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Pharmacological studies of *Pediococcus* on performance and some hematological and biochemical parameters in cattle

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ABSTRACT

The use of *Pediococcus* (23×10^6 CFU/calf/day) orally in drinking milk once daily for one month in Holstein calves have highly significant effect on blood picture ,liver function tests, kidney function test ,blood electrolytes and body weight in cow calves. There were a significant increase in total erythrocytes count , hemoglobin content, packed cell volume, mean corpuscular volume, mean corpuscular hemoglobin, mean corpuscular hemoglobin concentration, thrombocyte count, total leucocytes count, neutrophils counts, eosinophil, basophils, total protein and its fractions, level of alkaline phosphatase (ALP),level of sodium ,calcium ,phosphorus and body weight . On the other hand there were significant decrease in urea and potassium level in groups treated by *Pediococcus* compared to the control groups. Moreover there were a non-significant changes in lymphocytes, monocytes, creatinine, alanine transaminase (ALT), aspartate transaminase (AST), in the same groups compared to the control groups.

1. INTRODUCTION

In the light of population inflation in Egypt and increase the need for animal protein and milk and keep pace with global trends of the use of probiotics to improve the overall health of the animal, resulting in increased productivity and reproductive efficiency, we start our research plan.

Numerous studies have been conducted in an attempt to increase ruminant productivity by manipulating the rumen environment and to increase feed digestibility and nutrient utilization by the animals in order to supply sufficient nutrients to support a high level of milk production. One approach that has recently been widely investigated is the application of direct-fed microbial (DFM) preparations, in order to promote digestion and intestinal hygiene, enhance animal performance and reduce usage of antibiotics. The definition of DFM is very broad and may include specific and nonspecific yeast, fungi, bacteria, cell fragments, and filtrates. (Sobhy et al., 2014).

Probiotics, which are live cultures of harmless bacteria or yeast species that equilibrate intestinal micro flora to benefit the host (Fuller, 1989), have been demonstrated to be useful in maintaining the intestinal ecosystem and improving animal health. Because of a ban on the use of antibiotics, probiotics have been suggested as the most desirable alternative for livestock due to their beneficial effects. (Meng et al., 2010)

The most commonly used organisms in probiotic preparations are the lactic acid bacteria (*Lactobacillus*, *Streptococci*, *bifidobacteria* and *Pediococcus*). These are found in large numbers in the gut of healthy animals and do not appear to affect them adversely. Vibhute et al., (2011)

Pediococcus acidilactici and *Pediococcus Pentosaceus* are a species of Gram-positive cocci which prevent colonization of pathogens which has great effect to treat digestive disorders. *Pediococcus* exert antagonism against other microorganisms, including enteric pathogen through the production of lactic acid and secretion of bacteriocins known as pediocins. *Pediococcus* stimulates humoral immune response against infectious parasitic pathogens by possible competitive inhibition and pediocins production, which inhibit pathogenic bacteria and other Gram-positive spoilage. Haakensen et al., (2009)

This work was planned to evaluate some pharmacological effects of *Pediococcus* on hematological parameters, some liver and kidney functions, level of some electrolytes and on body weight gain in cow calves.

2. MATERIALS AND METHODS

1. Drugs: Max Boost powder Plus Sachet 100 gm. Powder prepared for oral administration contain:
-*Pediococcus Acidilactici* 1×10^6 cfu/g

- Pediococcus Pentosaceus* 1.3×10^6 cfu/g
- Dextrose Carrier

Manufactured & Exported by DVS Biolife limited – India
Elcon Nova-43 Degla Street-Mohandesen-Giza

Email: info@elconnova.com Reg. No: 5076 in 27/3/2013

Dose: 10 gm. / head / daily for 30 days orally that give 23×10^6 cfu / 10 gm. (Frizzo et al., 2010)

(CFU=COLONY FORMING UNITS)

2. Experimental calves:

Twenty Holstein calves at Damietta governorate in special dairy farm were divided into four groups (5 male and 5 females as a control group) and (5 male and 5 female under our experiment); all groups under the same hygiene management and the same nutrition (starter ration 21% - Al-Asema Group Company + milk before weaning while after weaning the ration were be 18% until reach 200-220 kg b.wt then 16% ration till reach moreover 350 kg b.wt. from the same company). Every calves under our experiment (10 calves) were take 10 gm. Maxboost* after fermentation in 100 ml water for 2 -3 hours then added to the milk one time per day for one month, then after 1 month give the drug for 3 successive days 20gm/head/once daily in drinking water after fermentation for 2-3 hours for five months. Body weight were determined in the beginning of the experiment and every week for 4 weeks then every 2 months for 10 months to the all groups, moreover the observation of the general health conditions.

3. Sampling

Two blood samples (the first sample for hematological studies and the second sample for serological studies) were collected from each animal in the four groups at zero day and after 15 days, 30 days and 45 days post the drugs administration, Stoffregen et al., 1997.

4. Hematological studies:

A-Mindray BC-2800:

Mindray Auto Hematology Analyzer BC 2800 is UN automated hematology analyzer designed for in vitro diagnostics use in blood cell counting and characterization.

