



Influence of the preplacement holding time and feeding hydration supplementation before placement on yolk sac utilization, the crop filling rate, feeding behavior and first-week broiler performance¹

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ABSTRACT This study investigated the effects of the broiler chick preplacement holding time and feeding hydration supplementation before placement on yolk sac utilization, the crop filling rate, feeding–drinking behavior and first-wk broiler performance. Broiler hatching eggs were obtained from a commercial broiler breeder flock of Ross 308 at 37 wk of age and incubated in a commercial hatchery. At 510 h of incubation, all chicks were removed from the hatcher and separated into cardboard chick boxes containing 80 chicks each. The chick boxes were randomly separated into two groups with either added commercial hydration supplementation (gel: Hydrogel-95) or the control (no gel). Then, the chicks were randomly distributed into 5 groups with different holding times across each hydration supplementation treatment (gel and control). The preplacement holding times were 6, 24, 48, 60, and 72 h from the pull time from the hatcher in the hatchery to placement in the broiler house on the farm, at which point the chicks were able to access feed and water. There were 10 subtreatment groups comprising 5 chick preplacement holding time groups × 2 hydration supplementation groups. There were 12 replicates (160 chicks per pen) per holding period × gel treatment, with a total of 19,200 chicks placed. The feed and water access time did not influence yolk sac utilization, but the absolute or relative residual yolk sac (g, %) decreased linearly with the duration after the pull time ($P < 0.001$). Longer preplacement holding times were associated with a higher percentage of chicks with full crops at 3 h after placement ($P < 0.001$). Chicks with the shortest (6 h) preplacement holding time had a lower percentage of feed-seeking activity compared to the 24, 48, and 72 h holding time groups at 3 h after placement ($P < 0.001$). The highest chick eating and

drinking activity was observed in the 72 h group at both 3 and 8 h after placement. Chick weight at placement was significantly reduced linearly with the duration after the pull time (0.106 g/h; $R^2 = 0.775$), and as expected, the highest and lowest BW were found in the 6 (41.51 g) and 72 h (34.50 g) preplacement holding time groups, respectively. However, BW and BW gain were higher in the 24 h group than in the other preplacement holding time groups ($P < 0.001$) at 7 d after placement. Mortality within the first 3 d after placement increased only when the preplacement holding time was extended to 72 h ($P = 0.002$). Mortality during 4 to 7 d postplacement was not affected by the holding time at all, but the 72-h holding time group still had statistically significantly higher mortality cumulatively from 0 to 7 d ($P = 0.024$). Neither BW nor mortality was affected by feeding the hydration supplement at placement, and the lack of effect persisted through 7 d after placement ($P > 0.05$). It can be concluded that the BW at 7 d after placement was greater in the 24 h holding time group than in shorter (6 h) or longer (48, 60, and 72 h) preplacement holding time groups. In the present study, a greater number of chicks were raised, and it was clearly demonstrated that mortality, as a direct indicator of flock health and welfare, was not affected by preplacement holding times up to and including a 60 h after take-off under thermal comfort conditions, but holding for a further 12 h to 72 h, mortality at 7 d of age after placement was increased. On the other hand, holding chicks in a short period (6 h) did not improve mortality and the BW at 7 d, suggesting that some delay to placement can be beneficial. In addition, feeding hydrogel during the preplacement holding period had no positive effect on BW gain and cumulative mortality during the first week of the growing period.

Key words: preplacement holding time, hydration supplement, residual yolk sac, body weight, mortality

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INTRODUCTION

The continuous genetic selection of broilers for growth has resulted in a reduced slaughter age; thus, early life conditions are critical for the development and performance of chicks later in life (Decuyper et al., 2001; Mitchell, 2009). Several studies have reported that both the posthatch handling time and environmental conditions are crucial for the development and performance of chicks later in life (Mitchell, 2009; Bergoug et al., 2013; Peebles et al., 2016; Hamissou Maman et al., 2019).

Under commercial situations, following chick removal from the incubator, access to feed and water is further delayed because of processes such as selecting, sexing, counting, vaccinating, packaging, and transporting from the hatchery to production facilities. The delay is usually very small in broiler hatcheries, but for high-generation breeding stocks, which are often farmed and hatched in remote locations to optimize disease control and biosecurity, the interval between pulling and placement times can be up to 60 h (Aviagen, 2021).

Newly hatched chicks contain a significant reservoir of residual yolk upon hatching, and this reservoir provides sufficient energy and water to keep the chicks in good condition for up to 3 d (EFSA Panel on Animal Health Welfare, 2011). For this reason, legislation usually allows longer journeys and does not mandate breaks for resting and feeding at the frequency required by older stock. For example, current EU legislation (Council of the European Union, 2005) specifies that day-old chicks can be transported for 24 h within the first 72 h of life without access to food or water. This recommendation is based on the fact that the chick's metabolic reserves stored in the yolk sac last up to 3 d.

