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Effect of neurotransmitters in drinking water and some medicinal flowers in broiler diet additives on body performances and some immune parameters

Nawal K. Shokry, and Mahbuba A. Mustafa

Animal Recourses Department, College of Agriculture Engineering Sci., Salahaddin University, Erbil, Iraq

* *Corresponding author: E-mail*: ***mahbuba.mustafa@su.edu.krd***

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ABSTRACT

This study was conducted to under seek the incidence of some neurotransmitters in drinking water and native medicinal flowers (Alecea Kurdica – Gule Hero and Chamomile) flowers as diet additives on body growth and immune of broiler. 720 one day unsexed hatched chicks will distributed into 12 treatments, each treatment with three replicates (20 chicks), as following: T0 (control -standard diet), [T1 (0.2 g Endorphin), T2 (0.2 g Dopamine) and T3 (0.2 g Serotonin)/ L drinking water], [T4 (0.5 kg Hero flower) and T5 (0.5 kg Chamomile flower)/ 100 kg diet], T6 (0.2 g Endorphin+0.5 kg Hero flower), T7 (0.2 g Dopamine+0.5 kg Hero flower), T8 (0.2 g Serotonin+ 0.5 kg Hero flower), T9 (0.2 g Endorphin+0.5 kg Chamomile flower), T10 (0.2 g/ Dopamine+0.5 kg Chamomile flower), T11= 0.2 g/ L Serotonin/ L drinking water + 0.5 kg Chamomile flower/ 100 kg diet. The addition of neurotransmitters and native flowers additives as immune modulator had significantly affected on body weight, body weight gain and production index and led to to significant increase ($P \leq 0.01$) in relative weights of spleen, Bursa of fabricius, Thymus and antibodies titer against ND and IB diseases, also decreased the stress ratio which represented by heterophil/ lymphocyte (H/L) especially in T3 and T11.

تأثير إضافة الناقلات العصبية في مياه الشرب و بعض االزهور الطبية الى عليقة فروج اللحم على الأداء الإنتاجي و الحالة المناعية

نوال كمال شكري ومحبوبة عبدالغني مصطفى
قسم الثروة الحيوانية ، كلية علوم الهندسة الزراعية ، جامعة صلاح الدين- أربيل / العراق

الخلاصة

أجريت هذه الدراسة لمعرفة مدى تأثير بعض الناقلات العصبية في مياه الشرب والزهور الطبية المحلية (البابونج و زهرة الخطمية) كمضافات غذائية على نمو الجسم والحالة المناعية لدجاج فروج اللحم. وزعت 720 فرخة فاقسة حديثا غير مجنسة بعمر يوم واحد على 12 معاملة: والى 3 مكررات (20 فرحة/ مرر) كما يلي: T0 (عليقية قياسية) ، [T1 (0.2 غم إندورفين) ، T2 (0.2 غم دوبامين) وT3 (0.2 غم سيروتونين)]/ لتر ماء شرب) ،[T4 (0.5 كغم زهرة الخطمية) و T5 (0.5 كغم زهرة البابونج)/100 كغم علف، T6 (0.2 غم إندورفين+ 0.5 كغم زهرة الخطمية) ، T7 (0.2 غم دوبامين+ 0.5 كغم زهرة الخطمية) ، T8 (0.2 غم سيروتونين+ 0.5 كغم زهرة الخطمية) ، T9 (0.2 غم إندورفين +0.5 كغم زهرة البابونج)، T10 (0.2 غم دوبامين+ 0.5 كغم زهرة البابونج) ، T11 (0.2 غم سيروتونين+ 0.5 كغم زهرة بابونج). كان لإضافة الناقلات العصبية وبعض الزهور الطبية المحلية كمحسن للمناعة له تأثير معنوي عالي ($P\leq0.01$) على وزن الجسم وزيادة وزن الجسم. ومؤشر الإنتاج وأدى ايضا إلى زيادة معنوية في الأوزان النسبية للطحال وجراب فابريشيا والغدة الزعترية والمعيار الحجمي للأجسام المضادة ضد مرضي النيوكاسل ND والتهاب الشعب الهوائية IB ، كما قلل من نسبة الإجهاد التي تمثلها الخلايا اللمتغايرة / الخلايا اللمفاوية (H / L) خاصة في T3 وT11 .

