

MORPHOLOGICAL, PHYSIOLOGICAL, AND IMMUNE RESPONSE OF NAKED NECK MALE CHICKENS FED CABBAGE LEAVES AS SELENIUM SOURCE REARED UNDER DIFFERENT PRODUCTION SYSTEMS

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ABSTRACT

The experiment was designed to investigate the impact of cabbage leaves as a selenium source on the morphological, physiological, and immune responses of Naked-Neck male chickens raised under different production systems. For this, 96 Naked Neck cockerels (6 weeks old) were raised, and maintained at the Avian Research and Training (ART) Centre, UVAS, Ravi Campus, Pattoki. These experimental birds were allocated into 2 production systems (Intensive and free range) and 4 feeding strategies (control feed, 0.3mg per kg selenium from cabbage source, 0.3mg per kg selenium from sodium selenite source, and 0.3mg per kg selenium from cabbage + sodium selenite source). A factorial ANOVA was used to analyze the collected data, followed by a comparison of significant means through Duncan's Multiple Range test. In the intensive production system, Naked Neck male birds exhibited greater morphometric traits and immune responses against Newcastle disease (ND) and Infectious Bronchitis (IB) compared to those reared in the free-range system; however, no difference was observed in the physiological response between the two systems. Regarding feeding strategies, greater morphometric traits, and immune responses against ND and IB were observed in birds reared on 0.3 mg per kg selenium from cabbage source feed treatment. On the other hand, higher bursa weight was found in those birds that were fed control feed and 0.3 mg per kg selenium from cabbage+ sodium selenite source-based feed treatment. Among physiological responses, higher heartbeat and respiratory rates were observed in birds raised on 0.3 mg per kg selenium from sodium selenite. Interactions between feeding strategies and housing systems showed significant variation among morphological, physiological, and immune responses. In conclusion, Naked Neck chickens exhibited excellent morphometric traits and a robust immune response when reared under an intensive production system supplemented with 0.3 mg per kg selenium from cabbage.

Key words: Naked neck, morphometrics, immune response, production systems.

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INTRODUCTION

In Pakistan, locally adapted breeds such as Aseel, Fayoumi, Desi, and Naked Neck chickens are commonly reared in backyard systems due to their hardiness, adaptability, and disease resistance (Jakhrani *et al.*, 2024; Ahmad *et al.*, 2014). These breeds produce meat and eggs with a rich nutritional profile (Bell and Weaver, 2002) and are well-suited to thrive in hot and humid conditions (Sahu *et al.*, 2015). Among the locally adapted indigenous chicken breeds of Pakistan (Aseel, Fayoumi, Naked Neck, and Desi), the Naked Neck chicken has gained significant popularity for its exceptional performance in tropical and sub-tropical environments (Sadeef *et al.*, 2015; Riaz *et al.*, 2024). This breed has shown promise in rural poultry farming due to its qualities, like heat tolerance and performance under extensive management systems. With increasing consumer awareness about health and nutrition, the demand for slow-growing and traditionally reared chickens has also risen (Tougan *et al.*, 2013; Ponte *et al.*, 2004). As a result, aligning with the increasing consumer demand for functional foods, researchers and producers are placing greater emphasis on optimizing poultry nutrition through the utilization of natural feed resources.

Trace minerals play vital roles in numerous physiological and metabolic processes, including enzyme activation, antioxidant defense, immune response, and growth regulation in both humans and animals. Among trace minerals, selenium stands out as an essential element for both human and animal health. Due to its multifaceted role in biological systems, it has gained considerable scientific and industrial importance. Selenium is widely used in the

pharmaceutical, food, and feed industries, where it plays a crucial role in supporting reproductive health (Kaur and Bansal, 2005), enhancing immune function (McKenzie *et al.*, 1998), and strengthening antioxidant defense mechanisms (Brenneisen *et al.*, 2005). Its biological function is mainly attributed to its role as a cofactor in selenoproteins such as glutathione peroxidase, which neutralize oxidative stress (Reis *et al.*, 2009; Hanschmann *et al.*, 2013). Selenium is available in two main sources: organic (e.g., selenomethionine in selenized yeast) and inorganic (e.g., sodium selenite or selenide). Studies have shown that the organic form is more bioavailable (Bodnar *et al.*, 2016) and less toxic (Tiwary *et al.*, 2006), making it a preferred option for dietary supplementation. Research has also revealed selenium's role in improving immune responses in poultry, including enhanced antibody production against diseases like Newcastle disease (Hegazy and Adachi, 2000; Hoffmann, 2007).