Offering hatchling supplements to chicks at the hatchery is a common practice, especially when the preplacement holding time is longer (Batal and Parsons, 2002; Mikesell, 2017). These supplements are thought to overcome the possible disadvantages of delayed access to feed and water posthatch. Products such as traditional solid feed, semisolid feed or liquid nutrients are used to promote the early growth and livability of poultry. The benefits of posthatch feeding supplementation (Oasis containing 70% water, 10% protein, 20% carbohydrate, and less than 1% fat; Novus International, Inc., St. Louis, MO) due to increased nutrient digestibility compared to chicks held without feed and water immediately after hatching have been reported by Knight and Dibner (1998), Noy and Sklan (1999), and Batal and Parsons (2002). Hydrogel-95 (Clear H2O, Portland, OR) is a novel green hatching gel supplement that contains 95% water and provides chicks with hydration and extra nutrients to offset the water and muscle loss incurred after a longer posthatch holding period.

It has been reported that holding chicks for an extended period of time after hatching can lead to dehydration, diminished yolk sac reserves (Hager and Beane, 1983), reduced body weight (BW) (Hager and Beane, 1983; Sklan et al., 2000; Careghi et al., 2005), and increased mortality before and after placement (Hamdy

et al., 1991; Dibner and Knight, 1999). Furthermore, reduced growth and increased early mortality in chicks held without access to feed and water were associated with dehydration and a shortage of available energy (Vieira and Moran, 1999). In contrast, holding chicks for 24 h or longer after hatching under suitable conditions has been reported to not clinically dehydrate chicks or affect live performance (Casteel et al., 1994; Stamps and Andrews, 1995; Joseph and Moran, 2005; Almeida et al., 2006; Lamot et al., 2014; Özlü et al., 2018, 2020). On the other hand, in the aforementioned previous studies, low numbers of chicks and replicates were used, and therefore, the mortality results might not be fully representative of the practice in the industry.

In the current study, a greater number of chicks (19,200) were raised in relatively large pens (1.50 × 2.75 m; 160 chicks/pen) to investigate the effects of shorter and longer preplacement holding times up to and including 72 h after the pull time and of feeding a hydration supplement (Hydrogel-95) before placement on yolk sac utilization, crop filling rate, feeding–drinking behavior, and first-wk BW and mortality.

MATERIALS AND METHODS

All procedures in the current study were approved by the Animal Ethics Committee of Poultry Research Institute, Ankara (2021/01).

Hatching Eggs and Incubation

Hatching eggs were obtained from a commercial broiler breeder flock of Ross 308 at 37 wk of age and stored for 2 d at 18°C and 75% relative humidity (RH). Then, the eggs were incubated in a single Avida model setter and hatcher (Chick Master Incubator Co., Medina, OH) in a commercial hatchery (Şenpiliç Inc. Adapazarı, Türkiye) A single-stage incubation program with a gradually decreasing set-point temperature of 38.1°C at embryonic day (E) 1 to 37.5°C at E19 was used. The hatcher began at a set-point temperature of 37.2°C at E19, which gradually decreased to 36.4°C at E21. RH was set to 53 ± 2% during the entire incubation process, and eggs were turned hourly by an angle of 45° until E19 of incubation.

Chick Management and Experimental Design

All chicks that had completed the hatching process were removed from the hatching baskets at 510 h of incubation. After the selection of second-grade chicks (splayed legs, unhealed navels or weak and physical abnormalities), the saleable chicks were counted into cardboard chick boxes, with 80 chicks in each box. Then, they received spray vaccinations for infectious bronchitis and Newcastle disease. The chick boxes were randomly separated into two groups. One group was given a-2 g/chick commercial hydration supplement that contained 95% water (Hydrogel-95, Clear H2O,

Table 1. Vent temperatures during preplacement holding period.

Treatment	Vent Temperature (°C)
Preplacement holding time (h)	
0 (Pull time)	39.65
6	39.62
24	39.68
48	39.68
60	39.69
72	39.58
SEM	0.041
Hydration supplementation treatment	
Control	39.68
Gel	39.62
SEM	0.026
<i>P</i> values	
Preplacement holding time (h)	0.221
Hydration supplementation treatment	0.083
Preplacement holding time × Hydration supplementation treatment	0.645

Portland, OR) at hatchery before transportation, and the other group was not provided with gel (control–no gel). The chicks were transported to the commercial broiler house for 4 h by road and were then held in the unlighted climate-controlled truck (H90, Heering, Vaassen, Holland) at $25 \pm 0.4^\circ\text{C}$ and $35 \pm 2\%$ RH until placement. The vent temperatures of 100 randomly selected chicks were measured at pull time and the chick placement times (10 chicks/box/treatment) with an infrared digital thermometer (IRT 4520, Thermoscan, Braun, Germany), as shown in [Table 1](#).

The chicks were randomly distributed into 5 groups with different preplacement holding times for each hydration supplementation treatment (gel and control). The preplacement holding times were 6, 24, 48, 60, and 72 h after the pull time. In total, 19,200 chicks were randomly assigned to 10 subtreatment groups (5 preplacement holding times × 2 hydration supplementation treatments during transportation and the holding period). During the first week of the growing period after placement, the 10 subtreatment groups were allocated into a randomized complete design, with each subtreatment including 12 replicate floor pens with 160 randomly selected mixed chicks per pen (120 total pens).

