

Research Article



Effects of Mesenteric Injection on the Physiological Traits of Broiler Chickens During the Hatching Window

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Abstract | This study aimed to evaluate the effects of mesenteric injection and administration method during the hatching window on early physiological responses and lipid metabolism in broiler chickens. A total of 600 one-day-old ROSS 308 chicks were allocated to a 2 × 5 factorial design, including two hatching windows (before and after 493 h) and five administration methods (mesenteric or intra-crop delivery of electrolytes or water, and a non-treated control). Blood lipid-related metabolites were assessed at 1 and 11 days of age. At hatch, a significant ($P < 0.05$) interaction between hatching window and administration method influenced serum lipid profiles. Water gavage resulted in higher glucose, triglycerides, very low-density lipoproteins (VLDL), and low-density lipoproteins (LDL) concentrations, indicating rapid post-hatch energy mobilization, whereas mesenteric electrolyte injection reduced undesirable fat indicators, reflecting improved ionic balance and metabolic regulation. The hatching window alone had no significant independent effect. By day 11, glucose levels were unaffected, while cholesterol fractions and triglycerides responded significantly ($P < 0.05$) to treatments. Electrolyte administration increased high-density lipoprotein (HDL) levels and reduced triglycerides (TG) and VLDL concentrations, suggesting enhanced lipid redistribution and metabolic stability during early growth. Overall, the findings demonstrate that the method of early post-hatch administration plays a more decisive role than hatching window in regulating lipid metabolism, with potential implications for improving metabolic health and early growth efficiency in broiler chickens.

Keywords | Mesenteric injection, Early post-hatch period, Lipid metabolism, Serum biochemical indices, Broiler chickens

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INTRODUCTION

Variation in hatching time among broiler chicks represents one of the major challenges in modern broiler production systems, as it directly influences early chick physiology, uniformity, health status, and subsequent productive performance (Such *et al.*, 2023). This phenomenon, commonly referred to as the ‘hatching window’, describes the time span between the emergence of

the first and last chicks within the same hatch and typically ranges from 24 to 36 hours (Boyner, 2023; Zuidhof, 2022). Several factors contribute to the width of the hatching window, including breeder age, egg storage duration, egg weight, incubation conditions, and incubator type, all of which interact to affect embryonic development and hatch synchrony (Abioja *et al.*, 2023; van de Ven *et al.*, 2021).

Chicks that hatch earlier within the hatching window often

experience prolonged fasting, dehydration, and delayed access to feed and water before placement, whereas late-hatched chicks may suffer from incomplete physiological maturation. These disparities result in uneven chick quality, impaired immune and metabolic development, and reduced flock uniformity, ultimately leading to poorer growth performance and higher early mortality rates (Kalaba *et al.*, 2023). The negative effects of delayed post-hatch nutrient and water access are particularly critical during the first days of life, a period characterized by rapid metabolic adaptation and high energy demand (Kalaba *et al.*, 2023).

Electrolyte balance plays a pivotal role in maintaining osmotic pressure, acid–base equilibrium, and cellular metabolism in newly hatched chicks. Disruptions in electrolyte homeostasis during the early post-hatch period can exacerbate dehydration, compromise nutrient absorption, and increase physiological stress (Sigolo *et al.*, 2021; Araujo *et al.*, 2022). Consequently, most nutritional interventions have focused on supplementing electrolytes through feed or drinking water. However, feed intake is typically low during the first week post-hatch, limiting the effectiveness of oral electrolyte supplementation and potentially delaying physiological recovery (Mgs *et al.*, 2023; Oketch *et al.*, 2023).

In this context, alternative early post-hatch intervention strategies that ensure rapid electrolyte availability may offer a valuable approach to support metabolic stability and early physiological adaptation (Jha *et al.*, 2019; Dang *et al.*, 2022). Mesenteric injection represents a controlled experimental method that enables direct delivery of electrolytes into the portal circulation, bypassing gastrointestinal limitations and allowing precise evaluation of early metabolic responses. Despite its experimental nature, this approach provides critical insights into the mechanisms underlying electrolyte regulation, lipid metabolism, and energy partitioning during the hatching window.

Therefore, the present study aimed to investigate the effects of mesenteric electrolyte injection at different hatching window intervals on early physiological traits and serum lipid metabolism in broiler chickens. By elucidating the interaction between hatching time and administration method, this study seeks to improve the understanding of early post-hatch metabolic regulation and to provide a physiological basis for developing practical nutritional and management strategies in broiler production.

