

## Growth and physiological responses of rabbit to dietary symbiotic supplementation and varying levels mixture of aromatic plant hay

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### ABSTRACT

Our study was conducted to assess the impact of Basil + Fennel hay-diets without or with symbiotic on growth performance, digestibility of nutrients, caecal microbial activity, volatile fatty acids estimation and some organs histology in rabbit. Six-week-old unsexed NZW rabbits were randomly distributed in a factorial (4 × 2) design arrangement with four dietary BH+FH levels were used at 0.0, 25, 50 or 75% of the diet rather than the percent of alfalfa hay in the control diet, and two symbiotic levels (0.0 or 0.5 g/kg diet). Results showed that digestibility coefficients of dry matter (DM), crude protein (CP), ether extract (EE) and NFE of 14-week old NZW rabbits were improved by feeding different levels of BH+FH. Treatments fed BH+FH-diets had a significantly effect on pH value, TVFA, and NH<sub>3</sub>-N values for cecal content of 14 weeks old NZW rabbits. Also, total bacterial count and lactobacillus counts were positively affected, but animals fed experimental diet had inferior count of E. Coli. Rabbits fed BH+FH-diets had significantly increased in CAT and SOD activities, however, a significant decrease in MDA levels were observed. The colony forming devices of coliform bacteria showed a significantly lower number in comparison control one. This study concluded that dietary levels of BH+FH may be safely used in rabbit diets as much as 75% in place of alfalfa hay. Conclusion: The present results indicate that growing rabbits had fed fennel and basil hay at 75% instead of alfalfa hay has a positive role on growth performance and cell reinforcement status notwithstanding its antibacterial impacts.

**Key words:** Rabbits, Digestibility, Cecal Activity, Organs Histology, Fennel, Basil Hay.

## INTRODUCTION

Weaning is the maximum critical period in rabbit breeding as it is related to a better chance of digestive disorders in weaning rabbits. An herbal feed preservative had a positive role on overall performance and health in weaned rabbits (Krieg et al., 2009). The impacts of some medicinal plants and their extracts on rabbit has been studied through many authors (Eiben et al., 2004; Soultos et al., 2009; Simonová et al., 2008; Gerencsér et al., 2012). Basil (*Ocimum spp.*), adapted to the Lamiaceae family, is pleasant and smells of a perennial shrub which grows in large areas around the world (Akgül, 1993; Baritoux et al., 1992). Basil is one of the species used for the profitable flavoring, which is grown in Middle Egypt to get seeds and leaves. The seeds and leaves contain 3-5 % essential oils

Baritoux et al. (1992).

Fennel (*Foeniculum vulgare*), the family *Lamiaceae*, is cultivated for seeds in Upper Egypt. The seeds contain 2-6 % essential oil which is utilized as human feed added substance for flavor (Nassar et al., 2010).

There are extensive numbers of feed added substances accessible for consideration in animal and poultry diets to enhance their productive performance. Fennel is known as a good resource of natural antioxidants (Shahat et al., 2011). Wild fennel has higher phenolic and flavonoid compounds and thus free radical protective activity, while Italian fennel shows the most protective level (Faudale et al., 2008). This herb is conventionally used as treatment for wind, colic, irritable bowel, kidneys, liver, spleen, lungs, breast enlargement, suppressing appetite, promoting menstruation, milk flow, improving digestive system and increasing urine flow (Delaram et al., 2011). Nweze and Ekwe (2012) observed that *Ocimum gratissimum* leaf extracts can be used to get better growth performance, stabilize the blood components and decrease the blood and gut micro-organisms for finishing broilers. Chemical investigations have

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shown that basil contains various active compounds, such as flavonoids, tannins, saponin, glycosides, terpenes and steroids (Pattnaik et al., 1997). Free radicals are continually produced as by-products beneath aerobic cellular metabolism and play a key role in pathological and physiological processes. Antioxidant defense system assists the organism to respond and fight against the excess amount of free radicals (Tan et al., 2012). Symbiotics (a mixture of prebiotics and probiotics), in fastidious, have been used as an antimicrobial agent for gastrointestinal improvement to encourage higher weight gains. *Saccharomyces cerevisiae* and  $\beta$ -glucan and its cell wall mannan oligosaccharides (MOS) are one such natural feed additive (Abo El-Maaty et al., 2014; Abo El-Maaty et al., 2017).

SOD detoxifies superoxide radicals and converts them to  $H_2O_2$  which is further converted to  $H_2O$  by catalase enzyme. *Ocimum sanctum* elevated the glutathione and antioxidant enzyme levels (SOD) and decrease lipid peroxidation, thereby suggesting that hypoglycemic effect of *Ocimum sanctum* may be linked and mediated through modulation of cellular antioxidant disobedience system (Sethi et al., 2004)

CAT and SOD are the two scavenging enzymes that remove toxic free radicals (Wohaieb and Godin, 1987). CAT (the enzymatic antioxidant-catalase) is concerned in the elimination of hydrogen peroxide in living cells and protects against hydroxyl radicals toxicity.

Zhang et al. (2005), Ozsoy (2011) and (Beski and Al-Sardary, 2015) reported that *Saccharomyces cerevisiae* contains equivalent amounts of protein, amino acids, and B vitamins as soybean. Symbiotic have positive effects on the productive performance of broilers thereby improving their hematological and intestinal histological aspects.

A few studies have been conducted on the use of Symbiotic in feeding of growing

rabbits. Therefore, the aim of the current research is to investigate the growth performance, carcass trait and selected blood constituents of weaning rabbits fed diets containing basil and fennel hay in absence or presence of a symbiotic.

## MATERIALS AND METHODS

This research was directed in the Rabbits Production Unit, Agricultural Research and Experiment Station, Faculty of Agriculture, Mansoura University. The compound analyses of the tested feedstuffs and the untried diets were carried out at the Laboratory of Poultry Production Department, Faculty of Agriculture, Mansoura University, Egypt. The main purpose of attending research examine the impact(s) of different BH and FH incorporating levels (0.0, 9, 18 and 27% representing 0.0, 25, 50, 75 % replacement levels from alfalfa hay) without or with a symbiotic (Bio-Yeast<sup>®</sup>) on the productive performance of fattening rabbits.

Impacts of these trails on carcass traits, some hemato-biochemical parameters and microscopic examination of both cecal and ileum of fattening rabbits, were also investigated.

### **Preparation and proximate analysis of dried fennel (*foeniculum vulgare*) and basil (*ocimum basilicum*) hay:**

Fennel (*Foeniculum vulgare*) and Basil (*Ocimum basilicum*) were sun dehydrated before they mixed with other feedstuffs to formulate the tested pelleted rations. The proximate analysis of basil hay DM, CP, EE, CF, Ash, NFE, Ca and TP are 90, 14.0, 23.5, 10, 20, 32.5, 0.60 and 0.10, respectively and fennel hay DM, CP, CF, EE, Ash, NFE, Ca and TP are 89.50, 6.00, 16.5, 8.0, 19.0, 50.5, 0.75 and 0.08, respectively as dry matter basis and samples basil hay (BH) and fennel hay (FH) were analyzed according to AOAC (2000).