Grow-Out Housing and Management

The grow-out period of the experiment was carried out in a commercial broiler house with automatic ventilating and heating systems. The brooding facilities were preheated for 24 h before chick placement to achieve a stable and uniform litter temperature. At placement, the air temperature was 30°C , and the temperature held constant at the end of the trial due to different placement time of the chicks. The chicks received a continuous light schedule (24 L: 0 D) throughout the experimental period. The chicks in each group were housed in floor pens (1.50 × 2.75 m) containing new wood litter shavings until 7 d from the day of placement. The initial chick density in each of the pens was

approximately 0.026 m^2 per bird. Both feed and water were available for ad libitum consumption throughout the experimental period. Water was provided via 2 nipple lines with 18 nipples in total in each pen. Feed was provided in two 33 cm pan feeders located in the center of each pen. Additionally, feed was provided on chick paper in all pens during the first 4 d after placement.

Starter diets produced in crumble form were fed throughout the experiment (0–7 d) to meet or exceed the demands of broiler chickens according to the recommendations of the breeding company ([Aviagen, 2019](#)).

Measurements

All on-farm measurements were performed at a defined number of days after the chicks were placed into the pens with free access to feed and water. The time elapsed since the chicks were removed from the hatcher thus varied by up to 3 d.

Mortality during the Preplacement Holding Period

All chick boxes were controlled, and dead birds were recorded to determine the percentage of mortality relative to the total chicks at each placement time.

Residual Yolk Sac Weight and Yolk-Free Body Mass

At the pull time, 80 randomly selected chicks were weighed and euthanized by cervical dislocation to measure the residual yolk sac weight. Subsequently, forty randomly selected chicks were collected, and the residual yolk sac weights were measured at each placement time (6, 24, 48, 60, and 72 h after the pull time) in each hydration supplementation treatment group (gel or control). Finally, to determine yolk sac utilization by chicks after feeding in both the gel and control groups, the residual yolk sac weight was also recorded in forty chicks from the 6 h placement group at each subsequent placement time (24, 48, 60, and 72 h). The yolk-free body mass (YFBM) was calculated as the chick weight minus the residual yolk weight. In the present study, residual yolk sac weights were determined in a total of 800 chicks.

Crop Filling and Feeding-Drinking Behavior

Crop filling was assessed in 30 randomly selected chicks from each pen at 3 h after placement. The chicks were gently lifted, and their crops were examined by palpation ([Aviagen, 2018b](#)) to determine crop filling progression, which was assigned to the following categories: (1) empty, (2) water only, (3) feed only, and (4) full, soft, and rounded (water and feed). Behavioral observations commenced within 3 h and 8 h after placement was completed. Chick behaviors were studied by scan sampling at the observation times to determine the eating and drinking drives of the chicks after placement in all

pens. The number of chicks performing either of the following behaviors was recorded: (1) eating from the feeder and chick paper or (2) drinking water from the nipples (Boyner et al., 2021).

Post Hatch Performance

BW were measured on a group basis (forty chicks in bulk) upon placement (0 d) and 7 d from the day of placement in each group. Body weight gain (BWG) from 0 to 7 d from placement was also calculated on a pen basis. Mortality in each pen was recorded six times a day.

Statistical Analyses

Separate analyses were performed to examine the data collected on mortality during the preplacement holding period, residual yolk sac weights (g, %), YFBM, crop filling, behavior, and live broiler performance.

For mortality during the preplacement holding period, a Z-test was employed to determine the existence of differences in two proportional mortality values in the hydration supplementation treatment groups and the preplacement holding time groups.

Data on the residual yolk sac weight (g, %), YFBM, crop filling, feeding behavior, BW, BWG and chick mortality were analyzed according to the following model: $Y_{ijk} = \mu + HST_i + PPHT_j + (HST \times PPHT)_{ij} + e_{ijk}$, where μ is the overall mean, HST_i is the hydration supplementation treatment ($i = \text{control or gel}$), $PPHT_j$ is the preplacement holding time of the chick ($j = 6, 24, 48, 60, \text{ or } 72 \text{ h}$), $(HST \times PPHT)_{ij}$ is the interaction between the hydration supplementation treatment and the preplacement holding time, and e_{ijk} is the residual error term. F tests were performed for the interactions and main effects of the hydration supplementation treatment and preplacement holding time with a 2×5 factorial scheme. Depending upon the nonsignificance of the interactions, Duncan's multiple tests were performed on the separate 2 means of the hydration supplementation treatment and 5 means of the preplacement holding time treatments. Additionally, data on yolk sac weights were analyzed according to the following model: $Y_{ijkl} = \mu + HST_i + PPHT_j + AFW_k + (HST \times PPHT)_{ij} + (HST \times AFW)_{ik} + (PPHT \times AFW)_{jk} + (HST \times PPHT \times AFW)_{ijk} + e_{ijkl}$, where AFW_k is the access to feed and water ($k = \text{fed/not fed}$), $(HST \times AFW)_{ik}$ is the interaction between the hydration supplementation treatment and access to feed and water, $(PPHT \times AFW)_{jk}$ is the interaction between the preplacement holding time and access to feed and water, $(HST \times PPHT \times AFW)_{ijk}$ is the interaction between the hydration supplementation treatment, preplacement holding time, and access to feed and water, and e_{ijkl} is the residual error term. In addition, comparisons of residual yolk sac weights between groups with differing access to feed and water (fed/not fed) were performed using one-way ANOVA for access to feed and water (fed or not fed) at each sampling time.

