

## The impact of some feed additives on behavior, welfare and performance of heat-stressed pigeon squabs.

Ramadan D. El Shoukary<sup>1</sup> and Mohamed A. Mousa<sup>2</sup>

<sup>1</sup>Department Animal Hygiene, Faculty of Veterinary medicine, Assuit University (New Valley branch) 71515, Egypt

<sup>2</sup>Department of nutrition and clinical nutrition, faculty of vet. Med., Sohag University, Egypt

Email:- dr\_m\_mousa@yahoo.com

### Abstract

This study aimed to assessment the impact of supplementing pigeon squab's diets with different feed additives on behavior, performance and some blood parameters during heat stress. One hundred and forty four, 4 wk-old age, unsexed pigeon squabs were used and divided in two groups. Thermo natural group squabs (TG) reared in 22 °C, were fed basal diet, while heat-stress group reared in 34 °C, and subdivided into 5 groups; heat-stress group was allotted to basal diet (HG) and four groups allotted to basal diet supplemented with different feed additives as natural feed additives (Moringaolifera (H1G) or fenugreek seeds (H2G)) and chemical feed additives (premix (H3G) or yeast(H4G)). Results showed that, Fenugreek seeds group (H2G) had more potent effect to alleviate the negative effects of heat stress on squab behaviors, performance, biochemical parameters and growth hormone, whereas the Moringaolifera, yeast and premix groups also have beneficial impact in alleviation the negative effect of heat stress, but less than Fenugreek seeds. In conclusion we could recommended the usage of 2 % Fenugreek seeds as feed additives to improve pigeon squab behavior, performance, welfare and biochemical parameters during heat stress condition.

**Key words:-** Fenugreek seeds; MoringaOlifera; Premix and Yeast, pigeon squab, feed additives, performance, behavior.

### Introduction

Pigeon squab's exposure to high ambient temperature induce an oxidative stress condition which reflected by increase of protein oxidation (increased protein carbonyls) and lipid peroxidation (increased MDA). And, increases in leptin blood metabolite such as (glucose; cholesterol and triglycerides), and increase H\|R ratio. (Al-Azraqi, 2008)

During heat stress bird reduced the heat production by decreasing locomotors activities (Pereira et al., 2007); and reducing the release of catabolic hormones ( $T_3$  and  $T_4$ ) (Kusnadi and Rahim, 2009) reducing of feed intake (Bartlett and Smith, 2003) resulting in decreasing weight gain (Soleimani, 2008). On the other hand, The bird increase the heat loses by increasing the respiration rate (Nadia 2003) and spend more time in drinking (Mack et al., 2013). Also, yawning behavior may be involved in thermoregulation during heat stress condition and act as a brain cooling mechanism (Gallup and Gallup 2008).

MoringaOlifera supplementation significantly improve feed intake and average body weight gain (Melesse et al., 2011) and increased total antioxidant capacity (KoutElkloub et al., 2015).

While, Fenugreek seeds having antimicrobial and antioxidant properties (Adil et al., 2015), also increase feed intake (Tariq et al., 2014) and improve broiler body weight (Qureshi et al., 2015). The production and reproduction of Baladipigeons, were improved by adding yeast to its commercial diets (Mariey, 2013). Also, premix plays a potent role in body metabolism activity which controls some important production parameters as increase body weight and changes in carcass cuts. (AbouKhashaba and Mariey, 2009)

The aims for this study are assessment the role and effects of some feed additives to compensate the negative effects of heat stress on pigeon squab, monitoring behavior, performance, welfare and some blood parameters.

## Materials and methods

### Squabs and feeding

One hundred and forty four, 4 wk-old age, unsexed pigeon squabs were purchased from a pigeon fancier, and were fed the basal diet according to AbouKhashaba and Mariey (2009) for 28 days (Table I). Rations and water were offered to squab ad-libitum along the study period from 1/6/2017 till 29/6/2017 in Educational Hospital, faculty of Vet.Med., Assiut University, Egypt.

**Table I.** Composition and chemical analyses of the basal diets.

Ingredients	Ingredients (%)	Chemical analyses	
Yellow corn	68.50	Crude protein, %	15.11
Soybean meal, 44 %	22.00	ME, Kcal/kg**	3201.36
Oil	5.20	Crude fiber, %	3.183
Limestone	1.40	Ether Extract, %	2.813
Bon meal	2.30	Calcium, %	1.346
Common salt (NaCl)	0.30	Available phosphorus, AP %	0.403
Vit. & Min. mix.*	0.30	Lysine, %	0.806
Total	100	Methionine, %	0.280
		Methionine + cysteine %	0.533
		C/P ratio	1/212

\*premix provide vitamins and minerals according the recommendation of AbouKhashaba and Mariey (2009).

\*\* Calculated according to NRC (1994).

### Experimental Design

#### A) Group 1 birds (Thermoneutral group) (TG)

Consist of 24 squabs (6 squab X 4 replicate) were housed in temperature-controlled units at 22<sup>o</sup>: 24<sup>o</sup> C this done by using air cooler and fed basal diet.

## B) Group 2 birds (Heat-stress groups)

Consist totally of 120 squabs divided into 5 groups each one (6 squabs X 4 replicate), that were kept in surrounding environmental temperature at 34<sup>o</sup>:36<sup>o</sup>C (summer season in Egypt) and fed basal diet only (heat-stress group) or fed basal diet containing natural feed additives groups (2% Moringaolifera leaves or 2% fenugreek seeds) or basal diet containing Chemical feed additives (0.4% premix or 0.2% yeast.)