MATERIALS AND METHODS

ELECTROLYTIC SOLUTION

An electrolytic solution (ES) was prepared according to

Imtiaz *et al.* (2023) by dissolving 3.5 g sodium chloride (NaCl), 1.5 g potassium chloride (KCl), and 2.9 g sodium citrate ($\text{Na}_3\text{C}_6\text{H}_5\text{O}_7$) in 100 mL of deionized water to obtain a total normality of 1.1372 N. The prepared solution was sterilized by autoclaving and stored under sterile conditions for 48 h prior to use. The same ES concentration was used for both mesenteric injection and intra-crop gavage. For administration, a sterile plastic tube connected to a 0.5 mL syringe was used to ensure accurate dosage and uniform delivery among chicks.

EXPERIMENTAL DESIGN

All experimental procedures were carried out in line with Animal Ethical Standards after getting institutional approval. A total of 600 one-day-old unsexed ROSS 308 broiler chicks (initial body weight 45 ± 1.5 g) were used in this experiment. ROSS 308 is a fast-growing commercial broiler strain, widely used in intensive poultry production systems due to its high growth rate, efficient feed conversion, and suitability for meat production (Aviagen, 2022). The experiment was arranged in a 2×5 factorial design, comprising:

FACTOR A (HATCHING WINDOW)

- Chicks hatched before 493 h of incubation
- Chicks hatched after 493 h of incubation

FACTOR B (ADMINISTRATION METHOD)

- Mesenteric injection with 0.5 mL electrolytic solution
- Mesenteric injection with 0.5 mL deionized water
- Intra-crop gavage with 0.5 mL electrolytic solution
- Intra-crop gavage with 0.5 mL deionized water
- Non-treated control (no injection or gavage)

Each treatment consisted of three replicates, with 20 chicks per replicate (60 chicks per treatment). Feed and water were provided *ad libitum*, and lighting and management practices were applied according to the ROSS 308 broiler management guide (Aviagen, 2022). The experimental diets (starter, grower, and finisher) were formulated to meet the nutritional requirements recommended for ROSS 308 broilers and are presented in Table 1 (Aviagen, 2022).

The 493 h incubation threshold was selected as a biological reference point separating early- and late-hatching chicks, consistent with previous studies indicating that chicks hatched earlier within the hatching window experience longer post-hatch fasting and dehydration compared to late-hatched chicks, leading to differences in metabolic adaptation and physiological status (van de Ven *et al.*, 2021; Such *et al.*, 2023).

Table 1: The diets used in the experiment.

Ingredients	Experimental diets (%)		
	Starter (1-10 days)	Grower (11-21 days)	Finisher (22-35 days)
Corn	52.8	58.65	62.4
Wheat	10	10	10
Soybean (48% protein)	29.8	24	20.5
Protein concentrate*	5	5	5
Sunflower oil	0.3	0.3	0.3
Limestone	1.14	1.21	1.22
Dicalcium phosphate	0.5	0.35	0.2
Methionine	0.17	0.17	0.13
Lysine	0.19	0.22	0.15
Salt (NaCl)	0.1	0.1	0.1
Total	100	100	100
Calculated chemical composition**			
Crud protein (%)	21.94	19.66	18.29
ME (Kcal/kg feed)	2940	2995	3035
Lysine (%)	1.39	1.26	1.11
Crude fibre (%)	2.73	2.64	2.58
Calcium (%)	0.9	0.88	0.83
Meth + Cys (%)	1.03	0.97	0.9
Available phosphor	0.44	0.41	0.38

*The protein concentrate utilized in the present study was of the Brocon type, originating from the Netherlands. Each kilogram of the concentrate provides 40% crude protein, 5% crude fat, 2.20% crude fiber, 5% calcium, 4.68% available phosphorus, 3.85% lysine, 4.12% methionine + cystine, 0.42% tryptophan, 2017 kcal/kg of metabolizable energy, and 1.70% threonine. Moreover, it contains a balanced mixture of vitamins and trace minerals formulated to meet the nutritional requirements of poultry for these essential elements. **The chemical composition of the diet was determined in accordance with the NRC (1994) standards.

BLOOD COLLECTION AND ANALYSIS

Blood samples were collected at 1 and 11 days of age. Two birds were randomly selected from each replicate, ensuring that their live weights were as close as possible to the replicate mean. Prior to sampling, birds were deprived of feed for 24 h while water was provided ad libitum.

Each bird was weighed and slaughtered by severing the jugular vein, allowing complete bleeding in a vertical position. Approximately 2 mL of blood was collected into sterile glass tubes (Bijou bottles). Serum biochemical analyses were conducted following standard procedures described by Kohn and Allen (1995) and Mouldin *et al.* (1996).

STATISTICAL ANALYSIS

To assess the significance of the main factors and their interactions on the selected parameters, percentage data were first transformed using the arcsine transformation. The transformed data were then analyzed using two-way analysis of variance (ANOVA). Statistical analyses were performed using the General Linear Model (GLM) procedure of SAS software (version 9.1; SAS Institute, 2004). Differences were considered significant at $P < 0.05$, $P < 0.01$, and $P < 0.001$. Results are presented as pooled means $\pm$ SEM.