### **Experimental Design:**

Ninety sex unsexed NZW rabbits, about 42 days of age, with the same initial live body

weights, were assigned, in to 4x2 factorial arrangement design with four experimental diets containing 0.0; 9; 18 and 27% of BH and FH (0.0; 25; 50 and 75% substituted by

contents of the tried diets are established in Table 1.

All rabbit groups were housed in a commercial wire net (50L×50W×45H cm)

**Table (1): Formulation and proximate analysis of the tested pelleted ration of weaning rabbits from 42 to 98 days old**

Feedstuffs	Basal diet	9% FH + BH	18% FH+BH	27% FH+BH
Yellow corn	17.0	15.5	13.0	12.0
Soybean meal, 44% CP	11.5	13.0	14.0	15.5
Wheat bran	16.0	16.0	17.5	16.95
Barley grain	15.0	15.0	15.0	15.0
Alfalfa hay	36.0	27.0	18.0	9.0
Fennel hay (FH)	----	4.5	9.00	13.50
Basil hay (BH)	----	4.5	9.00	13.50
Molasses	2.00	2.00	2.00	2.00
Limestone	1.00	1.00	1.00	1.00
Dicalcium phosphate	0.70	0.70	0.70	0.70
Vit. Min. Premix <sup>§</sup>	0.300	0.300	0.300	0.300
Sodium chloride	0.50	0.50	0.50	0.50
Total	100	100	100	100
Calculated analyses: As fed basis(NRC,1977) <sup>¶</sup>				
Digestible energy, kcal/kg	2502	2511	2505	2519
Crude protein, %	16.25	16.30	16.26	16.32
Crude fiber, %	13.57	13.00	12.55	11.92
Ether extract, %	2.58	3.14	3.71	4.26
Lysine, %	0.75	0.74	0.71	0.75
Methionine, %	0.21	0.21	0.21	0.21
Methionine + Cystine, %	0.53	0.51	0.49	0.47
Total phosphorus, %	0.59	0.58	0.58	0.57
Calcium, %	1.11	1.06	1.00	0.94
Determined analyses: As DM basis (AOAC, 2000)				
Dry matter, %	90.01	91.06	90.55	91.03
Organic matter, %	81.07	82.16	81.64	82.10
Crude protein, %	18.05	17.90	17.96	17.93
Crude fiber, %	15.08	14.28	13.86	13.10
Ether extract, %	2.87	3.45	4.09	4.68
Ash, %	8.94	8.90	8.91	8.93
Nitrogen free extract,%	55.09	55.47	55.18	55.36

<sup>§</sup>Each 3kg premix contained12,000,000IU Vit. A, 2,500,000IU Vit. D<sub>3</sub>,10,000mg Vit. E,2500mg VitK<sub>3</sub>,1000mg VitB<sub>1</sub>,4000mg Vit.B<sub>2</sub>,1500mg Vit. B<sub>6</sub>,10mg Vit. B<sub>12</sub>,10,000mg Pantothenic acid, 20,000mgNicotinic acid, 1000mg Folic acid, 50mgBiotin, 500mg Choline chloride, 60mg Manganese, 55mg Zinc, 100mg Selenium, 1000mg Iodine, 35mg Iron, 10mg Copper, 250mg Cobalt, and Carrier CaCO<sub>3</sub> to 3kg. <sup>¶</sup>Calculated

alfalfa hay) and tow levels of a symbiotic product (0.0 or 0.5 g/Kg diet). Eight tried pelleted rations were ready to meet all the broiler rabbit's essential nutrient requirements, as indicated by NRC (1977) and crude fiber fraction was determined in Regional Center for Food and Feed (RCFF). The nutrient substances and feedstuffs

of three rabbits each. The cages were given for a manual feeders and nipple drinkers, and fed their respective experimental rations from 42 to 98 days of age. All weaning rabbits were kept on the normal environmental, managerial and comparable veterinary all over the growth period from 6

to 14week of age. Fresh water and feed were available *ad lib.* basis.

Initial LBW (live body weights) of weaned animals were recorded at the starting of growing period (42 days old). Live body weight was recorded every four weeks, feed consumed (FC) and BWG (body weight gain) of experimental broiler rabbits were also maintained on a replicate group basis. Also, FCR (feed conversion ratio) and body weight gain were measured throughout the experimental period.

Mortality of weaned rabbits was recorded daily throughout the growth period.

#### **Bacterial count of cecal contents of growing rabbits:**

For cecal measurements, rabbits were immediately sacrificed; evacuated and cecal contents were collected. Coliform bacteria was measured on selective media and aerobically incubated at 37°/ 24 hr, as methods mentioned by Li (1991). Meanwhile, *Lactobacillus* counts were anaerobically evaluated on MSR agar. All microbial counts were assessed on methods described by Xia et al.(2004).

#### **Blood concentrations of growing rabbits:**

At the end of the growth period (14 weeks of age), 8 NZW rabbits from each group were haphazardly chosen and fasted for 16 hours, weighed and butchered.

At the time of slaughter, blood samples were taken from four growing rabbits in each experimental group, in clean heparinized tubes. Blood samples were centrifuged at 4000 rpm for 15 minutes. Plasma samples were isolated and stocked in -20°C until biochemical examination. The plasma samples was allocated for "triglycerides (TG), malondialdehyde (MDA), creatinine, T4 and T3 hormone as well as activity of plasma aspartate aminotransferase (AST), alanine aminotransferase (ALT), superoxide dismutase (SOD), catalase (CAT)" and determines using commercial kits.

#### **Histological observations:**

Illeal and cecal representative samples were fastidiously dissected and glued in sufficient volume of formalin solution (10%).

Permanent sections were prepared by using the paraffin method technique according to Junqueira et al. (1971).

Haematoxyline and eosin stains were used. Tissue sections of ileum and ceca were examined with light microscope and then subjected to a digital camera with a magnification power of X10.

