Comparative study of locally premix (its carrier *Malva parviflora* leaves meal) with imported premixes in diets on hematological bloodand Intestinal Microflora in broilers (Ross 308)

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Abstract

The study was carried out at the Animal Production Department, Faculty of Agriculture, University of Kufa to investigate the effect of feeding locally premixes comparable with imported premixes the broiler diets. Starter diets (23.3% C.P and 3021.0 kcal ME/kg) feed were gave to the birds from week 1 to week 3 of age, and finisher diets which contained 20.0% C.Pand 3204.0 kcal ME/kg were gave from 4thweek to 5th week of age. Five hundred seventy-six one day old Ross 308 chicks were randomly divided into eight groups with three replicate (24 bird /replicate)per group, Each group was subjected to the one of the following treatments: (T1)control contained 2.5% locally premix; (T2) 2.5%Provimi premix imported; (T3) 2.5%Turkish premixes imported; (T4) 2.5% Dutch premixes;(T5)1.25%locally+1.25%provimipremixes;(6)1.25%locally+1.25%Turkis;(7) 1.25%locally+1.25%Dutchpremixes;(8)0.625% from each one. Randomized Completely Block Design (RCBD) was used. Results showed that white blood cell, Hemoglobin, lymphocyte (L), Hetrophil (H) were improved significant (P≤0.05).The serum biochemical indices showed significant reductions values ofalkaline phosphatase(ALP),Aspartate aminotransferase (AST),phosphorus (P) compared with importedpremix.Harmful bacterial account were decreased significantly in local premix control. Additionally, beneficial bacteria were increased (p≤0.05) inlocal premix compared with imported. Based on the results obtained, the feeding of local premix(itscarrier*Malva parviflora* leavesmeal)2.5% of diet,the local premix gave promising results through the efficiency and ability that competed with the importer very strongly is recommended.

Key words: Broilers, Premix, *Malva parviflora,* ALP,AST, Bacteria, Biochemical traits.

Introduction

Many natural medicinal plants and herbs have attempted to benefit from what they contain of chemical compounds with different vital activities by extracting or isolating them. Many of these herbs grow in the wild, including what people consume, such as the mallow plant, due to its high nutritional benefits in addition to its medicinal uses. *Malva parviflora* is a leafy vegetable belonging to the family Malvaceae (35). It grows in most countries of the world, including Iraq, where it is found naturally in gardens, fields and roadsides. The plant is traditionally used in the treatment of all types of infections, especially its use as an anti-hemorrhoid and as a reliever for chest pain in children, in addition to its use as an anti-constipation substance (11 and 14). The plant contains varying amounts of minerals, the most important of which are zinc, copper, cadmium (18), calcium and iron (10), and phosphorous (27). It also contains vitamins, the most important of which are vitamins B3, B2, B1, E, C, A, in addition to salicylic acid (19 and 24). Flowers are used in the treatment of burns. The leaves extract by boiling is considered a nerve tonic and moisturizer for the skin (10). The data on the chemical analysis of the active substances anthocyanins; flavones; ferulic acid; hydroxycinnamic acid; sterols; sesquiterpenes (15and 26). The results of the studies showed that the leaves of plants are a good source of some phenolic compounds and antioxidant compounds (7). The plant has also been found to be effective against bacteria and fungi (23). In view of the nutritional and medical benefits that this plant possesses and the scarcity of research on its use in poultry feeds, Therefore, the aim of the above plan was to use this plant as a premix carrier (after collecting its leaves, drying them, chemically analyzing them, and producing an Iraqi premix with the same specifications as the importer, due to its abundant availability and very low cost) as a substitute for the importer in the diets to see the response of the birds by measuring some of the productive and economic traits that appear on broilers during the fattening periods.

Material and Methods

The study was conducted according to the International Guidelines for research involving animals (Directive2010/63 /EU (16), specially slaughtering birds according to the Islamic procedures.

Preparation of *Malva parviflora* weeds leavesmeal

This was carried out using the procedure of (25). Whole plants of weeds were collected from the college gardens in Najaf Governorate. The green plants were harvested freshly from the soil, roots and stems were cut manually ،leaves were transferred to the Animal Production Laboratory. They were washed and carefully inspection to remove all unwanted matters and sun-dried for about three days and once it was dried in the oven. They were then kept in apolythene sacks for further processing. Collections of the weed plant were carried out at one period of the year at the peak of the cold season, during February 2021.They were then dried in forced-oven at 40°C for about 24 h to a moisture content of about 10 %. The dried plants were then grinded using an attrition mill and sieved through a 1 mm sieve to obtain *Malva parviflora* L. weeds leaves powder which was then stored in

large plastic containers with tight-fitting lids until needed.

Chemical analysis measurement of essential ingredients of diets

Chemical composition of the *Malva parviflora* L weed leaves meal (MPM) was determined using the standard procedures of (5).All ingredient's diets determinations have been done in triplicates, including approximate chemical composition table (1) for all macro ingredient , mineral composition for *Malva parviflora* L.leaves meal (Table 2), bioactive compounds table(3) ,amino acids table(4),also made Approximate chemical analysis for all ingredients in dietsCorn,Wheat,Soybean meal(Table4).

