool ER9 . Gentamicin Only in samples number Erythromycin Stool 1,10,11 عيدالرحمن مبارك 10 Clindmycin Stool عادل خالد السهلي 11 عينات المرضى رقم اولاولاولوووووا واوا إيجابية لـSalmonella enteritidis عينات المرضى رقم او ١٠و١١ إيجابية لـ (Staph aureus Enterotoxins ملاحظات Zeinab Hadnei مدير المختبر المرجعي والصحة العامة