الكلمات المفتاحية: إضافات علفية ، دجاج فروج اللحم، نمو الجسم ، معايير المناعة.

INTRODUCTION

Neurotransmitters are chemical signaling and the nervous system's language, which is the primary means through which cells communicate with one another and contribute to the control of feeding behavior in avian species, control of movement, endocrine regulation, immune response and cardiovascular function (Bryant et. al., 2022). Dopamine is one of the principal neurotransmitters in the central nervous system known as "catecholamines" and to their pharmacological characteristics Dopamine has been shown to activate T cell function also led to modulating and suppressing immune functions also It has been implicated in the regulation of normal appetitive behaviors such as feeding, drinking, maternal, sexual behaviors and in pathological behaviors (McCutcheon et. al., 2020: Monge-Fuentes et. al., 2021). Endorphins are ligands for μ-opioid receptors and a natural painkiller, polypeptide, released by the hypothalamus and pituitary gland in pro-opiomelanocortin cells primarily as reaction to pain or stress, (Kaur et. al., 2020) according to McCormack and Denbow (1988) revealed that the ICV injection of β-endorphin lead to increase feed intake, drinking, and body temperature in layers chickens.

Currently, research is being directed toward the use of medicinal plants (feed additives or plant extracts) as antimicrobial, anti-inflammatory, and antiparasitic medications in poultry and its which contain some bioactive compounds with very effective antioxidant activities, resulting in improved feed efficacy, growth, and poultry health (Pliego et. al., 2022). Hero flowers, or *Alcea kurdica*, belong to the family Malvaceae, and are found in the Panjwen, Haj Omran, Garaguin Dokan, Qarahanjir, Derbendi Bazian, Sulaimani, Kani Spi, Kirkuk, Qara Tepe, Kifri, Jabal Hamrin, Baghdad,

Mosul, etc in Iraq (Qader and Awad, 2014). Flavonoids and phenolic compounds are the chemically active compounds in Hero flowers, and are used to treat tonsillitis, stomach ulcers, duodenal ulcers, pneumonia, urinary tract infections, and alopecia in the traditional medical system. It is also used in veterinary medicine to treat carminative coughs and infusions (Mohammadi et. al., 2016) According to Jabbar (2022), hero flowers improve body weight, lipid profile, and immune status in quails, which can be considered as a medicinal plant as well as a growth-enhancing agent in the poultry industry. Chamomile (*Matricaria chamomilla L.*) is a famous medicinal plant distributed worldwide. The active chemicals in chamomile are as sesquiterpenes, flavonoids, polyacetylenes, and coumarins and lead to improving diet palatability and enhancing appetite in poultry, thereby increasing feed intake and improving immunity and production. In the traditional medical use to treat the common cold, liver disorders, neuropsychiatric, and respiratory diseases, this plant is also commonly used to treat pain and infections, as well as skin, eye, and mouth disorders. (Hussein et. al., 2021; Beghoul et. al., 2021).

This study was conducted to investigate the impact of some neurotransmitters (endorphin, dopamine and serotonin) in drinking water and native flowers as diet additives (Gul-e-Hero and chamomile) flowers as immune modulators of broiler immune response on body growth, immune organs and immune titers.