Regression analyses were also performed to examine effects of preplacement holding time on Residual Yolk Sac (RYS) and BW.

All statistical analyses were performed with SAS version 9.1 (SAS Institute Inc, Cary, NC).

RESULTS

Mortality during the Preplacement Holding Period

No significant differences in mortality at the placement time were found between the 6 and 60 h preplacement holding times. Cumulative mortality (Figure 1), although very low, was higher for the 72 h preplacement holding time (0.244%) than for the 6 h (0.020%) and 24 h (0.039%) holding times ($P = 0.002$ and $P = 0.007$, respectively). On the other hand, the average mortality during the preplacement holding time was similar in the gel-fed and control groups (data not shown).

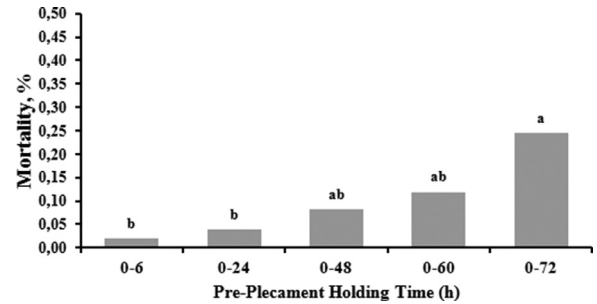


Figure 1. Cumulative mortality during the preplacement holding time. ^{a-b}Percentages of mortality with different superscripts differ significantly ($P < 0.05$).

RYS Weight and YFBM

The effect of the preplacement holding time and hydration supplementation treatment before placement on the absolute and relative residual yolk sac weight and YFBM at the time of placement is shown in Table 2.

In the current study, the absolute and relative RYS weights (g, %) decreased linearly with increasing time elapsed after the pull time ($P < 0.001$). The average RYS weight was 5.3 g (12.5%) at pull time and decreased to 4.5 g (10.9%) and 3.1 g (7.7%) at the 6 h and 24 h preplacement holding times. At 72 h after the pull time, the RYS weight was found to be 1.0 g (3.0%). Moreover, RYS utilization increased linearly ($Y = 4.86 - 0.0585 \times \text{preplacement holding time}$, $R^2 = 0.711$) by 0.0585 g/h of preplacement holding time.

The YFBM was affected by the preplacement holding time ($P < 0.001$). At placement, the YFBM values in the 6 h group and in chicks subjected to up to 48 h of holding after the pull time were similar, mainly because of similar yolk utilization during this period ($P > 0.05$). However, chicks held 60 or 72 h (after 48 h) possessed less ($P < 0.001$) YFBM than those in the 6 h holding time group (Table 2).

Table 2. RYS weights and YFBM of chicks.

Treatment	RYS ¹ Relative	RYS absolute	YFBM ²
	(%)	(g)	
Preplacement holding time (h)			
0 (Pull time)	12.45 ^a	5.339 ^a	37.26 ^a
6	10.87 ^b	4.496 ^b	36.82 ^{ab}
24	7.66 ^c	3.050 ^c	36.34 ^{ab}
48	4.80 ^d	1.825 ^d	35.95 ^{bc}
60	3.34 ^e	1.232 ^e	35.50 ^c
72	2.97 ^e	1.034 ^e	33.64 ^d
SEM	0.206	0.0865	0.251
Hydration supplementation treatment			
Control	5.80	2.270	35.76
Gel	6.06	2.384	35.58
SEM	0.130	0.0547	0.158
<i>P</i> values			
Preplacement holding time (h)	<0.001	<0.001	<0.001
Hydration supplementation treatment	0.169	0.142	0.418
Preplacement holding time × Hydration supplementation treatment	0.992	0.981	0.641

^{a-e}Means in a column with different superscripts differ significantly ($P \leq 0.05$).

¹RYS: Residual yolk sac weight at placement time.

²YFBM: Yolk-free body mass = chick weight – residual yolk sac weight at placement time.

Feeding Hydrogel-95 during the preplacement holding period did not affect the RYS weight or YFBM at placement.

In the current study, the RYS weights at 24, 48, 60, and 72 h were also recorded in an additional forty chicks with access to feed after 6 h to determine whether providing a nutrient-dense feed had any effect on the residual yolk utilization rate. There were no significant differences in the absolute RYS weight between chicks that were fed versus those held without access to feed for all preplacement holding times (24, 48, 60, and 72 h after the pull time) (Table 3). The RYS weights were 3.17 and 3.05 g in chicks with and without access to feed, respectively, at 24 h after the pull time. At 72 h after the pull time, which was the maximum preplacement holding

Table 3. Effects of access to feed and water on RYS weight (g and percentage) in chicks.