RESULTS AND DISCUSSIONS

EFFECT OF INJECTION TYPE AND CHICK REMOVAL TIME WITHIN THE HATCHING WINDOW ON SERUM LIPID PARAMETERS OF BROILER CHICKS AT 1-DAY OF AGE

Table 2 shows that the interaction between chick removal time within the hatching window and administration method had a significant ($P < 0.05$ or < 0.001 or < 0.0001) effect on most serum lipid traits at one day of age. In contrast, the hatching window factor alone did not exert a significant independent effect on any of the measured parameters, indicating that early metabolic responses were primarily driven by the method of administration rather than hatch timing.

A significant ($P < 0.0001$) interaction effect was observed for serum glucose (GLU) concentration. Chicks receiving water via intra-crop gavage, regardless of hatch timing, exhibited the highest GLU levels, reflecting rapid post-hatch energy mobilization. In contrast, control birds showed the lowest values. When administration methods were considered independently, water gavage resulted in significantly higher GLU concentrations than all other treatments.

This response may be attributed to improved hydration status and enhanced intestinal glucose absorption following water administration, which supports early metabolic adaptation after hatch (Shibata *et al.*, 2023). Electrolyte-based treatments showed intermediate GLU values, suggesting that ionic balance supports glucose utilization without inducing excessive hyperglycemia. Differences among studies may reflect variation in administration route, electrolyte composition, and chick physiological status (Pawłowska *et al.*, 2022; Mohamed *et al.*, 2025).

Serum cholesterol (CHOL) concentration was significantly ($P < 0.05$) affected by the interaction between hatch timing and administration method. Water gavage treatments generally resulted in higher CHOL values, whereas mesenteric electrolyte injection produced the lowest concentrations. When considered as a main effect, administration method significantly influenced CHOL, while hatch timing alone did not.

Table 2: Effect of injection type and chick removal time in the hatching window on some serum lipid parameters of broiler chicks at one day of age.

Treatments ⁽¹⁾		GLU (mg/100 ml)	CHOL (mg/100 ml)	HDL (mg/100 ml)	TG (mg/100 ml)	VLDL (mg/100 ml)	LDL (mg/100 ml)
Hatching windows factor							
Before 493 h		132.31	170.95	89.81	103.28	20.65	60.48
After 493 h		134.26	175.64	102.17	103.08	20.61	52.85
Probability		0.2193	0.5411	0.3013	0.9793	0.9793	0.3552
Administration factor							
Control		110.61 d	177.20 ab	105.00	96.87 bc	19.37 bc	52.82 b
MI with H ₂ O		116.27 c	160.61 b	103.18	166.14 a	33.22 a	24.21 c
MI with ES		127.74 b	158.00 b	80.40	81.33 cd	16.26 cd	61.34 b
Intra-crop with H ₂ O		184.39 a	189.79 a	83.30	61.79 d	12.35 d	94.14 a
Intra-crop ES		127.43 b	180.89 ab	108.07	109.80 b	21.96 b	50.86 b
Probability		0.0001	0.05	0.4271	0.0001	0.05	0.0006
Interaction (hatching windows × administration)							
Before 493 h	Control	104.80 f	194.19 a	96.93	95.07 bc	19.01 bc	78.24 abc
	MI with H ₂ O	122.80 cd	157.00 ab	100.00	132.27 b	26.45 b	30.55 d
	MI with ES	126.92 bc	147.81 b	77.63	111.73 b	22.35 b	47.83 bcd
	Intra-crop with H ₂ O	183.78 a	190.32 a	87.65	68.67 cd	13.73 cd	88.94 ab
	Intra-crop ES	123.28 cd	165.44 ab	86.84	108.67 b	21.73 b	56.87 bcd
After 493 h	Control	116.41 de	160.20 ab	113.07	98.67 bc	19.73 bc	27.40 d
	MI with H ₂ O	109.75 ef	164.22 ab	106.36	200.00 a	40.00 a	17.86 d
	MI with ES	128.57 bc	168.19 ab	83.16	50.93 d	10.18 d	74.84 abc
	Intra-crop with H ₂ O	185.01 a	189.27 a	78.94	54.90 d	10.98 d	99.34 a
	Intra-crop ES	131.59 b	196.33 a	129.30	110.93 b	22.19 b	44.85 cd
Probability		0.0001	0.05	0.6131	0.0001	0.0001	0.0018
Total mean		133.29	173.29	95.98	103.18	20.63	56.67
SEM ⁽²⁾		5.0063	4.3986	5.6489	8.1835	1.6367	5.9219

⁽¹⁾ Experimental Design: The hatching window (prior to 493 hours and after 493 hours) was considered the first factor, whereas the injection treatment (control, mesenteric water, mesenteric electrolyte, oral water, oral electrolyte) was regarded as the second factor. MI: Mesenteric injection; H₂O: Ionized water; ES: Electrolytic solution. ⁽²⁾ SEM: Values are presented as the Standard Error of the Mean. Note: Distinct letters within the same column denote statistically significant differences among treatment means at the 0.05 and 0.01 probability levels.

