International Journal of Agricultural Technology 2024 Vol. 20(6):2359-2378
Available online http://www.ijat-aatsea.com
ISSN 2630-0192 (Online)

Productive and physiological performance and carcass quality of feed-restricted broilers offered diet supplemented with organic selenium and Iraqi protein concentrate 6×1

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Kareem, D. S. and Al-Shammari, K. I. A. (2024). Productive and physiological performance and carcass quality of feed-restricted broilers offered diet supplemented with organic selenium and Iraqi protein concentrate 6×1. International Journal of Agricultural Technology 20(6):2359-2378.

Abstract Feed restriction (FR) regimes are reported as useful strategies in poultry feeding for economical and physiological considerations. From 1 to 5 weeks of broilers' rearing, the current results revealed that most of the FR groups involving quantitative FR (40% of diet) or temporal FR (12 hours/day) without feed additive or with dietary supplementation of 0.8 mg organic selenium (OS) and 25 g Iraqi protein concentrate 6×1 (IPC) per kg decreased ($p \leq 0.01$) feed intake with no effect on body weight, weight gain, relative growth rate and feed efficiency in comparison to control. Also, both temporal FR with dietary OS and IPC were lowered a total mortality ($p \leq 0.05$), registered a high carcass yield with giblets ($p \leq 0.05$) and maintained a body massiveness. Low abdominal fat ($p \leq 0.05$) was found to favor of quantitative FR with dietary OS and IPC and temporal FR with dietary OS. All FR groups fed OS and IPC except for quantitative FR group differed from control in carcass length and bone lengths of keel, thigh and drumstick and circumferences of thigh and breast as well as breast compactness. Each FR group was significantly changed in serum metabolites such as total protein, total cholesterol, glucose, uric acid, creatinine, alanine transaminase and alanine transaminase at 3 and 5 weeks of age. The gizzard and small intestine pH at 3 weeks and large intestine pH at 6 weeks were all decreased ($p \leq 0.05$) by quantitative FR with dietary OS as compared to control. In conclusion, a beneficially special mechanism for each FR group especially those fed diet supplemented with OS and IPC was observed to induce a positive change or maintenance the investigated variables without any deleterious effects on broilers' productivity.

Keywords: Dietary restriction, Physiology, Poultry, Protein, Selenium

Introduction

Modern poultry production is focused on genetic selection for broiler lines which characterized by susceptible fast growth rate and increased feed consumption that is increasingly synchronized with incidence of mortality and metabolic diseases (Saffar and Khajali, 2010; Davoodi-Omam *et al.*, 2019).

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Therefore, feed restriction (FR) is one of the economically adopted strategies to control excessive body weights and decrease feed intake. This will improve feed efficiency, lower abdominal fat deposition and maximize broiler health by providing high protection from mortality caused by skeletal disorders, bone deformities, ascites and sudden death syndrome (Sahraei, 2012; Alkhair, 2021). FC can be performed in feeding programs of poultry by different strategies such as physical approach by reducing quantity or quality of feed consumed and time of feeding and chemical approach by chemical manipulation of diet or controlling FC by various lighting schedules (Alkhair, 2021; Ebeid *et al.*, 2022a). Birds are able to recover their final body weight during compensatory growth phase after termination of FC which is mainly depended on severity of FC and genetic potential, age stage, species and gender of treated birds (Zubair and Leeson, 1996; Muhi and Al-Shammari, 2023). Many reports have previously indicated that implementation of different FC regimes might reflect on positive response during compensatory growth through maintaining growth performance, carcass quality (van der Klein *et al.*, 2017; Azis and Afriani, 2023) and blood constituents (Rahimi *et al.*, 2015). Moreover, FC might implicate in improving feeding behavior (Fondevila *et al.*, 2020), modifying immune response, stimulating microbial population of intestine, increasing digestive indices and absorptive capacity of intestine and improving meat properties (Ebeid *et al*., 2022 a,b) of broilers.

One of the most important feed additives in poultry diet is selenium (Se) which has been intensively attracted a lot of attention. Se is an essential trace element, plays an important role in antioxidant activity because of its chemical formation of glutathione peroxidase as one of essential selenoproteins family synthesized by Se (Surai, 2002 a,b). Se is present in organic and inorganic forms, and selenomethionine is more effective form of organic Se which is metabolized more quickly and efficiently in digestive system than inorganic one such as selenate or selenite (Suchý *et al.*, 2014). In poultry feeding, Se exerted a crucial influence to maximize growth performance and improve meat characteristics (Chuan-long *et al.*, 2021; Khan *et al.*, 2023) and reproductive variables with special emphasis on protection of embryonic growth and spermatogenesis from attract the deleterious free radicals (Surai, 2002b). In addition, Se affects positively to change hematological properties and spectacularly supports the intestinal integrity and immunity biomarkers of organism in broilers (Dalia *et al.*, 2020; Wickramasuriya *et al.*, 2023).

Protein concentrate is a feed material prepared from animal and plant origins with high level of proteins with acceptable content of energy and available amounts of minerals, which can be added or incorporated in poultry diet. In general, protein products are essential for live tissues growth, synthesis

the functional enzymes and hormones, repairing the damaged cells and supporting the overall physiological performance (Beski *et al.*, 2012). Numerous protein concentrates are potentially used for poultry feeding as partial alternatives for soybean meal with different levels in diet such as viable protein concentrates derived from potato (Tuśnio *et al.*, 2013), whey (Szczurek *et al.*, 2013), soy (Vasconcelos *et al.*, 2017), milk (Ahmadzai *et al.*, 2022) and cottonseed (Chen *et al.*, 2023). Some of these dietary products are able to inhibit the virulence and abundance of pathogens in digestive sections of gut (Kiarie *et al.*, 2021), stimulate the antioxidant capacity by lowering prooxidant constituents in serum (Chen *et al.*, 2023), change the intestinal histomorphology and promote the growth performance (Zhang *et al.*, 2022), immunological parameters and protein digestibility (Szczurek *et al.*, 2013) of broilers.