Among plants, members of the Brassica family, particularly cabbage (*Brassica oleracea* L.) have a potential natural supplier of organic selenium. The selenium content in such plants is influenced by the mineral availability in the soil (Olivera *et al.*, 2005). Cabbage has the ability to accumulate high selenium levels, including substantial amounts of seleno-methionine (Mechora *et al.*, 2012), and is also rich in minerals, crude protein, and phenolic compounds (Rosa and Heaney, 1996; Mustafa and Baurhoo, 2017; Sultana and Anwar, 2008). These bioactive components not only enhance the antioxidant defense systems but may also maintain the functions and structure of cells (Ji *et al.*, 2015).

Nowadays, commercial poultry farming predominantly employs a comprehensive and well-balanced diet; however, a free-range production system relies on household leftovers, vegetables, and grasses to meet dietary needs (Fanatico *et al.*, 2013). Previous studies have demonstrated that enriched or natural rearing environments can enhance exploratory behaviors and reduce stress responses in poultry (Bizeray *et al.*, 2002; Zahoor *et al.*, 2022). Although extensive research has been conducted on selenium supplementation in commercial broilers, limited attention has been given to the comparative effects of different selenium sources across diverse production systems and environmental conditions. Evidence remains scarce regarding the influence of selenium on the performance, health, and welfare of indigenous or backyard chicken breeds reared under semi-intensive or free-range conditions. In particular, studies focusing on Naked Neck chickens are lacking. Considering the constraints associated with conventional selenium sources in organic and free-range poultry systems, exploring alternative selenium-rich vegetables such as cabbage is essential. Such plant-based sources may not only provide organic selenium sustainably but also contribute additional functional and nutritional benefits. The present study was therefore designed to investigate how varying production systems and selenium-based feeding strategies affect the morphology, physiological traits, and immune responses of naked-neck chickens.

MATERIALS AND METHODS

The research was conducted at Avian Research and Training (ART), Centre, Department of Poultry Production, University of Veterinary and Animal Sciences, Ravi Campus, Pattoki, Pakistan, from March to July 2022. A total of 96 Naked Neck (6 weeks old) cockerels with an average body weight of 415 ± 28 g was reared and managed until 18 weeks of age. These experimental birds were allocated into 2 production systems (a) intensive and (b) free range, and four feeding strategies i.e., (a) commercial feed, (b) commercial feed with 0.3mg per kg Se from cabbage leaves, (c) commercial feed with 0.3mg per kg Se from sodium selenite (Sodium selenite feed grade, Chengou Sustar Feed Co., Ltd, China) and (d) commercial feed with 0.3mg per kg Se from cabbage+ sodium selenite source. Each treatment group contained 12 birds, distributed into 3 replicates of 4 birds each. The experimental treatments were organized in 2×4 factorial arrangements under a completely randomized design.

Bird's husbandry: The experimental birds were housed in well-ventilated facilities. In the intensive production system, the stocking density was initially maintained at 0.65 sq. ft per bird and gradually adjusted to a maximum of 1.5 sq. ft per bird with advancing age. For the free-range system, an outdoor area measuring 10m L $\times$ 2.99m W (providing 0.23m² per birds) was allocated in front of the house, however, the indoor facility measured 0.19m² (Ahmad *et al.* 2019). Clean drinking water was provided *ad libitum* throughout the experimental period. Birds had access to vegetation (spinach and lucerne) from 6:00 AM to 6:00 PM, which contributed approximately 75 % of their daily intake, while remaining 25% was met through commercial feed. Conversely, birds reared under the intensive system were fed exclusively with commercial feed supplemented with selenium additives. The composition and nutrient profile of the commercial feed are presented in Table 1 and 2.

Nutrient Profile of vegetables: The cabbage (*Brassica oleracea* L.), Spinach (*Spinacia oleracea* L.), and Lucerne (*Medicago sativa*) were grown on university premises. The proximate analysis of vegetables (cabbage, spinach, lucerne) was carried out as described by AOAC 2006, including moisture (Method No. 934-01) and ash contents (Method No. 942-05), crude fat (Method No. 920-39), protein (Method No. 984-13, and fiber (Method No. 978-10). The phenolic compounds were measured in vegetable extracts using the Folin-Ciocalteu reagent method as described by Mahboubi *et al.* (2015), and flavonoid contents were assessed by the method of Aluminum chloride colorimetric assay illustrated by

Mohammed and Manan (2015). Additionally, the quantification of phenolics and flavonoids using High Performance Liquid Chromatography (HPLC) (Shimadzu LC-20AC, Kyoto, Japan) was carried out following the procedures outlined by Sultana and Anwar (2008) and Sultana *et al.* (2012), respectively. Vitamin A and E were estimated in cabbage, spinach, and lucerne by implementing the technique of Siriamornpun *et al.* (2012) and Singh *et al.* (2007) respectively. Minerals like Ca, K, and Na were detected via Flame Photometer-410 (Sherwood Scientific Ltd., Cambridge, UK; method no 956.01) while Fe, Mg, Mn, Cu, and Zn were through Atomic Absorption Spectrophotometer (Hitachi Polarized Zeeman AAS, Z-8200, Japan; method no 975.03 and 991.11 (AOAC, 2006). For measuring the selenium content, the method described by Chatterjee *et al.* (2001) was adapted (Table 2).