Elevated CHOL levels in water-gavaged chicks may reflect limited hepatic regulation of lipid metabolism during the immediate post-hatch period. Conversely, electrolyte injection likely improved osmotic balance and hepatic lipid handling, facilitating cholesterol binding and clearance via lipoproteins, thereby reducing circulating CHOL (Jassim and Aqeel, 2017; Kalaba *et al.*, 2023).

Neither the main factors nor their interaction significantly ($P > 0.05$) affected high-density lipoprotein (HDL) concentrations at one day of age. This finding suggests that HDL metabolism may be less responsive to early post-hatch interventions and more dependent on longer-term nutritional and metabolic regulation, consistent with previous reports (Pawłowska *et al.*, 2022; Mohamed *et al.*,

2025).

A significant ($P < 0.0001$) interaction effect was detected for triglycerides (TG) and very low-density lipoprotein (VLDL) concentrations. Water gavage, particularly in chicks removed after 493 h, resulted in the highest values, whereas mesenteric electrolyte injection produced the lowest. As a main effect, administration method significantly influenced both traits, while hatch timing alone had no independent impact.

Higher TG and VLDL levels following water gavage may indicate enhanced fat absorption and rapid lipid mobilization to meet early energy demands. In contrast, electrolyte injection appeared to regulate lipid metabolism

more efficiently, reducing excessive triglyceride synthesis and transport. Such reductions are considered physiologically favorable, as elevated TG and VLDL are associated with increased fat deposition and metabolic stress in broilers (Safiyu *et al.*, 2023).

Serum low-density lipoprotein (LDL) concentration was significantly ($P = 0.0018$) influenced by the interaction between hatch timing and administration method. Water gavage treatments resulted in higher LDL levels, whereas mesenteric water and electrolyte injections yielded lower values. As a main effect, administration method significantly affected LDL concentration, while hatch timing alone did not.

Increased LDL levels following water gavage may reflect enhanced lipid transport to peripheral tissues to meet early energy requirements. Conversely, electrolyte injection may have supported a more balanced lipid distribution by improving osmotic pressure and hepatic lipid metabolism, thereby limiting excessive LDL accumulation (Shihab *et al.*, 2019; Oketch *et al.*, 2023).

Collectively, these findings indicate that early post-hatch metabolic responses in broiler chicks are predominantly determined by the administration method rather than by the hatching window alone. Water gavage promoted rapid energy mobilization but was associated with elevated lipid fractions, whereas mesenteric electrolyte injection contributed to improved metabolic regulation by reducing undesirable fat indicators. This interaction highlights the importance of early intervention strategies in shaping physiological adaptation during the first day after hatch.

EFFECT OF INJECTION TYPE AND CHICK REMOVAL TIME WITHIN THE HATCHING WINDOW ON SERUM LIPID PARAMETERS OF BROILER CHICKS AT 11 DAYS OF AGE

Table 3 indicates that neither the hatching window, the administration method, nor their interaction significantly ($P > 0.05$) affected serum GLU concentration at 11 days of age. This stability in GLU levels reflects the strict physiological regulation of glucose metabolism in poultry, which is primarily controlled by glucagon rather than insulin, maintaining blood glucose within a narrow range despite variations in early post-hatch interventions. Similar findings were reported by Chen *et al.* (2022) and Ölmez *et al.* (2023), whereas discrepancies with other studies may be attributed to differences in bird age, electrolyte composition, or environmental and nutritional conditions (Chen *et al.*, 2024; Yu *et al.*, 2022).

In contrast, serum CHOL concentration was significantly ($P < 0.05$) influenced by the interaction between hatching window and administration method. Chicks removed

before 493 h and subjected to mesenteric electrolyte injection exhibited the highest CHOL values, while mesenteric water injection resulted in the lowest levels. When considered independently, administration method had a significant effect on CHOL, whereas the hatching window alone did not.

This response can be explained by the direct delivery of electrolytes into the portal-hepatic circulation via mesenteric injection, allowing rapid hepatic uptake of sodium, potassium, and citrate ions. Citrate serves as a key precursor for acetyl-CoA production through ATP-citrate lyase, thereby enhancing cholesterol biosynthesis during periods of elevated hepatic metabolic activity (Feng *et al.*, 2020; Duarte Lau and Giugliano, 2023). Moreover, early chick removal coincides with increased hepatic lipid redistribution to support early growth, which may have amplified the observed interaction. Similar findings were reported by Sigolo *et al.* (2021), while contrasting results in other studies likely reflect differences in electrolyte balance and experimental conditions (Yu *et al.*, 2022; Ölmez *et al.*, 2023).

