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Effect of Dietary Supplemention with *Sargassum muticum* algae and *Pulicaria Undulate* (Rabol) on Growth Performance, Blood Parameters, Immune Response, Manure Characteristics and Ammonia Emissions of Broilers

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Abstract

This research aimed to explore the impact of different levels of dietary Sargassum muticum algae and Pulicaria undulata (Rabol) extract on various aspects including performance, immune response, blood parameters, as well as manure and ammonia emissions in broiler chickens. A sum of 315 one-day-olds, the average weight of the bodyis (37.37 ± 0.38) grams, were utilized for the study. These were divided into five groups, each receiving different feed additives incorporated daily into their diets. Growth performance, blood parameters, manure characteristics, and ammonia emissions were carefully monitored and analyzed. The results indicated that the group receiving the T5 diet exhibited the highest total feed intake (2493.40 g) and total gain (TG). Conversely, the group fed the T6 diet showed the lowest feed intake (3938.12 g), followed by the T7 group (4014.41 g). Additionally, the T5, T6, and T7 groups demonstrated superior total feed conversion ratios (TFCR), while the T7 group recorded the highest performance index (PI) and total performance index (TPI%) at 162.13% and 167.12%, respectively. Regarding manure characteristics, the T7 group showed the best results in terms of manure ash content, nitrogen levels, and pH values, with values of 22.03, 1.72, and 5.4, respectively. Furthermore, the T7 group exhibited significantly lower levels of manure ammonia emissions compared to the other groups ($P \leq 0.05$).Overall, dietary supplementation with 0.3% Pulicaria undulata extract per kilogram of feed (T5), or a combination of 1.0% Sargassum powder (100 g/kg feed) and 0.3% Pulicaria undulata extract per kilogram of feed (T6), or 1.5% Sargassum powder (150 g/kg feed) and 0.3% Pulicaria undulata extract per kilogram of feed (T7),

may contribute to protein protection from free radicals and potentially reduce protein consumption in immune globulin synthesis, thus benefiting broiler chicken health and performance.

Keywords: Broilers, *Sargassum muticum***,** *Pulicaria undulata* extract**,** Manure**,** Performance**.**

Introduction

In developing countries, the poultry production sectors are grappling with various issues. Among these challenges, one significant concern is the rising expense of feed, primarily attributed to the exorbitant prices of protein and energy sources **(Abbas, 2013).** Additionally, the industry faces the critical issue of pathogens developing antibiotic resistance due to the indiscriminate and excessive use of antibiotics. Consequently, there is a concerted effort among researchers to identify cost-effective, readily available, and safe alternative sources of protein and energy. Moreover, scientists are actively exploring natural antimicrobial ingredients as potential remedies for this pressing issue. In this concern, **Elnesr et al. (2023)** concluded that supplementation of milk thistle as a natural feed additive may affect the antioxidant, immunity, blood biochemistry, and productive performance in poultry. There is a clear and great interest in the moment of medicinal and aromatic plants as an important source of raw materials for the pharmaceutical treatment of many diseases and as an alternative to the use of chemicals. This is mainly due to the strong biological activity, where it is considered as safe, economical and powerful natural antioxidant. Many investigators reported that, secondary metabolites are known to have many the therapeutic activities against many diseases in human categories; therefore, traditional medicinal plants can be used for treating many diseases (**Ahmed and Ibrahim, 2018)**. Throughout history, humans have relied on plant products both for sustenance and medicinal purposes. Indeed, herbs and spices, known for their natural medicinal properties, have served as essential feed additives for livestock in agricultural practices **(Guo, 2003).** Among the ingredients, protein supplements are very expensive; therefore, it is necessary to look for alternative sources available locally for use as a protein supplement in poultry feed. According to certain studies, the Egyptian *Sargassum spp*., which was collected from the Red Sea's coasts, exhibits chemical components that are considered a promising natural source of antioxidants and anti-inflammatory substances. **(Fawzy et al., 2017; Fouda et al., 2019)**. Sargassum muticum is a brown edible algae. Its extract has several biological properties, including antimicrobial, antioxidant, and anti-inflammatory activities. **(Erum et al., 2017 and Catarino, et al., 2023)**. In essence, Pulicaria species' leaves are frequently employed to add flavor to culinary dishes and brew herbal infusions. Scientific investigations have revealed various advantageous characteristics associated with Pulicaria, notably its antibacterial and antispasmodic attributes, predominantly observed in P. undulata. Moreover, these plants have long-held traditional recognition for their insect-repelling properties and efficacy in easing symptoms of influenza and the common cold. Additionally, they have been utilized in traditional medicine for addressing ailments such as back pain, gastrointestinal issues, and inflammation **(Al-Hajj, et al., *2014*).** Pulicaria Undulata (Rabol) oil stands out as a promising herbal plant, primarily

attributed to its abundance in phenolic compounds and monoterpene hydrocarbons, coupled with its comparatively lower levels of sesquiterpene hydrocarbons. **Mossa et al.** (**1987**). The essential oil extracted from the aerial parts of Pulicaria Undulata demonstrated effectiveness against a broad spectrum of bacteria, including both gram-positive and gram-negative strains (**EL-Kamali et al., 1998**). The chemical composition of essential oils typically comprises terpenoids, phenylpropanoids, and their oxidation by-products. These constituents can vary in both quality and quantity within plants due to environmental factors, genetic variations, and the specific extraction methods utilized (**Bahgat, *et al.*, 2020).** The most widely used analytical method for essential oils analysis is gas chromatography which is far more sensitive than other chromatographic methods, such as TLC. *Pulicariaundulata*is a densely white – woolly branched erect herb. Leaves crowded, dentate, and oblong to linear at the apex, auriculate - amplexicaul at the base. Flower heads yellow solitary terminal or subterminal on peduncules **(Andrews, 1956**). The composition of the steam-distilled oil of the fresh aerial parts of *P. undulata*(L.) Kostel (from Saudi Arabia) was investigated by coupled GC/MS. Therefore, this report aimed to investigate how the addition of dietary Sargassum muticum algae and/or Pulicaria Undulate (Rabol) extracts influences growth performance and biochemical parameters. It emphasizes the importance of evaluating manure characteristics and ammonia emissions to mitigate nitrogen loss excreta.

Materials and Methods

Resources and Manners

The report was managed at EL RAHMA Fowls company farms in KOM OSHEEM, Fayoum Governorate, Egypt, and the Poultry Experimental Station at the Environmental Studies and Research Institute. The research spanned from October 2021 to November 2021. All experimental protocols adhered to the guidelines set forth by the local Investigational Animal-Care Commission and were agreedupon by the official ethics board of the Environmental Studies and Research Institute (ESRI), the University of Sadat City.

Collection and preparation of seaweed (*Sargassum muticum)*

Sargassum muticum was collected from Marsa Allam, Red Sea, Egypt and was directly brought to the workshop in malleablecarrierscovering water to stopthe decline. According to **(Azzazy et al., 2019)**, the identified algae was washed thoroughly with sterilized seawater to remove extraneous materials. The sample was shade-dried until a constant weight was obtained and ground into powder using a blender. The powdered samples were stored in airtight containers and kept in the refrigerator for future use.

Planning of algal excerpts

Seaweed dust was water-logged in glacial solvents, for example, methanol, At a ratio of 1 part substance to 3 parts volume by weight, and left undisturbed for a duration of 48 hours. Then the methanolic extract was prepared. First, the extract was filtered

through a Buchner funnel with *Whatman* No.1 filter paper. Next, the extracts using water were prepared by the same method and were filtrated, then disappeared to aridity under pressure using a vacuum evaporator at 50^{o}C. Next, the crude extracts were weighed, and Seven point five grams per one hundred gramssince methanol solvent was Five point six grams per one hundred grams of aqueous. Finally, then the extracts were tested against antibacterial and antifungal activities (**Yuvraj et al., 2016**). Twenty grams of leaves from the plant were individually extracted with 600 ml of 90% methanol, Ethanol and diethyl ether (separately) in an ultrasonic device at room temperature, The extracts were filtered and the residues were re-percolated three times and were eliminated from the solvent using a rotary evaporator and sample was preserved at 4°C until used.

