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## **Evaluation of antibiotic resistance in salmonella isolated from camel in a Najaf province**

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**Abstract**--Non-Typhoidal Salmonellosis (NTS) is a form of salmonellosis that infect human from host-non adapted serotypes of salmonella, these salmonella strains are distributed in the environment, water and food via warm blooded animals causing gastroenteritis illnesses mostly, accompanied by high antibiotic resistance profile. In Najaf, camel meat considered one of the most used meat for human consumption. Yet, studies on distribution of salmonella and antibiotic resistance is scarce. In this research, (32) isolates of salmonella were isolated from camels in the province of Al-Najaf, using ordinary bacteriological methods, according to internationally known protocols for the cultivation and characterization of this bacteria. Then, all positive isolates subjected by antibiotic sensitivity test. The results showed a very high multidrug resistance profile was noticed among the isolated salmonella.

**Keywords**--Salmonella, identification, antibiotics sensitive test.

## **Introduction**

Salmonella species are gram-negative, non-spore-forming, predominantly motile, rod-shaped bacteria of the Enterobacteriaceae family (Crump et al., 2015). They are facultative anaerobes and chemotropism that invade the gastrointestinal tract. Salmonella is universal in the environment and is normally found in animals and human (Loynachan, 2005). Although some Salmonella is species-specific, most of the Salmonella species occur universally in all animal species and man and have been the subject of extensive studies worldwide (Nadi et al., 2020). Salmonella has many serotypes (>2600) and is capable of causing clinical illness in animals (Eng et al., 2015). Salmonella enterica a primary cause of community-acquired blood flow infections in many low- and Middle -income countries and in industrialized countries, S. Typhimurium and S. Enteritidis have emerged as leading sources of foodborne disease, with a main effect on the economy and public health (Gast & Porter, 2020; Hadi et al., 2022).

Animals with salmonella infection frequently remain Asymptomatic; though, stressed animals can spread the disease (Song & Zhu, 2021; Medhat and Aljanaby, 2022a). Zoonotic spread happens by direct or secondary interaction with animals, milk intake, meat eating, and/or unclean water. Donation and be informed abortion as the main appearance of the famous camel disease known in the Middle East and northern Africa and demonstrated its possible relationship with an unclassified salmonella infection (Abbas & Omer, 2005; Hadi and Aljanaby, 2022a). The WHO and the FAO are two international systems speaking the problem of evolving Salmonella infections. Salmonella serotyping is the main tool to comprehend the epidemiology of Salmonella infections and is recurrently used to trace back causes of contamination throughout an outbreak (Meneses, 2010; Hadi and Aljanaby, 2022b). In this study using an antibiotic resistance test for isolated salmonella bacteria during the study period and determine which of the antibiotics are more resistant ( Medhat and Aljanaby, 2022b).

## **Materials and Methods**

### **Sample collection**

Collection of samples: using rectal swab to collect the sample that take from the rectum of the large intestine of camel and was put it directly in (9 ml) selenite F broth in a tube and keep this tube in a box contain ice and the samples can save in box ice until six-hour and then transport to the laboratory during this period (Clyde et al., 1997). About (5-10) were slaughter per day and samples were taken from all slaughter camels.

### **Isolation and identification of salmonella**

After incubation the tube that contains samples at 37°C for 24h and then culturing in Salmonella-Shigella Agar (SS Agar) and incubated at 37°C for 24h (Tie et al., 2018). The colonies of salmonella appear black colonies (lactose negative with H<sub>2</sub>S making) on an agar. Selective culturing, the samples were passaged onto xylose lysine Deoxycholate (XLD) agar and incubated at 37 °C for 24–48 h. The colony appearance Colorless or light pink colonies with darker

centers. The presumptive Salmonella isolates were identified by two confirmatory biochemical tests, the triple-sugar-iron (TSI) agar test and the urease.

### Antibiotics resistance investigation

All isolated salmonella culturing by using Muller-Hinton agar by using sterile swabs and take from 24-48h. The antibiotics which used as thirteen antibiotics for determination of the resistance isolated bacteria.

Table 1  
Show antibiotics classis

Antibiotic class	Antibiotic name and content	code	Inhibition zone diameter (mm)(CLSI 2020)		
			S	I	R
Penicillins	Ampicillin (10µg)	AMP	≥17	14-16 <sup>^</sup>	≤ 13
	Amoxicillin (20/10 µg)	AML	≥18	14-17 <sup>^</sup>	13≤
Aminoglycosides	Gentamicin (10 µg)	CN	≥ 15	13-14 <sup>^</sup>	≤12
	Amikacin (30 µg)	AK	≥17	15-16 <sup>^</sup>	≤14
Folate pathway antagonists	sulfamethoxazole23.7 5µg	SMX	≥ 16	11-15 <sup>^</sup>	≤10
	Trimethoprim 1.25 µg	TM	≥ 16	11-15 <sup>^</sup>	≤10
Cephalosporin	Cephalexin (15 µg)	CL	≥ 15	-	≤ 14
	Ceftriaxone (30 µg )	CRO	≥ 23	20- 22 <sup>^</sup>	≤19
Macrolides	Erythromycin (15 µg)	E	≥ 23	14- 22 <sup>^</sup>	≤1 3
Tetracyclines	Tetracycline (30 µg)	TE	≥ 19	15- 18 <sup>^</sup>	≤14
Fluoroquinolones	Ciprofloxacin (5 µg)	CIP	≥ 12	16- 20 <sup>^</sup>	≤15
Phenicols	Chloramphenicol (30 µg)	C	≥ 18	1317- <sup>^</sup>	≤12
Quinolone	Nalidixic acid (30 µg )	NA	≥19	14- 18 <sup>^</sup>	≤13

### MDR and XDR

MDR is one of the important parameters to assess the extent of antibiotic resistance in a bacterial population, various concepts referred to as the "Multidrug Resistance" (MDR), the current applied Extensive Drug Resistance (XDR), in order to define high resistant profile for some isolates.