Table 1 Approximate chemical composition for ingredient in diets

Ingredients	DM%	ASH%	CF%	EE%	CP%	NFE%	ME, Kcal/Kg
Malva parviflora L. leaves meal	6229	8.83	7.252	2.92	30.0	21.0	1999.8
Locally wheat grain	89.0	1.0	3.2	2.1	13.6	69.1	3150.0
Turkish yellow corn	90.0	2.1	2.3	3.0	8.5	74.1	3353.0
Soybean meal	92.0	10.3	6.1	2.3	48.0	25.3	2232.0

Table 2Mineral composition of *Malva parviflora*leaves meal

Mineral	Composition(ppm)
Phosphorus :P	926
Calcium :Ca	9326
Zinc (Zn)	1290
Selenium (Se)	920
Cobalt(Co)	0129
Magnesium(Mg)	2023
Manganese(Mn)	1223

Table 3Bioactive compounds in *Malva parviflora* weed leaves meal

Bioactive compounds	Value
Total alkaloid content %	323
Total phenolic content (mg Gallic/100gm)	26921
Total anthocyanin content(mg/100gm	1290
Total flavonoids content (mgRutin/100gm)	6929
Total glycoside content %	926

Table 4 Amino acids content in *Malva parviflora* weed leaves meal

AA	Tyrosine	Arginine	Aspartic	Glutamic	Glycine	Lucien
D.M%	2.69	1.26	1.58	2.00	3.05	3.07
A.A	Lysine	Methionine	Phenyl alanine	Serine	Valine	Asparagine
D.M%	3276	2296	0266	0269	2239	2236

Table 5 Composition of locally and imported premixes

component	Locally premix	Provimi premix	Turkish premix	Dutch premix
ME Kcal/Kg	0211	9311	9111	0691
Crude protein%	09	07	97	11.2
Crude fat %		1.5		
Moisture %	7.4	10	02	10
		Vitamins		
Vit. A(i.u)	480 000	931111	12.000.000	400 000
Vit.D3(i.u)	140000	221111	5.000.000	120000
Vit.E(i.u)	1333	3111	80.000	2111
Vit.K3(i.u)	100	033	3.200	021
Vit.B1(mg)	83	033	32211	091
Vit.B2(mg)	200	231	8.600	300
Vit.B6 (mg)	400.0	091	4.300	211
Vit.B12(mg)	0.7	0	07	0
Vit.B3(mg)	1333	0311	60.000	2111
Vit.B5(mg)	400	911	17.000	600
Folic acidB9 (mg)	50	93	2.200	91
BiotinB7 (mg)	3.3	9	221	9
Cholin B4(mg)	1	21111	300.000	1
		Minerals		
A val.P%	1.06	13.7%	14.6	1.04
Total P%		6.7%		
Total Ca		15.0%	15.6	
Na	-	4.8%	5.9	929
Cl		5.8%		
Mn(mg)	2666.7	3211	0212111	3211
Fe(mg)	1566.7	2911	912111	22911
Zn(mg)	2666.7	3331	0012111	2911
Cu(mg)	333.3	931	15.000	911
Co(mg)	8.3		—	1
I(mg)	50	93	1.250	2311
Se(mg)	6.7	01	311	02

BHT*	-	250 mg/Kg	Sepiolite1.075.00	1
Methionine%	06	7.8%	02261	8.5
Lysine%	01	9.3%	72111	5.4
Meth+Cys%	2.57	%723	12.5	6
Therionine%	1	0.4%	1.8	0.5
Phytase(u/kg)	1500	36211	0112111	-
Glucanase(u/kg)	-	211111	-	-
Amylase(u/kg)	3000	021111	-	-
xylanase (u/kg)	32000	31111	-	-
Protease(u/kg)	9111			
Multi enzyme**	-	-	0112111	-
Organic acids %	-	0.8	-	-

*BHT:mean antioxidant (Butylated Hydroxytoluene)

**Multienzyme :(Endo-1,4-beta-xylanase,6-phytase,Alpha – amylase,Protease,200 – 500 gm/Ton feed.Technozyme Multi,Germany

Table6 Starter Diets for all treatments

Ingredients	Treatments(1)							
	T1	T2	T3	T4	T5	T6	T7	T8
Corn	43.10	44.7	47.1	41.40	46.6	47.2	42.20	44.08
Soybean meal(48% CP)	36.50	39261	34.2	37.50	36261	39261	37.00	36.17
Wheat	3.000	03211	03211	3.000	03211	03211	3.000	13.00
Locally premix*	226	——	——	—	1.25	0226	0226	0.625
Jordanpremix(Provimi)	——	226	——	——	0226	—	——	12926
Turkish premix(BirsenKimya)	——	——	226	——	——	0226	——	12926
Dutch premix (Koudijs)	——	——	——	226	——	——	0226	12926
50% Locally+50% provimi	—	——	——	——	——	——	——	——
50%Locally+50%Turkish	——	——	——	——	——	——	——	——
50%Locally+50%Dutch	——	——	——	——	——	——	——	——
25% from each premix	——	——	——	——	——	——	——	——
Corn oil%	3.60	2.0	1.9	4.30	1.1	1.1	4.00	2.95
Salt%	0.3	0.3	0.3	0.3	0.3	0.3	0.3	0.3
Dicalcium phosphate %**	0.50	0.50	0.50	0.50	0.50	0.50	0.50	0.50
Limestone (Calcium carbonate)%	0.50	0.50	0.50	0.50	0.50	0.50	0.50	0.50
Total	100.0	100.0	100.0	100.0	100.0	100.0	100.0	100.0
Metabolizable Energy,Kcal/Kg	3021.2	3021.2	3021.0	3020.0	3023.0	3024.0	3023.0	3020.8
Crude Protein(CP)%	23.30	23.35	23.33	23.27	23.36	23.35	23.31	23.31