Preparation of *Pulicaria undulata* extract

The materials used in this study were obtained from the desert near Sadat City, Egypt. Fresh Pulicaria Undulata plants were carefully harvested under controlled conditions (Fig. 1). The entire plant of P. undulata was collected between March and May when it was in the flowering stage, following the procedure outlined by **Morton (1991)** he branches were detached from the plants, and the remaining parts were spread out to air dry under shade at room temperature for one week until they attained a crisp texture suitable for milling. Following this, the entire plants were ground into a fine meal using a hammer mill to produce Pulicaria Undulata powder, which was then integrated into the experimental diets.

Extracts were prepared by the method of **Bhavitaa et al. (2011)** with few modifications. The leaves of *P. undulata* were air-dried and powdered. 20g of the powder was taken with 100 ml of 70% methanol to prepare the methanolic extract. This was mixed in a closed flask for 72 h. Then it was filtered with a strainer and the remainder vanished to waterlessness, the weight of the water-soluble extractive value was obtained by calculating the difference in weight concerning powdered dried material and the heaviness after extract process (**Nishat Ansari and Divya Chandel, 2019).**

Fig. 1. The whole plant of *P. Undulata* were collected from March to May at flowering stage

Analysis of inorganic elements in Pulicaria undulate (Microwave digestion)

A0.5 g of crushed air-dried aerial parts of *P. undulata*was mixed with a 10 ml concentrated HNO3 in the beaker glass was placed inside a domestic Microwave oven. Sample was irradiated at a 900 W power for 10 min. Then, a 5 ml of concentrated HCl was added andirradiation was continued for another 5 min. After digestion, the vessel was cooled, filtered (Whatman No. 42 filter paper) and diluted with double distilled water to a final volume of 100 ml. Solution was used for elemental analysis by atomic absorption spectrophotometer model Philips PU 9100X and UV- Visible spectrophotometry were used to quantify metal levels and Phosphorus quantity, respectively.

Atomic absorption spectrophotometer was used for determination of Ca, Mg, K, Fe, Cr and Mn, the phosphor was estimated by UV- Visible spectrophotometer (at awavelength of 420 nm) according to the method described by **Ravandeh et al. (2011)**.

Trial proposal

A sum of 315 one-day-old chicks was acquired from the commercial flock of El DESOKY company's broiler breeder (**Indian River Strain**). All chicks were brooded together at the first two days. At the third day, chicks of each replicate (45 for each groups), having average body weight around the one day-old chick weight (37.37 ± 0.38), were used to measure their growth performance up to 42 day of age. Birds were allowed to free access to feed and water during the fattening period. The birds were kept in 21 floor pens ($3m^2$), which covered with wood shavings as litter material. Each pen was equipped with two hanging feeder and one drinker. The lighting cycle was 24 hrs. /d maintained. Vaccination program was applied during the growing period.All experimental birds were kept under the same managerial conditions.

The chicks received starter diet till 1 to 10 d of age, grower diet from 11 to 24 d of age and finisher diet from 25 to 42 d of age. The experimental diets (starter, grower and finisher) were formulated to cover the nutrient requirement of broiler chicks from 1 to 42 d according to breeder's company guideline IR strain (2018) and experimental condition. Chemical composition and chemical analysis of grow-out excremental and basal diets during fattening period (starter, grower and finisher) are shown in Table 1 and 2.

Prepare a premix containing the following for every three kilograms: Vitamin A twelve thousand international units, Vitamin D3 two thousand international units, Vitamin E forty milligrams, Vitamin K thirty-four milligrams, Vitamin B1 three milligrams, Vitamin B2 six milligrams, Vitamin B6 four milligrams, Vitamin B12 zero point zero three milligrams, Niacin thirty milligrams, Biotin zero point zero eight milligrams, Pantothenic acid twelve milligrams, Folic acid one point five milligrams, Choline chloride seven hundred milligrams, Manganese eighty milligrams, Copper ten milligrams, Selenium zero point two milligrams, Iron forty milligrams, Zinc seventy

milligrams, and Cobalt zero point two five milligrams.*** Calculated according **to A.O.A.C. (2006).**

Table 1. Elements and chemical arrangement of commercial diets.

Elements	**Start**	**Grow**	**Finish**
Ground yellow corn (8.5%).	57.260	62.220	69.055
Soybean meal (44.0%).	33.700	24.800	18.90
Corn gluten meal	3.750	6.000	6.00
Sun flower oil.	0.600	1.300	1.200
Lime stone	1.850	1.800	1.500
Mono calcium phosphate	1.400	1.200	1.100
L- lysine	0.320	1.800	1.500
Vitamin & Mineral premix	0.300	0.300	0.300
Sodium chloride (NaCl).	0.300	0.300	0.299
DL-Methionine	0.235	0.220	0.223
Sodium bicarbonate	0.220	0.220	0.218
L-Theronine	0.065	0.070	0.114
Total (kg)	**100.0**	**100.0**	**100.0**
Calculated diet composition			
Crude protein %.	22.440	20.47	18.450
Metabolizable energy (Kcal ME/Kg).	3003	3152	3222
Lysine %.	1.280	1.200	1.060
Methionine %.	0.580	0.560	0.540
Methionine + Cysteine%.	0.960	0.910	0.870
Calcium %.	1.050	0.980	0.840
Available phosphorus %.	0.500	0.450	0.420

*According to IR broiler performance guide requirements (2014) and NRC (1994).

Prepare a mixture comprising the following nutrients for every three kilograms: Twelve thousand international units of Vitamin A, two thousand international units of Vitamin D3, forty milligrams of Vitamin E, thirty-four milligrams of Vitamin K, three milligrams of Vitamin B1, six milligrams of Vitamin B2, four milligrams of Vitamin B6, zero point zero three milligrams of Vitamin B12, thirty milligrams of Niacin, zero point zero eight milligrams of Biotin, twelve milligrams of Pantothenic acid, one point five milligrams of Folic acid, seven hundred milligrams of Choline chloride, eighty milligrams of Manganese, ten milligrams of Copper, zero point two milligrams of Selenium, forty milligrams of Iron, seventy milligrams of Zinc, and zero point two five milligrams of Cobalt.

Table 2. Ingredients and chemical composition of control (-).

Ingredients*	**starter**	**grower**	**finisher**
Ground yellow corn (8.5%).	60.255	64.820	70.895
Soybean meal (44.0%).	33.700	27.00	21.300
Corn gluten meal	0.700	1.600	1.500
Sun flower oil.	0.600	2.00	2.00
Lime stone	1.900	1.850	1.550
Mono calcium phosphate	1.300	1.100	1.065
L- lysine	0.425	0.420	0.420
Vit & min premix	0.300	0.300	0.300
Sodium chloride (Nacl).	0.265	0.265	0.265
DL-Methionine	0.295	0.315	0.315
Sodium bicarbonate	0.220	0.230	0.230
L-Theronine	0.040	0.100	0.160
Total (Kg)	100.0	100.0	100.0
Calculated diet composition			
Crude protein %.	20.49	18.15	16.51
Metabolizable energy (Kcal ME/Kg).	3000	3153	3225
Lysine %.	1.380	1.200	1.09
Methionine %.	0.610	0.610	0.59
Methionine + Cysteine%.	0.960	0.800	0.89
Calcium %.	1.050	0.980	0.85
Available phosphorus %.	0.500	0.440	0.42

*According to IR broiler performance guide requirements (2014) and NRC (1994).

Treatments

T_1 :(Control+) :(commercial) received a dietary 23% CP starter and 19% CP Grower and finisher.

T_2: (Control-): received 20% CP starter and 16% CP Grower and finisher.;

T_3 : received **T_2**+ *0.3% Pulicaria Undulata* extract /kg feedin drinking water.

T_4: received **T_2**+ BHT 150 *ppm +15 ml Sargassum* algae extract /kg feedin drinking water.

T_5: received **T_2** + 0.5% *Sargassum* powder (50 g/kg feed)+*0.3% Pulicaria Undulata* extract /kg feedin drinking water.**,**

T_6: received **T_2**+ 1.0% *Sargassum* powder (100 g/kg feed).+*0.3% Pulicaria Undulata* extract /kg feedin drinking water.**,**

T_7: received **T_2**+ 1.5% *Sargassum* powder (150 g/kg feed)+*0.3% Pulicaria Undulata* extract /kg feedin drinking water.

Data collected
Data collected during the fattening period

The broilers' growth performance was analyzed by assessing their initial body weight and subsequent live body weight (LBW), body weight gain (BWG), feed intake (FI), feed conversion ratio (FCR), and mortality rate (MR) at one, seven, fourteen, twenty-one, twenty-eight, and thirty-five days of age for each experimental group.