### Behavioral parameters

Behavioral parameter were observed after introduction of bird to the experimental unit by 2 days (i.e. from 30 to 56 days old) the behaviors of the experimented bird were recorded by using video tap. The observation of pigeon squabs behavior was carried out via scanning technique (Fraser and Broom 1990). Observation time was ten minutes for three times at morning (8: 9 am); then (12:1 pm) and late afternoon (4 : 5pm) per day for 3 days \ week. So each replicate was observed 30 minutes daily. According to behavior ethogram. Table (III)

Behavior	Definition
<b>1) Ingestive behavior:-</b> a) Feeding b) Drinking <b>2) Comfort behavior:-</b> a) Foot pecking b) Wings and legs stretch c) Preening <b>3) Movement activity</b> a) standing b) Sitting  c) Walk <b>4) Panting behavior</b>  <b>5) Yawing behavior</b>  <b>6) Resting behavior</b> a) Crouching  b) Huddling	<b>Spudeit et al., 2013)</b> Squab's head observed inside the feeder. squab's head in contact with water <b>( Spiteri, 1975)</b> The pigeon pecks at its foot or the ground. Upward and downward Stretching of leg or wing.  Gentle pecking or scratching its own feathers. Both feet are in contact with the floor; no other body part is in contact ed. Most of the ventral region of the bird's body in contact with floor. No space is visible between the floor and the bird Bird is in the process of taking at least 2 steps, including scratching the litter.  Rapid movement of body wall or gasping during respiration.  a large gaping of the mouth accompanied by a deep inhalation and a shorter expiration of air  lying or sitting with breast on the floor, locking around with or without closed eyes with no other behavior  Three or more birds over lapped in a crouching position in an allelomimetic pattern.

## Squab performance

Live body weight (LBW), was measured in early morning (7 a.m) before feeding and drinking water. It is recorded weekly.

Body weight gain (BWG); calculated value of subtracting the average body weight at the beginning of study period from the body weight at end of the study period. In our work it is recorded weekly.

Feed intake (FI); The amount of diet that was eaten by each bird, at certain period. It is recorded weekly. In our work the feed intake per each group was estimated then divided to the total number of squabs to get average FI.

Feed conversion ratio (FCR); was measured according this equation, = feed intake / body weight gain. It was calculated weekly.

### Total aerobic microbial counts

The fecal contents of duodenum, jejunum, ileum and caecum were collected immediately after slaughter in screw capped sterile plastic cups, previously weighed, and the total aerobic count was done according to method of **Awan and Rahman, (2005)**.

## Squab welfare

At the end of our study 9 squabs from each group were randomly selected and transported to a test arena to do the following four conventional welfare indicators.

### Welfare indicators

#### a) Tonic Immobility (TI) test (Benoff and Siegel 1976)

- 1) Tonic immobility was induced by laying the bird down on its right side and gently restraining it by hand for 15 s. (Time of induction) When the bird straightened up in less than 10 s, it was restrained repeatedly. (Number of induction or attempts) when TI was not induced after three attempts, the duration of TI was considered 0 s.
- 2) Then the hand was withdrawn and the experimenter retreated approximately 1 meter and remained noiseless within the sight of the bird.
- 3) Duration of tonic immobility in second was measured from withdrawal of the hand until the bird straightened up. When the bird did not straighten up within 10 min, it was removed and given the maximal duration of 600 s.

Each bird was caught and sampled, one immediately after another. It was assumed that the catching and returning of birds did not disturb the other members of the flock.

#### b) Dermatitis according to Mayne (2005).

Footpad and hock dermatitis and breast blisters were scored on the birds selected. On a four-point scale (score 0 = no lesion; score 1 = pinhead, superficial lesions; score 2 = large or deep lesions, score 3 = ulcers, or scabs.) the scores of the left and right footpad were averaged, as were those of the hocks.

Data are collected as an aggregated index as summed all data and divided by four.

### Blood parameters

At the end of experiment at 56 day old age blood samples were collected from birds for hematology. The rest of blood was centrifuged at 3000 g for 10 min and plasma was separated and stored at -20° C for further analysis.

#### 1) Chemical blood analysis

Spectrophotometrically:- as Catalase activity; Total antioxidant capacity. (TAC); Glucose ; cholesterol; triglyceride level and Malondialdehyde (MDA which Estimation of Lipid peroxidation). While, enzyme immunoassay kit was used to estimate level of Protein carbonyls. (estimation of Protein oxidation), according to (AOAC, 2002)

#### 2) Hematological blood analysis:

The differential count of heterophils and lymphocytes percentage was carried out by light microscope using an immersion objective for blood smear stained by Gimsa stain.

### Statistical analysis

The study designed as incomplete randomized block design. One way ANOVA procedure was used for analyzed our data, SPSS Program 13.00 Software (SPSS Inc., Chicago, IL, USA). Means were compared by Duncan's test when a significant difference ( $P < 0.05$ ) was detected.

### Results

**Table (2) Effect of feed additives on some behavioral patterns of heat stressed squabs.**

Group    Items	22:24 °c	Heat stress				
	Basal diet					
	No additives		Natural antioxidant		Chemical antioxidant	
	Control (-)	Heat stress	Moringaolife ra	Fenugreeks seed	premix	Yeast
1) Ingestive behavior (act\30mint)						
Feeding behavior	4.7 ± 0.3 <sup>a</sup>	2.9 ± 0.1 <sup>b</sup>	3.9 ± 0.6 <sup>a</sup>	3.8 ± 0.1 <sup>a</sup>	3.1± 0.4 <sup>a,b</sup>	3.2 ± 0.8 <sup>a,b</sup>
Drinking behavior	3 ± 0.2 <sup>b</sup>	5 ± 0.4 <sup>a</sup>	2.4 ± 0.5 <sup>b</sup>	1.9 ± 0.7 <sup>b</sup>	2 ± 0.5 <sup>b</sup>	2.8 ± 0.5 <sup>b</sup>
F / D ratio	1.6 ± 0.1 <sup>a</sup>	0.6 ± 0.2 <sup>b</sup>	1.6 ± 0.5 <sup>a</sup>	2 ± 0.4 <sup>a</sup>	1.6 ± 0.3 <sup>a</sup>	1.1 ± 0.6 <sup>a,b</sup>
2) Movement activity(act\30mint)						
Walk	7 ± 0.4 <sup>a</sup>	4.1 ± 0.4 <sup>b</sup>	5.8 ± 1.3 <sup>a</sup>	5.2 ± 1.4 <sup>a,b</sup>	4.3 ± 0.6 <sup>b</sup>	6.9 ± 1.2 <sup>a</sup>
Standing	3.1 ± 0.3 <sup>a</sup>	1.2 ± 0.6 <sup>b,c</sup>	2 ± 0.1 <sup>b</sup>	1.7 ± 0.1 <sup>b</sup>	1.6 ± 0.9 <sup>b</sup>	1.9 ± 0.3 <sup>b</sup>
3) Resting behavior(act\30mint)						
Crouching	0.8 ±0.6 <sup>c</sup>	5 ± 0.1 <sup>a</sup>	2 ± 0.1 <sup>b</sup>	4.7 ± 0.1 <sup>a</sup>	4.2 ± 0.9 <sup>a</sup>	4.8 ± 0.3 <sup>a</sup>
Huddling	0.4 ±	0.2 ± 0.1	0.3 ± 0.1	0.2 ± 0.1	0.3 ± 0.1	0.3 ± 0.1