Parameters studied

Morphometric traits: Male birds were assessed weekly for their morphometric features during the development phase (6-18 weeks). Data were collected regarding body, keel, beak, comb, drumstick and shank length, as well as drumstick and shank circumference, and wing spread (Ahmad *et al.* 2019) with the help of a measuring tape (FT-070, China).

Physiological Response: The cloacal temperature (CT) of 96 male birds was assessed by utilizing a digital clinical thermometer (Certeza, Model , FT-707), which was inserted to a depth of 3cm for a duration of 2 minutes.

The infrared digital thermometer (Model, PC868) was used to measure the external temperatures of the head (Th), back (Tb), wings (Tw), and shanks (Ts). The measurements were obtained without making any physical contact with the skin, maintaining a distance of around 15 cm from the bird's body. The equation proposed by Malheiros *et al.* (2000) was utilized to calculate the mean body surface temperature (Tms):

$$Tms = 0.03Th + 0.70Tb + 0.15Ts + 0.12Tw$$

Heart rate (beats per minute) was assessed through the utilization of a Littman stethoscope, with the time being recorded using a stopwatch (Mutibvu *et al.*, 2017). The respiratory rate (breath per minute) of the bird was assessed by immobilizing it in an inverted posture, and the movement of the abdomen during respiration was observed for a period of 1 minute (Mutibvu *et al.*, 2017).

Immune Response: Immune assessment status (Salmonella, ND, and IB) of the experimental birds (3 male birds per treatment), along with the weight of the thymus, bursa, and spleen were determined. Fifteen days before slaughtering, the birds were vaccinated against Newcastle Disease (ND) and infectious bronchitis (IB), followed by an assessment of antibody titers for both diseases (Xie *et al.*, 2008).

Statistical analysis: Collected data were analyzed through factorial ANOVA using general linear model (GLM) procedure in SAS software (version 9.1). Treatment means were subsequently compared utilizing Duncan's Multiple Range test (Duncan, 1955) at a significance level of $P \leq 0.05$.

RESULTS

Birds exhibited significant differences in morphometric traits ($P \leq 0.05$) across various production systems, feeding strategies and their interactions. Among production systems, higher morphometric traits including; wing spread ($P < 0.0001$), body ($P < 0.0001$), keel ($P < 0.0001$), comb ($P < 0.0001$), beak ($P < 0.0001$) length, shank length ($P < 0.0001$) and circumference ($P < 0.0001$), drumstick length ($P < 0.0001$) and circumference ($P < 0.0001$) were observed in intensive production system as compared to free range system. Regarding feeding strategies, higher values for morphometric traits including wing spread ($P < 0.0001$), body ($P < 0.0001$), keel ($P < 0.0001$), beak ($P = 0.0002$), tendency for comb length ($P = 0.0868$), shank length ($P < 0.0001$) and circumference ($P < 0.0001$), drumstick length ($P < 0.0001$) and circumference ($P < 0.0001$) were observed in 0.3 mg per kg selenium from cabbage feed treatment. In interaction between production systems and feeding strategies, higher morphometric traits were observed in birds fed 0.3 mg per kg selenium from cabbage leaves in an intensive production system (Table 4).

There were no significant differences ($P \leq 0.05$) in the physiological response of birds across various production systems. Regarding feeding strategies, higher heartbeat ($P = 0.0455$) and respiratory rates ($P < 0.0001$) were observed in birds fed 0.3 mg per kg selenium from sodium selenite source; however, no variation was observed in head, back, wing, shank, and total mean body temperatures. Regarding the interaction between production systems and feeding strategies, higher back and total mean temperatures were observed in birds fed 0.3 mg per kg selenium from cabbage+ sodium selenite feed treatment in the intensive production system. A higher respiration rate was observed in birds fed 0.3 mg per kg selenium form sodium selenite in an intensive production system (Table 5).