Feed and water access ¹	Age at sampling, h			
	24	48	60	72
	(g)			
Fed	3.17	1.77	1.15	0.93
Not Fed	3.05	1.83	1.23	1.03
SEM	0.094	0.073	0.047	0.047
<i>P</i> value ²	0.363	0.616	0.261	0.127
	(%)			
Fed	6.56 ^b	3.00 ^b	1.75 ^b	1.25 ^b
Not Fed	7.66 ^a	4.80 ^a	3.33 ^a	2.97 ^a
SEM	0.204	0.161	0.101	0.108
<i>P</i> value ²	<0.001	<0.001	<0.001	<0.001

¹Fed group with access to feed and water at 6 h.

²*P* value for comparison of the 2 groups (fed/not fed; ^{a,b}: $P < 0.001$).

time, the RYS weights in chicks with access to feed and those that were not fed were 0.93 g and 1.03 g, respectively. However, relative RYS weight (%) decreased in chicks that were fed compare to those held without access to feed for the preplacement holding times (Table 3).

Crop Filling Rate

The effects of the preplacement holding time and hydration supplementation treatment on crop filling are shown in Table 4. No interactions between the preplacement holding time and hydration supplementation treatment were found for the crop filling rate at 3 h after placement. Feeding gel during the preplacement holding period had no impact on the crop filling assessment ($P > 0.05$), but the percentage of empty crops was significantly higher in the 6 h holding time group than in the other preplacement holding time groups ($P < 0.001$). In addition, in the 6 h holding time group, only 66.6% of chicks had full, soft and rounded (water and feed) crops, whereas the percentage of full chick crops increased to 81.1% in the 24 h holding time group at 3 h after placement. The highest (more than 90%) percentage of chicks with full crops was found in the 72 h preplacement holding time group at 3 h after placement ($P < 0.001$).

Table 4. Crop filling at 3 h after placement in chicks.

Treatment	Crop filling progression in chicks ¹				Total
	Empty	Water	Feed	Full, rounded	
	(%)				
Preplacement holding time (h)					
6	7.48 ^a	8.50	17.45 ^a	66.57 ^c	100.0
24	1.20 ^b	2.58	15.10 ^a	81.12 ^b	100.0
48	1.41 ^b	1.80	16.66 ^a	80.13 ^{ab}	100.0
60	1.90 ^b	5.08	7.77 ^b	85.25 ^{ab}	100.0
72	0.96 ^b	3.00	5.40 ^b	90.64 ^a	100.0
SEM	0.669	1.878	1.439	2.231	
Hydration supplementation treatment					
Control	2.49	4.54	13.14	79.83	100.0
Gel	2.69	3.84	11.82	81.65	100.0
SEM	0.423	1.188	0.910	1.468	
<i>P</i> values					
Preplacement holding time	<0.001	0.091	<0.001	<0.001	
Hydration supplementation treatment	0.740	0.678	0.307	0.383	
Preplacement time × Hydration supplementation treatment	0.483	0.376	0.453	0.372	

^{a-d}Percentages of chicks in a column with different superscripts differ significantly ($P \leq 0.05$).

¹Crop filling, which is divided into the following categories: empty, water only, feed only, and full, soft, and rounded (water and feed).

Behavioral Observations

The effects of the preplacement holding time and hydration supplementation treatment on observed

Table 5. Behavioral observations of chicks at 3 and 8 h after placement.

Treatment	Observation at 3 h ¹		Observation at 8 h	
	Eating	Drinking	Eating	Drinking
	(%)			
Preplacement holding time (h)				
6	20.11 ^c	3.81 ^c	19.87 ^c	4.30 ^b
24	28.39 ^b	7.21 ^a	12.08 ^d	1.64 ^d
48	26.80 ^b	4.98 ^{bc}	27.87 ^b	5.18 ^b
60	15.86 ^d	5.73 ^b	22.48 ^c	5.16 ^b
72	32.40 ^a	8.21 ^a	36.59 ^a	7.79 ^a
SEM	1.312	0.493	1.445	0.472
Hydration supplementation treatment				
Control	24.59	5.62	24.14	4.97
Gel	24.83	6.35	23.42	4.66
SEM	0.830	0.312	0.914	0.299
<i>P</i> values				
Preplacement holding time	<0.001	<0.001	<0.001	<0.001
Hydration supplementation treatment	0.832	0.096	0.580	0.460
Preplacement holding time × Hydration supplementation treatment	0.116	0.384	0.621	0.254

^{a-d}Percentages of chicks in a column with different superscripts differ significantly ($P \leq 0.05$).

¹Percentage of eating from the feeder and chicken paper or water drinking from the nipples behaviors at 3 h and 8 h after placement.

behaviors at 3 and 8 h after placement are shown in Table 5. No interactions between the preplacement holding time and hydration supplementation treatment were found for chick behaviors at any observation time after placement. There was no significant difference in chick behaviors after placement (3 and 8 h) between the hydration supplementation treatments ($P > 0.05$). The highest eating and drinking activity in chicks was observed in the 72 h group at both 3 and 8 h after placement. Chicks with the shortest (6 h) preplacement holding time had a lower percentage of feed- and water-seeking activity compared to the 24 h holding time group at 3 h after placement, but the opposite relation was observed at 8 h after placement ($P < 0.001$).

Body Weight

The effects of the preplacement holding time and hydration supplementation treatment on BW are shown in Table 6. No interactions between the preplacement holding time and hydration supplementation treatment were found for BW at placement or 7 d after placement.