The aim of this research was to highlight the efficacy of quantitative and temporal FR regimes with diet supplemented with organic selenium and Iraqi protein concentrate 6×1 from 2 to 3 weeks of age with investigation this practice after termination of FR on productive performance, carcass characteristics, blood biochemistry and gastrointestinal tract pH of broiler chickens for 5 weeks of age.

Materials and methods

Experimental scheme and treatment

The experiment was implemented in poultry farm and labs of Al-Musaib Technical College, Al-Furat Al-Awsat Technical University, Babylon, Iraq. In total, 420 one-day-old, unsexed chicks broilers Ross 308 were selected with an initial body weight (46.25 ±1.56 g). Birds were reared for 5 weeks in separate replicate pen (100× 150× 50 cm) under optimal management and hygienic conditions. The temperature was 34 °C during the initial 3 days and then was stepwise decreased to reach 25°C at 5 weeks with providing 20 hours of light per day during entire experiment. Birds were fed starter and finisher diets (NRC, 1994; Table 1) in accordance with management guide of broiler chicken308. Birds were fed *ad libitum* during 1st week and then exposed to 2 approaches of FR regimes with feed additives for 2 weeks starting from 2nd till 3rd week of age. Thereafter, birds were refed *ad libitum* without any feed additives for 2 weeks from 4th till 5th week of age as growth compensatory phase. Therefore, at 2nd week, chicks were randomly allocated into 7 groups with 3 replicates and 20 chicks in each. Birds in G1 group were offered *ad libitum* diet without feed additives whereas in G2, G3 and G4 groups, a quantitative FR (40%) was applied individually or with adding 0.8 mg organic selenium (OS) and 25 g Iraqi protein concentrate 6×1 (IPC) per kg of diet, respectively. Moreover, birds of G5, G6

and G7 groups were undergone a temporal FR (12 hours/day) individually or with adding 0.8 mg OS and 25 g IPC per kg diet, respectively.

Table 1. Diet composition and its chemical analysis

Items (%)	Starter (1-21 days)	Finisher (22-35 days)	Chemical analysis***	Starter (1-21 days)	Finisher (22-35 days)
Yellow corn	30.00	40.00	Crude protein (%)	23.00	20.00
Wheat	28.25	24.00	Metabolizable energy (kcal/kg)	3027.00	3195.30
Soybean meal	31.75	24.8	Lysine (%)	1.20	1.10
Protein concentrate	5.00	5.00	Methionine + cysteine (%)	0.85	0.78
Sunflower oil	2.90	4.40	Available phosphorus (%)	0.41	0.43
Limestone	0.90	0.60	Calcium (%)	0.86	0.94
Dicalcium phosphate	0.70	0.90	Se (mg/kg)	0.07	0.09
Salt	0.30	0.10			
Premix	0.20	0.20			
Total	100	100			

Feed supplements

Both OS and IPC were used as feed additives in diets during period of FR regimes. OS was obtained from commercial source in powder form as zinc-L selenomethionine (Zinpro Availa Se, USA) contains 4% selenium in its composition and used as anti-stress material. IPC powder was locally produced and obtained from College of Agricultural Engineering Sciences/University of Baghdad, Iraq and used as physiological modulator and metabolic stimulator. IPC supplement consists of 6 functional bioactive ingredients in its formulation which mainly involves 52% proteins purified from animal and plant origins and combination of immune proteins, detoxifiers, prebiotics, probiotics and other unidentified natural compounds. In addition, it contains 2990 kcal/kg metabolizable energy, 3.13% methionine, 3.92% lysine, 2.60% total phosphorus and 5.64% calcium.

Characteristics studied

Feed intake (FI) and mortality in each replicate of group were registered daily whereas averages of body weight (BW), body weight gain (BWG) and relative growth rate (RGR) were calculated for each bird weekly. From values of

FI and BWG, a feed conversion ratio (FCR) was determined with taken into consideration the number of dead birds. All these variables were presented periodically during FR (2-3 weeks), compensatory growth (4-5 weeks) and in cumulative (1-5 weeks) phases. The production efficiency factor (PEF) was calculated based on formula of Lemme *et al*. (2006).

At the end of the experiment at 5 weeks, all birds were deprived from feeding for 10 hours to empty the GIT content from digesta and stabilize the final BW. Thereafter, 4 birds per replicate, 3 females and 1 male (n= 12/ group) closed to the average BW of each respective experimental group were chosen to analyze the carcass quality. Before slaughter, birds were weighed thoroughly using high precision digital scale (Reshy, USA) to the nearest 0.1 g and then slaughtered using sharp knife under hygienic and biosecurity terms. All hot carcasses were washed and cleaned from feathers, internal viscera, giblets (heart, liver and gizzard) and abdominal fat and then weighed. The carcass yields with giblets or without giblets were calculated and relative abdominal fat content was obtained from dividing its absolute weight over average BW. Whole cleaned carcasses were kept in polyethylene bags for overnight in refrigerator at 4°C. In next day, cold carcasses were weighed and dissected to their main parts, involving breast, thighs and drumsticks to obtain weights of these components in relation to cold carcass weight. Also, these parts in carcass were deboned to register the relative carcass deboning. The procedure of Al-Hajo and Al-Fayadh (2007) was followed to measure dimensions of cold carcass body using tape measure with an accuracy of 1 mm. The length of carcass from posterior part of neck to caudal end was taken and also lengths of keel, thigh and drumstick bones were recorded precisely. For measuring the breast circumference, it was carried out by extending a tape measure around the breast, contacting the area of the upper wings tightly. Thigh circumference was taken from extending a tape measure in contact around the thigh. Carcass conformation indices involving body massiveness as percentage ratio of cold carcass weight in g to body length of cold carcass in cm was taken. Also, from percentage ratio of breast circumference in cm to body length of cold carcass in cm, a breast compactness was obtained.