Growth performance
Live body weight

Chicks were individually weighed at the start of the experiment (at 1 day of age) to the nearest gram in each group to avoid differences in weight at the start of the experiment and the weight was repeated every week till the close of the trial growing stage (42 d of age). Live body weight (LBW) for each group was totalled and divided on the number of chicks to obtain the average live-body-weight.

Body-weight-gain

Body weight gain -BWG- in the weight was calculated by subtracting the average initial LBW of a certain period of each replicate from the average final LBW of the same replicate and the No. of days to obtain the body weight gain. Therefore, BWG was calculated by the following equation: **BWG = (WX - W0)**
Where BWG is body weight gain, **WX**: is weight at any age, and **W0: is** weight at the previous age certain period in the same replication.

Feed intake

Feed was provided for each group alone by satisfied amount, and the residual feed was weekly collected, weighed and subtracted from the offered one to obtain feed intake on a group basis. Feed intake (FI) was calculated as the following equation:

FI (g/bird) = feed intake in a replication / No. of live birds in the same replication.

Feed conversion ratio (FCR)

This parameter was calculated as the amount of feed consumed per unit of weight gain. The feed conversion ratio (FCR) was calculated by using the following formula: FCR = Total FI (g) / BWG (g).

Performance index (PI)

The PI weekly and the whole experimental period was calculated for each replicate under each treatment giving to the calculation reported by **North (1981)** as follows:

$$PI\% = \frac{LBW(kg)}{FCR} \times 100$$

Plasma biochemical analysis
Plasma total proteins (TP)

Total plasma protein (g/dl) was determined by a calorimetric method using the Buriet method as described by **Cannon et al. (1974).**

Plasma albumin (A)

The concentration of plasma albumin (grams per deciliter) was determined using bromocresol green at a pH of four point two. The optical density of the standard or the samples was measured at a wavelength of six hundred twenty-eight nanometers.

Plasma globulins (G)

Plasma globulin concentration (g/dL) was calculated by deducting plasma albumin from total plasma proteins.

Albumin/Globulin ratio (A/G ratio): It was calculated as follows:

A/G ratio= plasma albumin (g/dl)/plasma globulins (g/dl)

Blood parameters

In order to evaluate the liver function under the effects of the physiological and nutritional factors, the plasma enzyme concentrations (u/l) of both alanine amino transferase (ALT) and aspartate amino transferase (AST) were assayed. Urea and lipid Profile include with (cholesterol – Triglycerides) were estimated by using commercial Kits according to the methods described by **Young (2001)** for Triglycerides and **Tietzard (1995)** for cholesterol.

Manure characteristicsand ammonia emissions determination

Samples were collected from all sections of one replicate, reaching the full depth of the litter, following a survey sampling of poultry litter. These samples were then thoroughly mixed to create a composite sample. This process was repeated weekly for each replicate. Equal amounts of composite from each replicate were combined and thoroughly mixed, and then one pint of the composite was placed in labelled freezer bags according to the designated protocol **Eladawy (2017)**. Manure pH was determined according to **Abousekken et al. (2018)** and **Bahgat et al. (2020).**

At the end of the fattening period, broiler chickens were weighed and feed consumption was recorded. Four-houraerial ammonia concentration samples were taken with Dräger long-term diffusion tubes at broiler chickens' height in 3 different

locations, and plastic odour bags were filled with room air according to **Kendall et al. (1989).**

Statistical analysis

An analysis of variance (ANOVA) was performed using the general linear model (GLM) methods described in SAS (two thousand nine). Distinctions between treatment averages were identified using Duncan's Multiple Range Test (Duncan, nineteen fifty-five). All conclusions regarding significance were drawn with a significance threshold of P less than or equal to zero point zero five. The statistical model utilized in the study was as follows:

$$Yij = M + Ti + Eij$$

Yij = individual observation,
M = overall mean,
Ti = treatment effect and
Eij = experimental error.

Results and Discussions

Proximate analysis of *Sargassum muticum* algae

The chemical composition of the studied algae is found in Table 3. The chemical composition showed that Sargassum muticum was CP; CF and ash (8.4; 17.9 and 28.1) % respectively (Table 3). Similar results were reported by Marín et al. (2009) who found that the chemical composition of Sargassum was 7.7; 6.4; 0.45 and 33.3% for CP; CF; EE and ash, respectively.While, Azad and Teo Zhi Xiang (2012) found that Sargassum sp. contains 13.85; 7.58; 0.48; 24.88 and 53.21% for CP; CF; Lipid; ash and NFE, respectively.

According to the information in Table 3, Sargassum muticum contains 8.7% crude protein, 11.12% crude fiber, good amounts of NDF (28.4%), ADF (20.73%), and only small amounts of lignin (4.5%) and fats (1.62%). Additionally, Sargassum muticum has high levels of complex carbohydrates and polysaccharides, as evidenced by its high Gross energy content (8.55MJ/kg). According to (Evans and Critchley, 2014) who reported that brown algae contain laminarin, sulphated fucose-containing polymers, and alginates.

Mineral elements of *Pulicaria Undulata*

Determination Mineral elementsof *Pulicaria Undulata*a ttempts to contribute knowledge of the nutritional properties of this plant. This may be due to various fractions of dissolved organic matter. Table 4 shows the percent of various metals in this plant. According to results, the highest mineral contents were Ca and K (2.975 and2.013). The present study suggested that the chemical composition and mineral elements in *Pulicaria Undulata* can be used it as an effective natural source of antioxidant and feed additives and also a good candidate for phytochemical and pharmacological investigations to discover new broad spectrum bioactive compounds. These results agree with those obtained by **Mehdi et al. (2011).** Also, **Al-Hajj et al. (2014)** determined mineral contents of *P. inuloides*by atomic absorption spectroscopy

and reported that highest levels of K, Mg, Na, Fe and Ca were found in *P. inuloides*to be 159.5, 29.5, 14.2, 13.875 and 5.225 mg/100 g respectively. They suggested that major minerals in *P. inuloides*were K, Mg, Na, Fe and Ca which can be considered as a good source of nutrition.

Table 3. Chemical Composition of *Sargassum muticum.*

Analysis	***Sargassum sp.*** [(1)]	***Sargassum muticum***
Crude protein (%)	8.5 ± 1.8 (10)	8.7±1.31
Crude fiber (%)	10.1 ± 2.4 (9)	11.12±2.11
NDF (%)	29.5 ± 3.4 (5)	28.4±2.71
ADF (%)	21.3 ± 4.4 (4)	20.73±2.85
Lignin (%)	4.5 (1.0–7.9)	3.97±1.89
Ether extract (%)	1.2 ± 0.9 (9)	1.62±1.1
Ash (%)	35.9 ± 12.8 (9)	34.81±7.33
Gross energy (MJ/kg)	9.1 (8.9–9.2)	8.55±1.15

[(1)]According toMakkar et al. (2015).

Table 4. Percentages of mineral elements in *Pulicaria Undulata.*

No	**Elements**	(%) **(Ravandeh et al., 2011)**	**%**
1	Potassium	1.969	2.013
2	Calcium	3.241	2.975
3	Magnesium	0.426	0.414
4	Iron	0.366	0.348
5	Manganese	0.152	0.149
6	Sodium	0.209	0.224
7	Copper	0.010	0.014
8	chromium	0.015	0.013
9	phosphor	0.204	0.197

Productive performanceand carcass traits

Live body weight (g)

Results presented in Table 5 shows the data of live body weight through the experimental broilers period. Latterly of 2nd week ,the outcomespresented that the highest live body weight was recorded in positive control and BHT 150 *ppm +15 ml Sargassum* algae extract /kg feed.in drinking water (T4) (290.93 and 290.11g) but the lowest value was observed in control-ve group (275.33g) at 14 days old. At 28 and 35 days of experiment and total LBW, broilers fed BHT 150 *ppm +15 ml Sargassum* algae extract /kg feed (T_4) and T_6 (1.0% *Sargassum* powder (100 g/kg feed) + *0.3% Pulicaria Undulata* extract /kg feedin drinking water); as well as those in control+ve group had significant ($P \leq 0.05$) higher live body weight than control-ve and other experimental groups (Table 5).