3. RESULTS

-1 The effect of oral administration of *Pediococcus* (23×10^6 CFU/animal) in drinking milk once daily for one month on total erythrocytes count, hemoglobin content and packed cell volume in cow calves as shown in Table (1.)

-2 The effect of oral administration of *Pediococcus* (23×10^6 CFU/animal) in drinking milk once daily for one month on some blood picture in cow calves as shown in Table (2.)

-3 The effect of oral administration of *Pediococcus* (23×10^6 CFU/animal) in drinking milk once daily for one month on total and differential leucocytes counts in cow calves as shown in Table (3.)

B-Differential leukocyte count (thousand/mm³): according to Feldman et al., (2000).

C- Serum Biochemical Analysis:

Biochemical serum analysis of total proteins, albumin, ALT, AST, ALP, Urea, creatinine, sodium, potassium, calcium and phosphorus.

(a) Liver function tests:

I-Determination of Total Protein: according to the method of (Doumas, 1975)

II-Determination of Albumin: according to Doumas et al., 1981

III-Serum globulin calculation: as described by Doumas and Biggs, 1972

IV-Determination of serum Transaminases (ALT) and (AST): according to Reitman and Frankel, 1957.

VI-Determination of serum alkaline phosphatase (ALP) according to Rosalki (1993)

(b) Kidney function tests:

I-Determination of serum Creatinine was determined calorimetrically according to Henry (1974)

II-Determination of Urea: was performed according to the method of Patton and Crouch 1977.

(c) Determination of electrolytes:

I-determination of serum Calcium: according to the method of Gindler and King, 1972.

II-Determination of serum phosphorus: according to the method of El-Merzabani, 1977.

III- Determination of serum sodium: according to the method of Henry et al., 1974.

IV-Determination of serum Potassium: according to the method Turbid metric Tetraphenylborate (TPB), Henry et al., 1974.

5. Statistical analysis as described by one way (ANOVA), using SPSS computer program (version. 20). (SPSS, 2015)

-4 The effect of oral administration of *Pediococcus* (23×10^6 CFU/animal) in drinking milk once daily for one month on creatinine and urea in cow calves as shown in Table (4)

-5 The effect of oral administration of *Pediococcus* (23×10^6 CFU/animal) in drinking milk once daily for one month on total protein and its fractions in cow calves as shown in Table (5.)

-6 The effect of oral administration of *Pediococcus* (23×10^6 CFU/animal) in drinking milk once daily for one month on alanine transaminase (ALT), aspartate transaminase (AST) and alkaline phosphatase (ALP) in cow calves as shown in Table (6.)

-7 The effect of oral administration of *Pediococcus* (23×10^6 CFU/animal) in drinking milk once daily for one month on blood electrolytes in cow calves as shown in Table (7.)

-8 The effect of oral administration of *Pediococcus* on body weight in cow calves as shown in Table (8).
(23×10^6 CFU/animal) in drinking milk once daily for one month

Table (1): The effect of oral administration of *Pediococcus* (23×10^6 CFU/animal) in drinking milk once daily for one month on some blood picture in cow calves.

Group	RBCs($n \times 10^6$ /ML)				Hb(gm./dl)				PCV (%)			
	Time (Week)				Time (Week)				Time (Week)			
	0	2 nd w	4 th w	6 th w	0	2 nd w	4 th w	6 th w	0	2 nd w	4 th w	6 th w
Control male	5.82 $\pm 0.54^b$	4.74 \pm 0.45 ^a	4.82 \pm 45 ^b	5.50 \pm 0.46 ^c	7.24 \pm 06 ^{bc}	7.00 \pm 45 ^a	8.66 \pm 27 ^a	8.14 \pm 20 ^a	22.30 $\pm 0.58^b$	22.88 $\pm 0.51^a$	22.36 \pm .63 ^b	23.43 \pm 68 ^{ab}
Treated male	7.43 \pm 32 ^a	6.69 \pm 0.34 ^b	7.63 \pm 0.41 ^a	7.47 \pm 0.49 ^{ab}	7.92 \pm 50 ^{ab}	7.36 \pm 50 ^a	8.12 \pm 60 ^{ab}	7.60 \pm 50 ^a	25.34 $\pm 1.73^a$	22.10 $\pm 1.54^a$	24.17 \pm .05 ^{ab}	25.40 \pm 04 ^{ab}
Control female	5.52 \pm 32 ^b	4.96 \pm 0.35 ^a	5.21 \pm 0.73 ^b	6.20 \pm 0.49 ^{bc}	6.76 \pm 27 ^c	7.72 \pm 13 ^a	7.30 \pm 51 ^b	7.14 \pm 17 ^a	25.42 $\pm 0.63^a$	23.92 $\pm 0.63^a$	24.16 \pm .43 ^{ab}	22.20 \pm 40 ^b
Treated female	7.75 \pm 24 ^a	7.08 \pm 0.18 ^b	7.65 \pm 0.22 ^a	7.72 \pm 0.43 ^a	8.64 \pm 33 ^a	7.86 \pm 27 ^a	8.26 \pm 20 ^{ab}	7.84 \pm 0 ^a	27.48 $\pm 1.78^a$	24.28 $\pm 1.04^a$	26.64 \pm .26 ^a	27.32 \pm 85 ^a

The different litters in the same column mean significance at ($p < 0.05$).