	0.1					
<b>4) Panting and Yawing behavior( act\30mint)</b>						
Panting	1.3 ±0.6 <sub>c</sub>	4 ± 0.1 <sup>a</sup>	2.8 ± 0.1 <sup>b</sup>	3.7 ± 0.1 <sup>a</sup>	3.2 ± 0.9 <sup>a</sup>	3.8 ± 0.3 <sup>a</sup>
Yawing	0.5±0.1 <sub>c</sub>	1.5 ± 0.1 <sup>a</sup>	0.6 ± 0.1 <sup>c</sup>	0.7 ± 0.1 <sup>b,c</sup>	0.9±0.9 <sup>b,c</sup>	0.8 ± 0.3 <sup>b,c</sup>
<b>5) Comfort behavior (act\30mint)</b>						
Preening	9 ± 0.9 <sub>b</sub>	6.1 ± 0.6 <sub>c</sub>	8.7 ± 1.3 <sup>b</sup>	8.6 ± 0.8 <sup>a</sup>	6.7 ± 0.4 <sub>b,c</sub>	7.8 ± 1.6 <sub>a,b</sub>
Stretching(wing&leg)	3.3 ± 0.5 <sup>a</sup>	1.8 ± 0.2 <sub>b</sub>	2.8 ± 0.9 <sup>a</sup>	2 ± 0.4 <sup>a</sup>	2.1 ± 0.5 <sup>a,b</sup>	3 ± 0.3 <sup>a</sup>
Head scratching	2.9 ± 0.9 <sup>a</sup>	0.9 ± 1.6 <sup>b</sup>	2.8 ± 1.1 <sup>a</sup>	2 ± 0.3 <sup>a</sup>	1.7 ± 0.6 <sup>a,b</sup>	2.1 ± 1.6 <sup>a</sup>
<sup>a,b,c</sup> Values with different superscripts in the same raw differ significantly (p < 0.05). Data are represented as means of behavior frequency for 30 mints per day for 12 days.						

**Table (3) Effect of feed additives on body performance and total aerobic count of heat stressed squabs.**

Group    Items	22:24 °c	Heat stress				
	Basal diet					
	No additives		Natural antioxidant		Chemical antioxidant	
	Control (-)	Heat stress	Moringaolifera	Fenugreeks seed	premix	Yeast
1) Performance parameter of single squab						
Feed intake (gm)	590 ± 12 <sup>a</sup>	540 ± 8.3 <sup>b</sup>	560 ± 13.6 <sup>a</sup>	575 ± 9.1 <sup>a</sup>	545 ± 11 <sup>a,b</sup>	550 ± 0.6 <sup>a,b</sup>
Body weight (gm)	175.5± 7 <sup>a</sup>	159 ± 6.4 <sup>b</sup>	166.3 ± 5.5 <sup>a</sup>	170 ± 9.5 <sup>a</sup>	162 ± 6 <sup>a,b</sup>	168 ± 3.9 <sup>a,b</sup>
Body gain (gm)	7.5± 0.3 <sup>a</sup>	6.4 ± 0.5 <sup>b</sup>	6.8 ± 0.7 <sup>b</sup>	7.2 ± 0.4 <sup>a</sup>	6.8 ± 0.3 <sup>b</sup>	7 ± 0.5 <sup>a,b</sup>
FCR	78.6±3.2 <sup>b</sup>	84.4 ± 8.5 <sup>a</sup>	82.4 ± 9.2 <sup>a</sup>	79.9 ± 2.5 <sup>b</sup>	80.2± 5.9 <sup>a,b</sup>	78.6 ±7.8 <sup>b</sup>
2) Total aerobic count ( cfu\ gram)						
Small intestine	5 ± 1.15 <sup>a</sup>	6 ± 1.15 <sup>a</sup>	2 ± 0.58 <sup>b,c</sup>	0.002 ± 0.001 <sup>c</sup>	5 ± 1.18 <sup>a,b</sup>	3 ± 1.73 <sup>b;c</sup>
Large intestine	4 ±0.58 <sup>a, b</sup>	4 ± 0.5 <sup>a,b</sup>	4 ±1.73 <sup>a, b</sup>	2 ± 1.15 <sup>a, b, c</sup>	6 ±1.73 <sup>a</sup>	2±0.58 <sup>b,c</sup>

<sup>a,b,c</sup>values with different superscripts in the same raw differ significantly at (p < 0.05).

Performance parameters of single squab data are represented as means of 4 weeks (28 days).

Total aerobic count data was multiplied by 10<sup>8</sup>cfu. (colony-forming units per gram).