Total Ca%	1.07	1.07	1.07	1.07	1.07	1.07	1.07	1.07
Available Phosphorus %	0.52	0.52	0.52	0.52	0.52	0.52	0.52	0.52
Crude Fiber(CF)%	3.86	3.86	3.86	3.86	3.86	3.86	3.86	3.86
Lysine %	1.43	1.43	1.43	1.43	1.43	1.43	1.43	1.43
Meth.+Cys.%	1.07	1.07	1.07	1.07	1.07	1.07	1.07	1.07
C/P Ratio Calorie/Protein Ratio	128.7	128.7	128.7	128.7	128.7	128.7	128.7	128.7

(1)T1:basal diet .(Control)contained2.5%ofdietlocallypremix(itscarrier*Malvaparviflora*leavesmeal; T2: 2.5%provimipremix; T3: 2.5%Turkishpremix; T4: 2.5% Dutchpremix; T5: 50%locally+50%provimi; T6: 50%locally+50%Turkish ; T7: 50%locally+50%Dutch; T8:0.625% from each four types premixes .

* Locally premix its carrier *Malvaparvi flora* leaves meal Jordan premix-Provimi, Turkish premix- BirsenKimya, Dutch premix-Koudijs

**DiCalcium Phosphate (Turkish) Contain:22% Inorganic Calcium,18%Inorganic Phosphorus.

Table 7 Finisher diets for all treatments

ingredients	Treatments(1)							
	T1	T2	T3	T4	T5	T6	T7	T8
Corn	50.60	52.20	63221	46.70	51.4	6029	50.0	51.2
Soybean meal (48% CP)	28.4	28.4	26.4	28.7	2329	27.4	28.7	27.9
Wheat	12.0	12.0	0321	13.0	12.0	13.0	12.3	12.7
Locally premix*	2.5	—	—	—	1.25	0226	0226	0.625
Jordan premix(Provimi)	—	226	—	—	0226	—	—	12926
Turkish premix(BirsenKimya)	—	—	226		—	0226	—	12926
Dutch premix (Koudijs)	—	—	—	226	—	—	0226	12926
50%Locally + 50%provimi	—	—	—	—	—	—	—	—
50%Locally + 50%Turkish	—	—	—	—	—	—	—	—
50%Locally + 50%Dutch	—	—	—	—	—	—	—	—
25% from each premix	—	—	—	—	—	—	—	—
Corn oil%	622	3.6	3.6	620	4.4	929	5.2	4.4

Salt%	123	123	123	123	123	123	123	123
Dicalcium phosphate %**	126	126	126	126	126	126	126	126
Limestone (Calcium carbonate)%	0.5	0.5	0.5	0.5	0.5	0.5	0.5	0.5
Total	100	100	100	100	100	100	100	100
Metabolizable Energy,Kcal/Kg	3204.0	3203.0	3203.0	3202.0	3204.0	3204.0	3204.0	3203.0
Crude Protein(CP) %	20.00	20.10	20.1	20.00	20.03	20.05	20.02	20.01
Total Ca%	1.00	1.00	1.00	1.00	1.00	1.00	1.00	1.00
Available Phosphorus %	0.48	0.48	0.48	0.48	0.48	0.48	0.48	0.48
Crude Fiber(CF) %	3.56	3269	3.56	3269	3.56	3269	3.56	3269
Lysine %	1.43	1.43	1.43	1.43	1.43	1.43	1.43	1.43
Meth.+Cys.%	1.07	1.07	1.07	1.07	1.07	1.07	1.07	1.07
C/P Ratio: Calorie/Protein Ratio	160.0	160.0	160.0	160.0	160.0	160.0	160.0	160.0

(1)T1:basal diet .(Control)contained2.5%ofdietlocallypremix(itscarrier*Malvaparviflora* leavesmeal; T2: 2.5%provimipremix; T3: 2.5%Turkishpremix; T4: 2.5% Dutchpremix; T5: 50%Locally+50%provimi; T6: 50%locally+50%Turkish; T7: 50%locally+50%Dutch; T8:0.625% from each four types premixes .

*Local premixes (itscarrier*Malvaparviflora*leavesmeal)

**DiCalcium Phosphate (Turkish) Contain:22% Inorganic Calcium,18%Inorganic Phosphorus.

Birds and Plan of Nutrition

A total of 576 one-day-old mixed-sex Ross 308 broiler birds were obtained from commercially hatched eggs (Al-Anwar Hatchery-Babylon). They were raised from day old at the Poultry farm of the Animal Production Department. Birds with one day-old-age were randomly allocated to 24floor pens (2 × 1. 5 m) with wood shavings (24 birds per pen). You have 8 treatments and 3 replicates, the floor pens were located in an open-sided house, and each pen was equipped with an AllWaterers are manual. The pen was considered as experimental unit for traits measurements. Eight treatments as following:

Tr1:(Control)contained2.5%ofdietlocallypremix(itscarrier*Malvaparviflora*weedleaves meal,Tr2:2.5%provimipremix,Tr3:2.5%Turkishpremix,Tr4:2.5%Dutchpremix,Tr5:1.25%locally+1.25%provimi,Tr6:1.25%locally+1.25%Turkish,Tr7:1.25%locally+1.25% Dutch,Tr8:0.625% from each fourtypes premixes. Levels the percentage composition of the experimental diets for the starter and finisher is shown in (Table I).These diets were formulated to be iso energetic and iso nitrogenous according to (28), nutrient requirements for broiler,in particular the recommendations for Ross

308 strain. The birds were reared and grown to market age 5 weeks. The birds were also given standard medication and prophylactic treatments as recommended by the Iraqi Veterinary Medical Association for this region.Birds were provided add libitum to feed and water, with constant illumination of 23 h of light and 1 h of dark per day during the entire growing period. The traits studied in the research are blood traits, some blood enzymes and microbial account in small intestine.

Measurements:

Hematological biochemical parametersevaluation

At 35 day of age blood sample were taken from the Brachial vein from six birds from each treatment (1 male and 1 female) / each replicate randomly. Blood sample were used for fresh blood count. Red Blood Cell (RBC) and White Blood Cell (WBC) were measured according to the method of Quinn*et al***.**(29). Packed Cell Volumes (PCV) were measured according toArcher (6). Hemoglobin was directly calculated depending on the PCV values using the equation described byCampbell(9)and H/L ratio were measured according to Varley *et al***.** (36).Forserum biochemical indices, blood sample was drawn and allowed to stand for an hour at room temperature (18 C^0) to serum collection. Serum was separated by centrifugation and stored at −20 C^0 for further analysis. Total protein and albumin were analyzed by a colorimetric method using commercial kits (Spinreact, Spain). Serum globulin was calculated by subtraction from total proteins.Blood serum cholesterol, triglycerides concentrations were determined according to the methods of Tietz (34),uric acid (31), using commercial kits (Spinreact, Spain). The concentration of high density lipoprotein cholesterol (HDL-c) in the serum was estimated by method of Warnick and Wood (37). The low density lipoprotein cholesterol (LDL-c) was estimated as the difference between total cholesterol and high density lipoprotein with triglyceride divide by five as the equations described by (21 and 38). Alkaline phosphatase (ALP) activity was determined according to the methods of Thomas (33).

Microbial Tests**:**

The microbial tests of ileum in small intestine measured by prepared media about Enterobacteriaceae using EMB according to Brook(8),for (differential) Eosin Methylene blue E coli according to Ouinn *et al*.(30), salmonella differential according to Fingegold and Baron(20).For *Lactobacillus* and *Shigella* Isolation used Litmus milk agar and MacConkey agar according to Gill(22).Multi tests have been done,Urease enzyme test according to Collee *et al*.(12), Indol test according to Cowan (13),Citrate utilization test according to Collee *et al*.(12),Methyl red test according to Fingegold and Baron(20),and Catalase test for any positive reaction.

Statistical analysis:

Statistical analysis were conducted using SAS(Version 6, SAS Institute, Cary, NC, USA) (32).Data collected were subjected to analysis of variance (ANOVA) by means of the General Linear Models (GLM) procedure, based on the Randomized Completely Block. Means

were compared using the Duncan's Multiple Range Test (17).

Results and Discussion

Hematological parameters are usually relatedto health status and are of diagnostic importance inclinical evaluation of the state of health. Blood parametersare good indicators of physiological, pathologicaland nutritional status of an animal and changes in hematologicalparameters have the potential of being used toelucidate the impact of nutritional factors and additivessupplied in diet on any living creature.

Tables 8, 9 show the effect of experimental diets on hematological traits of blood and differential white blood cells, respectively.RBC were not affected by the type of premix in the diet.PCV, had significantly higher ($p \leq 0.05$) percent in T6(34.30%) while the opposite trend was shown in T1,T5(31.50,31.60%),The last two, in turn, did not differ significantly with the rest of the treatments.Hb concentrates recorded the highestsignificantly value in T6(13.90g/dl) versus the lowest value (11.80g/dl) in T5, The latter, in turn, did not differ significantly with the rest of the treatments.Lymphocyteshad significantly highervalue in T3.T8(79.00, 77.54%)versus the lowest value in T7(64.52%) ,The latter, in turn, did not differ significantly with the rest of the treatments. Heterophilshad significantly higher value in T2 (5.23%) versus the lowest value in T5 (3.07%), The latter, in turn, did not differ significantly with the rest of the treatments.WBC had significantly higher value in control T1 (94.66) versus the lowest value in T2(85.33) ,The latter, in turn, did not differ significantly with the rest of the treatments.

The hematological values obtained in this study indicated no detrimental impact of *Malvaparvifloral*eavesmeal on RBC and WBC counts, hemoglobin content and hematocrit percentage2Reports on the effect of *Malvaparvi flora* leaves meal as carrier for local premixon blood hematological parameters are very scarce.

Table 9, show were not significant differences in some biochemical blood traits,Total protein,Albumin,Cholesterol,Triglycerides, LDL and HDL respectively, while there were significant differences ($p \leq 0.05$) in Glucose concentrate had significantly higher value in T1 (172.66mg/dl) versus the lowest value in T3(158.66mg/dl),The latter, in turn, did not differ significantly with the rest of the treatments.