(Mean \pm S.E) n= 5

Table (2): The effect of oral administration of *Pediococcus* (23×10^6 CFU/animal) in drinking milk once daily for one month on some blood picture in cow calves.

Items	Time (Week)	Control male	Treated male	Control female	Treated female
MCV(fl)	0	22.57 \pm 0.55 ^b	34.02 \pm 0.90 ^a	25.72 \pm 1.88 ^b	35.42 \pm 1.49 ^a
	2w	23.90 \pm 0.24 ^b	33.00 \pm 0.76 ^a	24.25 \pm 0.32 ^b	34.34 \pm 0.93 ^a
	4w	27.04 \pm 1.83 ^b	32.82 \pm 0.95 ^a	29.60 \pm 1.07 ^b	34.82 \pm 0.94 ^a
	6w	26.80 \pm 1.24 ^b	33.94 \pm 0.68 ^a	30.20 \pm 1.56 ^b	35.40 \pm 0.96 ^a
MCH (pg.)	0	8.40 \pm 0.92 ^b	10.78 \pm 0.25 ^a	6.80 \pm 0.37 ^c	11.08 \pm 0.20 ^a
	2w	7.00 \pm 0.45 ^b	10.92 \pm 0.32 ^a	6.80 \pm 0.58 ^b	11.04 \pm 0.18 ^a
	4w	6.20 \pm 0.58 ^b	10.10 \pm 0.21 ^a	7.80 \pm 0.37 ^b	10.76 \pm 0.17 ^a
	6w	7.80 \pm 0.37 ^b	10.10 \pm 0.25 ^a	7.80 \pm 0.37 ^b	10.10 \pm 0.17 ^a
MCHC (%)	0	27.00 \pm 0.70 ^c	31.92 \pm 1.17 ^a	27.20 \pm 0.66 ^{bc}	29.84 \pm 1.43 ^{ab}
	2w	26.00 \pm 0.70 ^b	33.32 \pm 1.21 ^a	25.80 \pm 0.92 ^b	32.44 \pm 1.06 ^a
	4w	25.20 \pm 0.73 ^c	30.22 \pm 0.37 ^a	28.20 \pm 0.37 ^b	31.12 \pm 0.84 ^a
	6w	25.20 \pm 0.97 ^b	30.06 \pm 0.95 ^a	25.60 \pm 0.60 ^b	28.72 \pm 0.35 ^a
Platelets($n \times 10^3$ /ml)	0	112.40 \pm 2.46 ^c	135.80 \pm 4.87 ^b	118.60 \pm 4.01 ^c	156.20 \pm 3.22 ^a
	2w	123.80 \pm 0.80 ^c	154.60 \pm 21.20 ^a	133.20 \pm 4.11 ^b	159.20 \pm 10.59 ^a
	4w	140.20 \pm 7.97 ^b	279.40 \pm 33.34 ^a	146.60 \pm 4.17 ^b	257.40 \pm 18.54 ^a
	6w	136.40 \pm 4.91 ^b	223.20 \pm 14.82 ^a	144.60 \pm 7.35 ^b	228.80 \pm 31.93 ^a

The different litters in the same column means significance at ($p < 0.05$)

(Mean \pm S.E) n= 5

Table (3): The effect of oral administration of *Pediococcus* (23×10^6 CFU/animal) in drinking milk once daily for one month on total and differential leucocytic counts in cow calves.