From the slaughtered birds, 1 male 2 females per replicate (n= 9/ group) were chosen to collect blood at 3 and 5 weeks. Blood was sampled from two sites of collection. First collection site was done by blood withdrawal from brachial vein before slaughtering using disposable syringe (5 ml). Second collection site of blood was from jugular vein immediately from the same birds after being slaughtered. Blood was put in gel separator tube and thereafter centrifuged at 3000 RPM for 15 minutes to separate serum. Blood samples were kept at –25°c until biochemical analyses in serum. A spectrophotometric method was run with following the analytical procedures of special diagnostic kits to investigate the

biochemical metabolites in serum. A commercial kit (Biolabo, French) was used to determine total protein (TP) and total cholesterol (TC) level based on protocol of Young (1995). A kit (Cromatest, Spain) was used to investigate the glucose (Young, 2000). For determination aspartate transferase (AST) and alanine transaminase (ALT), a respective kit (Randox, English) was used depending on method of Reitman and Frankel (1957). Moreover, levels of uric acid and creatinine were analyzed by following the instruction of kit (Biolabo, French) according to Burtis and Ashwood (1999).

At 3 and 5 weeks, from the same slaughtered birds in each replicated group (n=9/ group), birds were subjected to analysis of GIT pH. Following dissection, digesta were collected from 3 anatomical sites of GIT (gizzard, small intestine and large intestine) and put in Eppendorf tubes for determination the pH. This was carried out using litmus pH test strip paper (China) supplemented with complete pH level measuring pH from 1-14 after mixing 1g of digesta with 2 ml of distilled water based on procedure of Chaveerach *et al.* (2004). The changes in test strip color of sample must be corresponded to the pH scale color within 15 seconds and 2 readings were taken from each tested sample.

Statistical analysis

Data were analysed using statistical analysis system of SAS software (SAS, 2012) and means of variables under effect of respective group were compared by Duncan's multiple range test (Duncan, 1955) to find out any significant differences among tested groups. Probability level of ($P<0.05$) and ($P<0.01$) was considered statistically significant for all variables.

Results

Productive performance and carcass quality

G5-G7 had similar BW and BWG at 3 weeks and 2-3 weeks, respectively compared to G1 (Table 2). The groups (G4-G7) recorded similar BW and BWG at 5 weeks and 1-5 weeks, respectively compared to G1. No differences were found among groups pertaining BWG at 4-5 weeks.

Result showed that low FI ($p\leq0.01$) was achieved by all experimental groups as compared to G1 at 2-3 week with no significant difference among groups at 4-5 weeks (Table 3). However, G2–G5 showed lower FI ($p\leq0.01$) compared with G1 at 1-5 weeks. In FCR, it was not differed between G1 and other groups at 2-3 weeks with lack any differences was obvious among groups at 4-5 weeks and 1-5 weeks.

Table 2. Body weight (g) and body weight gain (g) of feed-restricted broiler chickens fed diet supplemented with OS and IPC (mean± standard error)

Groups	Body weight		Body weight gain		
	3 weeks	5 weeks	2-3 weeks	4-5 weeks	1-5 weeks
G1	753.10^{a} ± 31.34	2219.60^{a} ± 43.12	571.47^{a} ± 28.77	1466.50 ± 71.61	2173.32^{a} ± 53.12
G2	556.50^{b} ± 31.17	1889.80^{c} ± 18.95	373.90^{b} ± 33.79	1333.30 ± 14.70	1843.52^{c} ± 18.95
G3	597.73^{b} ± 34.16	1964.60^{c} ± 20.38	415.50^{b} ± 34.61	1366.80 ± 21.18	1918.32^{bc} ± 20.38
G4	615.73^{b} ± 12.18	2166.30^{ab} ± 24.93	433.50^{b} ± 4.86	1550.60 ± 54.58	2120.10^{ab} ± 64.93
G5	737.87^{a} ± 27.04	2201.20^{ab} ± 37.61	555.03^{a} ± 24.93	1463.30 ± 63.59	2154.92^{ab} ± 77.61
G6	645.57^{ab} ± 34.50	2003.90^{abc} ± 30.52	463.83^{ab} ± 65.58	1358.30 ± 14.02	1957.62^{abc} ± 50.52
G7	728.67^{a} ± 5.44	2167.20^{ab} ± 48.59	545.97^{a} ± 3.79	1438.50 ± 33.89	2121.00^{ab} ± 28.59
Significance	**	**	**	NS	**

Different superscript letters in the same column indicate to significant difference among groups, ** at ($p \leq 0.01$), NS: non-significant.

Table 3. Feed intake (g) and feed conversion ratio of feed-restricted broiler chickens fed diet supplemented with OS and IPC (mean± standard error)

Groups	Feed intake			Feed conversion ratio		
	2-3 weeks	4-5 weeks	1-5 weeks	2-3 weeks	4-5 weeks	1-5 weeks
G1	1005.00^{a} ± 13.52	2325.56 ± 29.54	3484.61^{a} ± 33.88	1.76^{ab} ± 0.06	1.59 ± 0.07	1.60 ± 0.03
G2	603.00^{c} ± 8.11	2337.06 ± 74.65	3091.23^{c} ± 72.52	1.63^{ab} ± 0.11	1.78 ± 0.20	1.69 ± 0.14
G3	603.80^{c} ± 8.12	2380.59 ± 33.06	3137.36^{bc} ± 25.32	1.47^{b} ± 0.11	1.74 ± 0.00	1.63 ± 0.02
G4	628.13^{c} ± 8.45	2371.00 ± 71.95	3157.08^{bc} ± 79.21	1.44^{b} ± 0.07	1.53 ± 0.03	1.49 ± 0.08
G5	896.33^{b} ± 9.87	2233.48 ± 69.74	3288.88^{bc} ± 74.21	1.62^{ab} ± 0.07	1.53 ± 0.10	1.53 ± 0.07
G6	874.95^{b} ± 2.85	2283.58 ± 48.74	3315.23^{ab} ± 46.47	1.97^{a} ± 0.31	1.69 ± 0.12	1.69 ± 0.02
G7	886.79^{b} ± 3.51	2257.88 ± 65.92	3302.02^{ab} ± 60.59	1.62^{ab} ± 0.04	1.57 ± 0.05	1.56 ± 0.03
Significance	**	NS	**	*	NS	NS

Different superscript letters in the same column indicate to significant difference among groups, * at ($p \leq 0.05$), ** at ($p \leq 0.01$), NS: non-significant.