Table 8.Effect of different treatments on some hematological parameters of 35 d old broiler

Treatments(1)	PCV%	Hb g/dl	Lymphocyte %	Heterophils %	WBC (1000/ml)	RBC (10^{12})/L
T1	31.50 ± 0.43^{b}	12.50 ± 0.74^{ab}	74.80 ±2.33^{ab}	4.68 ± 0.23^{ab}	94.66 ± 2.18^{a}	2.62 ± 0.28
T2	32.60 ± 1.02^{ab}	12.36 ± 0.24^{ab}	73.80± 5.25^{ab}	5.23± 0.21^{a}	85.33±1.76^{c}	2.11±0.06
T3	32.03±0.77^{b}	12.33±0.48^{ab}	79.00±0.00^{a}	4.66±0.00^{ab}	91.00±0.57^{abc}	2.59±0.18

T4	33.06±0.56^{ab}	13.13±0.34^{ab}	74.96±4.47^{ab}	4.50±0.69^{abc}	85.66±1.45^{c}	2.43±0.10
T5	30291±0.3^{b}	11.80±0.40^{b}	73.28±4.65^{ab}	3.07±0.07^{d}	88.66±2.60^{abc}	2.18±0.11
T6	34.30±0.62^{a}	13.90±0.32^{a}	73.25±3.22^{ab}	4.10±0.14^{abcd}	93.33±1.45^{ab}	2.39±0.30
T7	33.10±0.60^{ab}	12.96±0.18^{ab}	64.52±4.47^{b}	3.86±0.54^{abcd}	88.00±2.30^{bc}	2.45±0.15
T8	33.40±0.45^{ab}	12.96±1.10^{ab}	77.54±2.06^{a}	3.52±3.22^{cd}	87.33±2.02^{bc}	2.38±0.16
Significant level	*	*	*	**	*	N.S

(1)T1:basal diet .(Control)contained2.5%ofdietlocallypremix(itscarrier*Malvaparviflora*leavesmeal; T2: 2.5%provimipremix; T3: 2.5%Turkishpremix; T4: 2.5% Dutchpremix; T5:50%locally+50%provimi; T6:50%locally+50%Turkish; T7: 50%locally+50%Dutch; T8:0.625% from each four types premixes .

*The different letters within the same column indicate that there are significant differences between the averages at probability level (P <0.05)

**The different letters within the same column indicate that there are significant differences between the averages at probability level (P <0.01)

N.S:Non significant

Table 9. Effect of different treatments on some biochemicalparameters of 35 d old broilers

	Age(5)week Mean ± SE						
Treatments(1)	Total protein gm/dl	Albumin gm/dl	Cholesterol mg/dl	Triglycerides mg/dl	LDL mg/dl	HDL mg/dl	Glucose mg/dl
T1	4.2±0.25	1.10±1216	108.66±10.13	58.0±9.71	20.66±1.20	65.66±7220	172.66±5.45^{a}
T2	4.33±0.08	1.13±0.03	110.00±1.73	57.33±6219	21.3±0.33	60.33±3.48	169.00±2260^{ab}
T3	4.23±1221	1.06±0.06	71.86±35.25	56.00±4.16	21.66±0.66	64.00±3.78	158.66±**3.17**^{c}
T4	4.16±0.06	1.16±0.03	±0132993.28	65.00±3.78	22.66±1.20	57.66±2.96	167.00±1.52^{abc}
T5	4.46±0.08	1.06±0.03	109.66±2.33	67.33±2.33	21.33±0.88	62.33±4.33	162.33±2.33^{bc}
T6	4.33±1208	1.16±1213	113.33±1.76	72.00±6261	21.66±0.33	64.33±1.85	166.33±2.60^{abc}
T7	4.26±0.17	1.16±0.08	108.33±0.88	67.33±4.84	21.33±0.88	64.00±3.60	167.66±2.60^{abc}
T8	4.50±0.21	1.10±0.05	110.00±1.15	67.33±2.40	20.33±0.33	68.00±4.50	163.33±2.18^{abc}
Significant Level	N.S	N.S	N.S	N.S	N.S	N.S	*

(1)T1:basal diet .(Control)contained2.5%ofdietlocallypremix(itscarrier*Malvaparvifloral*eavesmeal; T2: 2.5%provimipremix; T3: 2.5%Turkishpremix; T4: 2.5% Dutchpremix; T5: 50%locally+50%provimi; T6:

50%locally+50%Turkish; T7: 50%locally+50%Dutch; T8:0.625% from each four types premixes .

*The different letters within the same column indicate that there are significant differences between the averages at probability level (P <0.05)

N.S:Non significant

Table 10 show value for some enzymes and minerals in blood serum of birds,ALPs had significantly (p≤0.05) higher value in control T8(1167.67 unit) versus the lowest value in T1(1082.00 unit) , The latter, in turn, did not differ significantly with T5(1132.00).ASTs had significantly (p≤0.05) higher value in T7(403.66 IU/L) versus the lowest value in T4(256.00), The latter, in turn, did not differ significantly with T1,T2,T3.ALTs had significantly (p≤0.05) higher value in T4(6.00 IU/L) versus the lowest value in T3(4.33 IU/L), The latter, in turn, did not differ significantly with the rest of the treatments.Phosphorous concentrationhad highsignificantly(p≤0.01) in T6, T7(4.00,4.20mg/dl)versus the lowest value in T2,T3,T1 (3.03,3.03,3.10mg/dl) respectively, but there were not significant differences between all treatments in Calcium concentration.

