

4. RESULTS

4.1. Identification of *Salmonella* species:

4.1.1. Morphological and colonial characteristics of *Salmonella* isolates:

In the present study, a total 1073 samples were studied for salmonellosis. Red colonies with black center on XLD agar media and white colonies with black center on S.S agar media were considered as *Salmonella* suspected colonies. Separated colonies were picked up on trypticase soya agar slants. All isolates were Gram negative and non sporulated bacilli.

4.1.1. Biochemical identification:

The purified suspected *Salmonella* isolates were examined using different biochemical reactions as indicted in Table (4).

Table (4): Biochemical identification of purified suspected *Salmonella* isolates.

Test	Result
Methyl red	+ ve
Voges Proskaur	- ve
Oxidase	- ve
H ₂ S production	+ ve
Citrate utilization	+ ve
Urea hydrolysis	- ve
Indole	+ ve
Lactose fermentation	- ve
Maltose fermentation	+ ve
Sorbitol fermentation	+ ve

Arabinose fermentation	+ ve
Dulcitol fermentation	Variable

+ ve= positive

- ve=negative

4.2. Results of incidence of *Salmonella* species in broiler chickens:

A total number of 1073 broiler chicken samples representing; 414 apparently healthy, 157 diseased and 502 dead birds were bacteriologically examined to detect the presence of *Salmonella* spp.

As shown in Table (5), the number of positive *Salmonella* samples that collected from four flocks containing 414 apparently healthy birds was 9 isolates that representing percentages of 2.2.

Table (6) shows the incidence of *Salmonella* spp. that isolated from 157 samples representing four diseased broiler chicken flocks with whitish watery diarrhea. The total positive *Salmonella* isolates were 15 with percentage of 9.6.

The incidence of *Salmonella* spp. that detected from collection of 502 dead broiler chicken samples is illustrated in Table (7). Seventeen isolates of *Salmonella* were detected that representing 3.4%.

Collectively, the incidence of *Salmonella* from the apparently healthy, diseased and dead broiler chickens in each flock was seen in Table (8). The percentages of positive samples in flock 1, 2, 3 and 4 were 2.76, 3.75, 5.32 and 4.12, respectively.

Table (5): Incidence of *Salmonella* spp. isolated from apparently healthy broiler chicken flocks.

Flocks	Type of samples	<i>Salmonella</i> positive samples		
		No. of total samples	No. of positive samples	%*
1	Cloacal swabs	39	2	5.12
	Gall bladder	30	0	0
	Yolk sac	13	0	0
	Spleen	30	0	0
	Liver	30	1	3.3
	Total	142	3	2.1
2	Cloacal swabs	35	1	2.9
	Gall bladder	13	0	0
	Yolk	8	0	0
	Spleen	13	0	0
	Liver	13	0	0
	Total	82	1	1.2
3	Cloacal swabs	31	1	3.2
	Gall bladder	15	0	0
	Yolk sac	9	1	11.1
	Spleen	15	0	0
	Liver	15	0	0
	Total	85	2	2.4
4	Cloacal swabs	37	2	5.4
	Gall bladder	21	0	0
	Yolk sac	5	0	0
	Spleen	21	0	0
	Liver	21	1	4.8
	Total	105	3	2.8
Total/all flocks		414	9	2.2

*: Calculated according to the total No. of each type of samples.

Table (6): Incidence of *Salmonella* spp. isolated from diseased broiler chicken flocks.

Flocks	Type of samples	<i>Salmonella</i> positive samples		
		No. of total samples	No. of positive samples	%*
1	Cloacal swabs	8	1	12.5
	Gall bladder	4	0	25
	Yolk sac	5	1	20
	Spleen	4	0	0
	Liver	4	1	20
	Total	25	3	12
2	Cloacal swabs	20	1	5
	Gall bladder	17	1	5.8
	Yolk	0	0	0
	Spleen	17	1	5.8
	Liver	17	2	11.7
	Total	71	5	7.04
3	Cloacal swabs	7	1	14.3
	Gall bladder	5	1	20
	Yolk sac	2	0	0
	Spleen	5	1	20
	Liver	5	1	20
	Total	24	4	16.6
4	Cloacal swabs	8	2	25
	Gall bladder	8	0	0
	Yolk sac	5	0	0
	Spleen	8	0	0
	Liver	8	1	12.5
	Total	37	3	8.1
Total/all flocks		157	15	9.6

*: Calculated according to the total No. of each type of samples.

Table (7): Incidence of *Salmonella* spp. isolated from dead broiler chicken flocks.

Flocks	Type of samples	<i>Salmonella</i> positive samples		
		No. of total samples	No. of positive samples	%*
1	Cloacal swabs	37	2	5.4
	Gall bladder	37	0	0
	Yolk sac	10	0	0
	Spleen	37	0	0
	Liver	37	1	5.4
	Total	158	3	1.9
2	Cloacal swabs	33	0	3.1
	Gall bladder	33	1	3.1
	Yolk	8	1	12.5
	Spleen	33	2	6.1
	Liver	33	1	3.1
	Total	140	5	3.4
3	Cloacal swabs	18	0	0
	Gall bladder	18	1	5.5
	Yolk sac	7	1	14.2
	Spleen	18	2	11.1
	Liver	18	0	0
	Total	79	4	5.1
4	Cloacal swabs	37	0	0
	Gall bladder	27	0	0
	Yolk sac	7	0	0
	Spleen	27	3	3.7
	Liver	27	2	3.7
	Total	125	5	4
Total/all flocks		502	17	3.4

* : Calculated according to the total No. of each type of samples.