Birds showed significant ($P \leq 0.05$) difference regarding immune responses across various production systems, feeding strategies and their interaction. Regarding production systems, intensive production systems showed significantly higher titers of Newcastle ($P < 0.0001$) and infectious bronchitis ($P < 0.0001$) viruses compared to free-range production systems. In terms of feeding strategies, higher Newcastle disease ($P < 0.0001$) and infectious bronchitis ($P < 0.0001$) virus titers were observed in birds fed 0.3 mg per kg selenium from a cabbage source. Moreover, all samples

of birds showed an absence of *Salmonella Pullorum* and *Mycoplasma Gallisepticum*. There were no differences observed in thymus and spleen weights of birds across the various treatment groups. However, a notable distinction was observed ($P \leq 0.05$) in the bursa weight of birds across various feeding strategies. Higher bursa weight ($P=0.0008$) was observed in birds fed control feed and 0.3 mg per kg selenium from cabbage+ sodium selenite source feed treatment. In interaction between production systems and feeding strategies, higher bursa weight was observed in birds fed control and 0.3 mg per kg selenium from cabbage+ sodium selenite feed treatments. Moreover, lower spleen weight was observed in birds fed 0.3 mg per kg selenium from a sodium selenite source in free range production system. Furthermore, higher titers against ND and IB were observed in birds fed 0.3 mg per kg selenium from cabbage leaves in an intensive production system (Table 6).

Table 1. Ingredients of commercial control, cabbage, sodium selenite, and cabbage+ sodium selenite grower rations

Ingredients	Control feed	Cabbage feed ²	Sodium selenite feed	Cabbage + sodium selenite feed
Maize 12%	30	27.17	30	24
Cabbage	--	12.60	--	6.3
Rice Tips	25.87	16.89	25.87	24
Rice Polish	6.00	--	6.00	8
SBM 44%	7.15	17.54	7.15	7
Wheat Bran	7.43	--	7.43	9
Lime Stone	--	8.41	--	1
Rapeseed Meal	3.00	1.67	3.00	--
Canola Meal	10.00	--	10.00	10
Animal Protein Concentrate 52%	--	4.00	--	--
Fish Meal 43%	5.50	5.00	5.50	5
Feather Meal	4.25	--	4.25	4.2
Sunflower Meal	--	3.00	--	--
Poultry Oil	--	2.50	--	--
Bone Ash	--	0.423	--	--
Chips	0.43	--	0.43	--
Soda	--	0.250	--	--
Sodium Selenite	--	--	0.3	0.15
D. C. P	--	--	--	0.3
Lysine Sulphate 70%	--	--	--	--
DLM 99%	--	0.241	--	--
L-Valine	--	--	--	--
Vitamin +Mineral Premix ¹	0.310	0.100	0.310	1.2
Choline Chloride	--	0.050	--	--
Betain HCl	--	0.050	--	--
Iso-Leucine	--	0.000	--	--
L-Tryptophan	--	0.009	--	--
L-Threonine	--	--	--	--
Salt	0.037	0.058	0.037	--
Protech	--	0.010	--	--
AXTR XAP	--	0.010	--	--
Total	100			

¹ Provided per kilogram of premix: Vitamin A=1200000 IU, Vitamin D3=500000 IU, Vitamin E=8000, Vitamin B12=1700 mcg, Biotin=22000 mcg, Menadione=330 mg, Thiamine=400 mg, Riboflavin=860 mg, Pantothenic acid=2000 mg, Pyridoxine=430 mg, Niacin=6500 mg, Folic acid=220 mg, Choline chloride=60000 mg, Iron sulfate=6000 mg, Copper sulfate=1000 mg

²Phytase=0.005 %

Table 2. Calculated nutrient composition of commercial control, cabbage, sodium selenite, and cabbage+ sodium selenite grower rations