A non-significant ($P = 0.5$) interaction effect was also observed for HDL. Oral electrolyte administration, irrespective of hatch timing, resulted in the highest HDL concentrations, whereas oral water treatments produced the lowest values. The hatching window alone did not exert a significant independent effect.

The increase in HDL following electrolyte administration may be attributed to improved acid-base balance and enhanced hepatic activity of lecithin-cholesterol acyltransferase (LCAT), which facilitates cholesterol esterification and reverse cholesterol transport. These findings are consistent with previous reports indicating improved HDL synthesis under optimized electrolyte conditions (Sigolo *et al.*, 2021; Manthei *et al.*, 2018, 2020).

Similarly, TG and VLDL concentrations were significantly ($P = 0.0033$) affected by the interaction between the two factors. Higher TG and VLDL values were observed in control and oral water treatments, whereas mesenteric electrolyte injection consistently resulted in the lowest concentrations. Administration method was the dominant factor influencing these traits, while the hatching window alone had no significant effect.

Reduced TG and VLDL levels following mesenteric electrolyte injection indicate improved lipid metabolic regulation and reduced hepatic lipid burden. Elevated TG and VLDL are generally considered undesirable indicators in poultry, as they are associated with excessive fat deposition and metabolic stress. Therefore, the observed reduction following electrolyte injection may be regarded

Table 3: Effect of injection type and chick removal time in the hatching window on some serum lipid parameters of broiler chicks at 11 days of age.

Treatments ⁽¹⁾		GLU (mg/100 ml)	CHOL (mg/100 ml)	HDL (mg/100 ml)	TG (mg/100 ml)	VLDL (mg/100 ml)	LDL (mg/100 ml)
Hatching windows factor							
Before 493 h		128.64	158.39	40.36	87.62	17.52	100.51
After 493 h		131.05	152.63	40.07	73.98	14.79	97.76
Probability		0.2758	0.1379	0.9090	0.0007	0.0007	0.4787
Administration factor							
Control		127.86	151.14 ab	38.15 ab	86.38 a	17.27 a	95.71 ab
MI with H ₂ O		128.13	148.17 b	39.91 ab	80.25 ab	16.05 ab	92.20 b
MI with ES		130.54	162.56 a	41.68 ab	71.71 b	14.34 b	106.53 a
Intra-crop with H ₂ O		133.36	152.34 ab	35.45 b	88.40 a	17.68 a	99.20 ab
Intra-crop ES		129.35	163.36 a	45.87 a	77.26 ab	15.45 ab	102.03 ab
Probability		0.5115	0.0405	0.05	0.0510	0.0510	0.05
Interaction (hatching windows × administration)							
Before 493 h	Control	127.23	156.67 abc	31.76 b	99.16 a	19.83 a	105.07 ab
	MI with H ₂ O	124.79	144.52 c	41.47 ab	83.58 ab	16.71 ab	86.33 b
	MI with ES	128.49	167.79 a	43.14 ab	75.87 b	15.17 b	109.47 a
	Intra-crop with H ₂ O	134.54	157.82 abc	38.95 ab	100.00 a	20.00 a	98.86 ab
	Intra-crop ES	128.17	165.17 ab	46.47 a	79.47 b	15.89 b	102.80 ab
After 493 h	Control	128.49	145.61 bc	44.54 ab	73.60 b	14.72 b	86.34 b
	MI with H ₂ O	131.47	151.82 abc	38.36 ab	76.92 b	15.38 b	98.07 ab
	MI with ES	132.60	157.32 abc	40.21 ab	67.55 b	13.51 b	103.60 ab
	Intra-crop with H ₂ O	132.18	146.86 bc	31.96 b	76.81 b	15.36 b	99.53 ab
	Intra-crop ES	130.53	161.55 abc	45.27 a	75.04 b	15.00 b	101.26 ab
Probability		0.6982	0.05	0.5	0.0033	0.0033	0.05
Total mean		129.85	155.51	40.21	80.80	16.16	99.13
SEM ⁽²⁾		1.0816	1.9987	1.3086	2.1239	0.4247	1.9790

⁽¹⁾ Experimental Design: The hatching window (prior to 493 hours and after 493 hours) was considered the first factor, whereas the injection treatment (control, mesenteric water, mesenteric electrolyte, oral water, oral electrolyte) was regarded as the second factor. MI: Mesenteric injection; H₂O: Ionized water; ES: Electrolytic solution. ⁽²⁾ SEM: Values are presented as the Standard Error of the Mean. Note: Distinct letters within the same column denote statistically significant differences among treatment means at the 0.05 and 0.01 probability levels.

as a beneficial physiological response (Sigolo *et al.*, 2021). Differences among published studies may reflect variations in administration route, electrolyte composition, and duration of exposure (Yu *et al.*, 2022).