#### **Statistical analysis**

"Data were statistically analyzed victimisation Two-way analysis of variance of SAS Program (SAS, 2004)". Important variations among means ( $P \leq 0.05$ ) were separated victimisation Duncan's New Multiple Ranges test (Duncan, 1955). Where:

$Y_{ijk}$  = Treats

$\mu$  = over all means

$T_i$  = Treatment ( $i = 1, 2, 3$  and 4) (0.0, 9, 18 or 27% FH and BH of ration

$A_j$  = effect of feed additive (synbiotic add with 0.0 and 0.5g/Kg diet)

$TA_{ij}$  = the effect of interaction between level of BH&FH and level of synbiotic

$e_{ijk}$  = error

## **RESULTS AND DISCUSSION**

#### **Growth Performance:**

The data for feed intake, body weight gain and feed conversion ratio of the rabbits are tabulated in Table (2). The impact of replacing dietary alfalfa hay by mixture of FH+BH levels without or with symbiotic supplementation had beneficial effects on growth performance of growing rabbits. These outcomes demonstrated that growth performance of 14 weeks-old growing NZW rabbits were positively influenced ( $P \leq 0.05$ ) by nourishing the incorporation of FH+BH containing diets (9, 18 and 27%) compression with their fed the control diet. These results recommended that the growing rabbits fed diets including FH+ BH had significant effect on feed intake. Furthermore, body weight gain was comparable amongst all treatments between different periods. There were significantly

differences in feed conversion ratio between treatments. The improved growth performance, coincide with the finding of Riyazi et al. (2015) who reviewed that rabbit

hay levels were improved growth promoter, body weight gain, and feed conversion ratio and reduced feed intake of fattening rabbits in this study. The enhancement in growth

**Table (2) Growth performance of growing NZW rabbits as affected by different dietary levels of both fennel and basil hay mixture without or with symbiotic treatment for 14 weeks of age.**

Item	BW6 (Kg)	BW10 (Kg)	BW14 (Kg)	BWG10 (Kg)	BWG14 (Kg)	TBWG (Kg)	FI10 (Kg)	FI14 (Kg)	TFI (Kg)	FCR10	FCR14	TFCR
BH and FH level (A)												
0% A1	0.760	1.46 <sup>c</sup>	2.31 <sup>b</sup>	0.70 <sup>c</sup>	0.86 <sup>b</sup>	1.55 <sup>c</sup>	2.43 <sup>a</sup>	3.32 <sup>a</sup>	5.74 <sup>a</sup>	3.48 <sup>a</sup>	3.88 <sup>a</sup>	3.70 <sup>a</sup>
9.0% A2	0.760	1.52 <sup>b</sup>	2.31 <sup>b</sup>	0.76 <sup>b</sup>	0.79 <sup>c</sup>	1.55 <sup>c</sup>	2.23 <sup>b</sup>	2.99 <sup>c</sup>	5.22 <sup>c</sup>	2.93 <sup>b</sup>	3.78 <sup>a</sup>	3.36 <sup>b</sup>
18.0% A3	0.760	1.58 <sup>a</sup>	2.58 <sup>a</sup>	0.82 <sup>a</sup>	0.99 <sup>a</sup>	1.82 <sup>a</sup>	2.44 <sup>a</sup>	3.17 <sup>b</sup>	5.60 <sup>b</sup>	2.96 <sup>b</sup>	3.17 <sup>b</sup>	3.08 <sup>c</sup>
27.0% A4	0.760	1.56 <sup>ab</sup>	2.55 <sup>a</sup>	0.80 <sup>ab</sup>	0.99 <sup>a</sup>	1.79 <sup>b</sup>	2.45 <sup>a</sup>	3.07 <sup>bc</sup>	5.52 <sup>b</sup>	3.08 <sup>b</sup>	3.09 <sup>b</sup>	3.08 <sup>c</sup>
SEM	0.001	0.014	0.012	0.010	0.018	0.04	0.04	0.04	0.01	0.07	0.06	0.04
Sig <sup>8</sup> .	Ns	***	***	***	***	***	**	***	***	***	***	***
synbiotic (B)												
B1 (0.0g/kg)	0.760	1.52	2.44	0.77	0.92	1.68	2.38	3.16	5.54	3.13	3.48	3.32
B2 (0.5 g/kg)	0.760	1.53	2.44	0.77	0.91	1.68	2.39	3.12	5.51	3.09	3.48	3.29
SEM	0.001	0.02	0.02	0.02	0.03	0.04	0.04	0.05	0.05	0.08	0.11	0.08
Sig <sup>8</sup> .	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS
AB Interaction												
A1B1	0.760	1.44	2.31	0.68	0.87	1.55	2.45	3.42	5.87	3.62	3.92	3.79
A1B2	0.760	1.48	2.32	0.72	0.84	1.56	2.40	3.21	5.62	3.33	3.84	3.60
A2B1	0.760	1.48	2.31	0.72	0.83	1.55	2.21	3.03	5.24	3.06	3.66	3.38
A2B2	0.760	1.56	2.32	0.80	0.76	1.56	2.25	2.96	5.21	2.81	3.90	3.34
A3B1	0.760	1.60	2.60	0.84	1.00	1.84	2.43	3.14	5.57	2.88	3.14	3.02
A3B2	0.760	1.56	2.56	0.80	1.00	1.80	2.44	3.20	5.64	3.04	3.20	3.13
A4B1	0.760	1.58	2.54	0.82	0.96	1.78	2.43	3.04	5.48	2.96	3.19	3.08
A4B2	0.760	1.53	2.56	0.77	1.03	1.81	2.47	3.09	5.56	3.20	3.00	3.08
SEM	0.002	0.01	0.02	0.01	0.02	0.02	0.06	0.05	0.02	0.07	0.07	0.04
Sig <sup>8</sup> .	NS	***	***	***	***	***	*	***	***	***	***	***

\*c:"means in the same column bearing different superscript are significantly different (P<0.05)" " NS: not significant,\*: significant at P<0.05., "SEM= Standard errors of the means

fed with basil produced advantageous impacts on growth performance traits of rabbits when contrasted with their control group. Benlemlih et al. (2014) found that rabbits fed with fennel added diet appeared to eat large quantities of feed and recorded a higher body weight and growth rate when contrasted with their control one throughout the first week of experiment.

On the other hand, adding dietary symbiotic had no significantly effect of growth promoter, BWG, feed intake and FCR of the rabbits. In addition, the interactions between symbiotic supplementation and dietary levels of mixture of fennel and basil

performance, noted herein, resulted from symbiotic addition may be due to the beneficial microorganisms which increased nutrient utilization.

Our recorded data harmonizes with those recorded by (Amber et al., 2014 and Huyghebaert et al., 2011) who established that a positive connection between productive performance of weaning rabbits and prebiotic-enhanced rations. Prebiotics create unfavorable condition for pathogenic role seems to be affected by class & type of the added prebiotic, hygienic states & diet piece of the animal. Prebiotics as a feed components have alleviative or metabolic

activity of a set count of interior *lactobacillus spp.*

### Digestibility traits of rabbits:

The impact of replacing dietary alfalfa hay by mixture of fennel and basil hay level without or with symbiotic supplementation on digestibility coefficients of growing rabbits are tabulated in Table 3. These outcome reviewed that nutrient digestibility of "dry matter, organic matter, crude protein, ether extract and nitrogen free extract" of 98 day

with their control group. Omer et al. (2013) observed that the highest value of OM, CF, EE digestibility's and DM values on dietary treatment compression with the control one. It may because of the beneficial effects on stimulation of digestive secretions such as saliva, digestive enzymes, bile, and mucus which measured a core mode of activity of phytobiotics, coincide with intestinal villi length and a lower passage rate of digesta, but it is very small with regard to practical nourishing conditions (Lee et al., 2003).