**Table (4) Effect of feed additives on welfare parameters of heat stressed squabs.**

Group	22:24 °C	Heat stress		
	Basal diet			
	No additives		Natural antioxidant	Chemical antioxidant

Items	Control (-)	Heat stress	Moringaolifer a	Fenugreeks seed	premix	Yeast
Tonic Immobility(s)	59 ± 0.66 <sup>c</sup>	85 ± 2.88 <sup>a</sup>	71 ± 0.88 <sup>b</sup>	70 ± 0.56 <sup>b</sup>	77 ± 0.52 <sup>b</sup>	65 ± 0.42 <sup>b,c</sup>
foot pad dermatitis	0.5 ± 0.01 <sup>c</sup>	2 ± 0.02 <sup>a</sup>	1 ± 0.002 <sup>b</sup>	0.67 ± 0.005 <sup>b</sup>	2 ± 0.02 <sup>a</sup>	1.33 ± 0.4 <sup>b</sup>
Hock dermatitis	0.6 ± 0.03 <sup>c</sup>	2 ± 00 <sup>a</sup>	1 ± 0.09 <sup>a,b</sup>	0.67 ± 0.3 <sup>b</sup>	1.67 ± 0.4 <sup>a</sup>	1 ± 0.75 <sup>a,b</sup>
Breast blisters	0.8 ± 0.05 <sup>c</sup>	2 ± 00 <sup>a</sup>	1.33 ± 0.33 <sup>a,b</sup>	1 ± 0.15 <sup>b</sup>	1.33 ± 0.3 <sup>a,b</sup>	1 ± 0.75 <sup>b</sup>
welfare index	15.4 ± 0.01 <sup>c</sup>	22.8 ± 0.05 <sup>a</sup>	18.5 ± 0.08 <sup>b</sup>	18.1 ± 0.75 <sup>b</sup>	20.5 ± 0.57 <sup>a</sup>	17.1 ± 0.57 <sup>b,c</sup>

<sup>a,b,c</sup>Values with different superscripts in the same raw differ significantly at (p < 0.05).

Welfare parameters of squab data are represented as means of 9 squabs per treatment.

**Table (5) Effect of feed additives on blood biochemical parameters of heat-stressed squabs.**

Group   Items	22:24 °c	Heat stress				
	Basal diet					
	No additives		Natural antioxidant		Chemical antioxidant	
	Control (- )	Heat stress	Moringaolifer a	Fenugreeks seed	premix	Yeast
1) Blood oxidative markers						
Malondialdehyde (μmol/L)	0.8 ± 0.01 <sup>c</sup>	2.7± 0.6 <sup>a</sup>	2 ± 0.9 <sup>b,c</sup>	1.9± 0.8 <sup>c</sup>	2.3± 0.05 <sup>a,b</sup>	2.2 ± 0.03 <sup>a,b</sup>
Protein carbonyls (nmol\L)	0.8 ± 0.03 <sup>c</sup>	1.8± 0.9 <sup>a</sup>	1.3± 0.7 <sup>b</sup>	1± 0.9 <sup>b,c</sup>	1.6 ± 0.08 <sup>a</sup>	1.6 ± 0.04 <sup>a</sup>
Total antioxidant capacity	0.29±0.01 <sup>a</sup>	0.06±0.00 <sup>2<sup>c</sup></sup>	0.19± 0.001 <sup>a</sup>	0.21 ± 0.06 <sup>a</sup>	0.12±0.00 <sup>3<sup>b</sup></sup>	0.13± 0.003 <sup>b</sup>
Catalase	1 ± 0.06 <sup>b</sup>	3 ±0.01 <sup>a</sup>	1.5 ± 0.01 <sup>b</sup>	1.33 ± 0.33 <sup>b</sup>	3.1 ± 0.57 <sup>a</sup>	1.66±0.33 <sup>b</sup>
2) Leptin metabolites						
Glucose (mg\dl)	233±13.8 <sup>b</sup>	280 ± 12 <sup>a</sup>	265 ± 12 <sup>a,b</sup>	245 ± 8 <sup>c</sup>	271 ± 7 <sup>a,b</sup>	269± 13 <sup>a,b</sup>
Cholesterol (mg\dl)	176± 9.6 <sup>b</sup>	188 ± 17 <sup>a</sup>	182 ± 11 <sup>a,b</sup>	169 ± 5 <sup>c</sup>	186 ± 8 <sup>a</sup>	181± 12 <sup>a,b</sup>
Triglyceride (mg\dl)	171± 6.7 <sup>c</sup>	189 ± 9 <sup>a</sup>	183 ± 9 <sup>a</sup>	181 ± 5 <sup>b</sup>	185 ± 9 <sup>a,b</sup>	183±16 <sup>a,b</sup>
3) Hormonal level						
T <sub>3</sub> levels	0.14± 0.02 <sup>b</sup>	0.07±0.01 <sup>c</sup>	0.5 ± 0.22 <sup>b</sup>	0.9 ± 0.001 <sup>a</sup>	0.25±0.07 <sup>b,c</sup>	0.14±0.03 <sup>c</sup>
T <sub>4</sub> levels	14 ± 2.3 <sup>a</sup>	10.7 ± 0.7 <sup>c</sup>	11.5 ± 0.28 <sup>b</sup>	13.3 ± 0.33 <sup>a,b</sup>	10.9±2.8 <sup>b,c</sup>	12 ± 0.02 <sup>b</sup>
Growth hormone	0.36 ±0.01 <sup>a</sup>	0.11±0.03 <sup>c</sup>	0.13 ±0.001 <sup>b,c</sup>	0.21 ± 0.02 <sup>a</sup>	0.13± 0.01 <sup>b,c</sup>	0.16± 0.03 <sup>b</sup>
4) Hematological blood parameters						
Heterophils (%)	42 ± 2 <sup>b</sup>	49 ± 3 <sup>a</sup>	47 ± 1 <sup>a</sup>	44 ± 2 <sup>a,b</sup>	46 ± 3 <sup>a</sup>	45 ± 2 <sup>a</sup>
Lymphocytes (%)	58 ± 3 <sup>a</sup>	51 ± 1 <sup>b</sup>	53 ± 2 <sup>a,b</sup>	56±2 <sup>a</sup>	54±1 <sup>a,b</sup>	55±1 <sup>a</sup>
H\L ratio	0.72±0.02	0.96± 0.02 <sup>a</sup>	0.89 ± 0.09 <sup>a</sup>	0.79 ± 0.03 <sup>b</sup>	0.85± 0.02 <sup>a</sup>	0.81± 0.07 <sup>b</sup>



	c					
<sup>a,b,c</sup> Values with different superscripts in the same raw differ significantly at (p < 0.05). Data are represented as means of 6 blood sample for each treatment group.						