In the current study, the initial BW at placement (d 0) clearly decreased with longer chick holding times before placement, and as expected, the highest and lowest BW values were observed in the 6 h (41.5 g) and 72 h (34.5 g) holding time groups ($P < 0.001$) at the placement time ($Y = 42.33 - 0.1057 \times \text{preplacement holding time}$; $R^2 = 0.775$). A similar trend was also found at 7 d, except the highest BW was found for the 24 h instead of 6 h preplacement holding time ($P < 0.001$). BW was not affected by the hydration supplementation treatment at placement or 7 d after placement (Table 6).

At 7 d after placement, chicks held 24 h after the pull time showed higher BW gain compared with the other groups (Figure 2; $P < 0.001$). Chicks held for the longest period after the pull time (72 h) showed the lowest growth during the first 7 d. However, even though BW

Table 6. BW of chicks from 0 to 7 d of placement.

Treatment	BW	
	0 d	7 d
	(g)	
Preplacement holding time (h)		
6	41.51 ^a	179.5 ^b
24	39.55 ^b	183.2 ^a
48	37.43 ^c	174.9 ^c
60	36.24 ^d	169.8 ^d
72	34.50 ^e	165.0 ^e
SEM	0.093	0.39
Hydration supplementation treatment		
Control	37.85	174.5
Gel	37.84	174.4
SEM	0.059	0.25
<i>P</i> values		
Preplacement holding time	<0.001	<0.001
Hydration supplementation treatment	0.930	0.699
Preplacement time × Hydration supplementation treatment	0.364	0.512

^{a-e}Means in a column with different superscripts differ significantly ($P \leq 0.05$).

was significantly greater in the 6 h than 48 h holding time group at 7 d after placement, BW gain (0–7 d) was similar in both groups.

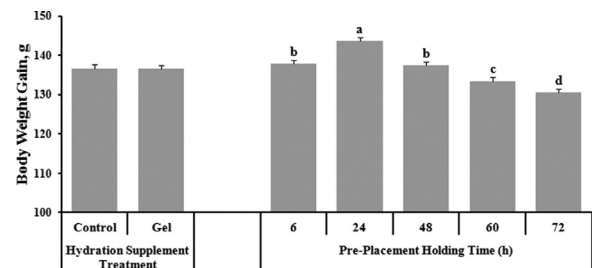


Figure 2. BWG in chicks from placement to the first 7 d after placement. ^{a-d}BWG in chicks in a section with different superscripts differs significantly ($P < 0.001$).

Mortality

There was no interaction ($P > 0.05$) between the preplacement holding time and hydration supplementation treatment for mortality during the first 7 d (Table 7).

Mortality was 1.4% in the 72 h group at 3 d, which was more than double that in the other preplacement holding time groups ($P = 0.002$). During the rest of the first week (4–7 d), mortality was similar among the groups (0.3–0.5%). However, the 72 h holding time group still had significantly higher mortality for the whole 7 d period (0–7 d) compared to the other preplacement holding time groups ($P = 0.024$). Additionally, feeding gel during the preplacement holding period had no impact at 7 d after placement.

Table 7. Mortality of chicks from placement to 7 d of placement.

Treatment	Mortality		
	0–3 d	4–7 d	0–7 d
	————— (%) —————		
Preplacement holding time (h)			
6	0.55 ^b	0.47	1.02 ^b
24	0.63 ^b	0.36	0.99 ^b
48	0.68 ^b	0.26	0.94 ^b
60	0.55 ^b	0.26	0.81 ^b
72	1.38 ^a	0.26	1.64 ^a
SEM	0.169	0.087	0.189
Hydration supplementation treatment			
Control	0.69	0.31	1.00
Gel	0.83	0.33	1.16
SEM	0.107	0.055	0.120
P values			
Preplacement holding time	0.002	0.340	0.024
Hydration supplementation treatment	0.372	0.790	0.358
Preplacement holding time × Hydration supplementation treatment	0.765	0.909	0.776

^{a-b}Percentages in a column with different superscripts differ significantly ($P \leq 0.05$).

DISCUSSION

Preplacement Holding Time

Chick body temperature, one of the important indicators of chick welfare, affects the quality of day old chicks and their subsequent broiler performance (Decuyper et al., 2001; Hamissou Maman et al., 2019). The optimum day old chick vent temperature should be 39.4°C to 40.5°C during posthatch holding period (Aviagen, 2021). In the present study, during the preplacement holding period, chicks were held under optimum conditions in which the vent temperature was kept in the ideal range (39.7 ± 0.58°C).