Table (8): The incidence of *Salmonella* spp. among different examined birds in each flock.

Flocks	Total No. of samples	No. of positive samples	% of positive * samples
1	325	9	2.76
2	293	11	3.75
3	188	10	5.32
4	267	11	4.12
Total/all flocks	1073	41	3.8

*: calculated according to the total No. of samples of each flock

4. 3. Results of serotyping of isolated *Salmonellae*:

The results of serotyping of *Salmonellae* strains are observed in Table (9). A total of 41 strains were serotyped as 13 (*S. Enteritidis*), 8 (*S. Infantis*), 6 (*S. Kentucky*), 3 (*S. Chiredzi*), 7 (*S. Typhimurium*) and 4 (*S. Tsevie*).

Table (10) and Figure (1) shows the percentages of *Salmonella* serovars. The results revealed that the percentages were 31.7, 19.5, 14.6, 7.3, 17.1 and 9.8 for *S. Enteritidis*, *S. Infantis*, *S. Kentucky*, *S. Chiredzi*, *S. Typhimurium* and *S. Tsevie*, respectively.

Table (9): Serotypes of *Salmonella* strains.

Serovars	No. of strain	Group	Antigenic structure		
			Somatic (O) antigen	Flagellar (H) antigen	
				Phase I	Phase II
<i>S. Enteritidis</i>	13	D1	9	g, m	-
<i>S. Infantis</i>	8	C ₁	6,7	R	1,5
<i>S. Kentucky</i>	6	C ₂	8,20	I	z ₆
<i>S. Chiredzi</i>	3	F	11	C	1,5
<i>S. Typhimurium</i>	7	B	4,5,12	I	1.2
<i>S. Tsevie</i>	4	B	4,5	I	e.n.z ₁₅
Total	41				

Table (10): The percentage of *Salmonella* serovars.

Serotype	No. of strain	%*
<i>S. Enteritidis</i>	13	31.7
<i>S. Infantis</i>	8	19.5
<i>S. Kentucky</i>	6	14.6
<i>S. Chiredzi</i>	3	7.3
<i>S. Typhimurium</i>	7	17.1
<i>S. Tsevie</i>	4	9.8

*: calculated according to the total No. of isolates

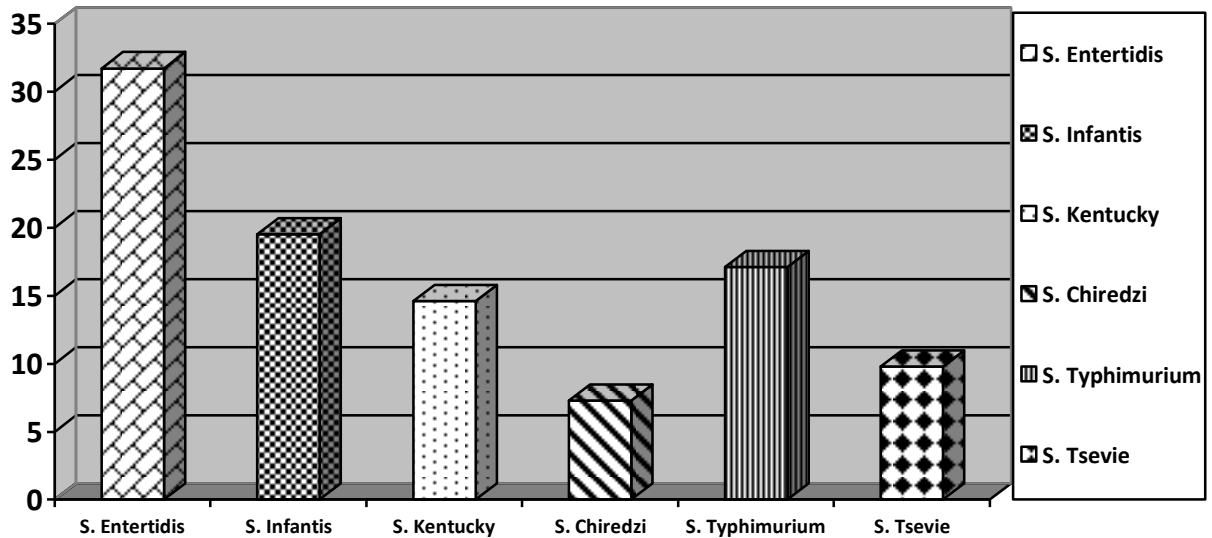


Figure (1): The percentage of *Salmonella* serovars.

Regarding the types, numbers and percentages of *Salmonella* serovars that isolated from each flock, Table (11) shows this result. It was observed that from flock (1), *S. Enteritidis*, *S. Typhimurium* and *S. Chiredzi* were detected with percentages of 2.2, 0.31 and 0.31, respectively. Flock (2) showed *S. Enteritidis* (1.4), *S. Infantis* (1.02), *S. Kentucky* (1.02) and *S. Tsevie* (0.341). In flock (3), *S. Enteritidis*, *S. Typhimurium*, *S. Kentucky* and *S. Tsevie* were the predominant servers in percentages 1.1, 2.1, 0.53 and 1.6, respectively, while *S. Chiredzi*, *S. Infantis*, *S. Kentucky* and *S. Typhimurium* were recovered in flock (4) in percentages of 0.75, 1.9, 0.75 and 0.75, respectively.