### **Implication of feed additives on behavior, performance, welfare and some blood parameters of heat-stressed pigeon squabs**

Data tabulated in Table (2&3&4) showed that, Feed additives used facing the negative impact of heat stress on drinking behavior; yawing and welfare index. While, Pigeon squab fed diet contain Moringaolifera leaves and fenugreek seeds facing the negative impact of heat stress on drinking behavior; feed \ drinking ratio; comfort behavior; feed intake; body weight; Total aerobic count of small intestine; blood oxidative markers; T<sub>3</sub> and T<sub>4</sub> level. Moreover, Using Fenugreek seeds only facing the negative impact of heat stress on body gain; FCR; leptin metabolite; growth hormone and H\|R ratio. And Moringaolifera leaves only facing the negative impact of heat stress on walk activity, crouching and panting.

On the other hand, Premix only facing the negative impact of heat stress on F\|D ratio. While, Yeast only facing the negative impact of heat stress on walk; comfort; total aerobic count; catalase; T<sub>4</sub>; growth hormone and H\|R ratio.

From previous data we concluded that, fenugreek seeds were the more effective than Moringaolifera leaves than yeast than premix in facing the negative impact of heat stress on squabs behavior; performance; welfare and some blood parameters.

### **Discussion**

The positive effect of Moringaolifera and fenugreek seeds on increasing feeding intake and body weight was agreed with previous finding of (Melesse et al., 2011 & Mousa et al., 2017 & Abou-Elezz et al., 2012 & Yattoo et al., 2012 & Kirubakaran et al., 2016). While, disagreed with (Onunkwo and George, 2015 & Juniaretal., 2008, El-Kaiaty et al., 2002 & Metin et al., 2013).

This finding may be related to some reasons such as; Moringaolifera leaves and fenugreek seeds facing the negative impact of heat stress on walk activity, crouching; panting; feed \ drinking ratio and yawing which reflected mainly by bird back to nearly normal behavior in feed intake leading to good body weight; also, Moringaolifera had low anti-nutritional compounds level and high total soluble protein makes it more suitable to feed by poultry (Kakengi et al., 2007); whereas Fenugreek seeds contain galactomannan which consider appetizer and stimulate digestion process (Alloui et al., 2012). Also, it was reported that it improve the feed intake and the body weight and broiler performance due to their content of both essential fatty acids and high quality proteins. (Hind et al., 2013)

The significant increase effect of fenugreek seeds on body gain was in agreement with finding of Magda Elbushra, (2012) Moreover, the significant decrease effect of fenugreek seeds and yeast on feed conversion ratio was previously finding by Tariq et al., (2014) and Abd El-Azeem, (2002) respectively. Also, fenugreek seeds have a significant positive effects on digestibility and absorption of nutrients, that is reflected as improvement in body weight gains. (EL-Mallah et al., 2005) also, its lead to significant increase in T<sub>3</sub> and T<sub>4</sub> and growth hormone level which in agreement with previous finding of (Azouz, 2001). This result may be related to the fenugreek



seeds may stimulate the thyroid gland directly as  $T_3$  and  $T_4$  of serum increased significantly (**Hassan, 2000**).

On the other hand, the positive effect of fenugreek on FCR may be related to the effective content of the essential oil in fenugreek, trigonellin that has antioxidant, antiviral, and antifungal activities (**Mazur et al., 1998**). While yeast cell wall components ( $\beta$ -glucans and  $\alpha$ -mannans) which provide greater villus height and size, also both contents have protect intestinal mucosa through decrease pathogens load or preventing it from attaching to villi this mainly decrease the pathogen load in intestine and guard the health status of mucosa, this mode of action lead to improved FCR. (**Zhang et al., 2005**)

Squabs fed diet containing Moringaolifera leaves or fenugreeks seed or yeast have the lowest number of total aerobic count of small intestine than the heat stress group this finding was agreed with previous finding of **Sahar et al., (2014)**; **Adil et al., (2015)** and **Rima et al., (2012)** respectively.

This may be due its antibacterial and antifungal activities and hepatoprotective effect of Moringa (**Bennett et al., 2003**) or it may be due to, fenugreek seeds having many therapeutic effects like antibacterial, antimicrobial (**Adil et al., 2015**) and it contains pterigospermin that has anti-bacterial and anti-fungal activities. While, yeasts mode of action is mainly depends on antagonism to microorganisms, through the nutrients competition; unsuitable medium for microorganisms growth via decrease pH; ethanol production; and secretion of mycocin that has antimicrobial activity (**Rima et al., 2012**).

A variety of indicators, including health, productivity, behavioral and physiological characteristics commonly used to reveal bird welfare. Behaviorally as drinking water, scratching, taking a dust bath and lying down are considered an important indicators of welfare of broiler. (**Pereira et al., 2005**).

All feed additives substance leading to improvement of squab's welfare during heat stressed condition this result may related to different reasons such as

All feed additives leading to decrease duration of tonic immobility as a test of fear (the longer the more fearful more worse of bird welfare (**Jones, 1996**) and increase A latency-to-lie (LTL) test which means decrease Leg weakness and decrease dermatitis which included because these represent 2 major welfare problems for broiler chickens, which can both be painful themselves and can lead to other welfare problems (**Bradshaw et al., 2002**).