Nutrients	Control feed	Cabbage feed	Sodium selenite feed	Cabbage + sodium selenite feed
Dry Matter	89.99	89.73	89.99	89.50
Metabolizable Energy	2790	2805	2790	2795
Crude Protein	19.00	20.29	19.00	19.52
Ether Extract	4.54	6.04	4.54	5.15
Crude Fiber	4.85	4.11	4.85	5.99
Ash	5.22	11.84	5.22	5.07
Calcium	1.00	1.01	1.00	0.94
Phosphorus	0.40	0.40	0.40	0.44
Sodium	0.19	0.18	0.19	0.17
Potassium	0.54	0.57	0.54	0.55
Chlorine	0.18	0.19	0.18	0.19
DEB	170	170	170	170
L-Acid	1.69	1.73	1.69	1.70
Nitrogen Free Extract	48.50	47.45	48.50	47.95
Dig. Methionine	0.51	0.53	0.51	0.37
Dig. Cystine	0.21	0.25	0.21	0.20
Dig. Methionine+ Cystine	0.78	0.76	0.78	0.69
Dig. Lysine	0.84	0.84	0.84	0.92
Dig. Threonine	0.61	0.62	0.61	0.59
Dig. Tryptophan	0.20	0.21	0.20	0.20
Dig. Arginine	1.2	1.17	1.2	1.18
Dig. Isoleucine	0.70	0.68	0.70	0.71
Dig. Leucine	1.29	1.27	1.29	1.28
Dig. Valine	0.82	0.79	0.82	0.80
Dig. Histidine	0.42	0.40	0.42	0.41
Dig. Phenylalanine	0.79	0.77	0.79	0.78
Dig. Phenylalanine+ Tryptophan	1.35	1.31	1.35	1.32
Dig. Glycine	0.60	0.58	0.60	0.62
Dig. Glycine+ Serine	1.21	1.19	1.21	1.18

Table 3. Compositional profile of cabbage, spinach, and lucerne

Components	Nutrients	Cabbage (<i>Brassica oleracea</i> <i>var. capitata</i>)	Spinach (<i>Spinacia oleracea</i> L)	Lucerne (<i>Medicago sativa</i>)
Proximate (%)	Dry matter	8.06	6.79	18.15
	Ether extract	3.74	4.5	1.88
	Crude protein	24.05	32.61	25.20
	Ash	10.31	24.65	14.85
	Fiber	11.77	9.44	23.65
Vitamins (Cabbage and Spinach=Fresh Weight Basis; Lucerne=Dry Weight Basis)	Vitamin A	0.08 (mg/100g)	137.07 (mcg/100g)	26 (mg/kg)
	Vitamin E	0.21 (mg/100g)	0.54 (mg/100g)	21 (mg/kg)
Antioxidants (cabbage=mg/100g fresh weight; Spinach and lucerne =TPC (mg GAE/g), TFC (mg QE/g))	Total Phenolic Content	55.21	39.12	0.80
	Total Flavonoid Content	32.35	21.05	0.065

High-Performance Liquid Chromatography (Phenolics and Flavonoids mg/g)	Gallic acid	0.187	0.148	0.175
	Chlorogenic acid	1.082	1.052	0.685
	Caffeic acid	0.165	--	0.537
	Sinapic acid	0.016	--	0.027
	Myricetin	1.056	0.816	--
	Quercetin	1.438	0.599	0.147
	Kaempferol	0.966	0.453	--
Minerals (cabbage and spinach=mg/100g fresh weight basis, lucerne=mg/kg dry weight basis)	Potassium	52.22	152.22	1.58 (g/100g)
	Calcium	19.55	27.55	3.25 (g/100g)
	Magnesium	21.55	20.35	4.35
	Sodium	9.65	17.65	0.78 (g/100g)
	Iron	0.70	0.72	89.15
	Zinc	0.30	0.12	70.25
	Copper	0.05	0.03	2.05
	Manganese	0.11	0.18	17.88
	Selenium (mg/kg D.M)	0.095	0.082	0.065

Table 4. Effect of feeding strategies on male morphometric traits of Naked Neck chicken among different production systems.