Table (11): The incidence of *Salmonella* serovars in the examined flocks.

Types of samples	No. of examined samples	<i>Salmonella</i> serovars		
		Serovars	No.	%*
Flock 1	325	<i>S. Entertidis</i>	7	2.2
		<i>S. Typhimurium</i>	1	0.31
		<i>S. Chiredzi</i>	1	0.31
Flock 2	293	<i>S. Entertidis</i>	4	1.4
		<i>S. Infantis</i>	3	1.02
		<i>S. Kentucky</i>	3	1.02
		<i>S. Tesvie</i>	1	0.34
Flock 3	188	<i>S. Entertidis</i>	2	1.1
		<i>S. Typhimurium</i>	4	2.1
		<i>S. Kentucky</i>	1	0.53
		<i>S. Tesvie</i>	3	1.6
Flock 4	267	<i>S. Chiredzi</i>	2	0.75
		<i>S. Infantis</i>	5	1.9
		<i>S. Kentucky</i>	2	0.75
		<i>S. Typhimurium</i>	2	0.75
Total	1073		41	3.8

* : calculated according to number of examined samples.

4.4. The result of the effect of the locally prepared *S. Enteritidis* bacterin and the probiotic treatment of broiler chickens:

4.4.1. The quality control tests of the locally prepared *S. Enteritidis* bacterin:

The result of the quality control tests of the locally prepared *S. Enteritidis* bacterin revealed that the bacterin was pure (contained only Gram negative short rods) as confirmed by Gram staining. The bacterin was completely inactivated as there was no growth of it on the selective media. Also, the bacterin was sterile without fungal or mycoplasmal growth as indicated by absence of growth on SDA and PPLO agar, respectively. No local reaction, signs on deaths were observed after bacterin inoculations in birds indicating its safety.

4.4.2. The effect of the locally prepared *S. Enteritidis* bacterin and the probiotic treatment against *S. Enteritidis* challenge in broiler chickens:

The results of experimental infection of broiler chickens at 20 days of age after vaccination with locally prepared *S. Enteritidis* bacterin and treatment with probiotic revealed that infected groups (2, 3 and 4) manifested signs of depression, anorexia and watery diarrhea 3 days post challenge.

The morbidity rates were 80, 30 and 20% in challenged non-treated, vaccinated and probiotic treated infected groups, respectively.

Table (12) reveals that the mortality rate was 30.67% in infected non-treated group, 5.33% in vaccinated infected birds and 12% in probiotic treated infected ones.

Dead birds were subjected to post mortem examination for specific *S. Enteritidis* lesions. The most predominant lesions were congestion of muscles and the internal organs and congestion of the intestinal mucosa with mucoid intestinal contents.

Table (12) and Figure (2) illustrates the protection rate of the locally prepared inactivated *S. Enteritidis* bacterin and the probiotic treatment against *S. Enteritidis* experimental infection in broiler chickens. The results cleared that

there was a significant ($p \leq 0.05$) difference between the infected not treated group and the vaccinated with bacterin and the treated with probiotic. The protection rate of the locally prepared **S. Enteritidis** bacterin and the probiotic were (82.61%) and (60.87%), respectively which was significantly ($p \leq 0.05$) higher than infected non treated birds (0%).

Table (12): The protection rate of the locally prepared inactivated *S. Enteritidis* bacterin and the probiotic treatment against *S. Enteritidis* infection in broiler chickens.

Groups	Total No. of birds	No. of dead birds/week post challenge			Total No. of dead birds	Mortality rate	Total No. of survived birds	Protection %
		1 st week	2 nd week	3 rd week				
Blank control	75	0	0	0	0	0	75	100 ^a
Infected, non-treated	75	17	5	1	23	30.67	52	0 ^d
Vaccinated and infected	75	3	1	0	4	5.33	71	82.61 ^b
Probiotic treated and infected	75	6	3	0	9	12	66	60.87 ^c

$$\text{Protection rate} = \frac{\text{Survived test-survived control positive}}{\text{Dead control positive}} \%$$

*different letters within the same column were significantly difference at (P≤0.05).