During late embryonic development, the residual yolk is enclosed within the abdominal cavity and provides immediate nutrition for maintenance and growth after hatching until exogenous feed is supplied (Romanoff, 1960). Both the fat energy content and the water reserves in the residual yolk are sufficient to meet chicks' requirements for approximately 3 d under optimum

temperature conditions (Freeman, 1984). In the current study, it was clearly demonstrated that the residual yolk sac weight (g) decreased linearly with increasing time after the pull time regardless of the feed access time. The weight of the RYS was not altered by early feeding but, if RYS is calculated as a percentage, the BW of the birds increased in the fed group as the feed consumed therefore, although the absolute RYS was same, the ratio of RYS to BW decreased and it appears that the fed chicks are using the RYS faster than those which had not eaten. The average RYS utilization was 42.8, 65.8, and 80.6% for 24, 48, and 72 h after the pull time, respectively. This finding was consistent with the findings of Aviagen (2021), which indicated that more than 40% of the RYS was used during the first 24 h after the pull time. Several studies (Noy et al., 1996; Speake et al., 1998; Noy and Sklan, 2001; El-Husseiny et al., 2008; Bhanja et al., 2009) have reported that the RYS weight in chicks with access to feed after hatching was reduced more rapidly than that in fasted birds, which is probably due to higher stimulation of gastrointestinal tract (GIT) activity (Noy and Sklan, 1996). However, other studies comparing immediate or delayed posthatch feed intake up to 72 h did not find differences in yolk utilization or residual yolk weights (Gonzales et al., 2003, 2008; Van den Brand et al., 2010; Özlü et al., 2020). In a recent study, Deines et al. (2021) showed that the absolute and relative RYS weights were not different between chicks with immediate and delayed access to feed and water at d 3 after hatching.

Feeding and drinking behaviors are 2 of the most important indicators of animal well-being and health (Hart, 1988). Moreover, crop filling rates are measured as an indirect means of assessing management during the brooding phase (Linhoss et al., 2021). Primary breeder guidelines (Aviagen, 2018a) recommend that within 2 and 4 h after placement, 75 and 80% of chicks should have a full, soft, and rounded crops, respectively. In the present study, the percentage of birds with full crops at 3 h reached the target crop filling recommendations (75–80%) in all preplacement holding time groups except the 6 h group. It appeared that some time before chick placement was required for the chicks to become motivated to engage in feed and water seeking activities at 3 h after placement based on comparison of the other groups to the 6 h group. This finding was consistent with the findings of Boyner et al. (2021), who found that the full crop percentage increased when chicks were held for an additional 6 h before placement. In this trial, the chicks held for 72 and 6 h were the most and least eager to find feed and water, respectively, with those held for 24, 48 and 60 h being quite similar, at least as demonstrated by their relative crop fill value at 3 h post placement. However, these differences in crop fill did not lead to equivalent differences in 7 d BW.

Some previous studies reported that a prolonged holding period (>24 h) without access to feed and water is associated with stress induction in chicks due to energy deficiency and dehydration (Pinchasov and Noy, 1993; Noy and Sklan, 1999). This phenomenon causes lower

viability and impaired growth, which negatively influence the final BW (Hager and Beane, 1983; Vieira and Moran, 1999; Juul-Madsen et al., 2004). However, several authors (Casteel et al., 1994; Baião et al., 1998; Bergoug et al., 2013; Hollemans et al., 2018) have reported that a longer interval between hatching and housing impaired the BW at placement and weight gain of broilers in the first week but did not change the final weight at the end of the grow-out period. According to Stamps and Andrews (1995), among chicks placed in a house on the same day at 24 and 48 h after hatching, the BW at placement was significantly lower in chicks with delayed placing to provide access to feed and water. At 7 d after placement, the 24 h holding time group was the heaviest of the 3 groups; however, this difference was not significant. In addition, Wang et al. (2014) indicated that broilers were able to compensate for 48 h of feed deprivation when they were able to access feed for a similar period of time at 35 d. Similarly, Özlü et al. (2020) reported that a posthatch holding period of up to 40 h after hatching under optimum conditions reduced the 7 d BW but had no detrimental effects on final live performance when the feeding periods were equal.

In the current study, the results indicated that BW was significantly reduced linearly with increasing duration after the pull time. At placement time, as expected, the highest and lowest BW values were found in the 6 (41.51 g) and 72 h (34.50 g) preplacement holding time groups, respectively. However, at 7 d after placement, chicks held for the shortest period (6 h) gained less BW than chicks held for 24 h after the pull time. Therefore, BW was greater in the 24 h (183.2 g) holding time group than in the 6 h (179.5 g) and the other preplacement holding time groups. In addition, BW gain between 0 and 7 d after placement was similar in the 6 h and 48 h preplacement holding time groups (138.0 vs. 137.6 g). A possible explanation for this result is that early yolk utilization could have produced a more metabolically mature chick that was ready to consume feed (Chamblee et al., 1992). In the current study, the residual yolk sac weight was greater in the 6 h preplacement holding time group (10.9%) than in the 24 h preplacement holding time group (7.7%), but YFBM was similar (36.8 vs. 36.3 g) in both groups when they were placed in the house. Therefore, a certain amount of yolk may need to be absorbed (<10%) before placement to induce active feed consumption and BW gain. This was also confirmed by the crop filling rates and feeding behavioral observations at 3 h after placement (Tables 4 and 5). Thus, the percentage of chicks with crops that were completely full of food and water and the percentage of eating and drinking behaviors at 3 h after placement were significantly lower in the 6 h preplacement holding time group than in the 24 h preplacement holding time group.