**Table (3) Nutrients Digestibility (%) of fattening NZW rabbits as affected by different dietary levels of fennel and basil hay mixture and symbiotic treatment for 14 weeks of age.**

Treatments	Digestibility coefficients (%)					
	DM (%)	OM (%)	CP (%)	CF (%)	EE (%)	NFE (%)
Mixture of fennel and basil hay level (A)						
0.00% A1	75.36 <sup>c</sup>	74.39 <sup>c</sup>	84.43 <sup>c</sup>	52.35 <sup>c</sup>	73.39 <sup>c</sup>	73.43 <sup>c</sup>
9.0% A2	77.65 <sup>b</sup>	76.68 <sup>b</sup>	86.72 <sup>b</sup>	54.64 <sup>b</sup>	75.68 <sup>b</sup>	75.72 <sup>b</sup>
18.0% A3	78.22 <sup>a</sup>	77.25 <sup>a</sup>	87.29 <sup>a</sup>	55.21 <sup>a</sup>	76.25 <sup>a</sup>	76.29 <sup>a</sup>
27.0%A4	78.09 <sup>ab</sup>	77.12 <sup>ab</sup>	87.16 <sup>ab</sup>	55.08 <sup>ab</sup>	76.12 <sup>ab</sup>	76.16 <sup>ab</sup>
SEM	0.169	0.169	0.169	0.169	0.169	0.169
Sig.	*	*	*	*	*	*
Synbiotic (B)						
B1 (0.0 g/kg)	76.69 <sup>b</sup>	75.72 <sup>b</sup>	85.76 <sup>b</sup>	53.68 <sup>b</sup>	74.72 <sup>b</sup>	74.76 <sup>b</sup>
B2 (1.0 g/kg)	77.96 <sup>a</sup>	76.99 <sup>a</sup>	87.03 <sup>a</sup>	54.95 <sup>a</sup>	75.99 <sup>a</sup>	76.03 <sup>a</sup>
SEM	0.119	0.119	0.119	0.119	0.119	0.119
Sig.	*	*	*	*	*	*
Interaction between A&B						
A1B1	74.38	73.41	83.45	51.37	72.41	72.45
A1B2	76.34	75.37	85.41	53.33	74.37	74.41
A2B1	77.09	76.12	86.16	54.08	75.12	75.16
A2B2	78.21	77.24	87.28	55.20	76.24	76.28
A3B1	77.61	76.64	86.68	54.60	75.64	75.68
A3B2	78.84	77.87	87.91	55.83	76.87	76.91
A4B1	77.69	76.72	86.76	54.68	75.72	75.76
A4B2	78.48	77.51	87.55	55.47	76.51	76.55
SEM	0.239	0.239	0.239	0.239	0.239	0.239
Sig.	NS	NS	NS	NS	NS	NS

<sup>a-c</sup> means in the same column bearing different superscript are significantly different (P<0.05) NS: not significant, \*: significant at P<0.005, SEM= Standard errors of the means

old growing NZW rabbits were useful effected (P≤0.05) by feeding the mixture of fennel and basil hay containing diets (25, 50 and 75%) compression with their control one. The enhanced nutrient digestibility, coincide with the finding of Mohamed et al., 2016, who recorded that rabbit fed with fennel or basil created useful effects on digestibility traits of rabbits in comparison

On the other hand, included dietary symbiotic had positively affect (p≤ 0.05) on nutrients digestibility of "DM, OM, CP, CF, EE or NFE" of weaning rabbits (Table 4). The useful effect extended by dietary symbiotic on nutrients digestibility by the trail rabbits might be due to its inspiring action on cecal microbial activity, and thus enhancing the digestion efficiency. Our results

agreement with Amber et al. (2014) observed that digestibility of DM, OM, CP, CF and NFE were significantly increased, despite that digestibility coefficient of EE was significantly reduced by supplementing. The increase in DCP may be due to the improvement of CP digestibility. The digestible energy (DE) did not significantly affect by experimental treatments. After feeding of probiotics, enhancements in feed efficiency and growth performance have been reviewed in broiler (Kabir et al., 2004; Samli et al., 2007; Mountzouris et al., 2007). In addition the interactions between supplemented symbiotic and dietary levels of mixture of fennel and basil hay levels, the mixture of fennel and basil hay levels by supplemented symbiotic interactions were

### Cecal microbial activity:

The impact of replacing dietary alfalfa hay by mixture of fennel and basil hay levels without or with symbiotic addition on cecal microbial activity and cecal fermentative activities of fattening rabbits are founded in Table 4. These results reported that TVFA's, NH<sub>3</sub>-N values, total bacterial count (TBC), and *Lactobacillus* count for cecum content of 14 weeks of age growing NZW rabbits were significantly improved ( $p \leq 0.05$ ), whereas pH value and total *E. Coli* count were reduced due to feeding the diets including (25, 50 or 75%) mixture of fennel and basil hay as compared with those of the control one. Our effects agreed with Rabie et al. (2011), who recorded lowered microbial count in cecal content of rabbits fed diets basil herb

**Table (4) Cecal microbial activity of growing NZW rabbits as affected by different dietary levels of fennel and basil hay mixture and symbiotic treatment for 14 weeks of age.**

Treatments	Cecal Fermentative Activities			Caecal Microbial Activity		
	PH Value	TVFA's mmol/100ml	NH <sub>3</sub> mg/100 dl	TBC	<i>Lacto</i> -count	<i>E. Coli</i> -count
<b>H and FH level (A)</b>						
0.00% A1	6.61 <sup>a</sup>	8.57 <sup>b</sup>	25.71 <sup>b</sup>	12.94 <sup>b</sup>	7.60 <sup>b</sup>	3.76 <sup>a</sup>
9.0% A2	6.41 <sup>b</sup>	8.74 <sup>a</sup>	27.55 <sup>a</sup>	13.52 <sup>a</sup>	9.10 <sup>a</sup>	3.45 <sup>b</sup>
18.0% A3	6.28 <sup>c</sup>	8.87 <sup>a</sup>	27.61 <sup>a</sup>	13.09 <sup>b</sup>	8.74 <sup>a</sup>	3.24 <sup>c</sup>
27.0% A4	6.33 <sup>c</sup>	8.73 <sup>a</sup>	25.64 <sup>b</sup>	13.51 <sup>a</sup>	9.10 <sup>a</sup>	3.45 <sup>ab</sup>
SEM	0.018	0.017	0.129	0.124	0.102	0.067
Sig.	*	*	*	*	*	*
<b>Symbiotic (B)</b>						
B1 (0.0 g/kg)	6.40	8.30 <sup>b</sup>	27.19 <sup>a</sup>	12.30 <sup>b</sup>	7.52 <sup>b</sup>	4.77 <sup>a</sup>
B2 (1.0 g/kg)	6.42	9.15 <sup>a</sup>	26.07 <sup>b</sup>	14.23 <sup>a</sup>	9.75 <sup>a</sup>	2.18 <sup>b</sup>
SEM	0.012	0.012	0.091	0.087	0.072	0.047
Sig.	NS	*	*	*	*	*
<b>Interaction between A&amp;B</b>						
A1B1	6.76	8.29	25.25	12.23	6.63	5.16
A1B2	6.47	8.86	26.17	13.64	8.57	2.36
A2B1	6.26	8.20	28.41	12.71	8.12	4.75
A2B2	6.57	9.27	26.68	14.33	10.07	2.15
A3B1	6.18	8.45	28.65	11.75	7.20	4.42
A3B2	6.37	9.29	26.58	14.43	10.29	2.07
A4B1	6.40	8.27	26.43	12.51	8.12	4.75
A4B2	6.25	9.19	24.84	14.52	10.07	2.15
SEM	0.025	0.024	0.182	0.175	0.144	0.095
Sig.	NS	*	*	*	*	*