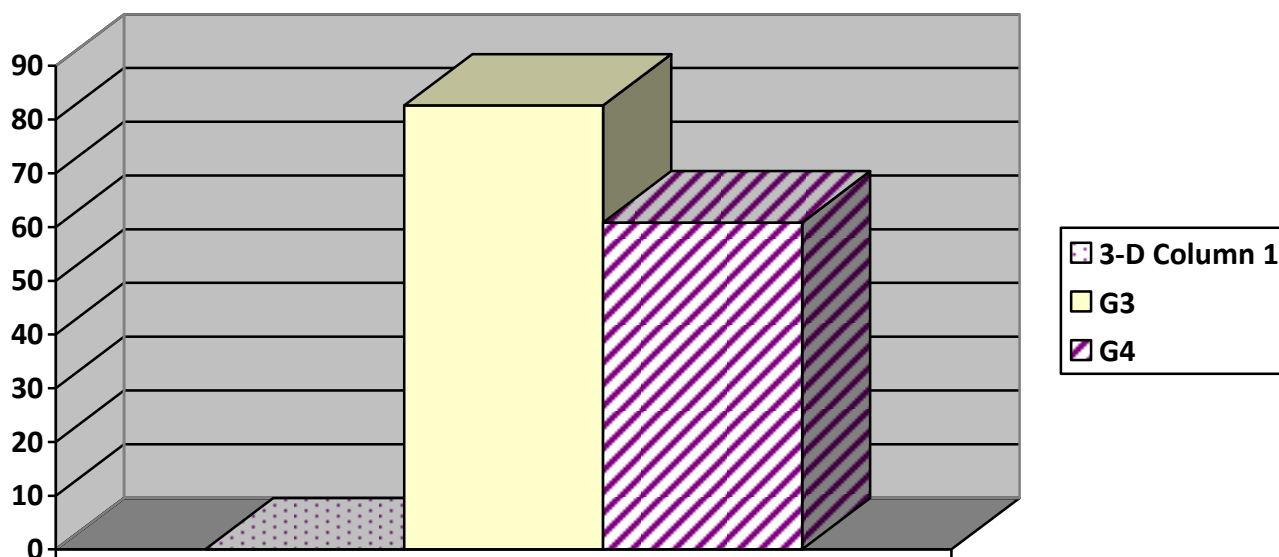


Figure (2): The protection rate of the locally prepared inactivated *S. Enteritidis* bacterin and the probiotic treatment against *S. Enteritidis* infection in broiler chickens.

4.4.3. The faecal shedding rate of *S. Enteritidis* in broiler chickens after vaccination with the locally prepared *S. Enteritidis* bacterin and treatment with probiotic:

Table (13) and Figure (3) demonstrates the faecal shedding rate of *S. Enteritidis* in broiler chickens after vaccination with locally prepared bacterin and treated with probiotic.

It was declared that there were significant ($p \leq 0.05$) differences between the vaccinated and probiotic treated birds and the infected non treated ones along 3 weeks observation period. Gradual significant ($p \leq 0.05$) decrease in the shedding rate was observed within each group until reaching the last week of observation period. The faecal shedding rate in the vaccinated birds collectively was 9.24% while in the probiotic ones was 17.5% which were significantly ($p \leq 0.05$) lower than infected non treated group (40.6%).

Table (13): The faecal shedding rate of *S. Enteritidis* in broiler chickens after vaccination with the locally prepared bacterin and treatment with probiotic.

Treatment group	Weeks post challenge						+ Ve/Total	%
	1 st wk		2 st wk		3 st wk			
	+ Ve/Total	%	+ Ve/Total	%	+Ve/Total	%		
Blank control	0/75	0^{Ad}	0/75	0^{Ad}	0/75	0^{A d}	0	0^d
Infected, non-treated	30/58	51.72^{Aa}	16/43	37.21^{Ba}	8/32	25^{Ca}	54/133	40.6^a
Vaccinated and infected	13/72	18.06^{Ac}	4/61	6.56^{Bc}	0/51	0.0^{Cc}	17/184	9.24^c
Probiotic treated and infected	18/69	26.08^{Ab}	10/56	17.86^{Bb}	2/46	4.35^{Cb}	30/171	17.5^b

*different small letters within the same column were significantly difference at (P≤0.05).

*different capital letters within the same row were significantly difference at (P≤0.05).

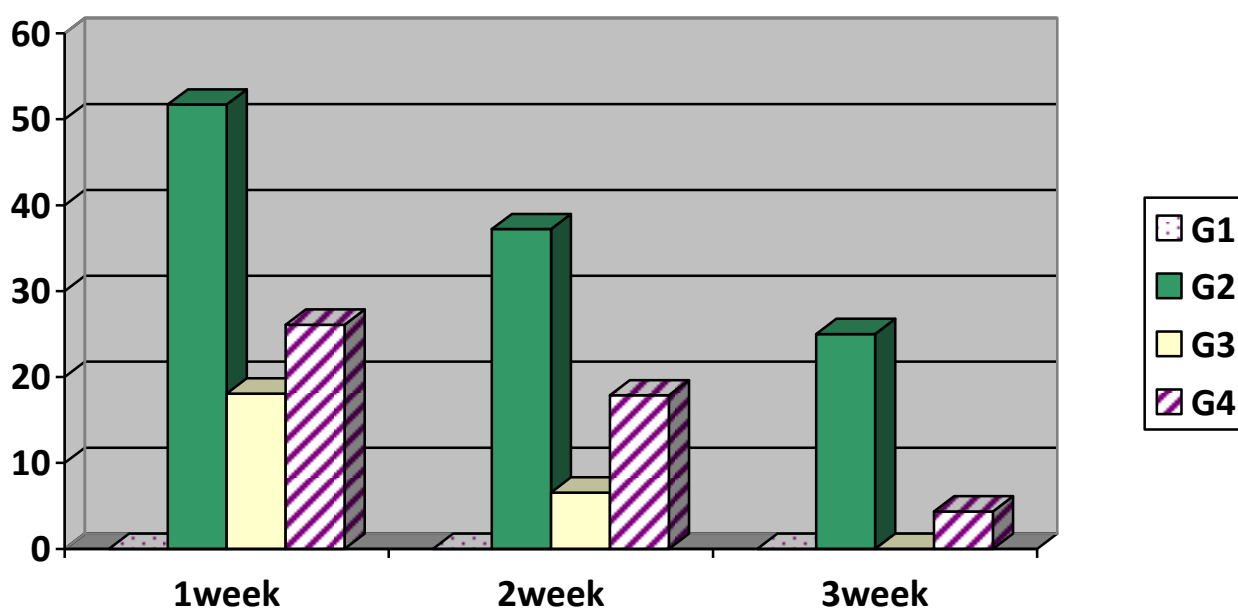


Figure (3): The faecal shedding rate of *S. Enteritidis* in broiler chickens after vaccination with the locally *S. Enteritidis* prepared bacterin and treatment with probiotic.