It is worth noting that elevated serum TG and VLDL concentrations are considered undesirable indicators in poultry, as they are associated with excessive fat deposition, which can adversely affect bird health and growth efficiency (Fan *et al.*, 2021). Accordingly, the reduction in TG and VLDL observed following mesenteric electrolyte injection may be regarded as a beneficial physiological response, reflecting improved ionic balance and more efficient regulation of lipid metabolism. This improvement likely

reduces hepatic metabolic burden and limits excessive fat deposition in tissues (Oketch *et al.*, 2023; Araujo *et al.*, 2022).

Regarding serum LDL, a significant ($P < 0.05$) interaction between hatching window and administration method was observed. Chicks removed before 493 h and subjected to mesenteric electrolyte injection exhibited the highest LDL values, whereas mesenteric water treatments generally resulted in lower concentrations. When considered independently, the administration method significantly affected LDL levels, while the hatching window alone had no significant effect.

The elevated LDL levels associated with mesenteric

electrolyte injection may be attributed to enhanced hepatic lipid metabolism and improved efficiency of lipoprotein-mediated lipid transport. Increased electrolyte availability likely improved osmotic balance and fluid homeostasis, thereby supporting hepatic synthesis and mobilization of LDL to meet the high energy demands of rapidly growing chicks (Oketch *et al.*, 2023; Araujo *et al.*, 2022). Variations among published studies suggest that the LDL response to electrolyte supplementation is influenced by bird genotype, physiological status, and nutritional conditions. For example, reduced LDL levels following electrolyte supplementation have been reported in Japanese quail (Ölmez *et al.*, 2023; Attia *et al.*, 2024), whereas no significant effects were observed in Rhode Island chickens (Pawłowska *et al.*, 2022). These discrepancies support the notion that the present findings represent a strain- and condition-specific response to mesenteric electrolyte administration.

Collectively, the results at 11 days of age demonstrate that administration method, rather than hatching window alone, plays a decisive role in regulating lipid metabolism during early growth. Mesenteric electrolyte injection promoted metabolic stability by reducing undesirable lipid fractions, whereas oral water and control treatments were associated with elevated TG and VLDL levels. These findings support the concept that targeted early post-hatch interventions can influence physiological adaptation and metabolic efficiency in broiler chickens.

CONCLUSIONS

The study concludes that the interaction between chick removal time and the method of administration plays a crucial role in regulating energy and lipid indicators in broiler blood. Water gavage contributed to increasing GLU, LDL, and VLDL, reflecting a rapid response to meet energy demands, while mesenteric electrolyte injection showed a positive role in reducing undesirable fat accumulation by improving ionic balance and supporting liver function. Electrolytes also contributed to raising HDL and enhancing the efficiency of reverse CHOL transport, whereas the hatching window factor alone showed no clear independent effect. These findings confirm that the method of administration is the most influential factor in regulating lipid metabolism in chicks during the early stages of growth. In addition to their effects on lipid metabolism, the observed physiological responses indicate that appropriate early post-hatch administration strategies may contribute to improving metabolic stability, liver function, and overall health status of broiler chicks, which can positively influence growth efficiency and productive performance under commercial rearing conditions.

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NOVELTY STATEMENT

This study is novel in evaluating mesenteric electrolyte injection as an early post-hatch intervention during the hatching window and elucidating its specific effects on lipid metabolism and physiological regulation in broiler chickens. The findings demonstrate that

AUTHOR'S CONTRIBUTION

Alaa Hasan Kadhim: Conceptualization, experimental design, data analysis, manuscript writing, and corresponding author.

Fadhil Rasool Abbas Al-Khafaji: Conducted the experiment, sample collection, and laboratory analyses.

Ziyad Tariq Mohammed Aldhanki: Statistical analysis, interpretation of results, and critical revision of the manuscript.

All authors read and approved the final manuscript.

GENERATIVE AI AND AI-ASSISTED TECHNOLOGY STATEMENT

The authors declare that no generative AI or AI-assisted technologies.

CONFLICT OF INTEREST

The authors have declared no conflict of interest.