\*-c: means in the same column bearing different superscript are significantly different ( $P < 0.05$ )

NS: not significant, \*: significant at  $P < 0.05$ , SEM= Standard errors of the means

not significant ( $P \leq 0.05$ ) for nutrients digestibility of "DM, OM, CP, CF, EE or NFE" examined of growing rabbits in this experimental research.

which has been found to have anti-infective function by inhibiting many pathogenic bacteria. Similarly, rabbits fed diets containing fennel and/or thyme recorded a

significant ( $P < 0.05$ ) lowering in CFU when corresponding with the others (Benlemlih et al., 2014). Fennel inhibited *Enterococcus* and *Clostridium Sp.* growth in rat's intestine and *Bacillus bifidus* growth. Moreover, confirmed the possibility of using the fennel in maintaining useful bacteria colony on rat's intestine (Omer et al., 2013).

On the other hand, pH of the caecal contents was not significantly affected by adding digestion to rabbit diets (Abd El-Hady et al., 2013). These results agreement with those reported by Krieg et al. (2009) who reviewed that VFA's was significantly affected by adding digestarom diet to growing rabbit's diets. Djouvinov et al. (1997) showed that concentrations of VFA's and  $\text{NH}_3\text{-N}$  in caecal contents were significantly affected when sheep fed diets contained peppermint byproducts compared with rabbits fed the control diet. Otherwise, symbiotic-added diets had useful effects ( $p \leq 0.05$ ) on cecal microbial activity and cecal fermentative activities of weaning rabbits are shown in Plate 8. Our results TVFA's levels, TBC and *Lactobacillus* counts are the superior significant values at ( $P \leq 0.05$ ) accompanied with the control group. The higher TBC counts of rabbits given symbiotic-enhanced diets might be connected to greater fermentation rate of microflora in the cecum. In the present research, no alteration was detected in concentrations of pH values of broiler NZW rabbits had fed symbiotic-added diets compared with the control one. Effects might have a useful action at low hygiene as they counteract the attack of potential pathogens to the intestinal tissue. a prebiotic is an indigestible food component that beneficial effects the host by selectively stimulating the activity and/or growth of one or a limited number of bacteria in the colon (Gibson and Roberfroid, 1995). The proposed modes of action of probiotics in poultry are as follows: 1) keeping up a beneficial microbial populace by competitive elimination and hostility (Fuller, 1989), 2)

enhancing feed admission and absorption (Nahanshon et al., 1992 and 1993), and changing bacterial metabolism (Cole et al., 1987; Jin et al., 1997). Chichlowski et al. (2007) observed that a probiotic including *lactobacilli Bifidobacterium thermophilus* and *Enterococcus faecium* improved the jejunal villus height and reduce the villus crypt depth rapprochement with salinomycin and control. Furthermore, shorter and thinner villi were associated' with toxins (Yason et al., 1987; Awad et al., 2006). On the other hand, higher villi were shown in the ileal of mature cock layers with slight.

Additionally the mixture of fennel and basil hay levels by enhanced symbiotic interactions increased significantly ( $P \leq 0.05$ ) cecal microbial community for each of TBC with pronounced positively effects on *Lactobacillus* population, in the meantime *E. coli* counts showed significant reduction ( $P \leq 0.05$ ). The examination of these interactions may give a sign that both symbiotic and mixture of fennel and basil hay levels addition exerted positively affect in this admiration. It was intriguing to take note that groups fed the fennel and basil-diet plus symbiotic displayed lower *E. coli* count and higher *lactobacillus* counts and TBC as appraisal with those fed the fennel and basil-diet alone or the control one.

Feeding diet included 9.0; 18.0 and 27% of FH and BH induced significant ( $p < 0.05$ ) decreased in plasma levels of triglycerides, AST, ALT and creatinine concentrations as indicators of liver and kidney functions in rabbits (Table 5). Plasma TG, ALT, creatinine and AST showed the best values or positive indication of rabbits fed the diets supplemented with increasing levels of BH and FH compared with control diet. The high level of BH and FH diet (27%) induced significant ( $p < 0.05$ ) decreases in the levels of TG, AST, ALT, and creatinine levels by 15.1, 11.8, 17.8 and 6.25%, respectively, and restored the levels of the AST and ALT enzymes to normal levels.



Similar results were obtained by Sobhy et al., (2015) who observed that feeding of tested fennel caused a significant decrease in

Our results showed that plasma malondialdehyde concentration, SOD and catalase enzyme activities were significantly

**Table (5) Plasma biochemical parameters of growing NZW rabbits as affected by different dietary levels of fennel and basil hay mixture and symbiotic treatment for 14 weeks of age.**