Treatment		Morphometric traits (cm)								
		WS	CL	BL	SL	SC	DL	DC	BDL	KL
Production systems										
Intensive		9.69 ^a	3.53 ^a	3.06 ^a	9.17 ^a	4.05 ^a	11.55 ^a	7.47 ^a	62.24 ^a	11.22 ^a
Free-range		9.12 ^b	3.35 ^b	2.77 ^b	8.51 ^b	3.79 ^b	10.60 ^b	7.08 ^b	58.81 ^b	10.13 ^b
Feeding strategies										
Control feed		9.47 ^b	3.45 ^{ab}	2.94 ^{ab}	8.97 ^b	3.99 ^b	11.09 ^b	7.25 ^b	61.01 ^b	10.78 ^b
Cabbage feed		9.84 ^a	3.47 ^a	3.07 ^a	9.32 ^a	4.12 ^a	11.63 ^a	7.64 ^a	62.95 ^a	11.25 ^a
Sodium selenite feed		9.04 ^c	3.35 ^b	2.73 ^c	8.49 ^c	3.82 ^c	10.56 ^c	7.11 ^c	59.10 ^c	10.33 ^c
Cabbage + sodium selenite feed		9.26 ^{bc}	3.48 ^a	2.91 ^b	8.58 ^c	3.74 ^c	11.00 ^b	7.10 ^c	59.02 ^c	10.33 ^c
Production systems × Feeding strategies										
Intensive	Control feed	9.82 ^b	3.58 ^{ab}	3.06 ^b	9.18 ^b	4.06 ^b	11.58 ^b	7.46 ^b	62.84 ^b	11.16 ^b
	Cabbage feed	10.36 ^a	3.64 ^a	3.34 ^a	9.74 ^a	4.28 ^a	12.24 ^a	8.12 ^a	66.32 ^a	11.92 ^a
	Sodium selenite feed	9.08 ^d	3.38 ^{cd}	2.86 ^{bc}	8.90 ^c	3.86 ^{cd}	11.06 ^c	7.20 ^c	60.10 ^c	10.94 ^b
	Cabbage + sodium selenite feed	9.50 ^{bc}	3.50 ^{abc}	2.96 ^{bc}	8.86 ^c	4.00 ^{bc}	11.30 ^{bc}	7.10 ^{cd}	59.68 ^{cd}	10.86 ^{bc}
Free-range	Control feed	9.12 ^d	3.32 ^d	2.82 ^c	8.76 ^c	3.92 ^{bcd}	10.60 ^c	7.04 ^{cd}	59.18 ^{cde}	10.40 ^d
	Cabbage feed	9.32 ^{cd}	3.30 ^d	2.80 ^c	8.90 ^c	3.96 ^{bc}	11.02 ^{cd}	7.16 ^{cd}	59.58 ^{cd}	10.58 ^{cd}
	Sodium selenite feed	9.00 ^d	3.32 ^d	2.60 ^d	8.08 ^d	3.78 ^d	10.06 ^f	7.02 ^d	58.10 ^e	9.72 ^e
	Cabbage + sodium selenite feed	9.02 ^d	3.46 ^{bcd}	2.86 ^{bc}	8.30 ^d	3.48 ^c	10.70 ^{de}	7.10 ^{cd}	58.36 ^{de}	9.80 ^e
SEM		0.08	0.03	0.04	0.08	0.04	0.11	0.06	0.44	0.11
ANOVA										
Production systems		<.0001	<.0001	<.0001	<.0001	<.0001	<.0001	<.0001	<.0001	<.0001
Feeding strategies		<.0001	0.0868	0.0002	<.0001	<.0001	<.0001	<.0001	<.0001	<.0001
Production systems × Feeding strategies		0.0021	0.0217	0.0167	0.076	0.0009	0.0666	<.0001	<.0001	0.0594

^{a-f}Superscripts on different means within column differ significantly at $P \leq 0.05$

Abbreviations: WS=Wing Spread; CL=Comb length; BL=Beak length; SL=Shank Length; SC=Shank Circumference; DL=Drumstick length; DC=Drumstick Circumference; BDL=Body Length; KL=Keel length

Table 5. Effect of feeding strategies on the male physiological response of Naked Neck chicken among different production system.

Treatment		Temperature (°F)					Rate (bpm)	
		HT	BT	WT	ST	TMT	HB	RR
Production systems								
Intensive		104.90	104.96	104.90	104.54	104.88	247.25	32.70
Free-range		104.98	104.91	104.99	105.03	104.93	248.85	32.15
Feeding strategies								
Control feed		104.76	104.83	104.97	105.05	104.88	244.70 ^b	32.60 ^b
Cabbage feed		105.07	104.81	104.72	104.67	104.79	245.60 ^b	31.10 ^c
Sodium selenite feed		104.86	104.83	105.06	104.85	104.86	260.40 ^a	34.00 ^a
Cabbage + sodium selenite feed		105.07	105.25	105.01	104.56	105.11	241.50 ^b	32.00 ^{bc}
Production systems × Feeding strategies								
Intensive	Control feed	104.76	104.80 ^b	105.02	104.74	104.82 ^{ab}	241.60	32.60 ^{bc}
	Cabbage feed	104.98	104.60 ^b	104.68	104.62	104.62 ^b	247.00	31.60 ^{cd}
	Sodium selenite feed	104.84	104.74 ^b	105.12	104.68	104.78 ^{ab}	260.80	34.40 ^a
	Cabbage + sodium selenite feed	105.02	105.68 ^a	104.76	104.12	105.32 ^a	239.60	32.20 ^{cd}
Free-range	Control feed	104.76	104.86 ^b	104.92	105.36	104.94 ^{ab}	247.80	32.60 ^{bc}
	Cabbage feed	105.16	105.02 ^b	104.76	104.72	104.95 ^{ab}	244.20	30.60 ^d
	Sodium selenite feed	104.88	104.92 ^b	105.00	105.02	104.94 ^{ab}	260.00	33.60 ^{ab}
	Cabbage + sodium selenite feed	105.12	104.82 ^b	105.26	105.00	104.91 ^{ab}	243.40	31.80 ^{cd}
SEM		0.07	0.09	0.07	0.14	0.06	2.52	0.25
ANOVA								
Production systems		0.6029	0.7547	0.5433	0.0950	0.6860	0.7446	0.1513
Feeding strategies		0.3866	0.1649	0.3775	0.6323	0.2994	0.0455	<.0001
Production systems × Feeding strategies		0.9775	0.0397	0.4215	0.7821	0.2054	0.9094	0.7885