MATERIALS AND METHODS

Present study was undertaken in Poultry Hall/ Grdarasha field /Animal Resources Dept./ College of Agricultural Engineering Sci./ University of Salahaddin-Erbil/ during the period (11-3 to 21-4/ 2021) to investigate the impact of some neurotransmitters (endorphin, dopamine and serotonin) in drinking water and native flowers as diet additives (Gul-e-Hero and chamomile) flowers as diet additives on body growth, antioxidant statue and some immune statues of broiler chickens, one day 720 unsexed hatched chicks (Ross-308) were distributed into 12 treatments (60 chicks for a treatment, each treatment contain three replicates with 20 chicks. T0 (control (standard diet), [T1 (0.2 g Endorphin), T2 (0.2 g Dopamine) and T3 (0.2 g Serotonin)/ L drinking water], [T4 (0.5 kg Hero flower) and T5 (0.5 kg Chamomile flower)/ 100 kg diet], T6 (0.2 g Endorphin + 0.5 kg Hero flower), T7 (0.2 g Dopamine + 0.5 kg Hero flower), T8 (0.2 g Serotonin+ 0.5 kg Hero flower), T9 (0.2 g Endorphin +0.5 kg Chamomile flower), T10 (0.2 g/ Dopamine + 0.5 kg Chamomile flower), T11: 0.2 g/ L Serotonin/ L drinking water + 0.5 kg Chamomile flower/ 100 kg diet.. The drinkers and a feeder were hanged in the center of pens, feed and water were *ad libitum* submitted. The experimental rations were produced in Evan company, contains starter (1- 10), grower (11-25), finisher (26-42) days: 3097, 2990, 2906 kcal/kg metabolizable energy and 22.97, 21.50, 20.78 % crude protein respectively. Chicks received 24 h light during 1st wk then gradually decreased to 20 L:4 D schedule. Also, birds were vaccinated against Gamboro at 7 and 14 days and Newcastle at 1, 14 and 28 days of rearing. The chemical analysis of total (antioxidant, phenolic and flavonoid) compounds in Alecea kurdica and chamomile were determined by High performance liquid (HPLC) chromatography as in Table 1.

Table 1. Some chemical analysis of Alecea Kurdica and Chamomile

	Alecea Kurdica (Gul-e-Hero)	**Chamomile**
Antioxidant (mg / 100 g)	116.05	114.30
Total phenolic (mg /100 g)	171.24	189.90
Flavonoids (µg CE/g)	1140.5	1613.8

Body performances: Initial body weight, body weight, weight gain, feed intake, Feed Conversion Ratio (FCR), mortality and production index (PI) were measured weekly until 42 days according to Olarotimi and Adu (2022).

WBC profile: Monocytes, Lymphocytes and Heterophil (%), and H/L ratio were determined according Shen and Patterson's method (1983) in whole fresh blood with EDTA at 42 days.

Immune organs and antibodies titer: Relative weights of spleen, bursa of fabricius and thymus glands were measured at (42 days) After slaughtering the birds according to Savón et. al., (2022). Blood collected in tubes from the jugular vein during the slaughter of birds to obtain serum by centrifuge to measure antibodies titer against Newcastle (ND) and Infectious Bronchitis (IB) diseases by ELISA.

Statistical analysis

All data were analyzed according to CRD (Complete Randomize Design) by SAS institute program (SAS, 2014), as per variance, significant differences among treatment means were determined by Duncan's multiple range tests at level 1 and 5%.

RESULTS AND DISCUSSION

The results in table (2) shown the addition of some neurotransmitters (dopamine, endorphin and serotonin) in drinking water and some native flowers additives (*Alecea Kurdica* – Gule Hero and chamomile) flowers and their mixture had significantly effects on body performance of broiler ($P \leq 0.01$) highest body weight, body weight gain and production index (PI) recorded, also improved in feed conversion ratio (FCR) in the water additive of T3 (serotonin), T8 (mixture of Serotonin and Hero flower), T10 (mixture of Dopamine and Chamomile flower) and T11 (mixture of Serotonin and Chamomile flower) treatments compared with control treatment (T0). While, the results of mortality percentages recorded significantly ($P \leq 0.01$) decrease in the all treatments of additives, especially inT1, T3, T4, T6 and T11. However, didn't seen any significant differences among the additive treatments and control in feed intake.

Table 2. The impact of some neurotransmitters in broiler drinking water and dietary some native flowers additives on body performance at 42 days.