Table 10 show significant differences (p≤0.05) for all treatments in ALP,AST,ALT enzymes respectively, higher value in T4 (1181.0 King.unit) and this in turn didn't different significantly with all rest treatments Except for control(T1), it was the lowest significant value among all treatments, as it was recorded(1082.0 King.unit)**,**while higher value in AST were recorded in T7(403.66 IU),T6 (374.66 IU),T8(368.66 IU),T5(360.66 IU) and The latter, in turn, did not differ significantly with T3(300.33 295.66 IU),T1(262.33 IU), and The latter, in turn, did not differ significantly with T4(256.0 IU),Also the same trends in ALTs values, The highest values recorded by the treatments T4(6.00 IU),T2(5.66IU), T5and T7(5.33IU),T8 and T1(5.00IU), The last two did not differ significantly with T6(4.66IU),T3(4.33IU). However, high significant differences (P≤0.01) appeared between treatments in phosphorous concentrations,he highest values recorded by T7(4.20 IU),T6(4.00 IU),T4 and T5(3.86 IU), The last two, in turn, did not differ significantly with T8(3.53 IU), and The latter, in turn, was morally superior to T1(3.10 IU) and T2,T3(3.03 IU). While there were no significant differences between all treatments in serum calcium concentration in our current study. These results agreements with(1 and 2) who showed the presence of medicinal capabilities in the leaves of the weeds of local plants when used as carriers in the production of local premixes that competed in their abilities with the Dutch premix in the cellular and biochemical characteristics of blood, The promising results of the local premix on the blood characteristics may be due to the

fact that the *Malva parviflora* weed leavesmeal contain bioactive compounds like phenolic content (mg Gallic.100^{-1}gm) 26921as well as flavonoid antioxidants like flavonoids content (mgRutin.100^{-1}gm) 6929,also glycoside content4.9 % which made the health status of the birds in the best way, which was physiologically reflected through the blood parameters.

Table 11 show Numbers of beneficial and harmful bacteria colonies in the ileumfor all treatments, As for the account of beneficial bacteria within the ileum segment in birds, the local premix alone or in half with Dutch premix showed significant superiority in the number of beneficial bacteria compared to the Jordanian or Turkish premix.It was significantly superior in the number of colonies of beneficial bacteria for both treatments T7(92.66 CFU/ml),T1 control(local premix) 92.00CFU/ml,T8(91.33 CFU/ml),The latter, in turn, did not differ significantly with T5(91.00),T6(89.00),T2and T3(88.00),T4(87.66), While we find that the number of colonies of harmful bacteria (*Salmonella* spp)was minimal in the intestines of birds that took a local premix alone or in combination with the rest of the three premixes.Higher significant values ($P \leq 0.01$) were recorded in T4(92.00),T7(90.66),T6(89.00),The latter, in turn, did not differ significantly with T2(88.66),T3(86.00),T5(85.66),The latter, in turndiffer significantly with control treatment (local premix) T1(73.66),Also the same trends with harmful bacteria (E-coli).We may attribute this explanation to the fact that the local premix ((its carrier *Malva parviflora*leavesmeal) contains pharmacologically active substances such as phenols(Gallic 256.0mg /100gm)and alkaloid content(3.5 %) which have a role in inhibiting the number of harmful bacteria and increasing the number of beneficial bacteria,(3 and 4)**.**

Table 10 effect of different treatments on some blood enzymes and Ca,P in blood serum of 35 d old broilers

Treatments[1]	ALP[2] (King. Armstrong unit)	AST(Iu/L)	ALT(Iu/L)	Ca (Mg/dl)	P(Mg/dl)
T1	1082.00±54.94^{b}	262.33±36.72^{cd}	5.00±0.57^{ab}	11.00±0.55	3.10±0.17^{c}
T2	1165.33±10.91^{a}	295.66±51.74^{bcd}	5.66±0.33^{ab}	10.6±0.29	3.03±0.08^{c}
T3	1159.00±21.07^{a}	300.33±44.36^{bc}	4.33±0.33^{b}	10.66±0.49	3.03±0.12^{c}
T4	1181.33±1.66^{a}	256.00±13.79^{d}	6.00±0.00^{a}	10.20±0.26	3.86±0.12^{ab}
T5	1132.00±19.05^{ab}	360.66±23.96^{abc}	5.33±0.66^{ab}	10.60±0.47	3.86±0.06^{ab}
T6	1174.33±4.37^{a}	374.66±23.90^{ab}	4.66±0.66^{ab}	10.30±0.37	4.00±0.05^{a}
T7	1167.67±14.83^{a}	403.66±5.89^{a}	5.33±0.66^{ab}	10.44±0.43	4.20±0.17^{a}
T8	1167.67±7.75^{a}	368.66±20.85^{ab}	5.00±0.00^{ab}	10.70±0.15	3.53±0.21^{b}
Significant level	*	*	*	NS	**

[1]T1:basal diet .(Control)contained2.5%ofdietlocallypremix(itscarrier*Malvaparviflora*leavesmeal; T2: 2.5%provimipremix; T3: 2.5%Turkishpremix; T4: 2.5% Dutchpremix; T5: 50%locally+50%provimi; T6: 50%locally+50%Turkish ; T7: 50%locally+50%Dutch; T8:0.625% from each four types premixes .