Items	Time (Week)	Control male	Treated male	Control female	Treated female
WBCs ($n \times 10^3$ /ml)	0	5.26 \pm 0.32 ^b	7.19 \pm 0.53 ^{ab}	6.00 \pm 0.41 ^b	8.67 \pm 1.14 ^a
	2 nd w	5.20 \pm 0.03 ^b	6.40 \pm 0.30 ^b	5.46 \pm 0.32 ^b	9.98 \pm 1.27 ^a
	4 th w	5.22 \pm 0.32 ^c	7.25 \pm 0.39 ^b	5.70 \pm 0.37 ^c	8.80 \pm 0.62 ^a
	6 th w	6.12 \pm 0.40 ^{ab}	7.24 \pm 1.02 ^a	4.40 \pm 0.43 ^b	7.68 \pm 0.78 ^a
Neutrophil (%)	0	51.40 \pm 2.16 ^b	57.00 \pm 2.47 ^a	55.60 \pm 0.51 ^{ab}	61.00 \pm 2.66 ^a
	2 nd w	54.40 \pm 1.16 ^a	55.00 \pm 3.58 ^a	52.20 \pm 0.37 ^a	50.40 \pm 2.77 ^a
	4 th w	54.60 \pm 0.86 ^a	50.60 \pm 4.41 ^a	58.40 \pm 1.36 ^a	52.60 \pm 2.08 ^a
	6 th w	56.00 \pm 1.48 ^a	53.20 \pm 2.70 ^{ab}	49.60 \pm 1.50 ^b	52.60 \pm 2.86 ^{ab}
Lymphocyte (%)	0	36.40 \pm 1.03 ^a	34.80 \pm 3.48 ^{ab}	39.40 \pm 1.76 ^a	31.20 \pm 2.63 ^b
	2 nd w	40.80 \pm 1.49 ^a	40.20 \pm 2.87 ^a	43.80 \pm 1.68 ^a	44.40 \pm 2.50 ^a
	4 th w	40.40 \pm 0.93 ^{ab}	42.40 \pm 3.97 ^a	37.00 \pm 0.93 ^b	42.40 \pm 2.77 ^a
	6 th w	39.60 \pm 0.68 ^a	40.80 \pm 2.96 ^a	44.00 \pm 1.08 ^a	41.00 \pm 2.55 ^a
Monocyte (%)	0	7.80 \pm 0.66 ^a	5.80 \pm 0.58 ^a	4.20 \pm 0.86 ^a	5.00 \pm 1.00 ^a
	2 nd w	3.60 \pm 0.50 ^a	3.40 \pm 0.60 ^a	3.00 \pm 0.89 ^a	4.00 \pm 0.70 ^a
	4 th w	3.80 \pm 0.66 ^a	4.60 \pm 0.50 ^a	3.60 \pm 0.40 ^a	3.60 \pm 0.50 ^a
	6 th w	3.80 \pm 0.37 ^a	4.00 \pm 0.31 ^a	5.80 \pm 0.37 ^a	4.00 \pm 0.70 ^a
Eosinophil (%)	0	2.00 \pm 0.00 ^{ab}	1.20 \pm 0.48 ^{ab}	0.40 \pm 0.40 ^b	1.60 \pm 0.40 ^a
	2w	0.80 \pm 0.20 ^a	1.00 \pm 0.31 ^a	0.40 \pm 0.24 ^a	0.60 \pm 0.24 ^a
	4w	0.80 \pm 0.37 ^{ab}	1.40 \pm 0.24 ^a	0.40 \pm 0.24 ^b	0.80 \pm 0.37 ^{ab}
	6w	0.60 \pm 0.40 ^b	1.20 \pm 0.20 ^{ab}	0.40 \pm 0.40 ^b	2.00 \pm 0.44 ^a
Basophil (%)	0	2.40 \pm 1.91 ^a	1.20 \pm 0.20 ^b	0.40 \pm 0.24 ^c	1.20 \pm 0.58 ^b
	2w	0.40 \pm 0.24 ^a	0.40 \pm 0.24 ^a	0.60 \pm 0.24 ^a	0.60 \pm 0.40 ^a
	4w	0.40 \pm 0.24 ^a	1.00 \pm 0.31 ^a	0.60 \pm 0.40 ^a	0.60 \pm 0.24 ^a
	6w	0.00 \pm 0.00 ^b	0.80 \pm 0.37 ^a	0.20 \pm 0.37 ^{ab}	0.40 \pm 0.24 ^{ab}

The different litters in the same column means significance at ($p < 0.05$)
(Mean \pm S.E) n= 5

Table (4): The effect of oral administration of *Pediococcus* (23×10^6 CFU/animal) in drinking milk once daily for one month on creatinine and urea in cow calves.

Items	Time (Week)	Control male	Treated male	Control female	Treated female
CR-S (mg/dl)	0	1.08 \pm 0.05 ^a	1.20 \pm 0.04 ^a	1.10 \pm 0.07 ^a	1.14 \pm 0.04 ^a
	2 nd w	1.12 \pm 0.05 ^a	1.22 \pm 0.02 ^a	1.10 \pm 0.06 ^a	1.22 \pm 0.02 ^a
	4 th w	1.00 \pm 0.13 ^a	1.06 \pm 0.02 ^a	0.72 \pm 0.11 ^b	1.08 \pm 0.03 ^a
	6 th w	0.88 \pm 0.05 ^a	0.94 \pm 0.02 ^a	0.90 \pm 0.04 ^a	1.02 \pm 0.06 ^a
Urea (mg/dl)	0	9.80 \pm 0.48 ^b	13.40 \pm 0.81 ^a	9.60 \pm 0.81 ^b	13.00 \pm 0.70 ^a
	2 nd w	10.60 \pm 0.40 ^b	14.20 \pm 0.37 ^a	10.40 \pm 0.40 ^b	15.40 \pm 0.60 ^a
	4 th w	14.40 \pm 0.92 ^a	9.00 \pm 0.44 ^b	16.00 \pm 0.70 ^a	10.80 \pm 0.80 ^b
	6 th w	18.60 \pm 0.60 ^a	11.40 \pm 0.50 ^b	17.60 \pm 0.67 ^a	12.80 \pm 0.58 ^b

The different litters in the same column means significance at ($p < 0.05$)
(Mean \pm S.E) n= 5

Table (5): The effect of oral administration of *Pediococcus* (23×10^6 CFU/animal) in drinking milk once daily for one month on total protein and their fractions in cow calves.

