

PREVALENCE OF *SALMONELLAE* IN APPARENT HEALTHY AND DIARRHEIC CATTLE

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ABSTRACT

Salmonella enterica in production animals is predominantly a public health hazard. However certain serovars can cause clinical disease in animals and mostly young animals are at risk. Bacteriological examination of 640 faecal samples collected from apparent healthy (250) and diarrhoeic cattle (390) for the presence of *Salmonella* organisms revealed that isolation of 12 *Salmonella* isolates with an overall incidence 1.87 %. Four isolates were recovered from apparent healthy cattle with a percentage rate 1.6 %, while 8 isolates were recovered from diarrhoeic cattle with a percentage rate 2.05%.

Bacteriological examination of faecal samples taken from the cow calves (140) and buffalo calves (115) suffering from diarrhea revealed the isolation of *Salmonella* organisms with an incidence 3.6% and 1.7% respectively.

Salmonella were isolated from diarrhoeic cow with an incidence 1.5% while no *Salmonella* organisms could be isolated from diarrhoeic buffalo.

Regarding the healthy cattle, *Salmonella* organisms were recovered from cow and buffalo with an incidence 2.2% and 1.1% respectively, while it was isolated from cow calves with an incidence 2%.

It was noticed that the highest percentage rate for *Salmonella* recovery was in diarrhoeic cow calves followed by healthy cow.

Serological identification of *Salmonella* isolates revealed that the most prevalent serovars were *S. Typhimrium*(2) & *S. Belgdam*(2) and *S. Lomita*(2).

Followed by *S. Larochelle*, *S. Virchow*, *S. Infantis*, *S. Derpy* & *S. Anatum* and *S. Berzany* one isolate for each.

INTRODUCTION

Salmonellosis is an infectious disease of humans and animals caused by organisms of the two species of *Salmonella enterica* and *Salmonella bongori*. *Salmonella enterica* is divided into six subspecies, which are distinguishing by certain biochemical characteristics, susceptibility to lysis by bacteriophage *felix o* and DNA relatedness. These subspecies are, *Salmonella enterica* subspecies *enterica*, *S. enterica* subspecies *salamae*, *S. enterica* subspecies *arizonae*, *S. enterica* subspecies *diarizonae*, *S. enterica* subspecies *houtenae* and *S. enterica* subspecies *indica*. However subspecies *enterica* serovars account for more than 99.5% of isolated *Salmonella* strains. (**Grimont and Weill, 2007**).

Bovine salmonellosis ,is a costly disease to dairy producer on account of mortality, treatment expenses ,reduced milk yield and weight loss/ decreased weight gain within the herd as well as the hazard of transmission to humans either through food chain or direct animal contact (**Callaway et al., 2005, and Lorenz et al., 2011**). Calf diarrhea is the commonest disease in young calves and is the greatest cause of death (**Heinrichs and Radostits, 2001 and Smith et al., 2014**).

Salmonella spp. infection occurs when a susceptible dairy cattle ingest feed or water that has been contaminated with faeces from animals shedding the organisms. Salmonellosis has a wide spectrum of manifestation in cattle asymptomatic ,mild clinical or fulminant bactermia/septicemia and endotoxmeic infection can occure The manifestation vary with virulence of the strain ,infections dose and immunity of the host (*Mcguirk and Peek, 2003*).

The objective goal in this work was to estimate the prevalence of *Salmonella*.

In apparent healthy and diarrhoeic cattle and provide an update recognition of *Salmonella* serovares and their association with clinical status and age.

MATERIAL AND METHODS

Sampling:

A separate disposable plastic glove was used to collect faecal sample from terminal portion of the rectum of each animal. Each glove was inverted after sampling then tied and labelled . Samples were sent to the lab with a minimum of delay.

A total of 640 faecal samples were collected from apparent healthy cattle (250) and diarrhoeic cattle (390).The apparent healthy were cow-calves (50)buffalo –calves(30) cow (90) and buffalo(80), and the diarrhoeic cattle cow- calves (140) buffalo –calves (115) cow(65)and buffalo (70). These cattle were raised in different farms in EL-Kalubia, EL-Giza and Kafr El-sheikh Governorates.

Bacteriological examination:

About 10gm of faeces was inoculated in to 90 ml of both Selenite-F-broth and tetrathionate broth incubated at 37 °C. for 16-18 hrs, and 24hrs, respectively.

Then a loopful from the incubated broth was streaked on the surface of *Salmonella* Shigella (S.S) agar and Xylose Lysine Deoxycolate (XLD) agar,

Incubated at 37 °C for 24 hrs., the suspected isolates were purified and identified according to Quinn *et al.*, (2002). Further biochemical identification were done using api 20E according to manufacture instruction.