(2) ALP (Alkaline phosphatase),AST(Aspartate aminotransferaseenzyme),ALT(Alanine aminotransferase)

*The different letters within the same column indicate that there are significant differences between the averages at probability level (P <0.05)

**The different letters within the same column indicate that there are significant differences between the averages at probability level (P <0.01)

N.S:Non significant

Table (11) Numbers of beneficial and harmful bacteria colonies in the ileum for all treatments

Treatments(1)	Age (5) wk. Mean ± SE *Lactobacillus* spp CFU/ml	*Salmonella* spp CFU/ml	*E-coli* CFU/ml
T1	92.00±1.15^{ab}	73.66±2.60^{c}	79.66±0.33^{c}
T2	88.00±0.57^{c}	88.66±2.60^{ab}	90.00±0.57^{ab}
T3	88.00±1.73^{c}	86.00±1.73^{abc}	84.66±2.02^{bc}
T4	87.66±1.45^{c}	92.00±1.73^{a}	91.66±1.45^{a}
T5	91.00±0.57^{abc}	85.66±2.02^{b}	91.66±0.88^{a}
T6	89.00±0.57^{bc}	89.00±0.00^{ab}	89.66±0.33^{ab}
T7	92.66±1.45^{ac}	90.66±1.45^{ab}	95.00±0.00^{a}
T8	91.33±0.33^{abc}	54.66±0.88^{d}	71.00±4.04^{d}
Significant level	*	**	**

(1)T1:basal diet .(Control)contained2.5%ofdietlocallypremix(itscarrier*Malvaparviflora*leavesmeal;T2:2.5%provimipremix;T3:2.5%Turkishpremix;T4:2.5%Dutchpremix;T5:50%locally+50%provimi; T6:50%locally+50%Turkish ; T7: 50%locally+50%Dutch; T8:0.625% from each four types premixes

*: The different letters within the same column indicate that there are significant differences between the averages at probability level (P <0.05).

**The different letters within the same column indicate that there are significant differences between the averages at probability level (P <0.01)

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References

1- **Al-Haddad,B.H.H.K.** 2020. Comparative of using locally prepared premix (its carrier leaves powder of (*Medicagohosipida*) in diets with imported premixes in the productive and physiological performance of broilers.M.Sc.Thesis. Faculty of Agriculture, University of Kufa. Iraq.

2- **Al-Haddad, B.H.H.K and A. M. A. Alkassar.**2020**.**Comparative of locally premix (Carrier *Medicago hispida* leafs meal) with imported premixes in

diets on the broiler performance.Plant Archives,20(Supplement 2):1880-1886.

3- **Alkassar,A.M.A.**2006**.** The effect of using the locally produced plant protein concentrates as compared to imported protein concentrates on broiler performance. Ph.D. Thesis. College of Agriculture. University of Baghdad.Iraq.

4- **Alkassar,A.M.A.** 2009. Production of pre-prepared (local) mixtures instead of imported animal protein concentrates in broiler diets.Ain Shams University Journal.Conference (4)pp: 2011-2019. Egypt.

5- **A.O.A.C.**1990**.**Official Methods of Analysis. 15th ed. Arlington (VA).Washington. USA.

6- Archer, R. K.1965**.**HaematologicalTechniques For use on Animals. Blackwell. Oxford.UK

7- Beghdad, M. C.;C.Benammar;F.Bensalah;F. Z. Sabri;M.BelarbiandChemat, F.2014**.** Antioxidant activity, phenolic and flavonoid content in leaves, flowers, stems and seeds of mallow (*Malva sylvestris* L.) from North Western of Algeria. Afr. J. Biotechnol., 13(3): 486-491.
DOI: 10.5897/AJB2013.12833

8- **Brook,G.F.; J.S.Butel and Morse,S. A.**2004**.**Jauietz,Melnick,andAdellberg s Medical Microbiology. 23rd ed. Printed in Singpore. pp.168-180.

9- **Campbell, T.**1995**.**Avian hematology (In: Cambell, T., ed. Avian Hematology and Cytology. 2nd ed. Iowa State Press. Iowa. USA.pp.3-156.).

10- **Chakravarty, H. L.**1976**.**Plant Wealth of Iraq (A Dictionary of Economic Plant). Ministry of Agriculture. Iraq.

11- **Classen, B.; F.Amelunxenand Blaschek, W.**2001**.**Ultrastructural Observations on the Rust Fungus *Puccinia malvacearum* in *Malva sylvestris* ssp. Mauritiana. Plant Biology, 3(4):437-442.
DOI: 10.1055/s-2001-16456.

12- **Collee,J.G.;A.G.Fraser and Marmion. B. P.**1996.Practical Medical Microbiology.14thed.Churchil Livingston.USA. pp.937.

13- **Cowan, S. T.** 1974**.**Cowan and Steel's Manual for the Identification of Medical Bacteria, 2nd Ed. Cambridge University Press. Cambridge.pp. 67-83.

14- **Couplan, F.**2003**.**Guide des plantessauvages comestibles et toxiques. Ed. Delachaux et Niestle. Collection. RegneVegtal.

15- **Cutillo, F.;F. D.Abrosca; M. D. Grecaand Florentino, A.** 2006**.**Terpenoids and phenolderivativees from *Malva sylvestris*. Phytochemistry, 67: 481-485.