Moringaolifera leaves and Fenugreek seeds facing the negative impact of heat stress on increased number of water intake and decreased number of feed intake and locomotors activity which resulted in worsening the welfare of birds (**Bozakova 2008**) so that, an increase in welfare index was seen.

Moringaolifera leaves and Fenugreek seeds facing the negative impact of heat stress on decreasing body weight which in linear relationship with resting and preening behavior (**Hocking et al., 1996**) which resulted in worsening the welfare of birds (**Bozakova 2008**) so that, an increase in welfare index was seen.

Moringaolifera leaves; Fenugreek seeds and yeast facing the negative impact of heat stress on comfort behaviors (A number of activities have been grouped together, these activities include wing / leg stretching, preening; body shaking and head scratching). These activities may serve functions as maintain plumage or stretch muscles in additional to improving physical

comfort (Black and Hughes, 1974) which are important indicators of welfare of broiler breeders. (Pereira et al., 2005)

Moringaolifera leaves facing the negative impact of heat stress on increased catalase and decreasing cholesterol level and total antioxidant capacity.

Antioxidant effect of Moringaoleifera leaf may be due to the presence of polyphenols, tannins, anthocyanin, glycosides and thiocarbamates which remove free radicals, activate antioxidant enzymes and inhibit oxidase. (Luqman et al., 2012).while, cholesterol lowering effect due to its high fiber content (Olugbemi et al., 2010)

Fenugreek seeds facing the negative impact of heat stress on increasing of H\|R ratio and leptin metabolites such as (glucose; cholesterol and triglycerides) which indicated to be a good quantitative measure of stress. (Al-Azraqi, 2008) this result was in agreement of (Abdul-Rahman, 2012)

The significant decreases in glucose and (cholesterol & triglycerides) level in fenugreek seeds were agreed with previous finding of (Adil et al., 2015). It may be due to the direct  $\alpha$ - cell stimulation by amino acid (4-hydroxyisoleucine) present in fenugreek seeds, which increases insulin secretion, thus improves glucose tolerance (Broca et al., 2000). On other side, presence of saponins and resins in fenugreek which inhibit bile acid and cholesterol absorption from intestine, thereby, decreasing cholesterol level in blood (Abaza, 2007).

Fenugreek seeds facing the negative impact of heat stress on increased catalase and decreasing total antioxidant capacity. This result was agreed with (Choudhary et al., 2001) and (Neetu et al., 2014) respectively.

This may be due to the fenugreek seeds are rich sources of lipids, protein, mucilage, calcium, dietary fiber B vitamins, Iron, protease inhibitors and several steroid saponins, tiny amounts of alkaloid, furostanol glycosides; steroidal peptide and flavonoids, especially naringenin. So that, it possess hypocholesterolaemic, hypoglycemic and antioxidative properties. (Ayah and Mahmoud., 2016)

Premix facing the negative impact of heat stress on increased catalase and decreasing total antioxidant capacity.

A significance increase in total antioxidant capacity in premix group was agreed with (Cheng et al., 2004). This may due to external sources of antioxidant nutrients vitamins E and C and the mineral selenium. Organic selenium is a natural seleno-amino acid (seleno methionine) which possesses antioxidant properties and improves resistance against oxidative stress and It is essential for adequate functioning of many enzymes such as, the antioxidant enzyme, glutathione peroxidase, which protects the cell against free radicals. (Schoonheere et al., 2009)

## Acknowledgements

We are grateful to the staff members of our departments for their gaudiness in finishing, writing and publishing of this article.

## References

- Abaza, I.M., 2007. Effect of using Fenugreek, chamomile and radish as feed additives on productive, reproductive and digestibility coefficients of laying hens. *Egyptianpoultryscience* 27, 199-218.
- Abd El-Azeem, F.A.M., 2002. Digeston, Neomycin and yeast supplementation in broiler diets under Egyptian summer conditions. *Egyptian Poultryscience* 22, 235: 257
- Abdul-Rahman S. Y., 2012. Effect of Fenugreek Seeds on Some Physiological Characters in Broiler Breeders Males. *Mesopotamia Journal of Agriculture*. 40, 114-121.
- AbouKhashaba H.A., and Y.A. Mariey., 2009. Nutritional and management studies on the pigeon: Effect of different levels of vitamins and minerals premix on productive and reproductive performance. *Egyptian Poultry Science*. 29, 947-969.
- Abou-Elezz K. F. M., L.S. Franco, R. S. Ricalde and J.F. Sanchez., 2012. The nutritional effect of *Moringaoleifera* fresh leaves as feed supplement on Rhode Island Red hen egg production and quality. *Tropical Animal Health Production* 44, 1035–1040.
- Adil S., S. Qureshi and R.A. Pattoo., 2015. A Review on Positive Effects of Fenugreek as Feed Additive in Poultry Production. *International Journal of Poultry Science* 14, 664-669.
- Al-Azraqi A.A., 2008. Pattern of Leptin Secretion and Oxidative Markers in Heat-Stressed Pigeons. *International Journal of Poultry Science* 7, 1174-1176.
- Alloui, N., Ben, S., Alloui, M.N. and Ibrir, F., 2012. Utilization of fenugreek (*Trigonellafoenum-graecum*) as growth promoter for broiler chickens. *Journal World's Poultry. Reserches*. 2, 25-27.
- Awan, J. A., and S. U. Rahman., 2005. *Microbiology Manual*. Unitech Communications, Faisalabad, Pakistan.
- Ayah R. H. and S. Mahmoud., 2016. A review on phytochemistry and pharmacological effects of *Trigonellafoenum-graecum*. *Advanced Herbal Medicine* 2, 61-67.
- Azouz, H.M.M., 2001. Effect of hot pepper and fenugreek seeds supplementation on broiler diets. Ph.D. Thesis. Faculty of Agric., Cairo university, Egypt.
- Bartlett J. R. and Smith M. O., 2003. Effects of different levels of zinc on the performance and immunocompetence of broilers under heat stress. *Poultry Science*. 82, 1580-1588.
- Bennett RN, Mellon FA, Foidl N et al., 2003. Profiling glucosinolates and phenolics in vegetative and reproductive tissues of the multi-purpose trees *Moringaoleifera* L. (Horseradish tree) and *Moringastenopetala* L. *Journal Agriculture FoodChemimastery* 51, 3546–3553.
- Benoff FH and Siegel PB., 1976. Genetic analyses of tonic immobility in young Japanese quail (*Coturnixcoturnixjaponica*). *Animal Learn Behavior* 4, 160-162.
- Berg, C., and G. S. Sanotra., 2003. Can a modified latency-to-lie test be used to validate gait-scoring results in commercial broilerflocks. *AnimalWelfare*. 12, 655–659.
- Black A.J. and Hughes B., 1974. Patterns of comfort behaviour and activity in domestic fowls: a comparison between cages and pens. *British Veterinary Journal* 130, 23-33. (Abstract) cited by [www.cabdirect.org](http://www.cabdirect.org).