REFERENCES

- Abioja MO, Ojoawo HT, Akinjute OF, Logunleko MO (2023). Influence of egg storage length and orientation on hatching traits and spread of hatch in Transylvanian naked neck chickens. *J. Anim. Physiol. Anim. Nutr.*, 107(4): 1083–1092. <https://doi.org/10.1111/jpn.13786>.
- Anyanwu VC, Ohanaka AUC, Alaoma OR, Aladi NO, Okeudo NJ (2024). Performance, hematological profile and serum biochemical composition of broiler chickens fed diets with different salt levels. *Int. J. Agric. Rural Dev.*, 27(1): 6828–6835.
- Araujo AC, Araújo RDS, Dourado LRB, Machado JS, Farias LA, de Sousa DM, de Sousa FCB, Biagiotti D, Bayão GFV, Sousa KRS (2022). Effects of dietary electrolyte balance on performance, energy balance, and expression of genes related to acid–base balance, absorption, and nutrient transport in broilers. *Trop. Anim. Hlth. Prod.*, 54: 165. <https://doi.org/10.1007/s11250-022-03165-z>
- Attia YA, Abdallah AA, Bovera F, Abd El-Hamid AEH, El-

- Naggar AS, Alhotan RA, Zaki RM (2024). Effect of dietary electrolyte balance and arginine-to-lysine ratio on hematological, antioxidant, and immunological traits in dual-purpose breeding hens under cyclic heat stress. *J. Therm. Biol.*, 121: 103835. <https://doi.org/10.1016/j.jtherbio.2024.103835>
- Aviagen (2022). ROSS 308 broiler performance objectives. <https://aviagen.com/tmea/brands/ross/products/ross-308>
- Boynner M (2023). A flying start: Adapted hatching and post-hatch feeding in broiler chickens. *Acta Univ. Agric. Sueciae*, 2023: 38. <https://doi.org/10.54612/a.35707n9nbs>
- Chen X, Zheng A, Chen Z, Pirzado SA, Wang Z, Chen J, Liu G (2024). Potassium diformate affects the growth and development of broilers by improving intestinal function and digestive enzyme activity. *Poult. Sci.*, 103(10): 104049. <https://doi.org/10.1016/j.psj.2024.104049>
- Chen Y, Wang Z, Yang Z, Yang Y, Yang J, Han H, Yang H (2022). Effects of different dietary sodium and chloride levels on performance, blood parameters, and excreta quality in goslings. *J. Anim. Physiol. Anim. Nutr.*, 106(1): 98–109. <https://doi.org/10.1111/jpn.13541>
- Dang DX, Zhou H, Lou Y, Li D (2022). Effects of in ovo feeding of methionine and/or disaccharide on post-hatching breast development, glycogen reserves, nutrients absorption parameters, and jejunum antioxidant indices in geese. *Front. Vet. Sci.*, 9: 944063. <https://doi.org/10.3389/fvets.2022.944063>
- Duarte Lau F, Giugliano RP (2023). Adenosine triphosphate citrate lyase and fatty acid synthesis inhibition: A narrative review. *JAMA Cardiol.*, 8(9): 879–887. <https://doi.org/10.1001/jamacardio.2023.2402>
- Fan R, Liu K, Zhou Z (2021). Abnormal lipid profile in fast-growing broilers with spontaneous femoral head necrosis. *Front. Physiol.*, 12: 685968. <https://doi.org/10.3389/fphys.2021.685968>
- Feng X, Zhang L, Xu S, Shen AZ (2020). ATP-citrate lyase (ACLY) in lipid metabolism and atherosclerosis: An updated review. *Progress Lipid Res.*, 77: 101006. <https://doi.org/10.1016/j.plipres.2019.101006>
- Imtiaz KY, Mukhtar N, Yousaf A, Iqbal J, Ahmad T (2023). Influence of in ovo administration of electrolytes during the perinatal period on subsequent broiler performance. *Adv. Life Sci.*, 9(4): 467–472. <https://doi.org/10.62940/als.v9i4.1285>
- Jassim EQ, Aqeel CH (2017). Effect of alkaline water and/or magnetic water on some physiological characteristics of broiler chickens. *J. Entomol. Zool. Stud.*, 5(5): 1643–1647.
- Jha R, Singh AK, Yadav S, Berrocoso JFD, Mishra B (2019). Early nutrition programming (in ovo and post-hatch feeding) as a strategy to modulate gut health of poultry. *Front. Vet. Sci.*, 6: 82. <https://doi.org/10.3389/fvets.2019.00082>
- Kalaba ZM, Ismail FSA, Kassem MF (2023). Physiological differences, physical characteristics, and plasma parameters of chicks hatched from multi-stage versus single-stage incubator systems. *J. Anim. Poult. Prod.*, 14: 119–124. <https://doi.org/10.21608/jappmu.2023.236889.1093>