4.4.4. Re-isolation rate of *S. Enteritidis* from different organs of sacrificed broiler chickens after vaccination with the locally prepared *S. Enteritidis* bacterin and treatment with probiotic:

The results of the re-isolation rate of *S. Enteritidis* from different organs of sacrificed broiler chickens after vaccination with the locally prepared bacterin and treatment with probiotic are seen in Table (14) and Figure (4). These results indicated that along the whole three weeks observation period, the highest significant ($p \leq 0.05$) re-isolation rate was in the infected non treated group (46.67%), but this percentage was significantly ($p \leq 0.05$) lower in the probiotic treated birds (21.67%) until it reached to its lowest significant ($p \leq 0.05$) level (11.67%) in the vaccinate group.

Table (14): The re-isolation rate of *S. Enteritidis* from different organs of sacrificed broiler chickens after vaccination with the locally prepared bacterin and treatment with probiotic.

Group	Re-isolation rate															Total (%)
	At the end of 1 st Wk post challenge					At the end of 2 nd Wk post challenge					At the end of 3 rd Wk post challenge					
	Examined Organ					Examined Organ					Examined Organ					
	Liver	Heart	Spleen	Cecum	Total	Liver	Heart	Spleen	Cecum	Total	Liver	Heart	Spleen	Cecum	Total	
Blank control	0	0	0	0	0 ^d	0	0	0	0	0 ^d	0	0	0	0	0 ^d	0 ^d
Infected, non-treated	7/10	5/10	6/10	8/10	26/40 65 ^{Aa}	4/10	4/10	5/10	6/10	19/40 47.5 ^{Ba}	2/10	1/10	4/10	4/10	11/40 27.5 ^{Ca}	56/120 46.67 ^a
Vaccinated and infected	2/10	2/10	2/10	3/10	9/40 22.5 ^{Ac}	1/10	0/10	1/10	2/10	4/40 10 ^{Bc}	0/10	0/10	0/10	1/10	1/40 2.5 ^{Cc}	14/120 11.67 ^c
Probiotic treated and infected	3/10	2/10	3/10	4/10	12/40 30 ^{Ab}	3/10	2/10	2/10	3/10	10/40 25 ^{Ab}	1/10	0/10	1/10	2/10	4/40 10 ^{Bb}	26/120 21.67 ^b

*different small letters within the same column were significantly difference at (P≤0.05).

*different capital letters within the same row were significantly difference at (P≤0.05).