Items	Time (Week)	Control male	Treated male	Control female	Treated female
TP(g/dl)	0	4.58±0.05 ^a	4.78±0.12 ^a	4.62±0.18 ^a	4.96±0.18 ^a
	2 nd w	4.72±0.15 ^b	5.30±0.14 ^a	4.98±0.04 ^{ab}	5.34±0.24 ^a
	4 th w	4.86±0.10 ^b	7.84±0.24 ^a	4.58±0.21 ^b	7.98±0.08 ^a
	6 th w	4.94±0.18 ^b	8.50±0.19 ^a	4.78±0.07 ^b	8.08±0.04 ^a
ALB (g/dl)	0	2.60±0.15 ^b	2.80±0.05 ^{ab}	2.88±0.06 ^a	2.90±0.06 ^a
	2 nd w	2.94±0.05 ^b	3.22±0.08 ^a	2.90±0.06 ^b	3.18±0.06 ^a
	4 th w	2.96±0.06 ^b	4.60±0.07 ^a	3.02±0.07 ^b	4.84±0.08 ^a
	6 th w	2.96±0.06 ^b	4.88±0.12 ^a	2.82±0.05 ^b	4.76±0.10 ^a
Globulin (g/dl)	0	1.98±0.15 ^a	1.98±0.12 ^a	1.74±0.22 ^a	2.06±0.20 ^a
	2 nd w	1.78±0.15 ^a	2.08±0.12 ^a	2.08±0.06 ^a	2.16±0.26 ^a
	4 th w	1.9±0.12 ^b	3.25±0.20 ^a	1.56±0.16 ^b	3.14±0.14 ^a
	6 th w	1.98±0.15 ^b	3.62±0.12 ^a	1.96±0.12 ^b	3.32±0.07 ^a

The different litters in the same column means significance at ($p < 0.05$)
(Mean ± S.E) n= 5

Table (6): The effect of oral administration of *Pediococcus* (23×10^6 CFU/animal) in drinking milk once daily for one month on some liver functions in cow calves.

Items	Time (Week)	Control male	Treated male	Control female	Treated female
ALT (IU/L)	0	25±4.33 ^a	25.6±3.94 ^a	20.2±2.13 ^a	24.4±3.11 ^a
	2 nd w	18.4±1.32 ^a	20±1.30 ^a	17.4±1.16 ^a	19.2±0.58 ^a
	4 th w	16±0.70 ^b	20±1.14 ^a	19.8±0.86 ^a	19.6±0.51 ^a
	6 th w	17.4±1.02 ^a	18.4±1.69 ^a	15.2±1.59 ^a	18.4±1.12 ^a
AST (IU/L)	0	29.80±0.73 ^a	29.00±2.16 ^a	31.20±1.59 ^a	34.00±3.08 ^a
	2 nd w	29.40±1.16 ^b	38.80±3.18 ^a	29.20±0.58 ^b	40.20±1.46 ^a
	4 th w	29.80±0.73 ^a	31.20±2.05 ^a	24.40±1.53 ^b	24.60±1.02 ^b
	6 th w	24.20±0.37 ^a	24.40±1.72 ^a	21.80±0.80 ^a	21.20±1.24 ^a
ALP(IU/L)	0	591.80±62.74 ^{ab}	591.00±76.87 ^{ab}	344.80±19.13 ^b	868.60±171.33 ^a
	2 nd w	531.80±23.60 ^b	461.80±42.90 ^b	438.00±4.31 ^b	776.00±79.59 ^a
	4 th w	327.20±46.91 ^{bc}	473.20±87.38 ^{ab}	264.80±37.09 ^c	631.00±33.51 ^a
	6 th w	223.80±20.74 ^b	300.60±46.59 ^{ab}	222.00±4.41 ^b	388.20±95.30 ^a

The different litters in the same column means significance at ($p < 0.05$)
(Mean ± S.E) n= 5

Table (7): The effect of oral administration of *Pediococcus* (23×10^6 CFU/animal) in drinking milk once daily for one month on blood electrolytes in cow calves.

Items	Time (Week)	Control male	Treated male	Control female	Treated female
Sodium(mEq/L)	0	97.8±7.33 ^a	105.2±8.88 ^a	86.6±6.01 ^a	103.4±9.71 ^a
	2 nd w	96±2.98 ^a	107.8±10.22 ^a	92.2±2.56 ^a	108.6±11.07 ^a
	4 th w	77.6±1.96 ^b	85.6±4.19 ^a	70.6±0.40 ^b	89.8±2.51 ^a
	6 th w	87.2±1.02 ^b	106.2±1.77 ^a	94±2.47 ^b	105.6±3.96 ^a
Potassium(mEq/L)	0	4±0.20 ^b	4.42±0.15 ^{ab}	4.88±0.22 ^a	4.96±0.37 ^a
	2 nd w	5.56±0.17 ^b	6.34±0.22 ^a	5.06±0.22 ^b	6.2±0.37 ^a
	4 th w	4.7±0.23 ^a	3.47±0.02 ^b	4.92±0.17 ^a	3.22±0.15 ^b
	6 th w	4.724±0.20 ^a	3.08±0.11 ^b	4.58±0.16 ^a	2.98±0.06 ^b
Calcium(mg/L)	0	10.18±0.18 ^a	9.78±0.16 ^a	10.32±0.27 ^a	9.7±0.27 ^a
	2 nd w	9.62±0.20 ^{ab}	9.94±0.28 ^a	9.34±0.06 ^b	10.04±0.12 ^a
	4 th w	8.7±0.20 ^b	9.61±0.27 ^a	9.24±0.19 ^{ab}	9.46±0.08 ^a
	6 th w	8.86±0.30 ^b	9.64±0.27 ^a	8.5±0.11 ^b	9.4±0.28 ^a
Phosphorus (mg/L)	0	7.84±0.20 ^b	8.42±0.25 ^{ab}	8.44±0.22 ^{ab}	8.92±0.37 ^a
	2 nd w	8.56±0.33 ^a	8.94±0.30 ^a	8.58±0.16 ^a	9.42±0.40 ^a
	4 th w	8.26±0.19 ^b	9.09±0.13 ^a	7.98±0.14 ^b	8.84±0.17 ^a
	6 th w	7.9±0.19 ^b	9.1±0.27 ^a	7.98±0.37 ^b	9.6±0.39 ^a