It revealed that RGR of G4-G7 and G3-G7 was identical compared with G1 at 2-3 weeks and 1-5 weeks, respectively with no differences in RGR were present among groups at 4-5 weeks (Table 4). Low mortality ($p \leq 0.05$) revealed in favor of G6 and G7 at 2-3 weeks and 1-5 weeks in comparison to G1 with no differences among groups at 4-5 weeks.

Table 4. Relative growth rate (%) and mortality (%) of feed-restricted broiler chickens fed diet supplemented with OS and IPC (mean± standard error)

Groups	Relative growth rate			Mortality		
	2-3 weeks	4-5 weeks	1-5 weeks	2-3 weeks	4-5 weeks	1-5 weeks
G1	$121.65^{a} \pm 2.37$	97.76 ± 4.23	$191.64^{a} \pm 0.15$	$3.33^{ab} \pm 0.67$	0.00 ± 0.00	$3.33^{ab} \pm 0.67$
G2	$99.94^{b} \pm 5.11$	107.39 ± 4.64	$190.04^{b} \pm 0.71$	$5.00^{a} \pm 1.86$	0.00 ± 0.00	$5.00^{a} \pm 1.86$
G3	$105.56^{b} \pm 4.94$	106.14 ± 3.64	$190.63^{ab} \pm 0.11$	$3.33^{ab} \pm 0.35$	0.00 ± 0.00	$3.33^{ab} \pm 0.35$
G4	$108.03^{ab} \pm 1.35$	110.48 ± 1.24	$191.47^{a} \pm 0.24$	$1.66^{bc} \pm 0.67$	0.00 ± 0.00	$1.66^{bc} \pm 0.67$
G5	$119.74^{a} \pm 1.37$	99.13 ± 2.21	$191.61^{a} \pm 0.29$	$3.33^{ab} \pm 0.64$	0.00 ± 0.00	$3.33^{ab} \pm 0.64$
G6	$110.33^{ab} \pm 7.83$	101.57 ± 8.26	$190.71^{ab} \pm 0.18$	$0.00^{c} \pm 0.00$	0.00 ± 0.00	$0.00^{c} \pm 0.00$
G7	$119.44^{a} \pm 0.20$	98.05 ± 3.038	$191.43^{a} \pm 0.38$	$0.00^{c} \pm 0.00$	0.00 ± 0.00	$0.00^{c} \pm 0.00$
Significance	*	NS	*	*	NS	*

Different superscript letters in the same column indicate to significant difference among groups, * at ($p \leq 0.05$), NS: non-significant.

G5 and G7 achieved similar value of PEF compared with G1 (Table 5). High carcass yield without giblets and carcass yield with giblets ($p \leq 0.05$) were shown in (G2, G6 and G7) and (G2 and G7), respectively. G3, G4 and G7 were obtained high breast weight ($p \leq 0.05$) with no differences among group in weights of thighs and drumsticks and carcass deboning. Moreover, abdominal fat was decreased ($p \leq 0.05$) by G3, G4 and G6 compared to G1.

The groups (G3-G7) increased lengths of carcass and drumstick ($p \leq 0.01$) with absence of differences among groups regarding to keel bone length and breast compactness (Table 6). All experimental groups were not differed from G1 in thigh bone length and thigh circumference. In comparison to G1. The groups of G4-G7 and G6- G7 revealed identical values ($p \leq 0.05$) of breast circumference and body massiveness, respectively.

Table 5. Production efficiency factor and carcass yield of feed-restricted broiler chickens fed diet supplemented with OS and IPC (mean± standard error)

Groups	PEF	Carcass yield (%)		Carcass components (%)			Carcass deboning (%)	Abdominal fat (%)
		Without giblets	With giblets	Breast	Thighs	Drum-sticks		
G1	362.86^{a} ± 16.84	72.27^{bc} ± 0.32	76.05^{bc} ± 1.52	37.67^{b}± 1.47	17.27 ± 1.21	13.95 ± 0.62	74.28 ± 1.91	1.56^{a} ± 0.25
G2	295.38^{c} ± 25.10	73.26^{a} ± 0.94	77.64^{a} ± 1.00	38.26^{ab} ± 1.71	16.88 ± 0.75	14.73 ± 0.75	74.02 ± 1.20	1.18^{ab} ± 0.02
G3	315.24^{b} ± 18.35	71.35^{c} ± 0.47	75.88^{c} ± 0.40	40.08^{a} ± 1.63	16.83 ± 0.90	13.48 ± 0.55	74.02 ± 0.71	0.91^{b} ± 0.06
G4	390.67^{b} ± 34.96	72.61^{ab} ± 0.73	77.13^{b} ± 0.44	39.72^{a} ± 0.97	17.85 ± 1.39	13.07 ± 0.48	75.43 ± 0.47	0.80^{b} ± 0.05
G5	379.87^{a} ± 33.08	72.42^{bc} ± 0.80	77.16^{ab} ± 0.56	37.61^{b} ± 1.83	17.58 ± 0.44	14.18 ± 0.36	75.15 ± 1.51	1.55^{a} ± 0.13
G6	321.36^{b} ± 13.04	73.27^{a} ± 0.09	77.37^{ab} ± 0.48	38.16^{ab} ± 0.86	17.18 ± 0.44	14.55 ± 0.12	74.89 ± 2.31	1.04^{b} ± 0.25
G7	378.19^{a} ± 25.63	73.74^{a} ± 0.23	78.23^{a} ± 1.23	39.99^{a} ± 1.82	16.23 ± 0.56	13.81 ± 0.96	75.28 ± 0.78	1.27^{ab} ± 0.19
Significance	*	*	*	*	NS	NS	NS	*

Different superscript letters in the same column indicate to significant difference among groups, * at ($p \leq 0.05$), NS: non-significant.