Treat Ments	BW (g)	BWG (g)	FI (g)	FCR	Mortality (%)	PI (%)
T0	2900 ^{c}	2858.2 ^{c}	5101.9 ^{a}	1.785 ^{a}	5.8 ^{a}	364.4 ^{c}
T1	3150 ^{b}	3108.2 ^{b}	5386.5 ^{a}	1.715 ^{b}	1.5 ^{c}	430.8 ^{b}
T2	3225 ^{ab}	3183.2 ^{ab}	5516.5 ^{a}	1.733 ^{b}	2.5 ^{bc}	432.1 ^{b}
T3	3319 ^{ab}	3277.2 ^{ab}	5030.5 ^{a}	1.535 ^{c}	1.5 ^{c}	507.1 ^{a}
T4	3075 ^{b}	3033.2 ^{b}	5165.5 ^{a}	1.703 ^{b}	1.5 ^{c}	423.5 ^{b}
T5	3325 ^{ab}	3323.2 ^{ab}	5483.3 ^{a}	1.650 ^{cb}	3.0 ^{bc}	465.4 ^{ab}
T6	3205 ^{ab}	3163.2 ^{ab}	5279.4 ^{a}	1.669 ^{cb}	2.0 ^{c}	448.1 ^{b}
T7	3275 ^{ab}	3233.2 ^{ab}	5334.8 ^{a}	1.650 ^{cb}	3.9 ^{b}	454.2 ^{b}
T8	3375 ^{ab}	3333.2 ^{ab}	5359.8 ^{a}	1.608 ^{cb}	3.0 ^{bc}	484.7 ^{ab}
T9	3229 ^{ab}	3187.2 ^{ab}	5054.9 ^{a}	1.586 ^{c}	2.5 ^{bc}	472.6 ^{ab}
T10	3293 ^{ab}	3251.2 ^{ab}	5127.1 ^{a}	1.577 ^{c}	4.5 ^{b}	474.8 ^{ab}
T11	3405 ^{a}	3363.2 ^{a}	5297.0 ^{a}	1.575 ^{c}	2.0 ^{c}	504.4 ^{a}
MSE	219	186	291	0.143	0.33	59.81
S.L	**	**	N.S	**	**	*

T0= control (standard diet), **T1**= 0.2 g Endorphin / L drinking water, **T2**= 0.2 g Dopamine / L drinking water, **T3**= 0.2 g Serotonin/ L drinking water, **T4**= 0.5 kg Hero flower/ 100 kg diet, **T5**= 0.5 kg Chamomile flower/ 100 kg diet, **T6** = 0.2 g Endorphin / L drinking water + 0.5 kg Hero flower, **T7**= 0.2 g/ L Dopamine + 0.5 kg Hero flower, **T8** = 0.2 g/ L Serotonin+ 0.5 kg Hero flower, **T9**= 0.2 g Endorphin/ L drinking water +0.5 kg Chamomile flower/ 100 kg diet, **T10**= 0.2 g/ L Dopamine/ L drinking water + 0.5 kg Chamomile flower/ 100 kg diet, **T11**= 0.2 g/ L Serotonin/ L drinking water + 0.5 kg Chamomile flower/ 100 kg diet. BW: body weight, BWG: body weight gain, FI: feed intake

$^{a, b}$ Mean values within the same row were significantly different (P≤0.01) & (P≤0.05)

Table (3) refers to the effect of adding dopamine, endorphin, serotonin, Hero flowers and chamomile and their mixture on broiler WBC profile and H/L ratio, the addition of neurotransmitters in drinking water and native flowers had in significant differences among all treatments of the study, while shows significant increases (P≤0.01) in lymphocytes percentage especially in the treatments (T8, T9, T10, T11) compared with control T0. Otherwise, the results show significantly (P≤0.01) decreases in heterophils percentage and H/L ratio mean less stress above the birds in the all treatments of the additive groups especially in the treatments (T3) of heterophil and T3 and T10) of H/L ratio compared with the control T0. Also, the addition of neurotransmitters and native flowers additives and their mixture as immune modulator led to significant increase (P≤0.01) in relative weights of spleen, Bursa of fabricius and Thymus in all additive's treatments, especially in T3, T7, T10 and T11 treatments compared to the control treatment T0 (Table 4).

Table 3. The impact of some neurotransmitters in broiler drinking water and some native flowers additives as immune modulator on WBC