Serological identification:

Suspected *Salmonella* isolates were carried out according to White Kuffmann- Le Minor scheme described by Grimont and Weill (2007) using diagnostic polyvalent and monovalent somatic O and flagellar H (phase I and phase II). antisera SIFIN Berlinear Allee 317-321,13088 Berline,Germany.

RESULTS

Salmonella organisms are non lactose fermenting colony with or without H₂S production. The reaction on TSI gave alkaline slant/acid butt with or without H₂S, urease production negative and lysine decarboxylation positive. The results of api 20E shown in table (1) and fig (1).

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Table (1): The biochemical characterization of genus *Salmonella* using the api 20E system

Biochemical Test.	Genus <i>Salmonella</i>
ONPG	-ve
ADH	+ve
LDC	+ve
ODC	+ve
CIT	+ve
H ₂ S	+ve
URE	-ve
TDA	-ve
IND	-ve
VP	-ve
GEL	-ve
GLU	+ve
MAN	+ve
INO	+ve
SOR	+ve
RHA	+ve
SAC	-ve
MAL	+ve
AMY	-ve
ARA	+ve
OXY	-ve
NO ₂	+ve



Fig.(1): Showing biochemical identification of *Salmonella* organism on api 20E.

Bacteriological examination of 640 faecal samples collected from healthy and diarrhoeic cattle revealed isolation of 12 *Salmonella* isolates with an overall incidence 1.87% from which 4 isolates were recovered from healthy cattle with a percentage rate 1.6 %, while 8 isolates were recovered from the diarrhoeic cattle with a percentage rate reaching 2.05% (table,2).

Table (2): Prevalence of *Salmonella* organisms in the apparent healthy and diarrhoeic cattle.

Animal	No of the positive	%*
Apparent Healthy (250)	4	1.6
Diarrhoeic cattle (390)	8	2.05
Total (640)	12	1.87**

*The percentage is calculated according to no of each clinical case.

**The percentage is calculated according to total number of cattle.

Bacteriology examination of faecal samples taken from the calves (cow 140 and buffalo 115) suffering from diarrhea revealed the isolation of *Salmonella* organisms with an incidence 3.6% and 1.7% respectively. meanwhile

Salmonella were isolated from cow with an incidence 1.5% while no *Salmonella* organisms could be isolated from diarrhoeic buffalo.

Regarding the healthy cattle, *Salmonella* organisms were recovered from cow and buffalo with an incidence 2.2% and 1.1% respectively, While it was isolated from cow calves with an incidence 2%. It was noticed that no *Salmonella* could be recovered from healthy buffalo calves. (table,3).

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Table (3): frequency distribution of *Salmonella* organisms relation to age in apparent healthy and diarrhoeic cattle.

Animal	Apparent healthy			Diarrhoeic animals			Total	
	No	+ve	%	No	+ve	%	no	%
Cow Calves	50	1	2	140	5	3.6	6/190	3.2
Buffalo calves	30	0	0	115	2	1.7	2/145	1.4
Cow	90	2	2.2	65	1	1.5	3/155	1.9
Buffalo	80	1	1.1	70	0	0	1/150	0.7
Total	250	4	1.6	390	8	2.05	12/640	1.87

* The percentage is calculated according to the no. of each clinical case.

** The percentage is calculated to the total no. of each kind of animal.

*** The percentage is calculated to the total no. of animal.

Serological identification using polyvalent and monovalent “O” and “H” antisera revealed that the most prevalent serovars was *S.Typhimurium* (2) & *S.Belgdam* (2) and *S.Lomita*(2). Followed by *S. Larochele*, *S. Virchow*, *S.Infantis* , *S.Derpy* & *S.Anatum* and *S.Berzany* one isolate for each.

Table (4): Antigenic structure of isolated *Salmonella* serovars.

Identified strain	AntigenicStructure		
	Somatic (o)	Flageller	
		Phase 1	Phase 2
Blegdam	9,12	g,m,q	-----
Lomita	6,7	e ,h	1,5
Typhimurium	1,4,[5],12	i	1,2
Larochele	6,7	e,h	1,2
Virchow	6,7,14	r	1,2
Berzany	1,4,12,27	d	1,6
Infantis	6,7,14	r	1,5
Derpy	1,4[5],12	f,g	[1,2]
Anatum	3,10	e,h	1,6

The results illustrated in table (5) showed that *S.Larochelle* and *S.Infantis* were recovered from apparent healthy cow while *S.Derpy* was recovered from apparent healthy buffalo.

It was noticed that *S.Belgdam* was isolated from both apparent healthy and diarrhoeic cow calves on the other hand *Salmonella* Typhimurium could be isolated from both diarrhoeic cow calves and cow. Moreover *S.Lomita*(2) and *S.Virchow*(1) were isolated from diarrhoeic cow calves.