^{a-d} Superscript on different means within column differ significantly at $P \leq 0.05$

Abbreviations: HT = Head Temperature; BT = Back Temperature; WT = Wing Temperature; ST = Shank Temperature; TMT = Total Mean Temperature; HB = Heart Beat; RR = Respiration Rate

Table 6. Effect of feeding strategies on male immune response of Naked Neck chicken among different production system.

Treatment		Weight (g)			ND	IB	SP	MG
		Thymus	Bursa	Spleen				
Production systems								
Intensive		1.85	0.75	2.68	4.63 ^a	3476.67 ^a	Negative	Negative
Free-range		1.77	0.77	2.54	4.33 ^b	3345.25 ^b	Negative	Negative
Feeding strategies								
Control feed		1.63	0.83 ^a	2.75	4.63 ^b	3428.83 ^b	Negative	Negative
Cabbage feed		1.97	0.70 ^b	2.62	4.86 ^a	3553.17 ^a	Negative	Negative
Sodium selenite feed		1.98	0.62 ^b	2.55	4.18 ^c	3300.50 ^c	Negative	Negative
Cabbage + sodium selenite feed		1.65	0.88 ^a	2.52	4.24 ^c	3361.33 ^c	Negative	Negative
Production systems × Feeding strategies								
Intensive	Control feed	1.63	0.73 ^b	2.73 ^a	4.78 ^b	3504.00 ^b	Negative	Negative
	Cabbage feed	2.07	0.67 ^b	2.77 ^a	5.23 ^a	3670.67 ^a	Negative	Negative
	Sodium selenite feed	1.97	0.77 ^{ab}	2.73 ^a	4.22 ^c	3307.00 ^d	Negative	Negative
	Cabbage + sodium selenite feed	1.73	0.83 ^{ab}	2.47 ^{ab}	4.29 ^{cde}	3425.00 ^{bc}	Negative	Negative
Free-range	Control feed	1.63	0.93 ^a	2.77 ^a	4.49 ^c	3353.67 ^{cd}	Negative	Negative
	Cabbage feed	1.87	0.73 ^b	2.47 ^{ab}	4.48 ^{cd}	3435.67 ^{bc}	Negative	Negative
	Sodium selenite feed	2.00	0.47 ^c	2.37 ^b	4.14 ^c	3294.00 ^d	Negative	Negative
	Cabbage + sodium selenite feed	1.57	0.93 ^a	2.57 ^{ab}	4.19 ^c	3297.67 ^d	Negative	Negative
SEM		0.06	0.03	0.04	0.08	26.81		

ANOVA							
Production systems	0.4569	0.6822	0.0870	<.0001	<.0001	-	-
Feeding strategies	0.0560	0.0008	0.1559	<.0001	<.0001	-	-
Production systems × Feeding strategies	0.2244	0.0005	0.0005	0.0029	0.0172	-	-

^{a-c} Superscript on different means within column differ significantly at $P \leq 0.05$

Abbreviations: ND = Newcastle Disease; IB = Infectious Bronchitis; SP = Salmonella pullorum; MG = Mycoplasma gallisepticum