Blood biochemistry

It indicated that only G6 and G7 obtained similar value in serum TP and AST compared with G1, respectively at 3 weeks (Table 7). The groups (G4-G7) decreased ($p \leq 0.01$) TC. In comparison to G1, all groups were high level ($p \leq 0.01$) in serum glucose. As compared with G1, similar level of ALT was found in G3, G4 and G6. Moreover, low level ($p \leq 0.01$) of uric acid was shown by G3-G6 and groups of G3, G5, G6 and G7 which decreased ($p \leq 0.01$) serum creatinine.

Table 6. Physical carcass quality of feed-restricted broiler chickens fed diet supplemented with OS and IPC (mean± standard error)

Groups	Length (cm)				Circumference (cm)		Breast compactness (cm/cm)	Body massiveness (g/cm)
	Carcass	Keel bone	Thigh bone	Drumstick bone	Breast	Thigh		
G1	23.7^{ab} ± 0.37	15.00 ± 0.57	7.60^{ab} ± 0.26	10.36^{a} ± 0.33	22.33^{a} ± 0.16	17.66^{ab} ± 0.33	0.94 ± 0.01	73.20^{a} ± 6.78
G2	22.46^{c} ± 1.03	14.00 ± 0.57	7.06^{b} ± 0.58	9.10^{b} ± 0.58	20.83^{b} ± 0.72	16.06^{b} ± 1.03	0.93 ± 0.09	66.77^{bc} ± 5.71
G3	23.30^{b} ± 1.99	14.66 ± 0.66	8.73^{a} ± 0.26	11.33^{a} ± 0.33	21.33^{b} ± 1.16	18.00^{a} ± 0.57	0.92 ± 0.08	63.81^{d}± 7.75
G4	24.06^{a} ± 0.06	15.16 ± 0.60	7.56^{ab} ± 0.23	11.50^{a} ± 0.50	21.83^{ab} ± 0.44	18.00^{a} ± 0.57	0.91 ± 0.02	63.97^{cd} ± 3.51
G5	25.00^{a} ± 0.57	15.33± 0.88	8.50^{a} ± 0.50	10.66^{a} ± 0.33	23.00^{a} ± 0.57	18.00^{a} ± 0.57	0.92 ± 0.03	63.04^{d} ± 4.76
G6	23.66^{ab} ± 0.33	14.00 ± 0.00	7.66^{ab} ± 0.33	10.83^{a} ± 0.16	22.16^{a} ± 0.88	17.66^{ab} ± 0.33	0.93 ± 0.04	70.24^{ab} ± 7.00
G7	23.10^{bc} ± 0.49	15.33 ± 0.88	8.33^{ab} ± 0.33	11.66^{a} ± 0.33	21.66^{ab} ± 0.33	17.33^{ab} ± 0.33	0.94 ± 0.02	74.01^{a} ± 4.18
Significance	**	NS	*	**	*	*	NS	*

Different superscript letters in the same column indicate to significant difference among groups, * at ($p \leq 0.05$), ** at ($p \leq 0.01$), NS: non-significant.

G4 increased serum TP ($p \leq 0.05$) compared to G1 at 5 weeks (Table 8). The decreased levels of TC and glucose ($p \leq 0.01$) were found in G2, G3, G4 and G7. G2, G4, G6 and G7 which was not differed from G1 in AST. G2 and G4 was not differed from G1 in creatinine. Low ALT ($p \leq 0.01$) was revealed in G4 and G7 whereas low uric acid ($p \leq 0.01$) was found in G2 and G3.

GIT pH level

G3 caused to drop gizzard and small intestine pH ($p \leq 0.05$) at 3 weeks and drop large intestine pH ($p \leq 0.05$) at 5 weeks as compared with G1 (Table 9). Lack of differences among groups was found in large intestine and small intestine pH at 3 and 5 weeks, respectively. G1 was not differed from all groups regarding to gizzard pH at 5 weeks.

Table 7. Biochemical constituents in blood serum of feed- restricted broiler chickens fed diet supplemented with OS and IPC at 3 weeks (mean± standard error)