- Bozakova N., 2008. Ethological aspects of chicken's welfare under different environmental conditions during summer time. *Ecology and Future - Bulgarian Journal of Ecological Science* 7, 29-33.
- Bradshaw R H, R D Kirkden and D M. Broom., 2002. A review of the aetiology and pathology of leg weakness in broilers in relation to welfare. *Avian and Poultry Biology Reviews* 13, 45-103.
- Broca C, Manteghetti M and R. Gross, 2000. 4-Hydroxyisoleucine: effects of synthetic and natural analogues on insulin secretion." *European Journal of Pharmacology* 45, 390: 339.
- Cheng, T. Guo, and J., Yuan, 2004. Effects of dietary supplementation of Amino Acids complex Zn, Cu, Mn on laying performance, immunity and antioxidation capacity in tissues. *China Poultry* 26, 18-21.
- Choudhary D., D. Chandra, S. Choudhary and R. K. Kale., 2001. Modulation of glyoxalase, glutathione S-transferase and antioxidant enzymes in the liver, spleen and erythrocytes of mice by dietary administration of fenugreek seeds. *Food and Toxicology* 39, 989-997.
- EL-kaiaty, A.M.; Soliman, A.Z.M.; and M.S., Hassan., 2002. The physiological and immunological effects of some natural feed additives in layer hen diets *Egyptian poultry Science* 22, 175-203.
- El-Mallah, G.M., S.A.M. Ibrahim and M.A.Z. Abdo., 2005. Garlic and fenugreek as feed additives to different levels of protein and energy in diets of growing turkeys. *Egyptian poultry Science* 25, 911-929.
- Fraser, A.F. and R.S. Broom., 1990. *Animal Behavior and welfare*. 3<sup>rd</sup> edition Thomson Litho. Ltd.
- Gallup, A. C. and, G.G. Jr Gallup., 2008. Yawning and thermoregulation. *Physiology and Behavior*. 95, 10-16.
- Hassan, M.S.H., 2000. Physiological studies on egg cholesterol and immunity in layers. Ph.D. thesis, Animal production Dept., Faculty of Agriculture, Cairo University.
- Hind, A.A., A.A. Saadia and M.E. Khalid., 2013. Effect of Different Natural Feed additives Compared to Antibiotic on Performance of Broiler Chicks under High Temperature Environment. *Bulletin of Environment, Pharmacology and Life Sciences* 2, 139-144.
- Hocking P.M.; Hughes S. and R.B. Jones., 1996. Behavioral comparison of layer and broiler fowl: Measuring fear responses. *Applied animal behavior science* 49, 321-333.
- Jones, R.B., 1996. Fear and adaptability in poultry: insights, implications and imperatives. *World's poultry science journal* 52, 131-174.
- Juniar I, Widodo E, and O Sjoefjan., 2008. Effect of Moringa oleifera leaf meal in feed on broiler production performance. *Jurnal Ilmu Peternakan Brawijaya* 18, 238-242.
- Kakengi, A.M.V., J.T. Kaijage, S.V. Sarwatt, S.K. Mutayoba, M.N. Shem and T. Fujiyama., 2007. Effect of Moringa oleifera leaf meal as a substitute for sunflower seed meal on performance of laying hens in Tanzania. *International poultry science journal* 9, 363-367.
- Kirubakaran A, Moorthy M, R, Chitra, and G Prabakar., 2016. Influence of combinations of fenugreek, garlic, and black pepper powder on production traits of the broilers." *Veterinary world*. 9, 470-474.