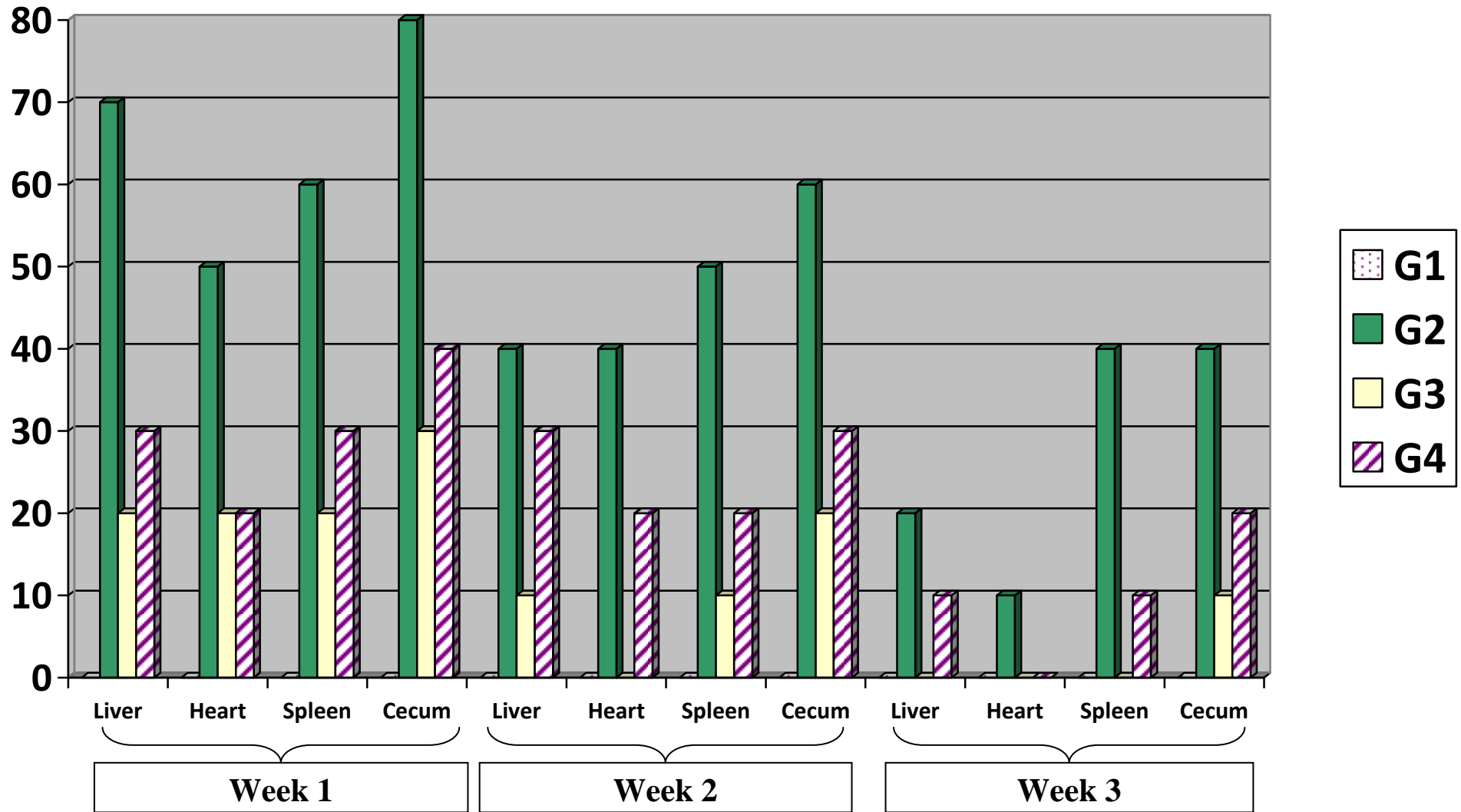


Figure (4): The re-isolation rate of *S. Enteritidis* from different organs of sacrificed broiler chickens after vaccination with the locally prepared bacterin and treatment with probiotic.

4.4.5. Average body weight, cumulative feed conversion and EPEF in broiler chickens after vaccination with the locally prepared *S. Enteritidis* bacterin and treatment with probiotic:

Concerning the results of the performance of each group after vaccination with the locally prepared *S. Enteritidis* bacterin and treatment with probiotic, they are detected in Table (15) and Figure (5).

The measured parameters were the average body weight, the cumulative feed conversion (CFC) and the European Production Efficiency Factor (EPEF).

It was showed that EPEF was significantly ($p \leq 0.05$) improved in the vaccinated and treated groups than infected non treated one along the whole coarse of the study (6 weeks of age). The best CFC rate was observed in the vaccinated and probiotic treated chickens (1.89 and 1.78, respectively), while the worst one was in the infected non treated one (2.43).

Table (15): Average body weight, cumulative feed conversion and European production efficiency factor in broiler chickens after vaccination with the locally prepared *S. Enteritidis* bacterin and treatment with probiotic.

Treatment Group	Average body weight/g						CFC	EPEF**
	Age/week							
	Before challenge			After challenge				
	1	2	3	4	5	6		
Blank control	133.40±6.30 ^a	291.1±5.21 ^a	563.75±11.86 ^b	744.12±24.77 ^b	984.1±42.21 ^b	1580.2±65.22 ^b	2.03	189.12
Infected, non-treated	128.11±5.31 ^a	290.1±8.679 ^a	501.1±18.0 ^c	699.22±19.63 ^c	801.50±13.50 ^c	1355.1±54.10 ^c	2.43	148.25
Vaccinated and infected.	134.73±2.77 ^a	293.1±8.7 ^a	589.1±18.21 ^{ab}	799.90±12.10 ^{ab}	1005.0±71.00 ^{ab}	1640.1±55.91 ^{ab}	1.89	205.44
probiotic treated and infected.	130.43±3.62 ^a	288.2±9.90 ^a	599.1±21.8 ^a	810.8±21.3 ^a	1100.1±55.3 ^a	1705±21.50 ^a	1.78	213.44
LSD	16.45	25.2	30.1	63.48	115.80	119.75		

CFC= Cumulative feed conversion

EPEF** = European Production Efficiency Factor. The higher the value, the better the performance.

LSD= Least significant difference as determined by Fisher's protected LSD procedures.

*Means within the column with different letters are significantly different ($p \leq 0.05$).