### Results and Discussion

Thirty two isolated bacteria was salmonella spp that give black colony on SS agar and XLD that resulting from formation of H<sub>2</sub>S and changing in the ground of

media from pink to yellow and pink to red respectively because of the fermentation of fructose and sucrose (A. & G., 2012).



Figure 1. Salmonella colonies on SS agar (A), XLD agar (B)

In TSI all isolates appear alkaline/ acid, black respectively with gas formation, yellow color resulting from fermentation glucose, black color result as formation of  $H_2S$  from iron of media and gas formation come from sucrose fermentation (Abdelwahab et al., 2019) (Lamas et al., 2018).



Figure 2. Salmonella on the TSI test

And in urea test all salmonella isolates were negative reaction (Gunasegaran et al., 2011) as in figure below



Figure 3. On Urease Salmonella showed negative results (no change in color)

### Antibiotics

We used thirteen antibiotics in this study for determination resistance of bacteria to antibiotic, three of antibiotics were highly susceptible for *Salmonella* is

(ceftriaxone CRO, amikacin AK and trimethoprim TM). Salmonella samples were 100% resist to six antibiotics (erythromycin E, amoxicillin AML, gentamycin CN, ampicillin AMP, tetracycline TE and cephalixin CL), while four antibiotics were moderately, resistant salmonella (Nalidixic acid NA, sulfamethoxazoline SMX, ciprofloxacin CIP and chloramphenicol C)( Figure 1).

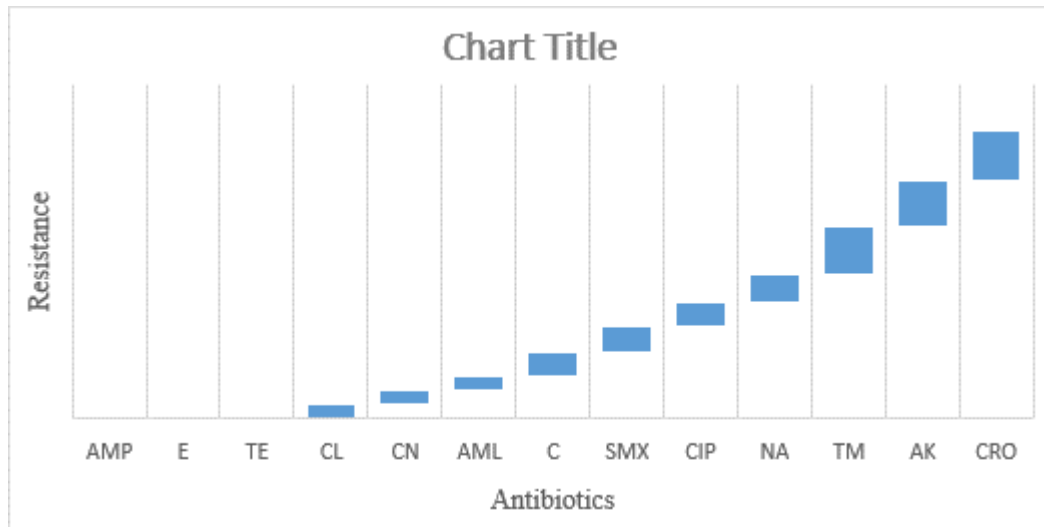


Figure 4. Antimicrobial Resistance profile, the row represent susceptible bacteria to antibiotics

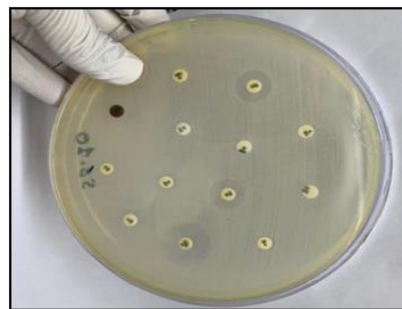


Figure 5. Inhibition zone antibiotic resistance

Table 2  
MDR and XDR

Multidrug resistance profile	No. (%) Of <i>Salmonella</i> isolates	No. of antibiotic classes (n=9)
MDR 18(56%)	2	3
	6	4
	5	5
	5	6
XDR 14(44%)	0	7
	7	8
	7	9

## Conclusion

The antibiotic resistance profile which has been adopted in this investigation presents a high MDR and XDR *Salmonella* phenotypes that circulating in camel in Al-Najaf province. Indicating a source of concern for the dissemination of antibiotic resistance trait in salmonella in camel, which might be also of an adverse effect on human health.

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