It is interesting to note that using dietary 0.5% *Sargassum* powder (50 g/kg feed) + *0.3% Pulicaria Undulata* extract /kg feedin drinking water. and (T_6) which received dietary 1.0% *Sargassum* powder (100 g/kg feed) + *0.3% Pulicaria Undulata* extract /kg feedin drinking water ($P \leq 0.05$) affected LBW was significantly increased due to different levels 0.5% *Sargassum* powder (50 g/kg feed) + *0.3% Pulicaria Undulata* extract /kg feedand (T_6) which received dietary 1.0% *Sargassum* powder (100 g/kg feed) + *0.3% Pulicaria Undulata* extract /kg feed, As a consequence it related with protective effect of *Pulicaria Undulata* where preserve intestine from coccidiosis **Allen et al., (1997)**, antioxidants and flavonoids Brisibe*et al.*, (2008). Broilers that received diet containing 1.0% *Sargassum* powder (100 g/kg feed) + *0.3% Pulicaria Undulata* extract /kg feedin drinking water had heavy body weights, and inhibitory effect against microorganisms such as Escherichia coli (**Lopes et al., 2008)** thus it had an improvement in the digestive factor.

Live body weight gain

Results of broiler's live body weight gain (LBWG) as affected by supplemented dietary levels of seaweeds and *Pulicaria undulate* (Rabol) extract areshown in Table (6). At two weeks of age, the valuesof LBWG recorded significantly ($P \leq 0.05$)that T_1(control +) T_4, (received basal diet + BHT 150 *ppm +15 ml Sargassum* algae extract /kg feed.in drinking water) the best LBWG values (253.12 and 252.61 g)compared with other tested groups. After 4 wks. of age groupsT_6 (fed basal diet + 1.0% *Sargassum* powder (100 g/kg feed) + *0.3% Pulicaria Undulata* extract /kg feedin drinking water); T_5 (received basal diet + 0.5% *Sargassum* powder (50 g/kg feed).+*0.3% Pulicaria Undulata* extract /kg feedin drinking water.) and T_1 (control +) significantly ($P \leq 0.05$) achieved the best LBWG (922.22; 904.89 and 923.07, respectively). Meanwhile, the group fed basal diet + BHT 150 *ppm +15 ml Sargassum* algae extract /kg feed.in drinking water) (T4) was the worst one (408.11g). After 4 weeks. of age, results indicated that T_6 and T_5 groups detected the best LBWG but the average were non-significant within other tested groups.

Table 6 shows that the group received basal diet + 0.5% *Sargassum* powder (50 g/kg feed) + *0.3% Pulicaria Undulata* extract /kg feedrecorded the best value (2493.40 g) in contrast to both the control and other experimental clusters.These results were in agreement with those obtained by **Ahmed and Ibrahim (2018)** who investigated that

secondary metabolites are known to have many therapeutic activities against many diseases in human categories, therefore, traditional medicinal plants can be used for treating many diseases. Similar results were registered by **Ahmad et al. (2015)**. The study conducted by [author's name] found that phytogenic feed additives contributed to enhanced apparent ileal digestibility of nutrients, which was assessed at 21, 35, and 42 days of age. Furthermore, the study evaluated the synergistic effects on growth rates resulting from oil supplementation. Various combinations of additives were also investigated, yielding significant findings **(Alcicek et al., 2003 and Denli et al., 2004).** Bioactive phenolic compounds in the *Astreacea* family (*Pulicaria Undulata*) are powerful antioxidants traditionally used in diets to improve growth performance and stimulate probiotics, increasing absorbing nutrients **(Ulewicz-Magulska and Wesolowski, 2018).**

Table 5. Live body weight of broilers as affected by supplemented dietary levels of *Sargassum muticum* algae and *pulicaria undulate* (Rabol) extracts (Means ±SE).

Item	**T_1**	**T_2**	**T_3**	**T_4**	**T_5**	**T_6**	**T_7**	**±SEM**	**Sig.**
I LBW	37.81	38.24	37.38	37.50	38.38	37.64	37.71	0.37	NS
LBW W_2	290.93^{a}	275.33^{c}	284.22^{ab}	290.11^{a}	285.56^{ab}	278.89^{bc}	282.89^{abc}	2.90	*
LBW W_4	1214.0^{a}	1084.9^{b}	1171.8^{a}	1206.3^{a}	1187.3^{a}	1201.1^{a}	1178.0^{a}	24.20	*
LBW W_6	2505.8^{a}	2330.7^{b}	2393.7^{ab}	2451.9^{ab}	2531.8^{a}	2547.1^{a}	2467.9^{ab}	56.36	*
FLBW	2511.89^{a}	2331.56^{b}	2404.11^{ab}	2451.89^{ab}	2551.11^{a}	2567.11^{a}	2486.11^{ab}	55.6	*

a, b, c, and d = values within the similarline with different superscripts indicate significant differences ($P < 0.05$). NS = not significant SE= standard error.**Treatments were as follows:

T1 (Control+): Commercial diet containing 23% CP in starter and 19% CP in grower and finisher phases,T2 (Control-): Basal diet with 20% CP in starter and 16% CP in grower and finisher phases, T3: Basal diet from T2 supplemented with 0.3% Pulicaria Undulata extract per kg feed in drinking water, T4: Basal diet from T2 supplemented with 150 ppm BHT and 15 ml Sargassum algae extract per kg feed in drinking water, T5: Basal diet from T2 supplemented with 0.5% Sargassum powder (50 g/kg feed) and 0.3% Pulicaria Undulata extract per kg feed in drinking water, T6: Basal diet from T2 supplemented with 1.0% Sargassum powder (100 g/kg feed) and 0.3% Pulicaria Undulata extract per kg feed in drinking water, T7: Basal diet from T2 supplemented with 1.5% Sargassum powder (150 g/kg feed) and 0.3% Pulicaria Undulata extract per kg feed in drinking water, *** ILBW: Initial live body Weight, LBW: Live body weight, W2, W4 and W6 = meansweeks and **FLBW**: Final live body weight .

Table 6. Live body weight gain of broilers as affected by supplemented dietary levels of *Sargassum muticum* algae and *Pulicaria undulate* (Rabol) extracts (Means ±SE).

Item	T_1	T_2	T_3	T_4	T_5	T_6	T_7	±SEM	Sig.
G2	253.12^{a}	237.10^{c}	246.84^{ab}	252.61^{a}	247.18^{ab}	241.24^{bc}	245.17^{abc}	2.95	*
G4	923.07^{a}	809.56^{b}	887.56^{a}	408.11^{c}	904.89^{a}	922.22^{a}	899.60^{a}	23.2	*
G6	1291.78	1245.78	1221.89	1271.67	1344.47	1346.00	1333.40	50.1	NS
TG	2467.97^{a}	2292.43^{b}	2356.3^{ab}	2415.26^{ab}	2493.40^{a}	2509.47^{a}	2431.86^{ab}	56.0	*

a, b, c, and d = values within a line with other superscripts differ major ($P < 0.05$). NS = not significant. SE = standard error. **T1: Control+ (commercial): received a dietary 23% CP starter and 19% CP grower and finisher. T2: Control-: received 20% CP starter and 16% CP grower and finisher. T3: received T2 + 0.3% Pulicaria Undulata extract/kg feed in drinking water. T4: received T2 + BHT 150 ppm + 15 ml Sargassum algae extract/kg feed in drinking water. T5: received T2 + 0.5% Sargassum powder (50 g/kg feed) + 0.3% Pulicaria Undulata extract/kg feed in drinking water. T6: received T2 + 1.0% Sargassum powder (100 g/kg feed) + 0.3% Pulicaria Undulata extract/kg feed in drinking water. T7: received T2 + 1.5% Sargassum powder (150 g/kg feed) + 0.3% Pulicaria Undulata extract/kg feed in drinking water.***G1, G2and G8 = meansperformance index -TG = meanstotalgain.

Feed intake (FI)

Results of feed intake (FI) as affected by supplemented dietary levels ofseaweeds and *Pulicaria undulate*(Rabol) extract are presented in Table (7). Findings of feed intake (FI) after 4 weeks of the fattening period showed that broilers fed T_6 group (supplemented 1.0% *Sargassum* powder (100 g/kg feed).+*0.3% Pulicaria Undulata* extract /kg feedin drinking water) significantly ($P \leq 0.05$) consumed the lowest amount of feed (1208.46 g) followed by groups fed diet T_5 (supplemented 0.5% *Sargassum* powder (50 g/kg feed).+*0.3% Pulicaria Undulata* extract /kg feedin drinking water) (1293.84 g) and group fed T_7 (supplemented 1.5% *Sargassum* powder (150 g/kg feed) + *0.3% Pulicaria Undulata* extract /kg feedin drinking water) (1309.06g) , Meanwhile , broilers group fed T_2 diet(control (-), consumed the highest feed (g) (1373.33^{a}). It was expected to be the largest proportion of feed consumption associated with regulator and other trial groups.