Groups	TP (g/dl)	TC (mg/dl)	glucose (mg/dl)	AST (U/L)	ALT (U/L)	Uric acid (mg/dl)	Creatinine (mg/dl)
G1	$2.60^{a\pm}$ 0.00	$140.66^{c}\pm$ 1.33	$180.00^{g}\pm$ 0.00	$25.00^{d}\pm$ 0.00	$5.00^{c}\pm$ 0.00	$7.60^{b}\pm$ 0.00	$0.44^{b}\pm$ 0.00
G2	$1.73^{f}\pm$ 0.03	$171.00^{a}\pm$ 0.00	$193.66^{a}\pm$ 0.33	$29.00^{a}\pm$ 0.000	$6.66^{a}\pm$ 0.333	$8.43^{a}\pm$ 0.03	$0.49^{b}\pm$ 0.00
G3	$2.36^{b}\pm$ 0.03	$157.66^{b}\pm$ 0.66	$188.33^{d}\pm$ 0.33	$26.00^{c}\pm$ 0.00	$4.00^{c}\pm$ 0.00	$6.50^{c}\pm$ 0.00	$0.42^{d}\pm$ 0.00
G4	$2.23^{c}\pm$ 0.03	$119.00^{e}\pm$ 2.30	$186.00^{e}\pm$ 0.00	$26.00^{c}\pm$ 0.00	$4.33^{c}\pm$ 0.00	$5.10^{e}\pm$ 0.00	$0.45^{b}\pm$ 0.00
G5	$1.86^{e}\pm$ 0.03	$107.66^{f}\pm$ 2.02	$191.00^{b}\pm$ 0.00	$27.00^{b}\pm$ 0.00	$7.00^{a}\pm$ 0.00	$5.10^{e}\pm$ 0.00	$0.40^{e}\pm$ 0.00
G6	$2.56^{a}\pm$ 0.03	$116.33^{e}\pm$ 1.33	$189.33^{c}\pm$ 0.33	$26.00^{c}\pm$ 0.00	$4.00^{c}\pm$ 0.00	$5.60^{d}\pm$ 0.01	$0.43^{c}\pm$ 0.00
G7	$2.03^{d}\pm$ 0.03	$129.66^{d}\pm$ 1.33	$182.66^{f}\pm$ 0.33	$25.00^{d}\pm$ 0.00	$6.00^{b}\pm$ 0.00	$7.60^{b}\pm$ 0.00	$0.42^{d}\pm$ 0.00
Signifi-cance	**	**	**	**	**	**	**

Different superscript letters in the same column indicate to significant difference among groups, ** at ($p\leq0.01$).

Table 8. Biochemical constituents in blood serum of feed- restricted broiler chickens fed diet supplemented with OS and IPC at 5 weeks (mean± standard error)

Groups	TP (g/dl)	TC (mg/dl)	Glucose (mg/dl)	AST (U/L)	ALT (U/L)	Uric acid (mg/dl)	Creatinine (mg/dl)
G1	$2.76^{b}\pm$ 0.03	$135.66^{b}\pm$ 0.84	$162.00^{c}\pm$ 0.00	$17.00^{c}\pm$ 0.00	$6.33^{c}\pm$ 0.13	$5.10^{e}\pm$ 0.00	$0.31^{e}\pm$ 0.00
G2	$2.26^{c}\pm$ 0.03	$122.66^{c}\pm$ 0.33	$144.00^{e}\pm$ 1.00	$16.66^{c}\pm$ 0.33	$7.00^{b}\pm$ 0.00	$4.46^{f}\pm$ 0.03	$0.35^{cde}\pm$ 0.00
G3	$2.50^{bc}\pm$ 0.35	$100.66^{d}\pm$ 0.33	$136.00^{f}\pm$ 0.57	$18.33^{b}\pm$ 0.66	$6.00^{cd}\pm$ 0.00	$4.50^{f}\pm$ 0.00	$0.36^{cd}\pm$ 0.00
G4	$3.23^{a}\pm$ 0.08	$127.00^{c}\pm$ 0.00	$145.00^{e}\pm$ 0.00	$16.00^{c}\pm$ 0.00	$5.00^{e}\pm$ 0.00	$7.30^{a}\pm$ 0.00	$0.33^{e}\pm$ 0.03
G5	$2.80^{ab}\pm$ 0.00	$149.66^{a}\pm$ 0.33	$169.66^{b}\pm$ 0.33	$20.33^{a}\pm$ 0.66	$8.00^{a}\pm$ 0.00	$5.30^{d}\pm$ 0.00	$0.52^{a}\pm$ 0.00
G6	$2.66^{bc}\pm$ 0.03	$136.00^{b}\pm$ 0.00	$174.66^{a}\pm$ 0.33	$17.00^{c}\pm$ 0.00	$5.66^{cde}\pm$ 0.23	$6.60^{b}\pm$ 0.00	$0.45^{b}\pm$ 0.00
G7	$2.43^{bc}\pm$ 0.12	$122.00^{c}\pm$ 0.57	$159.33^{d}\pm$ 0.33	$17.00^{c}\pm$ 0.00	$5.33^{de}\pm$ 0.33	$5.90^{c}\pm$ 0.00	$0.39^{c}\pm$ 0.00
Signifi-cance	*	**	**	**	**	**	**

Different superscript letters in the same column indicate to significant difference among groups, * at ($p\leq0.05$), ** at ($p\leq0.01$).

Table 9. Gastrointestinal tract pH of feed-restricted broiler chickens fed diet supplemented with OS and IPC (mean± standard error)

Groups	Gizzard	Small intestine	Large intestine	Gizzard	Small intestine	Large intestine
		3 weeks			5 weeks	
G1	$3.50^{a} \pm 0.50$	$5.00^{a} \pm 0.00$	6.50 ± 0.50	$3.00^{ab} \pm 0.00$	4.66 ± 0.66	$6.00^{a} \pm 1.00$
G2	$2.50^{ab} \pm 0.50$	$4.00^{ab} \pm 0.00$	6.50 ± 0.50	$3.66^{a} \pm 0.88$	5.00 ± 1.00	$5.66^{ab} \pm 1.20$
G3	$2.00^{b} \pm 0.00$	$3.50^{b} \pm 0.50$	6.50 ± 0.50	$4.00^{a} \pm 0.57$	5.00 ± 0.57	$4.33^{b} \pm 0.88$
G4	$2.50^{ab} \pm 0.50$	$4.50^{a} \pm 0.50$	6.50 ± 0.50	$3.33^{a} \pm 0.88$	4.00 ± 0.57	$5.66^{ab} \pm 1.33$
G5	$2.50^{ab} \pm 0.50$	$4.50^{a} \pm 0.50$	6.00 ± 1.00	$3.33^{a} \pm 0.33$	4.33 ± 0.33	$6.33^{a} \pm 0.66$
G6	$3.00^{a} \pm 1.00$	$5.00^{a} \pm 0.00$	6.50 ± 0.50	$3.33^{a} \pm 0.33$	4.66 ± 0.33	$5.66^{ab} \pm 0.33$
G7	$2.50^{ab} \pm 0.50$	$5.00^{a} \pm 1.00$	6.50 ± 0.50	$2.33^{b} \pm 0.33$	4.66 ± 0.33	$6.66^{a} \pm 0.33$
Signifi-cance	*	*	NS	*	NS	*

Different superscript letters in the same column indicate to significant difference among groups, * at ($p \leq 0.05$), NS: non-significant.