S.Anatum and *S.Berzany* were isolated from buffalo calves while no *Salmonella* could be isolated from diarrhoeic buffalo.

Table (5): Serological identification of *Salmonella* isolates in relation to age and cattle status.

Animal	Apparent healthy			Diarrhoeic animal			Total	
	Serovars	no	%*	Serovars	no	%*	no	%**
Cow calves	<i>S.Belgdam</i>	1/1	100	<i>S.Lomita</i>	2/5	40	6	50
				<i>S.Belgdam</i>	1/5	20		
				<i>S.Typhimurium</i>	1/5	20		
				<i>S.Virchow</i>	1/5	20		
Buffalo calves	-----	0	0	<i>S.Anatum</i>	1/2	50	2	16.7
				<i>S.Berzany</i>	1/2	50		
Cow	<i>S.Larochelle</i>	1/2	50	<i>S.Typhimurium</i>	1/1	100	3	25
	<i>S.Infantis</i>	1/2	50					
Buffalo	<i>S.Derpy</i>	1/1	100	-----	0	0	1	8.3

*The percentage is calculated according to number of *Salmonella* serovars recovered in each kind of animal

** The percentage is calculated according to the total no of isolates (12).

DISSCUSSION

Salmonella infection occurs worldwide in both developed and developing countries and are major contributors to morbidity and economic costs (*Antoine et al., 2008*). *Salmonella* organisms are frequently isolated from environmental sources that serve as a relay for the bacteria and play a major role in its spread between different hosts (*Lilje bjelke et al., 2005*). However there are limited national surveillance programs for *Salmonella* in cattle compared to poultry.

The study was conducted on 640 cattle, (250 apparent healthy and 390 had enteritis and diarrhea). As shown in table (2). Twelve *Salmonella* isolates were recovered from cattle with an overall incidence 1.87%. Four isolates were recovered from apparent healthy cattle with a percentage rate 1.6%. This finding is lower than that reported by (*Berg, 2011*) who said that 2.6% were positive for *Salmonella* with microbiological culture and declared that *Salmonella* infection can be present without clinical signs in cow /calves and (*Aleslamboly, Y., 2011*) who recovered *Salmonella* from rectal swabs of apparently healthy cattle with a percentage rate 4.4%.

It was illustrated in table (3) that *Salmonella* organisms were recovered from 5 out of 140 cow calves had diarrhoeic with an incidence 3.6% while 2 out of 115 were recovered from buffalo calves with an incidence 1.7%. A higher rate were recovered by *Zaki (1994)* who estimated the prevalence of *Salmonella* fecal culture in diarrhoeic calves to be 8.9%. On the other hand *Sobhi (1997)* found that 14 fecal samples were positive for *Salmonella* out of 351 with an incidence 4% that including 4 *Salmonella* isolates recovered from diarrhoeic calves and the remaining 10 were from apparent healthy calves.

Salmonella shedding in cows occurred in 31% of 105 daires (**Huston et al., 2002**). one isolate could be recovered from 80 apparently healthy buffalo and no isolates could be recovered from diarrhoeic one. It was noticed that the rate of *Salmonella* isolation was some what higher in cow calves than buffalo calves 3.2% versus 1.4% and cow than buffalo 1.9% versus , 0.7 respectively .**Vanselow et al. (2007)** reported that dairy cattle were significantly more likely to shed *Salmonella* in faeces than pasture beef cattle. The variable results of *Salmonella* incidence here could be attributed to several factors including difference in standard of nutrition, presence of stress factors, medication used and time of sampling as *Salmonella* organisms are intermittently excreted by the affected animal in addition to collecting of faecal samples during the chronic phase also lead negative culture results.

Serological identification of *Salmonella* isolates revealed that the most prevalent *Salmonella* serovars were Typhimurium (2) & Blegdam (2) and Lomita (2) followed by *S.Larochelle*, *S.Virchow*, *S.Infantis*, *S.Anatum*, *S.Derpy* and *S.Berzany* (one isolates for each). As illustrated in table (5) it was noticed that *S.Typhimurium* was recovered from both diarrhoeic calf and cow and *S.Belgdam* was recovered from both diarrhoeic calf and apparently healthy cow while *S.Lomita* were recovered from 2 diarrhoeic cow calves these may be due to cycling and shedding .In Egypt *S.Typhimurium* and *S.Anatum* were recovered by (**Zaki, 1994;** **Sobhi, 1997 and Ammar, 2014**), while *S.Typhimurium* and *S.Enteritidis* were recovered by (**Aleslamboly, Y., 2011**).

Although bovine salmonellosis affects cattle of all ages it was clear that calves are more susceptible to infection than adults.It is also known that newborn calves could be infected with salmonellae at time of parturition or sooner after birth. Subclinical infected dairy cattle act as asymptomatic shedders and could be source of infection for both animal and human.

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