Treat.	TG g/dl	AST <sup>1</sup> U/L	ALT <sup>2</sup> U/L	Creat <sup>3</sup> mg/dl	MDA <sup>4</sup> nmol/ml	SOD <sup>5</sup> nmol/ml	CAT <sup>6</sup> U/ml/h	T3 <sup>7</sup> ng/dl	T4 ng/dl	T3/T4
BH and FH level (A)										
0.00% A1	54.46 <sup>a</sup>	56.73 <sup>a</sup>	13.46 <sup>a</sup>	1.12 <sup>a</sup>	37.97 <sup>a</sup>	67.62 <sup>a</sup>	36.77 <sup>a</sup>	4.27	18.9	22.90
9.0% A2	50.09 <sup>b</sup>	54.43 <sup>a</sup>	11.75 <sup>b</sup>	.97 <sup>b</sup>	35.45 <sup>ab</sup>	71.02 <sup>ab</sup>	40.23 <sup>ab</sup>	4.54	19.16	23.79
18.0% A3	49.77 <sup>b</sup>	51.03 <sup>b</sup>	11.72 <sup>b</sup>	.97 <sup>b</sup>	32.79 <sup>bc</sup>	72.65 <sup>ab</sup>	42.63 <sup>b</sup>	4.30	18.25	23.55
27.0% A4	46.33 <sup>b</sup>	50.03 <sup>b</sup>	11.05 <sup>b</sup>	1.05 <sup>ab</sup>	30.04 <sup>c</sup>	75.48 <sup>b</sup>	45.03 <sup>c</sup>	4.37	18.32	24.16
SEM	1.3	1.09	0.48	0.04	1.39	1.68	1.14	0.17	0.25	0.27
Sig.	**	***	**	*	**	*	***	NS	NS	NS
symbiotic (B)										
(0.0 g/kg) B1	51.61	54.57 <sup>a</sup>	12.59 <sup>a</sup>	1.05	35.62 <sup>a</sup>	70.68 <sup>b</sup>	39.88	4.25	19.10	22.36
(0.5 g/kg) B2	48.71	51.53 <sup>b</sup>	11.40 <sup>b</sup>	1.01	32.50 <sup>b</sup>	72.71 <sup>a</sup>	42.46	4.49	18.22	24.84
SEM	1.15	0.99	0.38	0.03	1.21	1.39	1.13	0.11	0.41	0.75
Sig.	NS	*	*	NS	*	*	NS	NS	NS	NS
AB Interaction										
A1B1	56.75	58.48	14.58	1.23	39.07	65.37	34.83	3.89	19.22	20.37
A1B2	52.18	54.98	12.34	1.02	36.87	69.87	38.70	4.65	18.58	25.44
A2B1	51.40	56.52	12.18	.93	37.08	69.77	38.53	4.63	20.01	23.18
A2B2	48.77	52.34	11.32	1.01	33.81	72.27	41.93	4.45	18.30	24.40
A3B1	50.20	51.88	12.55	1.00	34.45	73.17	42.17	4.13	17.86	23.08
A3B2	49.33	50.17	10.89	.95	31.12	72.13	43.10	4.48	18.64	24.02
A4B1	48.10	51.41	11.05	1.04	31.87	74.40	43.97	4.36	19.29	22.80
A4B2	44.55	48.64	11.04	1.06	28.21	76.57	46.10	4.37	17.35	25.51
SEM	1.7	1.31	0.56	0.05	1.9	2.45	1.5	0.21	0.30	0.59
Sig.	**	***	**	*	*	*	**	*	NS	NS

<sup>a-c</sup>: means in the same column bearing different superscript are significantly different ( $P < 0.05$ )

NS: not significant, \*: significant at  $P < 0.05$ , SEM= Standard errors of the means

triglyceride values comparing with the control group. Other studies by Abdel-Azeem (2006) and Farok et al. (2011) approved decreasing in plasma creatinine level when feeding with fennel.

However, Abdel-Azeem et al. (2010) showed that plasma ALT concentration was not significantly affected by adding fennel hay to growing rabbit diets. Sherlock (1975) reported that AST and ALT levels reflect the impairment of liver function when their levels increase. The elevated blood creatinine is correlated with an increased protein catabolism in the mammalian body (Butani et al., 2002).

influenced by dietary treatments (Table 6). A positive effect of FH and BH-diets on the antioxidant defenses system was observed compared to control group, in terms of a significant ( $p < 0.05$ ) decrease in plasma MDA level and a significant increase in the activity of CAT and SOD in comparison with the control rabbits. In agreement with Sobhy et al. (2015) who found that feeding of tested fennel caused a significant decrease in MDA values comparing with the control. Since, MDA (lipid peroxidase) is a lipid degradation product (Romeo et al., 2002) that is formed by peroxidation of

unsaturated fatty acid and is used as a biological marker of oxidative.

Our results come in agreement with Ansari et al. (2017) who found that MDA concentration in control group shows a significant increase compared to all groups. In addition, catalase activity in control group shows a significant decrease compared to all groups. In agree with our results Sadeghpour et al. (2015) observed that fennel extract can decrease the serum level of oxidative factors in experimental groups compared with the control one. The present study also agreed with Gharaghani et al. (2015) showed that fennel consumption significantly decreased MDA levels. Our data come in agree with Sadek et al. (2015) who reviewed that Supplemented groups with basil has antioxidant activity revealed in significant increases in SOD, CAT in all examined tissues compared with the control one. Also, Kahilo et al. (2015) revealed that the value of MDA significantly decreased in group containing basil and they concluded that the feeding of basil has a good effect on antioxidant enzyme concentration and acts as immunostimulant. It is well known that lipid peroxidation is one of the most important destructive effects of free radicals which destroy cell membrane. Thus, unsaturated fatty oxidation leads to decreased membrane fluidity and loss of its structure and function (Hararguchi et al., 1996). This may results in many disease pathogenesis. Also, lipid peroxidation causes a loss in membrane integrity and a change in related enzymes. MDA is the final product of lipid peroxidation (Joyeux et al., 1990).

The antioxidant enzymes GPX and CAT are considered to be indicators of antioxidant status (Flores-Mateo et al., 2009). Fennel has physiologic antioxidant activities including the radical scavenging effect, inhibition of hydrogen peroxides  $H_2O_2$  and  $Fe(2+)$ -chelating activities where it can minimize free radical which initiate the chain reactions of lipid peroxidation (EL and Karakaya, 2004). The antioxidant effect of *Foeniculum*

*vulgare* was evaluated by Zaahkouk et al. (2016) who showed a significant increase in GSH and CAT accompanied by a decrease in Malondialdehyde (MDA) in diabetic rats treated with fennel when compared with diabetic group. This may be due to the presence of phenols and flavonoids, which may have a major role in reducing oxidative stress associated with diabetes (Anitha et al., 2014 and Parsaeyan, 2016).

On the other hand, our data showed that T3 hormone and T3/T4 ratio were slightly enhanced in treated groups especially with symbiotic groups. Similar results were obtained by Abdel-Malak et al. (1995); Ibrahim, et al. (1998) and Abd El-Latif et al. (2002) stated that adding thyme or fennel to Japanese quail diets enhanced T3 hormone level.

On the other side, there were non-significant differences between symbiotic treatments and control values. Plasma ALT and AST levels as affected by symbiotic supplementation, regardless of BH and FH levels were significantly lower for rabbits fed the symbiotic-supplemented diet compared with the control group. However, plasma creatinine, T3, T4 hormones, T3/T4 ratio and TG levels were not affected by different dietary treatments. Additionally, effects of symbiotic supplementation on the antioxidant defense system were generally minor. However, Feeding rabbits with symbiotic supplementation reduced plasma MDA levels and significantly increase activity of plasma SOD and CAT enzyme as numerically but the differences locked the significant level.

On the other hand, rabbits fed the diets supplemented with increasing levels of BH and FH with or without symbiotic especially with symbiotic significant decreased values of plasma TG, creatinine concentration, AST and ALT activities compared to control treat.

In addition, co-administration of different levels of FH and BH without or with symbiotic increased significantly the elevated SOD and CAT plasma level observed after

administration. All treated groups significantly ( $P < 0.05$ ) reserved the lower values of catalase induced by FH and BH without or with symbiotic supplementation-diets administration indicating their antioxidant activities. Also, significant ( $p < 0.05$ ) lower in plasma MDA levels and higher values of MDA indicate the oxidative stress in control group. The decrease in MDA level was completely attenuated by higher level of FH and BH with symbiotic followed by higher level of FH and BH without though symbiotic did not vary considerably compared with control one.