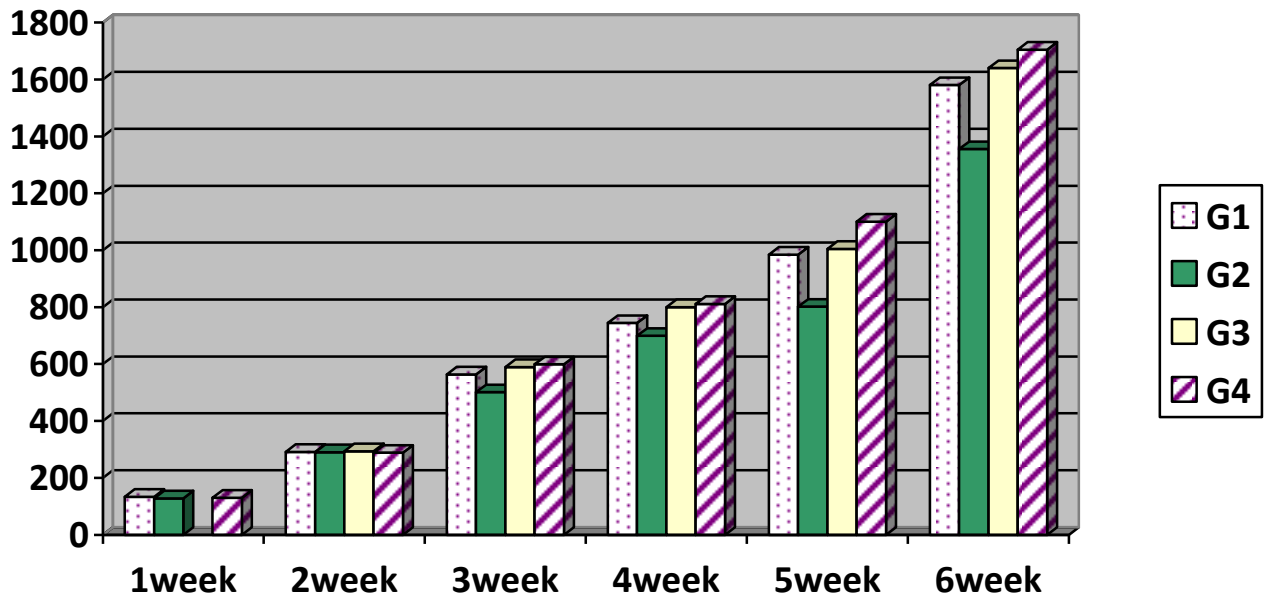


Figure (5): Average body weight, cumulative feed conversion and European production efficiency factor in broiler chickens after vaccination with the locally prepared *S. Enteritidis* bacterin and treatment with probiotic.

4.4.6. Micro-agglutination antibody titers in the sera of broiler chickens after vaccination with the locally prepared *S. Enteritidis* bacterin and treatment with probiotic:

Table (16) clarifies the titer of antibodies against *S. Enteritidis* after vaccination with the locally prepared bacterin or treatment with probiotic using the micro-agglutination (MA) test. After the 1st dose of vaccine and treatment with the probiotic, the geometric mean titer (GMT) of antibodies increased to reach 65 in vaccinated group and 60.6 in probiotic group. After booster dose of the vaccine (before challenge), the GMT increased to reach 98 and 74.5 in vaccinated and probiotic treated birds, respectively.

One week after *S. Enteritidis* challenge, the GMT increased to reach 60.6, 211.1 and 113.1 in the infected non-treated, vaccinated and probiotic treated groups, respectively.

Two weeks after challenge, the GMT of antibodies increased to 130, 226.2 and 197 in infected non-treated, vaccinated and probiotic treated groups, respectively.

At the 3rd week post *S. Enteritidis* challenge, the antibodies GMT increased in the infected non-treated chickens and vaccinated ones to 139.3 and 242.5, respectively, but it declined in the probiotic treated birds to 171. **discussion** هذا الكلام معاد كما هو هي ال

Table (16): Micro-agglutination antibody titers in sera of broiler chickens after vaccination with the locally prepared *S. Enteritidis* bacterin and treatment with probiotic.

Age/ Days	Interval	Group	No. of tested samples	Antibody titers at different serial dilution								%*
				0	20	40	80	160	320	640	1280	
1	Before 1 st vaccination dose	G1	10	10	-	-	-	-	-	-	-	0
		G2	10	10	-	-	-	-	-	-	-	0
		G4	10	10	-	-	-	-	-	-	-	0
		G3	10	10	-	-	-	-	-	-	-	0
10	Before booster vaccination dose	G1	10	10	-	-	-	-	-	-	-	0
		G2	10	10	-	-	-	-	-	-	-	0
		G3	10	-	-	3	7	-	-	-	-	65
		G4	10	-	-	4	6	-	-	-	-	60.6
20	Before challenge	G1	10	10	-	-	-	-	-	-	-	0
		G2	10	10	-	-	-	-	-	-	-	0
		G3	10	-	-	-	8	1	1	-	-	98
		G4	10	-	-	2	7	1	-	-	-	74.5
27	1 st week post challenge	G1	10	10	-	-	-	-	-	-	-	0
		G2	10	-	-	4	6	-	-	-	-	60.6
		G3	10	-	-	-	-	7	2	1	-	211.1
		G4	10	-	-	2	1	7	-	-	-	113.1
34	2 nd week post challenge	G1	10	10	-	-	-	-	-	-	-	0
		G2	10	-	-	-	3	7	-	-	-	130
		G3	10	-	-	-	-	6	3	1	-	226.2
		G4	10	-	-	-	2	3	5	-	-	197
41	3 rd week post challenge	G1	10	10	-	-	-	-	-	-	-	0
		G2	10	-	-	-	2	8	-	-	-	139.3
		G3	10	-	-	-	-	6	2	2	-	242.5
		G4	10	-	-	-	3	3	4	-	-	171

*: Numbers of birds showed antibody titers at the highest dilution.

G1=

G2=

G3=

G4=

4.4.7. Measurement of antibodies in the sera of broiler chickens after vaccination with the locally prepared *S. Enteritidis* bacterin and treatment with probiotic using ELISA test:

For evaluation of the humoral immune response of broiler chickens after vaccination with the locally prepared *S. Enteritidis* bacterin and treatment with probiotic, the ELISA test was conducted and the mean optical density (OD) values was detected. As shown in Table (17) and Figure (6), the mean optical density values were significant ($P \leq 0.05$) increased to reach 1.614 and 0.561 in vaccinated group and probiotic group, respectively (after 1st dose of vaccination and treatment with probiotic).