Discussion

After termination of FR at 2-3 weeks and throughout the whole experiment at 1-5 weeks, groups of temporal FR (G5-G7) were followed by G4 which successfully maintained their BW, BWG, RGR to synchronize with decreased FI and unchanged FCR, reduced mortality and stable PEF value as compared to G1. FC regimes with dietary OS and IPC supported productive profile of feed restricted birds. More noticeable results were found to absence any alterations in growth performance after termination the compensatory growth phase at 4-5 weeks. This might explain that birds after FR applications will adopt to recover their performance very quickly (Omam *et al.*, 2019; Alkhair, 2021). In the meanwhile, there improved in carcass yields and relative weight of breast with

lowered abdominal fat in treated group-dependent manner. Moreover, FR caused a stability in physical dimensions of carcass. Birds exposed to restricted diet with specific additives could enhance digestibility process and absorption rate of nutrients from low feed consumed due to activate the histomorphology, immune function and antioxidant capacity of intestinal sections (Dastar *et al.*, 2017; Ebeid *et al*., 2022a). Some previous results were consistent and inconsistent with the current data. For instance, Jahanpour *et al*. (2015) suggested that quantitative FR at 75 and 50% of FI starting from 8 till 14 or 21 days of age had no affect BW, relative weights of wing, leg and breast and abdominal fat whereas weight of carcass in feed restricted birds (75%) at 8-21 days was higher than control in broilers. Identical result was reported by der Klein *et al*. (2017) that consuming 90, 80 or 70% of FI or 95, 90, 85 or 80% of FI during 2 and 3 weeks, respectively in broiler did not affect FCR and BW at 14, 21 and 35 days, and also no changing in weights of breast and legs with low cumulative FI for birds consumed 90, 85, or 80% of FI at 35 days. The result is partly agreed with Fondevila *et al*. (2020) who concluded that exposing broiler to temporal FR for 4, 6, or 8 hours per day from 7 to 19 days of age exhibited low daily FI and BW gain with no effect on FCR depending on length of the FR period. Also, the data are partly in compliance with Muhi and Al-Shammari (2023), who indicated that FR at 40% or fasting for 12 hours per day from 2 to 3 weeks of age caused decreased values of FI, BW, FCR and mortality and increased PEF without effects on carcass yield, carcass parts and abdominal fat of broilers at 35 days. Similarly, it was found that broiler given FI for 2 and 4 hours from 7-14 or 7-21 days caused no changing in BW, carcass yield, weights of carcass parts with low abdominal fat at 42 days (Azis and Afriani, 2023). Dietary OS additive during FR periods might succeed to alleviate the harmful consequences on productive variables of broilers after FR application. This can be translated into positive importance of OS in boosting the antioxidant activity in avian body system against multiple environmental stressors (Surai, 2002a; Suchý *et al.*, 2014). Consumption of OS in diet might protect broilers from BW depression and improving feed efficiency because OS characterizes with high retention and deposition in tissues and affects villi height and overall digestibility with potential impact on activation the serum immunoglobulins (Dalia *et al.*, 2020). No previous data are available regarding to using OS sources as antioxidant element during of FR periods in poultry research findings. However, many reports revealed the antioxidant role of OS in blood, tissues and meat of chickens. During heat stress, Mohamed and Toson (2022) concluded that feeding broilers up to 5 weeks with 0.6 or 0.9 mg of OS/kg diet improved nutrient digestibility, and as a result, BW, BWG, FCR, carcass weight was enhanced with no effect on FI and decreasing in abdominal fat. Also, during pathological stress with Eimeria challenge, when broilers fed 0.3 mg/kg

selenized yeast as OS did not change BW, BWG, FCR and FI up to 24 days, although there was a high expression of tight junction in intestinal tissues as marker of gut health due to higher availability of OS in serum and tissues compared to control and 0.3 mg/kg inorganic Se (Wickramasuriya *et al.*, 2023). Moreover, maintaining in growth performance and carcass properties following FR in G6 then G3 were recently in line with results of Zhao *et al.* (2023), who reported that FI, FCR and BW were all not changed at 42 days for broilers received diet containing 0.6 mg/kg OS derived from yeast compared to control and sodium selenium under non stressful conditions. Presumably, maintaining in certain productive variables of G4 and G7 might be created from the feeding influence with IPC and its content from synbiotic. Dastar *et al.* (2017) found that temporal FR for 6 hours per day from 10 to 38 days of age depressed BWG and lowered FI of broilers but when birds received 2 g prebiotic or its mixture with 0.1 g of probiotic as synbiotic per kg of diet decreased FI, FCR, carcass yield and abdominal fat with increased BWG from 0 to 42 days. According to results documented by Saffar and Khajali (2010), that application temporal FR at 08-12 hours and 13-17 hours for 5-11 days or fasting on days 9 and 11 for 24 hours with or without consuming 1 g of probiotic per litter drinking water did not influence BW, BWG, FCR, carcass yield and weights of breast and thighs at 49 days with no changing also in FI and mortality of high altitude reared-male broilers. Probably that using protein concentrates enhances the ability to absorb nutrients very quickly through intestinal maturation and its microbial composition and antioxidant capacity (Beski *et al.*, 2012; Kiarie *et al*., 2021; Ahmadzai *et al*., 2022). Zhang *et al.* (2022) proved that were increasing levels of beneficial growth in ceacal bacteria with high muscle thickness and villus height of jejunum and ileum and high duodenal goblet cells in male broilers fed diet fortified with 12% soybean concentrate during the first 10 days compared to control. Also, Szczurek *et al.* (2013) conveyed that adding 8 or 32 g per kg whey protein concentrate improved nutrient absorption rate, lowered heterophile/lymphocyte ratio and malondialdehyde level in the liver and meat and consequently decreased FI and FCR and increased BGW and carcass yield of broilers at 42 days. Consistent with the present data, Chen *et al.* (2023) showed that was maintaining in normal growth and decreasing in FI and FCR of broiler received diet with 4 % of cottonseed protein concentrate.