Asimilar trend was observed among the total feed intake during the hole fattening period(g /chick) where's T_6 group (supplemented 1.0% *Sargassum* powder (100 g/kg feed) + *0.3% Pulicaria Undulata* extract /kg feedin drinking water) significantly ($P \leq 0.05$) recorded the lowest amount of feed (3938.12 g) followed by group fed T_7 (supplemented 1.5% *Sargassum* powder (150 g/kg feed) +*0.3% Pulicaria Undulata* extract /kg feedin drinking water) (4014.41g) and group fed diet T_5 (supplemented 0.5% *Sargassum* powder (50 g/kg feed)+ *0.3% Pulicaria Undulata* extract /kg feedin drinking water) (4020.58 g)**,** compared with control and other experimental groups.(Table 6). These reductionsin feed intake may be due to some improvements of essential oils in the internal environment of broilers'digestive tract.

Outcomes are in harmony with individuals reported by **Tadjong et al. (2017)** who stated that feed intake was affected by oregano essential oil in broiler diets. Therefore, the performance of the broilers seen in the current study may have been enhanced following the addition of AA extract in the diets, probably because of its high protein content and the presence of essential minerals such as sodium, potassium, zinc and manganese, amino acids and vitamins **(Brisibe et al., 2008).**

On the other hand, these results are inconsistent with those reported by **El-Deek et al. (2011)** indicated that using different levels of algae in a broiler diet increased feed intake compared to control treatment. Also, **Erum et al. (2017)** found that substituting 5% *Sargassum muticum* increased feed consumption of birds.

Table 7. Feed intake (g)/2wks of broilers as affected by supplemented dietary levels of *Sargassum muticum* algae and *Pulicaria undulate* (Rabol) extracts (Means ±SE).

Item	T_1	T_2	T_3	T_4	T_5	T_6	T_7	±SEM	Sig.
FI2	408.88^{b}	412.83^{b}	424.90^{a}	428.61^{a}	427.06^{a}	425.28^{a}	425.28^{a}	3.10	*
FI4	1345.54^{b}	1373.33^{a}	1342.73^{b}	1342.72^{b}	1293.84^{d}	1208.46^{e}	1309.06^{c}	4.82	*
FI6	2304.47^{b}	2352.08^{a}	2299.67^{b}	2299.64^{b}	2299.67^{b}	2299.64^{b}	2280.06^{c}	1.57	*
TFI	4058.90^{b}	4138.24^{a}	4067.31^{b}	4070.98^{b}	4020.58^{c}	3938.12^{d}	4014.41^{c}	6.01	*

a, b, c, and d = values within a row with different superscripts differ major ($P < 0.05$). NS = not meaningfully. SE = standard error. ** T1: Control+ (commercial) received a dietary 23% CP starter and 19% CP grower and finisher. T2: Control- received 20% CP starter and 16% CP grower and finisher. T3: received T2 + 0.3% Pulicaria Undulata extract/kg feed in drinking water. T4: received T2 + BHT 150 ppm + 15 ml Sargassum algae extract/kg feed in drinking water. T5: received T2 + 0.5% Sargassum powder (50 g/kg feed) + 0.3% Pulicaria Undulata extract/kg feed in drinking water. T6: received T2 + 1.0% Sargassum powder (100 g/kg feed) + 0.3% Pulicaria Undulata extract/kg feed in drinking water. T7: received T2 + 1.5% Sargassum powder (150 g/kg feed) + 0.3% Pulicaria Undulata extract/kg feed in drinking water. ***W0, W1, and W8 = means weeks - TFI= means total feed intake

Feed conversion ratio (FCR)

The findings presented in Table 8 revealed that the Feed conversion ratio (FCR) of broilers was influenced by the supplemented dietary levels of Seaweeds and Pulicaria undulate (Rabol) extract after 5 weeks of the fattening period, and the total Feed conversion ratio (TFCR) was significantly impacted ($P \leq 0.05$).recorded the best values with T_5,T_6 and T_7groups being (1.79; 1.80; 1.76) and (1.63; 1.59 and 1.60), respectively,while, T_2 group ($control^-$) significantly ($P \leq 0.05$) achieved the worst one (1.99 and 1.83)compared with $control^+$ and other experimental groups. These FCR improvements among T_5, T_6 and T_7 groups may be due to the improvements in feed intake and live body weight gain of these groups caused by feed additives were supplemented to these groups.These results agree with the findings obtained by **Al-Banna et al. (2005)** whoreported that the control group consumed higher feed intake than therabbits fed on *Ulva lactuca* and the group fed on *Gntromorpha intestinalis*. Also, **Rossi et al. (2020)** found that the average daily feed intake was lower in the group fedon a diet supplemented with 0.3% of brown seaweed and plant polyphenols than the other groups. Also, **El-Banna (2003) and El-Banna et al. (2005)** reported that adding seaweed supplementation to the diet of growing rabbits improved the feed conversion ratio possibly because including macroalgae in the rabbit's diet improves feeding efficiency by enhancing gut integrity, nutrient absorption and infection resistance, which improves the productive performance of the growing rabbits. Also, **Rossi et al. (2020)** reported that FCR was positively affected ($P<0.001$) in groups fed on brown seaweed and plant polyphenols with (0.3% and 0.6%). The same trend was detected by **Abu Hafsa et al. (2021)** who found that rabbits fed the *Ulva lactuca* (UL) diet had the highest FCR may be due to the

adequate amounts of Zn in the feed. On the other hand, **Fan et al. (2021)** found that the feed conversion ratio (feed intake / egg mass) decreased in the group that was fed 5% *Sargassum* compared to the rest of the groups.

On the other hand, **El-Deek et al. (2011)** indicated that using different levels of dietary*Sargassum spp.*in broiler finisher diet did not improved the FCR comparing to the control ones.Theyfed brown Sargassum species from the Red Sea shore to laying hens during 20–30 weeks at 1–12% dietary level and detected it had no deleterious effect on BW, egg weight, egg production, FCR.

Table 8. Feed conversion ratio (FCR) of broilers as affected by supplemented dietary levels of *Sargassum muticum* algae and *Pulicaria undulate* (Rabol) extracts. (Means ±SE).

Item	**T_1**	**T_2**	**T_3**	**T_4**	**T_5**	**T_6**	**T_7**	**±SEM**	**Sig.**
FCR w2	1.11^{cd}	1.74^{ab}	1.729^{ab}	1.71^{b}	1.74^{ab}	1.79^{a}	1.75^{ab}	0.03	*
FCR w4	1.49	1.75	1.53	1.49	1.42	1.33	1.58	0.18	NS
FCR w6	1.86^{ab}	1.99^{a}	1.97^{a}	1.89^{ab}	1.79^{ab}	1.80^{ab}	1.76^{b}	0.07	*
TFCR	1.67^{bc}	1.83^{a}	1.74^{b}	1.68^{bc}	1.63^{c}	1.59^{c}	1.60^{c}	0.03	*

a, b, c, and d = The values within the similarline exhibited variationssuperscripts that differ meaningfully ($P < 0.05$). NS = not significant. SE = standard error. ** T1: Control+ (commercial) received a dietary 23% CP starter and 19% CP grower and finisher. T2: Control- received 20% CP starter and 16% CP grower and finisher. T3: received T2 + 0.3% Pulicaria Undulata extract/kg feed in drinking water. T4: received T2 + BHT 150 ppm + 15 ml Sargassum algae extract/kg feed in drinking water. T5: received T2 + 0.5% Sargassum powder (50 g/kg feed) + 0.3% Pulicaria Undulata extract/kg feed in drinking water. T6: received T2 + 1.0% Sargassum powder (100 g/kg feed) + 0.3% Pulicaria Undulata extract/kg feed in drinking water. T7: received T2 + 1.5% Sargassum powder (150 g/kg feed) + 0.3% Pulicaria Undulata extract/kg feed in drinking water.***W2, W4and W6 = weeks -TFCR= totalfeed conversion ratio.