The different litters in the same column means significance at ($p < 0.05$)
(Mean ± S.E) n= 5

Table (8): The effect of oral administration of *Pediococcus* (23×10^6 CFU/animal) in drinking milk once daily for one month on bodyweight in cow calves.

Items	Time (Week)	Control male	Treated male	Control female	Treated female
Body weight(kg)	0	83.2±1.39 ^a	76.6±5.98 ^a	79.6±2.18 ^a	76.2±6.37 ^a
	2 nd w	82.2±2.83 ^a	92.4±8.70 ^a	87±.836 ^a	92.6±4.61 ^a
	4 th w	100.2±3.28 ^b	122±8.45 ^a	100±1.70 ^b	116±5.099 ^a
	6 th w	105±1.61 ^b	134.2±9.91 ^a	103.8±2.00 ^b	133.6±6.70 ^a
	14 th w	175±2.98 ^b	196±9.66 ^a	163.6±2.69 ^b	180±6.71 ^{ab}
	22 th w	271±2.68 ^b	329±3.31 ^a	224.8±5.54 ^c	260.6±3.85 ^b
	37 th w	359.6±1.75 ^c	419.6±9.17 ^a	333.8±6.64 ^d	387±2.53 ^b
	47 th w	402.4±4.58 ^b	477.6±8.35 ^a	359.4±2.54 ^c	399±9.00 ^b

The different litters in the same column means significance at ($p < 0.05$)
(Mean ± S.E) n= 5

4. DISCUSSION

Our results showed a significant increase in total erythrocytes count in treated groups compared to control groups at zero and fourth weeks, while a significant decrease in treated groups compared to control groups was recorded at second week. Also data reflected a significant increase in

treated female group compared to control groups and a significant increase in treated male group compared to control male group at sixth week.

But there is a significant increase in hemoglobin content in treated female group compared to control groups at zero week, also showed a significant increase in treated male group compared to control female group. While a non-significant

changes was recorded at second and sixth week's. Moreover, showed a significant increase in control male group compared to control female group at fourth week.

The significant increase in packed cell volume at zero and fourth weeks in treated female group compared to control male group. While a non-significant changes was recorded at second week, but recorded a significant increase in treated female group compared to control female group at sixth week. Table (1)

Our data was in agreement with Salahuddin et al., (2013) and Shareef and Dabbagh (2009) who concluded that, the feeding of commercial ration with different level of probiotic in mice improved Total erythrocyte count and hemoglobin concentration. Probiotics supplementation helps in improving normal hematology.

Moreover Doaa and Moshira (2015) stated that, rabbits received mixture of pre and probiotic recorded the highest value of RBCS count, PCV, Hb concentration.

But our data disagree with Sarah et al., (2017) reported that lactating Holstein-Friesian cows received oral probiotic administration (containing *Lactobacillus acidophilus*, *Saccharomyces cerevisiae*, *Enterococcus faecium*, *Aspergillus oryza* and *Bacillus subtilis*) had no effect on PCV.

Looking to Table(2) there is a significant increase in mean corpuscular volume in treated groups compared to control groups at zero, second, fourth and sixth weeks.

Result evaluated a significant increase in mean corpuscular hemoglobin in treated groups compared to control groups at zero, second, fourth and sixth week, but at zero week only showed a significant decrease in control female group compared to control male group.

Moreover there is a significant increase in mean corpuscular hemoglobin concentration in treated male group compared to control groups at zero week, but showed a significant decrease in control male group compared to treated groups. Data recorded a significant increase in treated groups compared to control groups at second, fourth and sixth weeks, but at fourth week only showed a significant decrease in control male group compared to other groups.

The present data reflected a significant increase in thrombocyte count at zero, second, fourth and sixth week in treated groups compared to the control groups, but at zero week only showed a significant increase in treated male group compared to control groups and at second week only showed a significant increase in control female group compared to control male group.

Table(3) showed a significant increase in total leucocytes counts in treated female group compared to control groups at zero week. While showed a significant increase at second and fourth week in treated female group

compared to the others groups but recorded at fourth group only a significant increase in treated male group compared to control groups. Data showed a significant decrease in control female group compared to other groups at sixth week.