16- **Directive 2010/63/EU of the European Parliament and of the Council of 22 September** 2010 on the protection of animals used for scientific purposes Text with EEA

relevancehttp://data.europa.eu/eli/dir/2010/63/ojMalik et al.,2013

17- **Duncan. B.D.** 1955**.** Multiple range and multiple F-test: Biometrics, 1142.

18- **El-Rjoob, A. O. and M. N. Omari.2009.**Heavy metals contamination in *Malva Parviflora* L. (Malvaceae) grown in soils near the Irbid-Amman Highway. J. Env. Appl. Sci., 4(4): 433-441.

19- **Esteves, P. F.; A.Sato; A.Esquibel; A.Mariaand and Campos-Buzzi, F.**2009**.**Antinociceptive Activity of *Malva sylvestris*L. Latin American Journal of Pharmacy, 28 (3):456- 459.

20- **Fingegold, S and E. Baron.**1986**.**Bailey and Scott's diagnosticmicrobiology.(C.V.MosbyCo,pp : 210-215).

21- **Friedewald, W. T.;R. J. Levy, and Fredrickson, D. S.** 1972**.** Estimation of the concentration of low density lipoprotein cholesterol in plasma without use of the preparative ultracentrifuge. Cline chem., 18(6):499- 502.

22- **Gill,D.M.**1982**.** Bacterial toxin,a table of lethal amount microbial. Microbiol. Rev., 46(1):86–94.doi: 10.1128/mr.46.1.86-94.1982

23- Islam, M.; E.Ali,;M. A. Saeed;M.Jamshaidand Khan, M. J. J. 2010. Antimicrobial and Irritant activities of the extracts of *Malva parviflora* L., *Malvastrumcorom* and *Elianum* L. and *Amaranthus viridis* L. a preliminary investigation. Pak. J. Pharmacol., 20-23(1and2): 3-6.

24- **Mabrouk, I.;G. Bako and Abubakar, M. A.**2009**.**Antioxidant effect of ethanolic seed extract of *Hibiscus sabdariffa* Linn (Malvaceae) alleviate the toxicity induced by chronic administration of sodium nitrate on some hematological parameters in wistars rats. Adv. J. Food Sci. Technol.,1(1):39-42.

25- **Malik,A.A.;A.Aremu;B.A.Ayanwale and Abdulmojeed.T.**2013**.**Growth performance and nutrient digestibility of pullet chicks fed graded levels of Water Hyacinth [*Eichhorniacrassipes* (Martius) Solms-Laubach] meal diets at the starter phase (0-8 weeks). International Journal of Advanced Research, 1(9):46-54.

26- .**Mihaylova, D.;A. Popova;R.Denkova;I.Alexievaand Krastanov, A.**2015**.***In vitro* antioxidant and antimicrobial activity of extracts of Bulgarian *Malva sylvestris*. LAnnuaire de l'Université de Sofia "St. KlimentOhridski" Faculte de Biologie, 100(4):41-48. First National Conference of Biotechnology, Sofia. Bulgaria.

27- Nagata, T. andY,Ebizuka.2002**.**Biotechnology in Agriculture and Forestry 51. Verlag Berlin Heidelberg. Germany

28- .**National Research Council.**1994**.** Nutrient Requirement of Poultry. 9th ed.National Academy Press. Washington. D. C. USA.

29- **Natt, M. P. and C. A. Herrick .**1952**.** A new blood diluents for counting the erythrocytes and leucocytes of the chicken .Poult. Sci., 31:735-738.

30- **Quinn,P.J.; B.Markey.; M.E.Carter; W.J.C.Donnelly; F.C.Leonard and Magh, D.**2002.Veterinary Microbiology and Microbial disease.the Blackwell Publishing, Company Iowa.,U.S.A.pp .39-50.

31- **Ross, J. G.,C.Christic; W.G. Halliday and Jones, R. M.**1978.Hematological and blood chemistry (comparison values) for clinical pathology. Poult. Vet. Rec., 102:29-31.

32- **SAS,** 2001. SAS/STAT User's Guide for Personal Computers. Release 7.0 SAS Institute Inc., Cary, N. USA.

33- **Thomas, L.** 1995.Alkaline Phosphatase (In. Thomas L. edition. Labor and Diagnose Marburystudents' edition 4:50 – 51.).

34- .**Tietz, N. W.**1999.Textbook of Clinical Chemistry.3rd ed. Burtis, E.R. Ash Wood, W.B. Saunders Company. Philadelphia. pp. 616.

35- Vanassche,O;Z.Golubenko;E.Pshenichnov;I.Arzanova;V.Uzbekov;E.Sultanova; S.Salikhov;H. J. Williams; J. H Reibenspies;L. S. Puckhaberand Stipanovic. R. D.2006.Malvone A, a phytoalexin found in *Malva sylvestris* (family Malvaceae). Phytochemistry, 67(21):2376-2379. DOI: 10.1016/j.phytochem.2006.08.010

36- **Varley, H., A.H.G. Owenlock and Bell, M.**1980.Practical Clinical Biochemistry. Vol. 1. Williams and Heinemann Medical books Ltd. London. England.

37- **Warnick, G. R. and P. D. Wood.**1995.National cholesterol education programrecommendations for measurement of high-density lipoprotein cholesterol: Executive summary. Clin. Chem., 41(10):1427-1433.

38- **Wilson, P. W.1998.**Why treated dyslipidemia. Saudi Med. J., 19(4): 3776-3781.