Performance index (PI %)

Results of the Performance Index (PI) at 2, 4, and 5 weeks, as well as the total PI% over the entire 35-day experimental period, influenced by supplemented dietary levels of seaweeds and Pulicaria undulate (Rabol) extract are presented in Table (9). The data on PI (%) indicated that chicks in group T7 exhibited the highest PI value after 5 weeks of the fattening period and for the total experimental duration (35 days) (162.13% and 167.12%, respectively). In contrast, group T2 recorded the lowest PI value (126.39% and 131.07%) as depicted in Table 9, compared to the control and other treatment groups. A similar trend was observed in previous studies conducted by **Kopecky et al. (2012)** for the diet added with organic acids (**Krishan and Narang, 2014).** The beneficial effects of essential oils have been identified, including the enhancement of enzyme secretion linked to food digestion, stimulation of appetite, and activation of the immune response.

Table 9. Performance index (PI %) of broilers as affected by supplemented dietary levels of *Sargassum muticum* algae and *Pulicaria undulate* (Rabol) extracts (Means ±SE).

Item	T1	T2	T3	T4	T5	T6	T7	±SEM	Sig.
PI2	18.39^{a}	15.95^{c}	16.62^{bc}	17.21^{b}	16.62^{bc}	15.72^{c}	16.43^{cb}	0.40	*
PI4	84.42^{ab}	65.28^{c}	78.17^{b}	83.49^{ab}	87.63^{ab}	92.59^{a}	86.58^{ab}	3.17	*
PI6	143.14^{abc}	126.39^{c}	129.66^{bc}	146.56^{abc}	150.56^{ab}	152.64^{ab}	162.13^{a}	7.53	*
TPI	154.56^{ab}	131.07^{c}	140.66^{bc}	154.98^{ab}	159.78^{a}	165.46^{a}	167.12^{a}	5.30	*

*a, b, c, and d = The values within the similar line exhibited variationssuperscripts that differ meaningfully (P < 0.05). NS = not significant. SE = standard error. ****T**$_1$ **:(Control+) :**(commercial) received a dietary 23% CP starter and 19% CP Grower and finisher. **; T**$_2$**: (Control-):** received 20% CP starter 16% CP Grower and finisher**; T**$_3$: received **T**$_2$ + *0.3% Pulicaria Undulata* extract /kg feedin drinking water. **; T**$_4$**:** received **T**$_2$ + BHT 150 *ppm +15 ml Sargassum* algae extract /kg feed.in drinking water.;**T**$_5$: received **T**$_2$+ 0.5% *Sargassum* powder (50 g/kg feed).+*0.3% Pulicaria Undulata* extract /kg feedin drinking water.;**T**$_6$: received **T**$_2$+ 1.0% *Sargassum* powder (100 g/kg feed).+*0.3% Pulicaria Undulata* extract /kg feedin drinking water; **T**$_7$: received **T**$_2$+ 1.5% *Sargassum* powder (150 g/kg feed).+*0.3% Pulicaria Undulata* extract /kg feedin drinking water. ***PI2, PI4and PI6 = meansperformance index -TPI= meanstotalperformance index.

The Blood Chemistry

The Effect of various dietary levels of *Sargassum muticum* algae and *Pulicaria undulate* (Rabol) extracton blood chemistry after 5 weeks of the experimental period is presented in Table 10. Data indicated that the broilers group fed T_7: received a basal diet consisting of 20% crude protein (CP) for the starter phase and 16% CP for the grower and finisher phases + 1.5% Sargassum powder (150 g/kg feed).+0.3% *Pulicaria Undulata* extract /kg feed) meaningfully (p<00.050) achieved The finest serum blood principles of Entire Protein (TP), Albumin (A) and globulin (G) being (6.01; 3.33 and 2.68 mg/dl, respectively).

Meanwhile, results mentioned that there were no significant differences between T_3; T_5; T_6 and T_7 among A/G ratio values being (1.1;1.74; 1.25 and 1.24, respectively) compared with control and other experimental clusters. Drawing from these outcomes, It can be decided that birds feed on foods supplemented with 0.3% *Pulicaria Undulata* extract /kg feed**or** 1.0% *Sargassum* powder (100 g/kg feed) + 0.3% *Pulicaria Undulata* extract /kg feed **or** 1.5% *Sargassum* powder (150 g/kg feed) + 0.3% *Pulicaria Undulata* extract /kg feed **or**1.5% *Sargassum* powder (150 g/kg feed) + 0.3% *Pulicaria Undulata* extract /kg feed was the best immunity compared with the control birds group. The supplementation of 0.3% *Pulicaria Undulata* extract /kg feed**or** 1.0% Sargassum powder (100 g/kg feed).+0.3% *Pulicaria Undulata* extract /kg feed **or** 1.5% *Sargassum* powder (150 g/kg feed).+0.3% *Pulicaria Undulata* extract /kg feed may affect either gram negative or positive bacteria and consequently, the immune cost will be decreased**.**

Results were in agreement with the same trend obtained by **(Ahmed et al., 2018)** ,studied the effect of *Moringa Oleifera* a natural antioxidant as a feed additive on blood chemical composition and immunity of broilers. Dietary supplementation with a mixture of herbs notably enhanced growth performance compared to the control group. Furthermore, it resulted in a significant decrease in plasma total lipids and a simultaneous increase in high-density lipoprotein (HDL) cholesterol levels.

Table 10. Blood Chemistry as affected by supplemented dietary levels of *Sargassum muticum* algae and *pulicaria undulate* (Rabol) extracts (Means ±SE).

Blood parameters*	**Treatments**							**Sig.**
	Control T_1	**T_2**	**T_3**	**T_4**	**T_5**	**T_6**	**T_7**	
Some blood constitutes								
TP	2.71^{b}±0.12	2.7^{b}±0.12	4.66^{a}±0.12	4.52^{a}±0.12	4.61^{a}±0.12	5.5^{b}±0.14	6.01^{a}±0.14	*
Alb	2.16^{d}±0.08	2.01^{d}±0.08	2.44^{c}±0.08	3.55^{b}±0.08	2.93^{a}±0.08	3.05^{b}±2.76	3.33^{a}±2.76	*
Glub.	0.55^{b}±0.10	0.69^{c}±0.10	2.22^{b}±0.07	0.96^{c}±0.09	1.68^{cd}±0.07	2.45^{ab}±0.11	2.68^{a}±0.13	*
A/ G ratio	3.93^{a}±0.07	2.91^{ab}±0.10	1.10^{c}±0.09	3.70^{a}±0.65	1.74^{c}±0.36	1.25^{c}±0.17	1.24^{c}±0.14	*
Some plasma metabolites								
TL (mg/dl)	521.74^{b}±11.5	565.09^{a}±11.5	492.75^{b}±11.5	434.78^{c}±11.5	275.36^{d}±11.5	246.38^{c}±10.9	218.84^{d}±11.0	*
TG (mg/dl)	3.61^{a}±0.07	3.32^{b}±0.07	1.75^{c}±0.07	1.36^{d}±0.07	1.27^{d}±0.07	0.85^{b}±0.06	0.66^{c}±0.06	*
Total cholesterol (mg/dl)	138.25^{a}±0.6	137.2^{a}±0.61	120.7^{b}±0.61	121.85^{b}±0.6	116.39^{c}±0.6	111.66^{c}±0.6	110.56^{c}±0.6	*
HDL cholesterol (mg/dl)	55.16^{a}±0.86	60.0^{a}±1.54	39.37^{b}±0.54	52.22^{a}±1.82	38.23^{b}±1.50	30.91^{bc}±0.3	23.60^{c}±2.67	*
LDL cholesterol(mg/ dl)	43.73^{ab}±3.3	53.62^{a}±6.26	45.40^{a}±5.63	35.55^{ab}±2.2	27.46^{b}±6.21	21.41^{c}±8.26	20.57^{c}±2.98	*
Some plasma enzymes activity and total antioxidant								
ALT/GPT	39^{c}±1.26	35.6^{c}±1.26	48^{b}±1.27	50^{b}±1.28	57^{a}±1.28	57^{a}±1.27	48^{b}±1.27	*
AST/GOT	41^{c}±1.66	36.73^{c}±1.66	36^{c}±1.66	68^{b}±1.66	94^{a}±1.66	70^{a}±1.42	54^{b}±1.42	*
TAC	0.10^{c}±0.02	0.11^{c}±0.02	0.39^{b}±0.02	0.75^{a}±0.02	0.77^{a}±0.02	0.77^{a}±0.02	0.77^{a}±0.02	*