After booster dose of the vaccine (before challenge) the mean OD values were significantly ($P \leq 0.05$) increased to reach 2.543 in vaccinated group and 0.953 in probiotic treated birds.

After challenge, the mean OD values were 2.281, 2.340 and 2.486 at the 1st, 2nd and 3rd weeks post challenge, respectively.

Regarding the probiotic treated birds, the mean OD values increased significantly ($P \leq 0.05$) to reach 1.574, 1.845 and 1.779 at the 1st, 2nd and 3rd weeks post challenge, respectively.

By the challenge time, the mean OD value was 0.245. After challenge, the mean optical density values were 1.781, 1.457, 1.274 at the 1st, 2nd and 3rd weeks post challenge, respectively.

The statistical analysis of the obtained data by the ANOVA test indicated that there was significant ($P \leq 0.05$) difference between the vaccinated, probiotic treated and non treated infected groups at different intervals.

Table (17): ELISA mean optical density of antibodies in sera of broiler chickens after vaccination with the locally prepared *S. Enteritidis* bacterin and treatment with probiotic.

Age	Intervals	Blankcontrol group	Infected, non-treated	Vaccinated and infected	Probiotic treated	LSD
1	Before 1 st vaccination dose	0.234±0.064 ^{Aa}	0.234±0.052 ^{Ca}	0.234±0.032 ^{Ca}	0.234±0.039 ^{Da}	0.044
10	Before booster vaccination dose	0.212±0.031 ^{Ac}	0.212±0.027 ^{Cc}	1.614±0.249 ^{Ba}	0.561 ±0.034 ^{Cb}	0.115
20	Before challenge	0.245±0.035 ^{Ac}	0.245±0.024 ^{Cc}	2.543±0.427 ^{Aa}	0.953 ±0.078 ^{Bb}	0.198
27	1 st week post challenge	0.232±0.042 ^{Ac}	1.781±0.396 ^{Ab}	2.281±0.366 ^{Aa}	1.574±0.375 ^{Ab}	0.298
34	2 nd week post challenge	0.231±0.050 ^{Ac}	1.457±0.346 ^{Bb}	2.340±0.294 ^{Aa}	1.845±0.452 ^{Aa}	0.291
41	3 rd week post challenge	0.241±0.033 ^{Ad}	1.274±0.339 ^{Bc}	2.486±0.444 ^{Aa}	1.779±0.458 ^{Ab}	0.328
LSD		0.039	0.230	0.298	0.274	0.274

*Different small letters within the same row were significantly difference at (P≤0.05).

*Different capital letters within the same column were significantly difference at (P≤0.05).

N.B. Control group was negative at the 41days.

+ve result = > 0.65

-ve result < 0.65

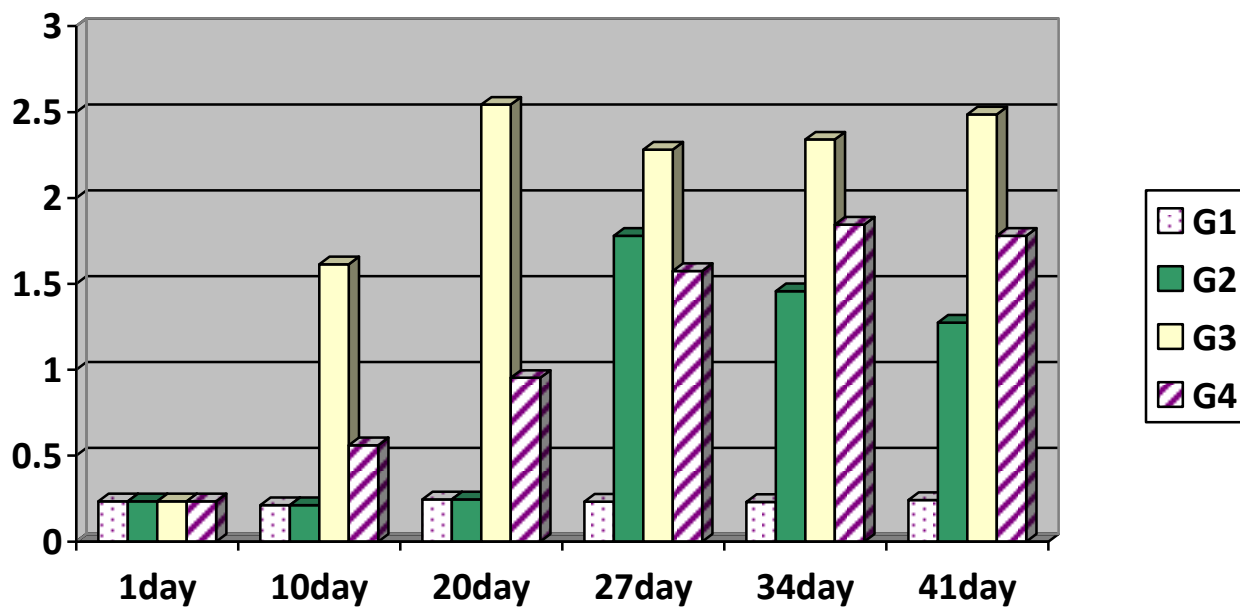


Figure (6): ELISA mean optical density of antibodies in sera of broiler chickens after vaccination with the locally prepared *S. Enteritidis* bacterin and treatment with probiotic.