#### **Ileum histology:**

Histological investigation of ileal sections for the experimental is introduced in Plates 1 to 8.

It is obvious from these sections that the villi shape and size were significantly affected by treatments. Great variations were observed in the diameter and tall of the intestinal villi, coincided with changes in the mucosa layer thickness. The villi in the control one (plate 1) were short and narrow diameters with little mucus-secreting cell. Also, an increase in the villi number (per microscopic field area) and size were observed especially for rabbits fed high level of BH and FH without or with symbiotic. It appears that supplemented levels of BH and FH especially with symbiotic caused an increase in villi diameter with many well-developed crypts in the sub-mucosa layer. Many crypts were detected in the base of villi contradictory in number and size located more than the sub-mucosal layer inside the lamina propria. These crypts are considered the mainly numerous tubular glands in high levels of FH and BH sections irrespective of symbiotic supplementation compared with the control section.

It is generally known that the crypts of Lieberkuhn have the ability to secrete fluids containing different substances essential for improving the interior micro-environment of the intestinal segments (Hodges, 1974). Mucus is secreted in large amounts by goblet cells of the crypts and villi. The

crypts fluids are nearly pure among a neutral pH (6.5 to 7.5). These secretions might improve the productive performance via enhancing the activity of most digestive enzymes. These histological observations suggest that the use of FH and BH with or without symbiotic could promote the histological structure of the alimentary tract in the weaning rabbits. This characterization is in harmony with the reported by Parker and Armstrong (1987), who showed that feeding an antibiotic-containing diet to growing pigs can result in elongated villi and a lower crypt: villus ratio, which are indicative of a lower rate of enterocyte- migration from the crypt to the villus. Goblet cells are responsible for the secretion of mucin that is used for the mucinous lining of the intestinal epithelium (Schneeman, 1982). Hence, a higher density of goblet cells may result in an increase in the secretion of mucin. Intestinal microbes would possibly have an impact on goblet cell dynamics via release of bioactive compounds or indirect activation of the immune system (Bienenstock and Befus, 1980). Dietary FH and BH with symbiotic resulted in a decreased proliferation of goblet cells into the surface of the villus membrane. This is indicative of a decreased host reliance on mucus secretion for protection. In the study of Ferket et al. (2002), contrary to expectations, antibiotic therapy increased goblet cell numbers. Decreasing numbers of viable Gram-positive bacteria, such as *Lactobacilli* and *Bifidobacteria* might also increase the presence of Gram-negative species. An increase in these kinds of microbes may also actually necessitate the need for greater mucus production and hence more goblet cells (Edens et al., 1997).

#### **Caecal histological observations:**

Microscopic examination of caecal sections as affected by BH+FH with or without symbiotic supplementation showed some changes due to applied treatments (plates 9 to 16). Generally, the cecal wall in the

experimental rabbits was found to be comparable to that the opposite intestinal segments; but, there were many special changes in the cecal histology. The histological section can dictate that the control section is composed of small lymph nodules and few tubular glands compared with the other experimental sections.

Externally, the muscular layer (M) of caecum section of the control treatment group is composed of a thin longitudinal muscle layer. The villi are being well developed in the length and breadth. But crypts of lieberkuhn (Cr) being larger in size. The villi are very short and blunt while the underlying tunica is infiltrated with lymphoid tissue with many small lymphocytes and more large lymphocytes by supplemented high levels of BH and FH with symbiotic. There are dramatic changes in the size and shape of caecal villi; number and size of crypts and the presence of the lymphocytes in the tunica propria region.

Among those changes, the muscularis mucosa layer was not completely advanced, although had exterior longitudinal and interior circular muscles which strengthen the cecal wall. Cecal sections show that the villi have been properly agreed in numerous plicae with different shapes and sizes mainly within the sections from broiler rabbits group that had been fed the high levels of FH and BH with symbiotic.

The crypts are characterized with small and short tubular glands and occupy mainly of the lamina propria layer among the bases of the villi and the muscularis mucosa. Also, there was numerous nodular cells accretion of that cells and both blood vessels and nerves in the lamina propria layer. It is intricate to interpret these histological changes based on the nutritional treatments. Nevertheless, the muscular layer and the size and shape of the villi among their mucosal epithelium lining in the FH and BH supplementation plus symbiotic-fed rabbits might corroborate the feasible influence of examination group on the activity of caecal fermentation, production of VFA'S, a

lessening in the bacterial ecosystem and the pH value in the ceca of rabbits, which might also elucidate the optimistic effects of symbiotic and FH and BH supplementation on the productive performance of weaning rabbits.

Several investigators have premeditated the intestinal morphology in poultry throughout the last decade (Uni et al., 1999; Geyra et al., 2001). No detailed data are available in the literature on the effects of FH and BH supplementation on the gut morphology of rabbits. It generally accepted that intestinal microflora reduce nutrient absorption by rising the thickness of gastrointestinal tract, digesta passage rate, and moreover can raise nutrient requirements of the host by increasing nutrient turnover rate of the gut mucosa and by competing with the host for a portion of the dietary energy and protein (Ravindran et al., 1984; Apajalahti et al., 2004). The reduction of gastrointestinal bacteria may also ease the competition for vital nutrients between the bird and the microbes (Ferket, 1991). The crypt can be considered as the villus factory, and a large crypt refers to a fast tissue turnover and a high demand for new tissue (Yason et al., 1987). The villus : crypt ratio is an indicator of the likely digestive capacity of the small intestine. Enhance in this ratio is an indicator to a raise in digestion and absorption (Montagne, 2003).



Plate 1: T.S. In the ileum from the Control Treatment (X10)

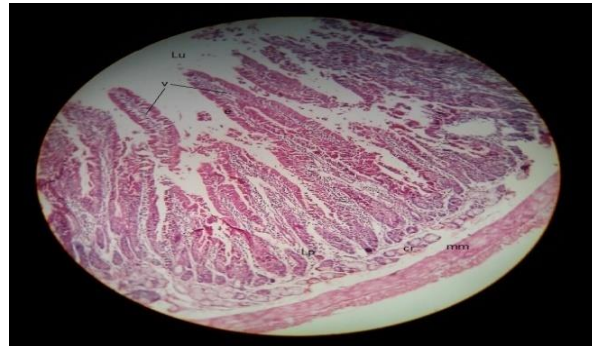


Plate2: T.S. In the ileum from the 25.0%FH +BH Treatment (X10)

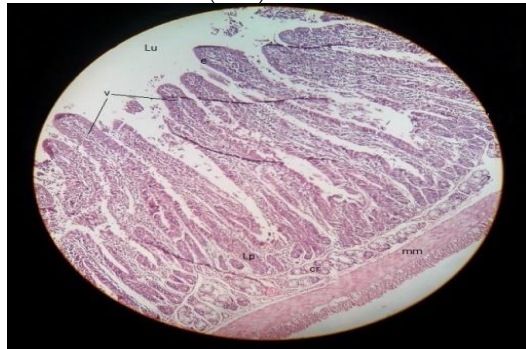


Plate3: T.S. In the ileum from the 50.0%FH +BH Treatment (X10)