T_1 :(Control+) :(commercial) received a dietary 23% CP and 19% CP Grower and finisher. **; T_2: (Control-):** received 20% CP starter and 16% CP Cultivator and finisher**; T_3**: received **T_2** + *0.3% Pulicaria* Undulata extract /kg feedin drinking water. **; T_4:** received **T_2** + BHT 150 ppm +15 ml Sargassum algae extract /kg feed.in drinking water.;**T_5**: received **T_2**+ 0.5% Sargassum powder (50 g/kg feed).+0.3% Pulicaria Undulata extract /kg feedin drinking water.;**T_6**: received **T_2**+ 1.0% Sargassum powder (100 g/kg feed).+0.3% Pulicaria Undulata extract /kg feedin drinking water; **T_7**: received **T_2**+ 1.5% Sargassum powder (150 g/kg feed).+0.3% Pulicaria Undulata extract /kg feed in drinking water. ***TP Total Protein, Alb:Albumin; G: globuline; A/G ratio: Albumin / globuline ratio; TL: Total Lipids; TG :Triglycerides , HDL: Cholesterol, ALT/GPT:Alanine, AST/GOTAspartateand TAC: Total Antioxidant Capacity.

Additionally, the herb mixture supplementation led to raised levels of plasma total protein and antibody titers against Newcastle disease virus, both prior to and following infection. These findings indicate that incorporating dietary herb mixture supplementation has beneficial effects on growth performance, antioxidative properties, and humoral protection in broiler chickens.Accordingly birds fed diets supplemented *Sargassummuticum* algae and/or *pulicaria undulate* (Rabol) extracts were best immunity compared with control birds group. Proof of this globulin level has been used as indicator of immune responses and source of antibody production. **Griminger (1976)** stated that in bird's high globulin level and low A/G ratio indicated more disease resistance and immune response.

From our observed in this study, we can indicate that this feed additive may save the protein by protecting the protein from free radical and/or decrease protein consumed in immune globulin synthesis (immune cost), this additive decrease globulin synthesis and consequently saved protein which is directed towards growth. We indicated that birds group less globulin (immune cost) is the same group showed the best performance on all trails studied at all periods during this experiment (**Abousekken, 2015 and Abousekken et al., 2018**).

Among results of some plasma metabolites, data presented in Table 11 showed that broiler groups fed T5; T6 and T7 diets meaningfully ($P \leq 0.05$) achieved The finest standards of total lipids (TL), triglycerides (TG), total cholesterol (Cho), HDL cholesterol and LDL cholesterol values in blood plasma of broilers compared with control and other experimental groups. That,s means that the supplementation of 1.0% Sargassum powder (100 g/kg feed) + 0.3% Pulicaria Undulata extract /kg feed or 1.5% Sargassum powder (150 g/kg feed).+ 0.3% Pulicaria Undulata extract /kg feedor 1.5% Sargassum powder (150 g/kg feed) + 0.3% Pulicaria Undulata extract /kg feed had improved the metabolic reactions in the body as a result of improving the digestive tract environment. These results were in agreement with those obtained by **Arslan and Saatic (2003)** whoreported that SBP-diets decreased blood TL and total cholesterol (Cho), values in geese as well as HDL cholesterol values in broiler (**Razdan and Pettersson, 1993 and 1994**).

In another study, **Teteh et al. (2016)** The triglyceride concentration in M1 (1%) was observed to be higher compared to M2 (2%). Elevated triglyceride levels in M1 could potentially be attributed to estrogen synthesis from sterols present in Moringa oleifera leaves, while in M2, increased intake of estrogenic substances by hens may reduce their production, although the potential effects of anti-nutritive substances cannot be discounted.

On the other hand, **Abousekken et al. (2013)** found that plasma cholesterol was not significantlyaffected by BHT or sugar beet pulp (SBP) ethanolic extract at two-level supplementation, although bird groups had received feed additives were lower cholesterol levels. Regarding TL,they found that birds that received an ethanolic extract of SBP at 0.5% had significantly lower compared with control birds,these resultswere due to the ferulic acid effect according to **Ardiansyah et al. (2008)** who reported that a single administration of ferulic acid (9.5mg/kg) may lower blood pressure in rat,also total cholesterol and triglyceride level was found lower.

Among the effect of dietary levels of Sargassum muticum algae and Pulicaria *undulate* (Rabol) extractson some plasma enzymes activity and total antioxidant, results

illustrated in Table 10 showed that birds received that broiler groups fed T_5; T_6 and T_7 diets significantly ($P \leq 0.05$) achievedthe optimal levels of Alanine (ALT/GPT);Aspartate (AST/GOT) and total antioxidant capacity (TAC) being T_5 (57, 57 and 48); T_6 (94, 70, 54 and T_7 (77, 77 and 77), respectively compared with control and other experimental groups.

In this concern, **Salem et al., (2008)** showed slight changes in AST and ALT levels occurred by adding AV®1500 to the diets of Golden Montazah chicks.

Adedapo et al. (2009) reported a significant increase in the levels of ALT and AST for rats fed diets containing 400 mg/kg and 1600 mg/kg doses of extracts of *Moringa oleifera* leaves but **Ologhobo et al. (2014)** did not follow a similar trend The highest mean values of AST and ALT were noticed in his study and that may suggest damage to the liver, as ALT is known to increase in liver disease. However, the increase was not significantly different from the control diet.

The Manure Characteristics after the fattening experiment period

Results of the manure characteristics as affected by supplemented dietary levels of Sargassummuticum algae and pulicaria undulate Rabol) extract in broiler chickens are presented in Table 11. It was observed that broiler groups fed diets T4 (supplemented 150 ppm BHT +15 ml Sargassum algae extract /kg feed T_5 (supplemented 0.5% Sargassum powder (50 g/kg feed) + 0.3% Pulicaria Undulata extract /kg feedin drinking water) andT_6 (supplemented 1.0% Sargassum powder (100 g/kg feed) + 0.3% Pulicaria Undulata extract /kg feed) significantly ($P \leq 0.05$) reduced Manure nitrogen content. compared with control T_2 and T_3groups.Meanwhile, the best manure characteristics were achieved with the broilers group fed the T_7 diet which supplemented 1.5% Sargassum powder (150 g/kg feed) + 0.3% Pulicaria Undulata extract /kg feedsignificantly ($P \leq 0.05$) achievedthe best values of manureash;nitrogencontents and PH value being (22.03; 1.72 and 5.4) compared with control and other experimental groups. These results agree with those obtained by **Abousekken et al. (2018).** Broilers in Group 4 (T4), which were provided with the basal diet supplemented with 3g of Pulicariaundulata powder per kg of diet, significantly ($P \leq 0.05$) achieved the most favourable reductions in nitrogen (N) levels compared to the positive control, according to **Abousekken et al. (2018)**.

The same trend was detected among the effect of various dietarylevels of *Sargassummuticum* algae and *Pulicaria undulate* (Rabol) extract on Manure contents of Phosphors (mg/dl) where the results of group fed T_7 diet (supplemented 1.5% *Sargassum* powder (150 g/kg feed) + *0.3% Pulicaria Undulata* extract /kg feedin drinking water) meaningfully ($P \leq 0.05$) accomplishedthe topstandards of manure Phosphorus contents (54.54mg/dl).

The results regarding manure pH values at 35 days of age, influenced by the utilization of natural or synthetic antioxidants in broiler chickens, are outlined in Table 10. The data demonstrated that T7 significantly ($P \leq 0.05$) attained the most favourable manure pH values compared to both the control and other experimental groups. Conversely, the least desirable manure pH values were observed in T1.

Table 11. Manure components proportionof broilers as affected by supplemented dietary levels of *Sargassum muticum* algae and *Pulicaria undulate* (Rabol) extracts (Means ±SE).