Same data also reported that Doaa and Moshira (2015) who stated that, rabbits received mixture of pre and probiotic recorded the highest value of Total Leucocyte Count. Moreover Al-Saad et al.,(2014) also showed a significant increase in number of white Blood Cells(WBC) in blood samples of probiotic and organic acids groups compared to antibiotic group, It seems that the microbial interactions and effects on local immune stimulation results increase of white blood cells and immunity these results are accordance with Endings of Zareshahneh et al.,(2007) and EFSA(2010-b) but are not accordance with the findings of Shareef and A-Dabbagh(2009).

Our results detected a significant increase in neutrophil percent in treated groups compared to the control male group at zero week, while at second and fourth week showed a non-significant changes in all groups. Data evaluated a significant increase in control male group compared to control female group at sixth week. While lymphocyte showed a significant increase in control groups compared to treated female groups at zero week but a non-significant changes in lymphocyte percent at second and sixth weeks in all groups, while in other side there are a significant increase in treated groups compared to control female group at fourth week.

These data agree with Doaa and Moshira (2015) stated a high value in heterophils and lymphocytes number in rabbits received mixture of pre and probiotic. Same data obtained by Sarah et al., (2017) who evaluated that oral probiotic administration (containing *Lactobacillus acidophilus*, *Saccharomyces cerevisiae*, *Enterococcus faecium*, *Aspergillus oryza* and *Bacillus subtilis*) on lactating Holstein-Friesian cows had a significant increase in per cent lymphocyte count, and a significant decrease in per cent neutrophil count in probiotic-treated animals.

Our results detected a non-significant changes in monocyte percent at zero, second, fourth and sixth weeks in all groups. Our data mirrored a significant increase in eosinophil percent at zero week in treated female group compared to control female group, and detected a non-significant change in all groups at second week, while a significant increase was recorded in treated male group compared to control female group at fourth week. But at sixth week detected a significant increase in treated female group compared to control groups.

Result evaluated a significant increase in basophil percent in control male group compared to other groups and a significant increase in treated groups compared to control female groups at zero week and a non-significant changes at second and fourth week in all groups, but showed a significant

increase in treated male group compared to control male group at sixth week.

In Table (4) there were a non-significant changes in creatinine at zero, second and sixth weeks in all groups, but a significant decrease was recorded in control female group compared to the others groups at fourth week. The results showed a significant increase in urea at zero and second weeks in treated groups compared to control groups, but showed a significant decrease in treated groups compared to control groups at fourth and sixth weeks.

Our data was in agreement with Kehoe et al., (2008) who detected that calves supplemented with purified nucleotides had higher in BUN-to-creatinine ratio and decreased in kidney functions. Not only Patterson and Burkholder (2003) recorded lower serum cholesterol Decrease ammonia and urea excretion and lower skatol, indole and phenol in cattle supplemented with probiotics, but also Doaa and Moshira (2015) who stated that, rabbits received mixture of pre and probiotic recorded a significant decrease in total cholesterol, creatinine and urea. Dietary supplementation of prebiotic and probiotic and their mixture improves kidney functions. However Saleem et al., (2017) concluded that, lambs that received PRO (a commercial probiotic containing a mixture of two strains of *Pediococcus*, *Pediococcus acidilactici* and *Pediococcus Pentosaceus*, with dextrose as the carrier compound) in post-weaning diet appeared to show a significant decrease in blood urea and cholesterol concentration.

In Table (5) there were non-significant changes in total protein at zero week in all groups, but showed a significant decrease in control male compared to the other groups at second week. Moreover a significant increase was detected in treated groups compared to control groups at fourth and sixth week.

In the same ground our results were supported by the results of Ezema (2014) who recorded that, Probiotic supplementation in broiler chicken significantly increased total proteins. But Sarah et al., (2017) reported that oral probiotic administration (containing *Lactobacillus acidophilus*, *Saccharomyces cerevisiae*, *Enterococcus faecium*, *Aspergillus oryza* and *Bacillus subtilis*) on lactating Holstein-Friesian cows had no effect on total protein concentration in plasma.

Our results showed a significant decrease in albumin in control male group compared to control and treated female groups at zero week, but at second, fourth and sixth weeks showed a significant increase in treated groups compared to control groups. There were a non-significant changes in globulin in all groups at zero and second weeks, but detected a significant increase in treated groups compared to control groups at fourth and sixth weeks.

Our results showed non-significant changes in serum alanine transaminase ALT in all groups at zero, second and

sixth weeks while data showed a significant decrease in control male group compared to the others groups at fourth week, Table (6). The present work reflected that non-significant changes at zero and sixth weeks in serum aspartate transaminase (AST) in all groups, while a significant increase was evaluated at second week in treated groups compared to control groups.

Result also detected a significant increase in control and treated male groups compared to control and treated female groups, also was recorded a non-significant changes between treated male group and control male group, and between treated female group and control female group at fourth week.