DISCUSSION

The current study's findings demonstrated variations in morphometric traits, physiological and immune responses in Naked Neck chickens across various production systems and feeding approaches. Regarding morphometric traits of birds, it was observed that those reared under the intensive production system exhibited higher body measurements compared to those in the free-range system. This difference can be attributed not only to the improved management practices and controlled environmental conditions in the intensive system but also to the provision of a balanced and nutritionally complete diet, which supports better growth and body development than the variable and often limited feed resources available in free-range conditions. Ahmad *et al.* (2019) also observed notable differences in body size weight, and keel length among crossbred chickens raised in various housing environments. Contrarily, Campbell *et al.* (2017) found that the foraging opportunities were more limited in the intensive system in comparison to the free-range system. Another study reported that birds raised under semi-intensive production systems showed a steady growth in body morphometrics due to their access to both formulated feed and natural foraging resources, which together ensure a balanced nutrient intake and promote uniform body development (Nandi *et al.*, 2017). Concerning feeding strategies, higher body measurements including wing spread, drumstick length and circumference, body and keel length were observed in birds fed 0.3 mg per kg selenium from cabbage leaves. This may be due to the presence of a certain proportion of organic selenium in cabbage, which gave protection from damage during excess production of free radicals, hence a better utility of nutrients. Similar observations have been reported by Khan *et al.* (2022), who found improved performance in birds receiving organic selenium sources. Furthermore, organic selenium including plant derived and selenomethionine forms, has been shown to enhance antioxidant enzyme activity (e.g., GPx), tissue selenium deposition, and growth or carcass traits compared with inorganic selenium, thereby supporting superior somatic development (Surai *et al.*, 2017). Others reported no significant variations in shank length among the different feeding regimes (Mutayoba *et al.*, 2012). Moreover, selenium deficiency is directly associated with the overall performance of animals (Weiss *et al.*, 1990). According to a study conducted by Boostani *et al.* (2015), it was found that the growth performance of poultry could be influenced by selenium. In the interaction between production systems and feeding strategies, birds fed selenium from cabbage leaves in an intensive system exhibited higher morphometric traits, likely due to the controlled conditions and optimized nutrition that enhanced nutrient utilization; additionally, selenium improved the antioxidant status and growth of these birds. These findings align with Khan *et al.*, (2025), who reported higher performance in naked neck chickens under controlled housing with selenium-supplemented diets. The physiological response of male birds did not differ significantly among production systems. A contradictory finding was reported by Yakubu *et al.* (2018), housing systems affect the physiological traits of Sasso laying hens. Concerning the feeding strategies, present results showed significantly higher heartbeat and respiratory rates in male birds that were fed on 0.3 mg per kg selenium from sodium selenite. This may be attributed to the insufficient selenium in the diet, as selenite can convert into a volatile form and evaporate, reducing its availability for absorption by the body. Furthermore, the research carried out by Attia *et al.* (2017), manipulating the composition of feed had a significant impact on the physiological characteristics of broiler chickens.

Regarding immune response, birds from intensive production systems showed higher titer against Newcastle disease and Infectious Bronchitis virus when compared to free range. This may be due to the availability of controlled environmental conditions in intensive production system, most probably due to reduced ambient stress in intensive system. In contrast, in a previous study, the free range system exhibited a notably elevated antibody titer against the Newcastle disease virus (Shini, 2003). Another study revealed a reduction in antibody levels against the Newcastle disease virus in the intensive farming system, which was attributed to the stocking density of the birds (Buijs *et al.*, 2009). Similarly, Kucukyilmaz *et al.* (2012) reported differences in antibody generation across various housing environments. In terms of feeding strategies, higher Newcastle disease, and infectious bronchitis virus titer were observed in birds reared on 0.3 mg per kg selenium from cabbage source feeding treatment. The potential reason behind this occurrence could be attributed to the functional characteristic of cabbage, which can regulate the detrimental impact of free radicals utilizing non-enzymatic and enzymatic antioxidant defense mechanisms. Kieliszek and Blazejak (2016)

reported that the selenium diet directly increases the activity of the immune system. Hegazy and Adachi, (2000) also reported that the supplemented selenium had a positive response toward the formation of antibodies against Newcastle disease in chicken as selenium can merge with selen-o-protein and trigger the immune system (Hoffmann, 2007). Salmonella Pullorum and Mycoplasma Gallisepticum did not show any variations among production systems and feeding strategies. Raza *et al.* (2018) also reported that feeding regimes did not impact any variation regarding antibody titers against *Mycoplasma gallisepticum*, Infectious Bronchitis, and Newcastle disease. Rehman *et al.* (2017) also reported that Salmonella Pullorum and Mycoplasma Gallisepticum did not differ among different production systems. Regarding interactions, the higher ND and IB titers observed in birds fed selenium from cabbage leaves under an intensive system may be due to better nutrient availability and enhanced antioxidant status, supporting stronger immune responses.

Conclusions: The findings presented above lead to the conclusion that the Naked Neck male chicken performed better regarding morphometrics traits as well as a physiological and immune response under an intensive production system with 0.3 mg per kg selenium from the cabbage source.

Author's contribution: UK conducted this study as part of her Ph.D. research work under the supervision of JH, AM, and AK. UK did the data collection and write the manuscript. JH helped in the statistical analysis and formatting of the manuscript. JH, AM, and AK helped in reviewing the manuscript. All authors read and approve the final manuscript.

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