Treat Ments	White blood cells percentages (%)			H/ L ratio
	Monocytes	Heterophils (H)	Lymphocytes (L)	
T0	2.03 [a]	26.90 [a]	52.43 [c]	0.513 [a]
T1	1.92 [a]	18.03 [b]	59.10 [b]	0.305 [b]
T2	1.93 [a]	18.35 [b]	60.75 [b]	0.302 [b]
T3	1.91[a]	14.52 [d]	64.25 [ab]	0.226 [c]
T4	1.79 [a]	17.59 [b]	60.87 [b]	0.289 [b]
T5	1.97 [a]	19.33 [b]	63.79 [ab]	0.303 [b]
T6	1.92 [a]	15.29 [cd]	63.43 [ab]	0.241[bc]
T7	1.80 [a]	17.79 [bc]	64.21[ab]	0.277 [b]
T8	1.93 [a]	16.88 [bc]	66.19 [a]	0.255 [bc]
T9	1.81 [a]	16.06 [c]	65.27 [a]	0.246 [bc]
T10	1.83 [a]	15.61[cd]	66.43 [a]	0.235 [c]
T11	1.89 [a]	17.33[bc]	64.91[a]	0.267 [bc]
MSE	0.115	2.07	3.18	0.0139
S.L	N.S	**	**	**

T0= control (standard diet), **T1**= 0.2 g Endorphin / L drinking water, **T2**= 0.2 g Dopamine / L drinking water, **T3**= 0.2 g Serotonin/ L drinking water, **T4**= 0.5 kg Hero flower/ 100 kg diet, **T5**= 0.5 kg Chamomile flower/ 100 kg diet, **T6** = 0.2 g Endorphin / L drinking water + 0.5 kg Hero flower, **T7**= 0.2 g/ L Dopamine + 0.5 kg Hero flower, **T8** = 0.2 g/ L Serotonin+ 0.5 kg Hero flower, **T9**= 0.2 g Endorphin/ L drinking water +0.5 kg Chamomile flower/ 100 kg diet, **T10**= 0.2 g/ L Dopamine/ L drinking water + 0.5 kg Chamomile flower/ 100 kg diet, **T11**= 0.2 g/ L Serotonin/ L drinking water + 0.5 kg Chamomile flower/ 100 kg diet.

[a, b] Mean values within the same row were significantly different (P≤0.01) & (P≤0.05)

Table 4. The impact of some neurotransmitters in broiler drinking water and some native flowers additives as immune modulator on immune organs relative weight

Treatments	Immune organs relative weights (%)		
	Spleen	Bursa of fabricius	Thymus
T0	0.0413 [b]	0.0426 [c]	0.0555 [c]
T1	0.0519 [a]	0.0550 [b]	0.0818 [b]
T2	0.0541[a]	0.0524 [b]	0.0808 [b]
T3	0.0569 [a]	0.0655 [a]	0.0908 [a]
T4	0.0518 [a]	0.0575 [ab]	0.0879 [ab]
T5	0.0513 [a]	0.0558 [b]	0.0868 [ab]
T6	0.0556 [a]	0.0594 [ab]	0.0869 [ab]
T7	0.0523 [a]	0.0603 [a]	0.0894 [ab]
T8	0.0528 [a]	0.0516 [b]	0.0981 [a]
T9	0.0529 [a]	0.0598 [ab]	0.0913 [a]
T10	0.0526 [a]	0.0636 [a]	0.0871 [ab]
T11	0.0598 [a]	0.0603 [a]	0.0966 [a]
MSE	0.00317	0.00349	0.00422
S.L	*	**	**

T0= control (standard diet), **T1**= 0.2 g Endorphin / L drinking water, **T2**= 0.2 g Dopamine / L drinking water, **T3**= 0.2 g Serotonin/ L drinking water, **T4**= 0.5 kg Hero flower/ 100 kg diet, **T5**= 0.5 kg Chamomile flower/ 100 kg diet, **T6** = 0.2 g Endorphin / L drinking water + 0.5 kg Hero flower, **T7**= 0.2 g/ L Dopamine + 0.5 kg Hero flower, **T8** = 0.2 g/ L Serotonin+ 0.5 kg Hero flower, **T9**= 0.2 g Endorphin/ L drinking water +0.5 kg Chamomile flower/ 100 kg diet, **T10**= 0.2 g/ L Dopamine/ L drinking water + 0.5 kg Chamomile flower/ 100 kg diet, **T11**= 0.2 g/ L Serotonin/ L drinking water + 0.5 kg Chamomile flower/ 100 kg diet. [a, b] Mean values within the same row were significantly different (P≤0.01) & (P≤0.05)

Results in Figures (1 and 2). refers to the effect of adding neurotransmitters (dopamine, endorphin and serotonin) in drinking water and some native flowers (Hero, chamomile and their mixture) on broiler serum antibodies titer against Newcastle (ND) and Infectious Bronchitis (IB) diseases by ELISA had significantly (P≤0.01) improvement in the additive's treatments compared with the control T0, especially in T5 and T11 of ND, also T9 and T11 of IB achieved superiorly in immune titer.