Our data detected a significant increase in serum alkaline phosphatase (ALP) in treated female group compared to control female group at zero week, while at second week showed a significant increase in treated female group compared to other groups. Data showed a significant increase in treated female group compared to control groups at fourth and sixth week, but showed only at fourth week a significant decrease in control female group compared to treated groups. (Table (6))

Similar findings were reported with Xing et al., (2006). who stated that consumption of *Bifidobacterium Catenulate* and *Lactobacillus Fermentum* improves liver functions. Also the use of *Lactobacillus plantarum* and *Bifidobacterium infantis* in rats induced liver injury improved. Osman et al., (2007). Not only administration of heat-killed *Lactobacillus brevis* (dosage of 100 or 500 mg/kg once a day) after 35 days was inhibited an increase in serum ALT and AST levels in alcoholic liver disease using ethanol-containing diet-fed mice Segawa et al., (2008), but also there were an inhibition of TNF- α and sterol regulatory element-binding protein (SREBPs) up-regulation by *Lactobacillus brevis*, which in turn may decrease serum ALT levels. Furthermore, elevated ALT, a sign of hepatocyte damage results from damaged biological membranes. Larson et al., (2008)

The decrease in serum ALT levels was seen with *Bifidobacterium pseudocatenulatum*, *Bifidobacterium longum* and *Bifidobacterium longum* (10^8 – 10^9 CFU) after 7 weeks in high fat diet-induced obese rats. An HM et al., (2011). These data agree with the results of Doaa and Moshira (2015) found a significant decrease in alanine amino transferase (ALT), aspartate amino transferase (AST) and improves in cell-mediated immune response and liver function in rabbits received mixture of pre and probiotic.

In Table (7) results showed non-significant changes in sodium in all groups at zero and second weeks, while at fourth and sixth weeks showed a significant increase in treated groups compared to control groups.

Our results match with the results of Schroeder et al., (2004) who concluded that, *Boulardii* has specific duration-dependent effects on the secretory response of the pig jejunal mucosa which developed during 8-day treatment and significant increase Na⁺ and Cl⁻ but disappeared during further application. Thus, this study supports the concept that probiotics may exert beneficial effects in the gastrointestinal tract. Similar findings were seen with consumption of strain *Enterococcus faecium* EK13 (dosage 10⁹ CFU/ml) in piglets for 14 days Stropfova et al., (2006).

In Table (7) also we found a significant decrease in Potassium in control male group compared to control and treated female groups at zero week, while a significant increase was recorded in treated groups compared to control groups at second week, but detected a significant increase in control groups compared to treated groups at fourth and sixth weeks. Our results showed a non-significant changes in Calcium in all groups at zero week, while detected a significant increase in treated groups compared to control female group at second week. A significant increase was recorded in treated groups compared to control male group at fourth week, but at sixth week reflected a significant increase in treated groups compared to control groups. However Phosphorus showed a significant increase in treated female group compared to control male group at zero week, while at second week showed a non-significant changes in all group, moreover a significant increase was detected in treated groups compared to control groups at fourth and sixth weeks.

Our results in agreement with Patterson and Burkholder (2003) who detected that, probiotics and prebiotics improve mineral absorption. Also Ateequr et al., (2012) who showed that administration of the probiotic mixture (containing the 8 bacterial strains *Streptococcus thermophilus*, *Bifidobacterium breve*, *Bifidobacterium longum*, *Bifidobacterium infantis*, *Lactobacillus acidophilus*, *Lactobacillus plantarum*, *Lactobacillus paracasei* and *Lactobacillus delbrueckii* sub sp. *Bulgaricus*) parallel with the clindamycin therapy had a beneficial and stabilizing effect on the intestinal metabolic homeostasis by decreasing toxic metabolites and protecting the endogenic microbiota from destruction. Probiotics could be a reasonable strategy in prevention of antibiotic associated disturbances of the intestinal homeostasis and disorders.

Our results showed non-significant changes in body weight in all groups at zero and second week, while showed a significant increase in treated groups compared to control groups at fourth and sixth weeks. But at 14th week there is a significant increase in treated male group compared to control groups in Table (8). There is a significant increase in treated male group compared to other groups and a significant decrease in control female group compared to other groups at 22th and 47th week, while showed a significant increase in treated male group compared to other groups and a

significant increase in treated female group compared to other control groups at 37th week.

Our results in agreement with Frizzo et al., (2011) who said that, lactic acid bacteria (LAB) supplementation increased body weight gain (BWG) and improve feed efficiency in calves. And confirmed with Hiruta (2015) indicated that, the usage of probiotics in ruminants stimulate forage intake, and the increase in forage intake can result in improved live weight gain. Moreover application of probiotics in receiving cattle, increase in daily gain, increase in feed consumption, and improvement in feed gain and steers fed a probiotics had greater final weight, ADG, DM intake, hot carcass weight, carcass-adjusted ADG and improved feed efficiency.

Also our results confirmed with Bhatt et al., (2016) concluded that, probiotics supplementation, more specifically *Lactobacillus acidophilus* (at 10⁷ CFU/g concentrate) in growing Chinchilla rabbits improved digestibility and utilization of nutrients, BW gain and feed conversion ratio. Not only agree with Ezema (2014) who said that, Probiotic supplementation in broiler chicken significantly increased weight gain, but also agree with Corcionivoschi et al., (2010) reported that, probiotics increased food intake, and body weight. Manipulating ruminal fermentation and ruminant productivity.

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