- KoutElkloub, M. EL. Moustafa, Riry, F.H. Shata, Mousa, M.A.M., Hanan, A.H.Alghonimy and S.F Youssef., 2015. Effect of using Moringaoleifera leaf meal on Performance of japanese quail. *Egyptian Poultry science* 35, 1095-1108.
- Kusnadi, E. and F. Rahim., 2009. Performance and plasma triiodothyronine of broilers as result effects of heat stressed in tropic area. *Media Peternakan* 32, 155-161.
- Luqman S, Srivastava S, Kumar R, Maurya AK, and D.Chanda., 2012. Experimental assessment of Moringaoleifera leaf and fruit for its antistress, antioxidant and scavenging potential using In vitro and In vivo assays. *Evidence-Based Complementary and Alternative Medicine* 2012, 1-12.
- Mack, L.A.; Felver-Gant, J.N.; Dennis, R.L. and H.W.Cheng., 2013. Genetic variation alter production and behavioral responses following heat stress in 2 strains of laying hens. *Poultry Science* 2, 285–294.
- Magda Elbushra., 2012. Effect of Dietary Fenugreek Seeds (*Trigonellafoenum*) as Natural Feed Addition on Broiler Chicks Performance. *Journal of Science and Technology* 13, 22-25.
- Mariey Y.A., 2013. Nutritional and management studies on the pigeon: effect of dietary yeast supplementation on productive and reproductive performance of pigeon. *Egyptian Poultry Science Journal* 33, 349-356.
- Mayne, R.K., 2005. A review of the etiology and possible causative factors of food pad dermatitis in growing turkeys and broilers. *World's Poultry Science Journal*. 61, 256–267.
- Mazur, W.M., Duke, J.A., Wahala, K., Rasku, S. and H., Adlercreutz., 1998. Isoflavonoids and lignins in legumes: Nutritional and health aspects in human. *Journal of Nutrition Biochemistry* 9, 193-200.
- Melesse .A., W. Tiruneh and T. Negesse., 2011. effects of feeding Moringastenopetala leaf meal on nutrient intake and growth performance of rhode island red chicks under tropical climate. *Tropical and Subtropical Agroecosystems* 14, 485- 492.
- Metin D., Z. Erdoğan, A. Duru, A. Küçükgül, V. Düzgüner, D. Alpaslan and A.Şahin., 2013. Effect of Seed Powder of a Herbal Legume Fenugreek (*Trigonellafoenum-graceum* L.) on Growth Performance, Body Components, Digestive Parts, and Blood Parameters of Broiler Chicks. *Pakistan Journal Zoololgy* 45, 1007-1014.
- Mousa, A.M., Osman, AS, Hala A. 2017." Performance, immunology and biochemical parameters of Moringa oleifera and/or Cichorium intybus addition to broiler chicken ration " *Journal of Veterinary Medicine and Animal Health (JVMAH)*, ISSN: 2141-2529 , Volume 9, Issue 10, , 255-263.
- Nadia, M.A., 2003. A study of some physiological, productive and reproductive parameters of Japanese quail under stress condition. PhD.thesis Department of poultry production .Faculty of Agriculture, El- Fayoum.Cairo University.
- National Research Council (NRC.).,1994. Nutrient requirements of poultry 9 Rev. Ed.National Academy th Press, Washington, D.C., USA
- Neetu P., N. Pant, J.P. Singh, and S. Agrawal., 2014.Antioxidant activity of *Trigonellafoenumgraecum*L.using various in vitro models. *International Journal of Herbal Medicine* 2, 53-57.

- Olugbemi, T. S., Mutayoba, S. K. and F. P. Lekule., 2010. Moringa oleifera leaf meal as a hypocholesterolemic agent in laying hen diets. *Livestock Research for Rural Development* 22, 1-6.
- Onunkwo, D.N and O. S. George., 2015. Effects of Moringa Oleifera Leaf Meal on the Growth Performance and Carcass Characteristics of Broiler Birds. *Journal of Agriculture and Veterinary Science* 8, 63-66.
- Pereira DF, Naas A., Salgado DA, and B. C. Gaspar CR and N.LJ. Penha., 2007. Correlations among behavior, Performance and Environment in Broiler Breeders using Multivariate Analysis. *Brazilian Journal of Poultry Science*, 9, 207-213.
- Pereira, D.F.; Nääs, I.A.; Romanini, C.E.B.; Salgado, D.D.; G.O.T Pereira., 2005. Welfare pointers in function of behavior reactions of broiler breeders. *Engenharia Agrícola Jaboticabal* 25, 308-314.
- Qureshi, S., M.T. Bandy, S. Adil, I. Shakeel and Z.H. Munshi., 2015. Effect of dandelion leaves and fenugreek seeds with or without enzyme addition on performance and blood biochemistry of broiler chicken and evaluation of their in vitro antibacterial activity. *The Indian journal of animal sciences* 85, 1248-1254.
- Rima Hatoum, S. Labrie and F. Ismail., 2012. Antimicrobial and probiotic properties of yeasts: from fundamental to novel applications. *Frontiers in Microbiology* Volume 3, 421-428
- Sahar M. Kheir, Kafi S K and E. Haitham., 2014. The antimicrobial activity and phytochemical characteristic of Moringa oleifera seeds, leaves, and flowers. *World Journal of Pharmaceutical Research*. 4, 258-271.
- Schoonheere, O. Dotreppe, J. Pincemail, L. Istasse and J. L. Hornick., 2009. Dietary incorporation of feedstuffs naturally high in organic selenium for racing pigeons (*Columba livia*): effects on plasma antioxidant markers after a standardized simulation of a flying effort. *Journal of Animal Physiology and Animal Nutrition* 93, 325–330.
- Soleimani A.F., Kasim A., A.R. Alimon and I. Zulkifli., 2008. Durability of induced heat tolerance by short term heat challenge at broilers marketing age. *Pakistan journal of biology sciences* 11, 2163-2166.
- Spiteri, N. J., 1975. Social especially agonistic behavior in the pigeon. Durham theses, Durham University." at: <http://etheses.dur.ac.uk/8902/>
- SPSS., 2001. SPSS user's guide statistics Version 10. Copyright SPSS Inc., USA.
- Spudeit W. Anderson, N. S. Sulzbach, M. de A. Bittencourt, A.M. Duarte, H. Liang, C. Oliveira, and J.M.-Neto., 2013. The behavioral satiety sequence in pigeons (*Columba livia*). Description and development of a method for quantitative analysis. *Physiology and Behavior* 122, 62–71.
- Tariq Mamoun, M. A. Mukhtar and M. H. Tabidi., 2014. Effect of fenugreek seed powder on the performance, carcass characteristics and some blood serum attributes. *Advance Research in Agriculture and Veterinary Science* 1, 6-11.
- Yatoo, M.A., R.K. Sharma and N. Khan., 2012. Effect of Fenugreek and Black Cumin seeds as Feed Additives on Blood Biochemical Profile and Performance of Broilers. *Indian Journal of Animal Nutrition* 29, 174- 178.

Zhang, A. W., B. D. Lee, S. K. Lee, K. W. Lee, G. H. An, K. B. Song, and C. H. Lee., 2005. Effects of yeast (*Saccharomyces cerevisiae*) cell components on growth performance, meat quality, and ileal mucosa development of broiler chicks. *Poultry Science* 84, 1015–1021.