- Kohn RA, Allen MS (1995). Enrichment of proteolytic activity relative to nitrogen in rumen preparations for *in vitro* studies. *Anim. Feed Sci. Technol.*, 52(1–2): 1–14. [https://doi.org/10.1016/0377-8401\(94\)00711-H](https://doi.org/10.1016/0377-8401(94)00711-H)
- Manthei KA, Patra D, Wilson CJ, Fawaz MV, Piersimoni L, Shenkar JC, Tesmer JJ (2020). Structural analysis of lecithin–cholesterol acyltransferase bound to high-density lipoprotein particles. *Commun. Biol.*, 3: 28. <https://doi.org/10.1038/s42003-019-0749-z>
- Manthei KA, Yang SM, Baljinnayam B, Chang L, Glukhova A, Yuan W, Tesmer JJ (2018). Molecular basis for activation of lecithin–cholesterol acyltransferase by compounds that increase HDL cholesterol. *eLife*, 7: e41604. <https://doi.org/10.7554/eLife.41604>
- Mgs ERCP, Mgs LARO, Ordóñez LNJ (2023). Effect of different electrolyte levels on growth and fattening of COB 500 broiler chicks. *HIV Nurs.*, 23(2): 637–644.
- Mohamed A, Khalil M, Soliman F, El-Sabroun K (2025). Effect of drinking ionized water on productive performance, physiological status, and carcass characteristics of broiler chicks. *Animals*, 15(2): 229. <https://doi.org/10.3390/ani15020229>
- Mouldin RE, Goldale OA, Engeman RM, Goodall MJ, Craves RK, Johnston JJ (1996). Determination of zinc phosphide residues by gas chromatography. *J. Agric. Food Chem.*, 44: 189–191. <https://doi.org/10.1021/jf9501498>
- Oketch EO, Wickramasuriya SS, Oh S, Choi JS, Heo JM (2023). Physiology of lipid digestion and absorption in poultry: Implications for early nutrition. *J. Anim. Physiol. Anim. Nutr.*, 107(6): 1429–1443. <https://doi.org/10.1111/jpn.13859>
- Ölmez M, Şahin T, Aras ŞY, Karadağoglu Ö, Sari EK, Büyükbaki HG, Şerbetçi I (2023). Effects of coated calcium sodium butyrate on growth performance and biochemical traits of quail. *Turk. J. Vet. Anim. Sci.*, 47(2): 167–175. <https://doi.org/10.55730/1300-0128.4282>
- Pawłowska J, Sosnowka-Czajka E, Skomorucha I (2022). Effect of in ovo electrolyte injection site on biochemical blood parameters and chick quality. *Animals*, 12(4): 532. <https://doi.org/10.3390/ani12040532>
- Safiyu KK, Akinsola KL, Ibedu AJ, Ariwodo CE, Onabanjo RS, Sogunle OM (2023). Effect of citrus–coconut electrolyte blends on growth performance and haemato-biochemical traits of broilers. *Trop. Anim. Health Prod.*, 55: 56. <https://doi.org/10.1007/s11250-023-03463-0>
- SAS Institute Inc. (2004). SAS User's Guide: Statistics (6th ed.). Cary, NC, USA.
- Shibata M, Takahashi T, Kozakai T, Shindo J, Kurose Y (2023). Development of active jejunal glucose absorption in broiler chickens. *Poult. Sci.*, 102(8): 102804. <https://doi.org/10.1016/j.psj.2023.102804>
- Shihab IM, Ezzat HN, Hussein MA (2019). Effect of ionized water on productive and physiological performance of Japanese quail. *Poult. Sci.*, 98(10): 5146–5151. <https://doi.org/10.3382/ps/pey590>
- Sigolo S, Ahmadian A, Seidavi A, Gallo A, Prandini A (2021). Effects of dietary electrolyte balance on growth performance, carcass traits, blood parameters, and immune responses of broilers. *J. Appl. Anim. Res.*, 49(1): 472–478. <https://doi.org/10.1080/09712119.2021.2009840>
- Such N, Schermann K, Pál L, Menyhárt L, Farkas V, Csitári G, Dublec K (2023). Hatching time modifies production traits and early cecal microbiota development in broiler chickens. *Animals*, 13(17): 2712. <https://doi.org/10.3390/ani13172712>
- van de Ven LJF, van Wagenberg AV, Kemp B (2021). Hatching window, chick quality, and broiler performance: A review. *Animal*, 15(1): 100025.
- Yu H, Azzam MM, Wang YB, Lin XJ, Alqhtani AH, Al-Abdullatif AA, Jiang SQ (2022). Dietary sodium and

- chloride requirements for slow-growing broilers during the finisher phase. *J. Appl. Poult. Res.*, 31(2): 100243. <https://doi.org/10.1016/j.japr.2022.100243>
- Zhang Z, Wen L, Jiang L (2021). Nanofluidics for osmotic energy conversion. *Nat. Rev. Mater.*, 6(7): 622–639. <https://doi.org/10.1016/j.enconman.2021.114846>
- Zuidhof MJ (2022). Embryonic development, hatch window, and chick quality in broiler production. *World's Poult. Sci. J.*, 78(3): 567–580.