Manure components proportion	**Treatments**							**Sig.**
	Control T_1	**T_2**	**T_3**	**T_4**	**T_5**	**T_6**	**T_7**	
Ash (%)	23.14^{b}±0.65	22.17^{c}±0.65	27.49^{a}±0.65	25.64^{a}±0.65	23.41^{bc}±0.65	28.89^{a}±0.65	22.03^{c}±0.65	*
Nitrogen (%)	3.95^{a}±0.09	2.93^{c}±0.09	3.14^{b}±0.09	2.95^{b}±0.09	2.6^{c}±0.09	2.15^{c}±0.09	1.72^{d}±0.09	*
Phosphors (mg/dl)	66.14^{a}±1.56	63.17^{ab}±1.56	59.99^{bc}±1.56	55.62^{c}±1.56	57.35^{c}±1.56	56.63^{c}±1.56	54.54^{d}±1.56	*
PH value	8.9^{a}±0.1	7.1^{b}±0.1	7.1^{b}±0.1	7.5^{b}±0.1	6.7^{bc}±0.1	5.5^{c}±0.1	5.4^{c}±0.1	*

a, b, c, and d indicate significant differences within the same row, with distinct superscripts denoting statistical significance at the *P≤0.05* level. *NS* signifies non-significance, and *SE* denotes standard error. ****T_1 :(Control+) :(** commercial) received a dietary 23% CP starter and 19% CP Grower and finisher. **; T_2: (Control-):** received 20% CP starter and 16% CP Grower and finisher**; T_3** : received **T_2** + *0.3% Pulicaria Undulata* extract /kg feedin drinking water. **; T_4 :** received **T_2** + BHT 150 *ppm +15 ml Sargassum* algae extract /kg feed.in drinking water.;**T_5**: received **T_2**+ 0.5% *Sargassum* powder (50 g/kg feed).*+0.3% Pulicaria Undulata* extract /kg feedin drinking water.**;T_6** : received **T_2**+ 1.0% *Sargassum* powder (100 g/kg feed).*+0.3% Pulicaria Undulata* extract /kg feedin drinking water; **T_7** : received **T_2**+ 1.5% *Sargassum* powder (150 g/kg feed).*+0.3% Pulicaria Undulata* extract /kg feedin drinking water.

These findings were consistent with those reported by **Shriver et al. (2003)** have observed that reducing crude protein (CP) with amino acid supplementation leads to decreased nitrogen (N) excretion without affecting growth performance. The antimicrobial activity of organic acids is attributed to their ability to lower pH and their lipid solubility in the undissociated form, enabling them to penetrate microbial cells. The pH levels in different segments of the gastrointestinal tract (GIT) influence specific microbial populations and also impact the digestibility and absorptive capacity of various nutrients.

El Adawy (2017) observed a linear relationship between manure characteristics and the increase in dietary Moringa Leaf Protein (MOLP) and Moringa Leaf Extract (MOLX) with laying hens. Additionally, similar trends were observed in groups fed with 5% and 7.5% dietary Moringa Leaf Meal Powder, where significantly worse values (P≤0.05) were recorded likened to the regulatorcluster.

It should be noted that when commercially available amino acids are integrated into diet formulation, there's an approximate reduction of 10% in nitrogen excretion for every 1 percentage point decrease in dietary crude protein **(Shriver et al., 2003)**.

Lenis (1993) and Hartung and Phillips (1994) has indicated that reducing dietary protein excess can result in decreased nitrogen excretion, leading to extensive investigations into determining the precise amino acid needs of pigs and poultry. Furthermore, these studies have noted that diets enriched with feed-grade amino acids produce performance outcomes similar to those fed with intact protein sources. Moreover, integrating feed-grade amino acids into the diet reduces feed expenses compared to relying solely on intact protein sources. As a result, the incorporation of

feed-grade amino acids allows producers to achieve comparable animal performance, reduce feed costs, and alleviate the environmental consequences of nitrogen excretion.

Cromwell and Coffey (1993) has established that lowering crude protein levels in the diet by 2 percentage points by adding commercially available lysine led to a reduction in nitrogen excretion by 17 to 23%. Subsequent studies revealed that additional reductions in dietary protein of 3 to 4 percentage points, achieved by incorporating feed-grade lysine, methionine, threonine, and tryptophan, resulted in a 35% decrease in nitrogen excretion **(Carter et al., 1996).**

In a comparable trial using corn-soybean meal-based diets, it was found that reducing crude protein by 3 percentage points led to a 28% reduction in nitrogen excretion. **(Sutton et al., 1996).** Findings **of Latshaw and Zhao (2011**) indicated that in laying hens, the addition of lysine and methionine to feed to decrease dietary protein by 4 percentage points resulted in a 30% reduction in faecal nitrogen content. These results align with previous studies suggesting that enhanced utilization of commerciallyavailable amino acids led to a 45% reduction in nitrogen excretion in laying hens **(Keshavarz and Austic, 2004).**

Nahm et al. (2011) has found that supplementing poultry diets with antioxidants and enzymes can lead to a reduction in nitrogen excretion by up to 40%. Additionally, supplementation has been observed to decrease intestinal viscosity, enhance metabolizable energy levels, and improve amino acid digestibility.

Ammonia emissions after fattening experimental period

The results of ammonia emissions determination after the experimental fattening period are summarized in Table 12. Analysis of manure ammonia content after 4 and 6 weeks of the fattening period revealed that broiler groups supplemented with 1.0% Sargassum powder (100 g/kg feed) and 0.3% Pulicaria Undulata extract per kilogram of feed (T6), as well as broiler groups fed a diet supplemented with 1.5% Sargassum powder (150 g/kg feed) and 0.3% Pulicaria Undulata extract per kilogram of feed in drinking water (T7), significantly ($P \leq 0.05$) achieved the lowest values of manure ammonia emissions, measuring (11.00; 10.50 ppm) and (10.03; 9.10 ppm) respectively, compared to the control group and the other experimental groups.

As laying hens, **Latshaw and Zhao (2011)** reported that by supplementing feed with lysine and methionine to reduce dietary protein by 4 percentage points, a notable decrease of 30% in nitrogen content in the faeces was observed. These results are consistent with previous studies suggesting that enhanced utilization of commercially available amino acids led to a substantial reduction of 45% in nitrogen excretion (**Keshavarz and Austic, 2004**).

Table 12. Effect of dietary levels of *Sargassum muticum* algae and *Pulicaria undulate* (Rabol) extractson ammonia emissions (Means ±SE).

Ammonia con. (ppm)	**Treatments**							Sig.
	(Control) T_1	**T_2**	**T_3**	**T_4**	**T_5**	**T_6**	**T_7**	
Ammonia ppm) 4 w	23.7^{a}±0.06	15.2^{b}±0.06	12.7^{c}±0.06	12.0^{c}±0.06	11.3^{c}±0.06	11.0^{cd}±0.08	10.5^{d}±0.08	*
Ammonia (ppm) 6w	25.7^{a}±0.07	17.2^{b}±0.07	13.7^{c}±0.06	11.1^{c}±0.07	10.2^{cd}±0.06	10.03^{d}±0.05	09.1^{d}±0.05	*

a, b, c, and d indicate significant differences (P < 0.05) among values in the same row. NS denotes no significant difference. SE represents standard error. ****T_1 :(Control+) :**(commercial) received a dietary 23% CP starter and 19% CP Grower and finisher. **; T_2: (Control-):** received 20% CP starter and 16% CP Grower and finisher**; T_3**: received **T_2** + *0.3% Pulicaria Undulata* extract /kg feedin drinking water. **; T_4:** received **T_2** + BHT 150 *ppm +15 ml Sargassum* algae extract /kg feed.in drinking water.;**T_5**: received **T_2**+ 0.5% *Sargassum* powder (50 g/kg feed).+*0.3% Pulicaria Undulata* extract /kg feedin drinking water.**;T_6**: received **T_2**+ 1.0% *Sargassum* powder (100 g/kg feed).+*0.3% Pulicaria Undulata* extract /kg feedin drinking water; **T_7**: received **T_2**+ 1.5% *Sargassum* powder (150 g/kg feed).+*0.3% Pulicaria Undulata* extract /kg feedin drinking water.

Conclusions

It can be concluded that dietary supplementation with 0.3% *PulicariaUndulata* extract /kg feed **(T_5) or** 1.0% *Sargassum* powder (100 g/kg feed) + 0.3% *PulicariaUndulata* extract /kg feed **(T_6)** or 1.5% *Sargassum* powder (150 g/kg feed) + 0.3% *PulicariaUndulata* extract /kg feed **(T_7)** may save the protein by protecting it from free radicals and/or decrease protein consumed in immune globulin synthesis (immune cost), this additive decreasesglobulin synthesis and consequently saves protein**.**

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