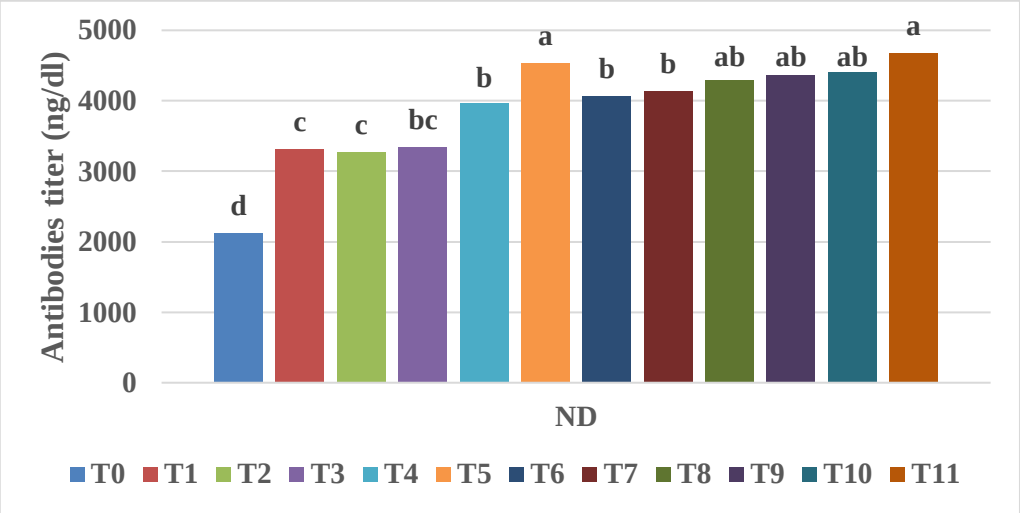

Figure 1. The impact of some neurotransmitters in broiler drinking water and some native flowers additives as immune modulator on antibiotic titer against Newcastle (ND).

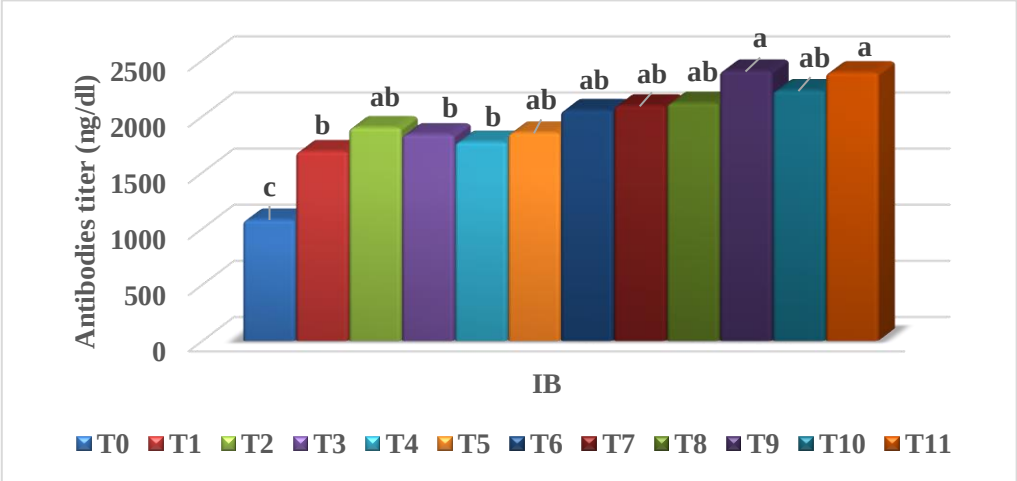

Figure 2. The impact of some neurotransmitters in broiler drinking water and some native flowers additives as immune modulator on antibiotic titer against Infectious Bronchitis (IB) diseases by ELISA.