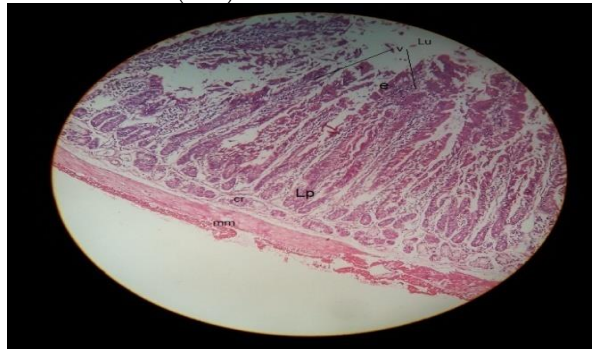


Plate 4: T.S. In the ileum from the 75.0%FH +BH Treatment (X10)



Plate 5: T.S. In the ileum from the Control+Symbiotic Treatment (X10)

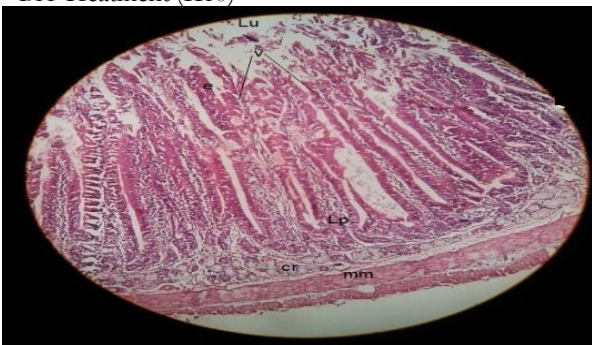


Plate 6: T.S. In the ileum from the (25.0%FH+BH)+SymbioticTreatment (X10)

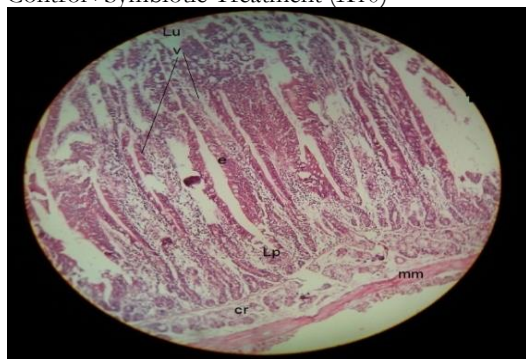


Plate 7: T.S. In the ileum from the (50.0%FH+BH)+SymbioticTretment (X10)



Plate 8: T.S. In the ileum from the (75.0%FH+BH)+SymbioticTreatet (X10)





Plate9: T.S.In the Cecal from the control Treatment (X10)

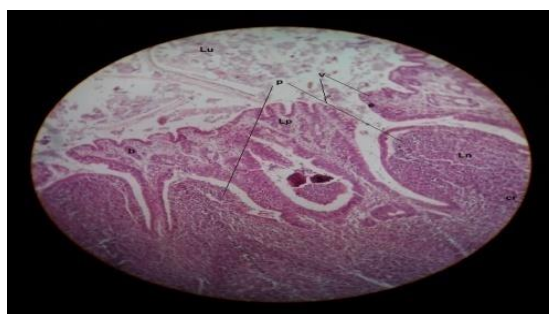


Plate10: T.S.In the Cecal from the 25.0%FH +BH Treatment (X10)



Plate11: T.S.In the Cecal from the 50.0%FH +BH Treatment (X10)

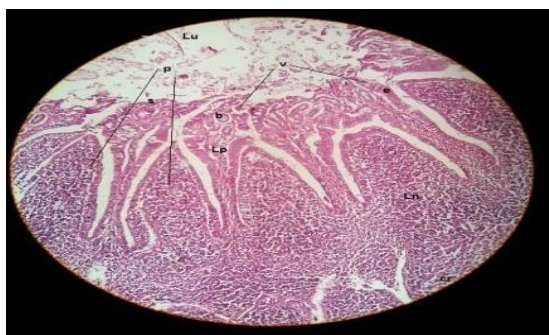


Plate12: T.S.In the Cecal from the 75.0%FH +BH Treatment (X10)

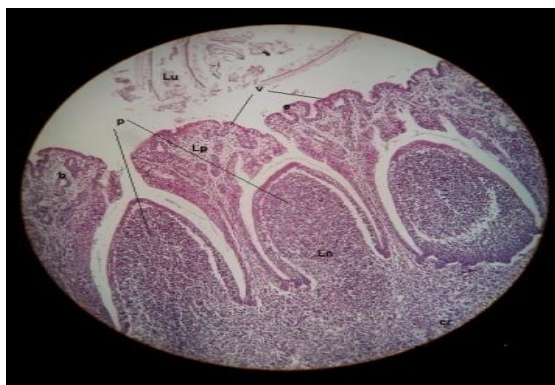


Plate13: T.S.In the Cecal from the Control+Symbiotic Treatment (X10)



Plate14: T.S.In the Cecal from the (25.0%FH+BH)+Symbiotic Treatment (X10)

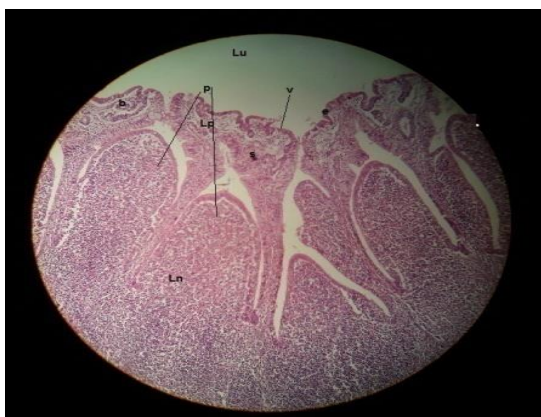


Plate15: T.S.In the Cecal from the (50.0%FH+BH)+Symbiotic Treatment (X10)

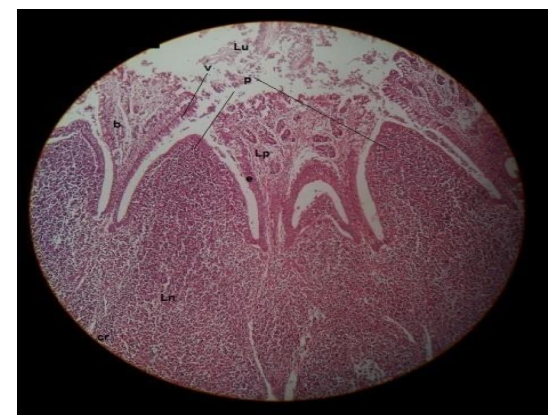


Plate16: T.S.In the Cecal from the (75.0%FH+BH)+Symbiotic Treatment (X10)

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