Animal health can be monitored by any alterations in biochemical metabolites of liver and kidney functions under influence of diet manipulations. Various outcomes among authors were previously reported which were basically related to type, period and intensity of FR regimes and its reaction with age of experienced birds (Zubair and Leeson, 1996). Inconsistent result by Tůmová *et al.* (2019) who found that serum TP, TC and glucose levels were not altered

weekly and at 35 days during and after quantitative FR in both males and females broilers received diet restricted at 20 and 35% of free feeding from 7 to 14 days old. Other data recorded that was no changing in blood glucose, TP, uric acid, TC, AST and ALT at 42 days of broilers fed restricted diet at 5, 10, 15 and 20% for 1 or 2 weeks on days 8-14 or 8-21 of age, respectively (Davoodi-Omam *et al.*, 2019) or during feeding on restricted diet at 15 and 30% for 1 week followed by re-feeding different levels of energy and protein from 15 to 42 days of age (Rahimi *et al.*, 2015). In previous study of Boostani *et al.* (2010) showed that temporal FR for 8 hours per day in 3 periods (7-21, 14-28 and 21-35 days) altered serum glucose and TC with no altering the TP based on period of FR of stressed broiler by ascites. The most prominent effect in investigations of these variables in current data was in FR with OS and IPC after FR and re-alimentation periods. Also, each group had a special mechanism based on FI type to induce positive changing of our investigated parameters. OS can be easily deposited in serum and tissues and thus protects liver as main organ for metabolism from oxidative damage. This was perhaps correlated to interesting role of OS in stimulation the antioxidant capacity in blood and muscles (Chuan-long *et al*., 2021) via higher gene expression of antioxidative enzymes and selenoproteins involving glutathione peroxidase, thioredoxin reductase, iodothyronine deiodinase (Dalia *et al.*, 2017), catalase and superoxide dismutase (Ibrahim *et al*., 2019) in liver of broilers. Similarly, there was lowering in concentrations of creatinine, ALT and AST, albeit no changing in TP of broilers' serum at 6 weeks depending on feeding 3 bacterial sources as OS at 0.3 mg/ kg (Dalia *et al.*, 2017). Differently, it was found that serum TP, glucose, uric acid and TC were not changed by adding 0.3 mg/kg OS, inorganic Se and nano-Se in male broilers stressed by tert-butyl hydroperoxide (Boostani *et al*., 2015) or in naked neck chickens under non-stress condition (Khan *et al.*, 2023). Likewise, the strong activity of IPC in some biochemical variables is probably due to its influential composition which exhibited antioxidant activity in blood and tissues (Chen *et al.*, 2023) during and after FR periods. In accordance with findings of Ashour *et al*. (2019), who proved that broilers' health improved by increasing serum antioxidant indicators during feeding on dietary whey protein concentrate at 0.15, 0.20 and 0.25% which strongly reflected on lowering creatinine and TC levels without effects on serum AST and ALT. Different results to us showed that was no changing in serum AST and TP by adding protein concentrates of potato at 5, 10 or 15% (Tuśnio *et al.*, 2013) and soy at 3, 6 and 9% (Vasconcelos *et al.*, 2017) in broiler diets.

The positive decrease in GIT pH for G3 might be associated with increasing retention time of the digesta which caused high acidity in these digestive sections affected by FR with enhancing in digestive adaptation after compensatory growth period (Zubair and Leeson, 1996). Fondevila *et al.* (2020) pointed that

quantitative FR encourages growth of microflora population such as *Lactobacillus spp* in crop section with enhanced levels in products of microbial fermentation with secretion of hydrochloric acid in the proventriculus which consequently might cause a lowering in GIT pH. Therefore, G3 might decrease GIT pH through modulating the gut microbial ecosystem by reducing the pathogenic bacteria and increasing the healthy one (Ebeid *et al.*, 2022a). In contrast, Dastar *et al.* (2017) reported that crop pH increased at 42 days of age compared to control in broilers exposed to FR for 6 hours daily at 10-38 days. In addition, intestinal homeostasis and its association with microbial balance and antioxidation protection are Se-dependent (Surai, 2002a) and this would explain the reduced GIT pH in G3 group.

It is obvious that using both quantitative FR at 40% and temporal FR for 12 hours per day in diets supplemented with 0.8 mg OS and 25 g IPC per kg lasted from 2 to 3 weeks of broilers' age was able to induce several changes in variables studied. At 35 days, all FR groups especially during supporting by these feed supplements decreased FI with no mostly harmful effect on final growth indicators, feed efficiency and physical characteristic of carcass. The temporal FR is better than quantitative FR in lowering mortality and maintaining growth indicators with increasing carcass yield. Moreover, the changing in biochemical indicators of liver and kidney functions with stable most of the GIT pH among groups were observed. In general, the productive and physiological response to FR was based on treated group-dependent manner. Therefore, the currently suggested programs of FR might be applied as beneficial strategies for broilers feeding.

Acknowledgements

The authors would like to express their special thanks and sincere gratitude for Prof. Dr. Saad Abd Al-Hussien Naji, University of Baghdad, Iraq for free supporting of research of the MSc degree of 1st author with a new tested feed additive, Iraqi protein concentrate 6×1.

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(Received: 27 April 2024, Revised: 13 September 2024, Accepted: 7 November 2024)