T0= control (standard diet), **T1**= 0.2 g Endorphin / L drinking water, **T2**= 0.2 g Dopamine / L drinking water, **T3**= 0.2 g Serotonin/ L drinking water, **T4**= 0.5 kg Hero flower/ 100 kg diet, **T5**= 0.5 kg Chamomile flower/ 100 kg diet, **T6** = 0.2 g Endorphin / L drinking water + 0.5 kg Hero flower, **T7**= 0.2 g/ L Dopamine + 0.5 kg Hero flower, **T8** = 0.2 g/ L Serotonin+ 0.5 kg Hero flower, **T9**= 0.2 g Endorphin/ L drinking water +0.5 kg Chamomile flower/ 100 kg diet, **T10**= 0.2 g/ L Dopamine/ L drinking water + 0.5 kg Chamomile flower/ 100 kg diet, **T11**= 0.2 g/ L Serotonin/ L drinking water + 0.5 kg Chamomile flower/ 100 kg diet.

[a, b] Mean values within the same row were significantly different (P≤0.01) & (P≤0.05)

Our results agreement with McCormack and Denbow (1989) revealed that the ICV injection of neurotransmitters led to increase feed intake, drinking, and body temperature in layers chickens and with Hussein *et al.,* (2021) discovered that chamomile led to increase broiler appetites and stimulate production performance, also Jabbar (2022) found that hero flowers improve body weight, and immune status in poultry. The antioxidant capability, total phenol, total flavonoid, contents in the flowers of *Alecea Kurdica* and chamomile (Table 1) had effectiveness role in improving body growth and immune statues represented by immune organs and antibodies titer against ND and IB diseases, The increasing of antioxidant capability due decreases of free radicals and reactive oxygen species (ROS) in biology is producing a medical revolution that promises a new age of health and disease management (Aruoma, 2003). It is ironic that oxygen, an element indispensable for life (Mohammed and Ibrahim, 2004), under certain situations has deleterious effects on the human body (Bagchi and Puri, 1998). Most of the potentially harmful effects of oxygen are due to the formation and activity of a number of chemical compounds, known as ROS, which have a tendency to donate oxygen to other substances. Free radicals and antioxidants have become commonly used terms in modern discussions of disease mechanisms (Aruoma, 1994). The natural plants additives are nontoxic natural compounds with antioxidative activity has been intensified in recent years. In addition to endogenous antioxidant defense systems, consumption of dietary and plant-derived antioxidants appears to be a suitable alternative. Dietary and other components of plants form a major source of antioxidants (Lobo et al., 2010). Therefore, as it is briefly explained, neurotransmitters and herbs plants as a nutrition additives plays an important role in the broiler production and led to improve the secretions of saliva and bile from the gastrointestinal tract, mucous, liver and pancreatic secretions, because of their content of the effective compounds and these compounds have affected on the broiler metabolic representation, which leads to the improvement of most digestive enzymes secretions such as amylase and trypsin and this benefit to maximize of the nutrients available in the bird feed by increasing the permeability of the mucosa in the intestine and thus raise the absorption of nutrients , which works to remove heat stress and improve the health status of the broiler also effect on the proliferation of satellite cells (muscle cells) that is considered the precursor of myogenic cells, which affects increasing of growth and development of skeletal musculature (Hussein *et al.,* 2022; Beghoul *et al.,* 2022), also, the Dopamine, serotonin and Endorphin led to improvement of eating behavioural and appetite thus they were increased in body weight and body weight gain compared with control treatment, also the additives led to improve in H/L ratio and relative weight of Immune organs increase the immunity of cells and increases the fighting ability of macrophage cells and enhances the activity of T cells, which is responsible for immunity (Mohammedi *et al.,* 2016).

CONCLUSIONS

Natural antioxidants are recently reported to be useful to health. Antioxidants prevent free radical induced tissue damage by preventing the formation of radicals, scavenging them, or by promoting their decomposition, so the addition of neurotransmitters (dopamine, endorphin and serotonin) in drinking water and native flowers additives (Hero and chamomile flowers) and their mixture had significantly improve on body weight, body weight gain, FRC, immune organs relative weights and broiler immune at the end of study represented by antibodies titer against New Castle (ND) and Infectious Bronchitis (IB), decreased the mortality percentage, which finally led to more economic profit of the present stock.

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