Several previous studies reported that extended holding before placement increased early rearing mortality (Hamdy et al., 1991; Pinchasov and Noy, 1993; Dibner and Knight, 1999; Yi et al., 2005) due to feed and water inaccessibility resulting in dehydration and a shortage of available energy (Vieira and Moran, 1999). Furthermore,

Stamps and Andrews (1995) divided chicks into three groups that were either placed in floor pens on the same day after the pull time or held in chick boxes in the hallway of the house for an additional 24 h or 48 h. The percentage of mortality from the day of placement to 7 d was higher in chicks that were placed 48 h after the pull time than in chicks that were placed in the house on the same day or 24 h after the pull time. In addition, a meta-analysis of data from multiple studies showed that 48 h or longer of posthatch feed and water deprivation had long-term effects on total mortality at 42 d of age (De Jong et al., 2016). However, most of the experiments had low numbers of replicates and relatively small pen sizes. Perhaps more importantly, in many of the experiments, the 'delayed' chicks were held in unsuitable conditions (boxed within the heated poultry house), which could be expected to raise early mortality. In a recent study, (Dişa et al., 2022) with relatively large pen sizes (200 chicks/pen), sending the late hatch chicks to the broiler house shortly after hatching (7 h) increased the mortality at 41 d, unlike holding early hatched chicks for a relatively long time (50 h) in the hatcher.

In the current study, relatively high numbers of replicates (12 pens/subtreatment) with suitably sized pens (160 chicks/pen) allowed accurate treatment separation. Therefore, statistically significant results were found for mortality even though the number of dead chicks was very low during the first week.

Mortality, as a direct indicator of the welfare, should not exceed 1 to 1.5% during the first 7 d when a good quality chick is provided with proper nutrition and brooding management (Alcorn, 2008). In the current study, the mortality was not affected by holding times up to 60 h and ranged from 0.8 to 1.0% at 7 d after placement. However, holding for a further 12 h to 72 h, mortality was increased (1.64%) and this increased in mortality occurred only during the first 3 d (1.38%). In addition, mortality at 7 d was numerically lower in the 60 h preplacement holding time group than 6 h preplacement holding time group (0.81 vs. 1.02%). This finding was consistent with those of Casteel et al. (1994), Corless and Sell (1999), Daşkıran et al. (2012), Özlü et al. (2020) and Dişa et al. (2022) that sending the chicks earlier (6 h) did not reduce the first week mortality.

In the current study, the percentage of cumulative mortality was not high (0.24%) during 72 h preplacement holding period (Figure 1). However, mortality increased by more than 2 times during the last 12 h of the holding period (60–72 h) and was significantly higher at placement time in the 72 h preplacement holding time group than in the 6 and 24 h preplacement holding time groups ($P < 0.05$). This trend was also seen at 7 d, with mortality at 7 d being greater in the 72 h holding time group than in the other preplacement holding time groups ($P = 0.024$). This finding can be explained by the higher weight loss and lower YFBM between the 60 and 72 h holding times. Thus, YFBM was reduced by 4.7% from pull to 60 h but was reduced to 9.7% when the holding time reached 72 h, which was 2 times higher than that at 60 h. Dehydration is caused

by a lower YFBM due to immoderate water loss, which can negatively affect chicks' physiological and metabolic statuses (Peebles et al., 2004; Incharoen et al., 2015). On the other hand, in the current study, the crop filling rates and feeding and drinking behavioral observations of chicks held 72 h after the pull time did not indicate signs of dehydration at 3 h after placement.

Feeding Hydration Supplements before Placement

Hatching supplements have been used in both chicken and turkey hatcheries, particularly during longer preplacement holding periods, either to allow newly hatched chicks access to nutrients or to eliminate the risk of dehydration prior to placement with feed and water in production facilities. Several previous studies have reported that chicks and poults with access to semi-solid hydrated nutritional hatching supplements (Oasis) versus those fasted during the posthatch holding period had improved growth parameters (Knight and Dibner, 1998; Noy and Sklan, 1999; Batal and Parsons, 2002; Mozdziak et al., 2002). In another study, feeding Hydrogel-95 (containing 95% water) to emu chicks posthatch and during the brooding period had beneficial effects on growth performance (Lowman and Parkhurst, 2014). Furthermore, the use of Aqua Agar as a water replacement during the 24 h posthatch holding period improved the growth performance of young broilers at 21 d of age compared with the control group (Incharoen et al., 2015). The beneficial effects of hatching supplements are explained by chicks having immediate access to feed, regardless of the form of early nutrition. This leads to the hypothesis that not the kind of feed but the stimulatory effect of any feed on GIT secretions, growth factors or neuronal factors initiates the further development of the GIT and therefore gives an advantage to chicks provided early feed access (Willemsen et al., 2010). In contrast to these previous findings, in the current study, feeding Hydrogel-95 in chick boxes during the preplacement holding period had no impact on BW at 7 d. In addition, mortality at placement and at 7 d of age after placement was not affected by using hatching supplements during the preplacement holding period, which is similar to previous studies (Noy and Sklan, 1999; Batal and Parsons, 2002; Henderson et al., 2008; Lowman and Parkhurst, 2014).

CONCLUSIONS

It was concluded that there were no significant differences in the absolute RYS weight between chicks that were fed versus those held without access to feed with a maximum holding period of 72 h after the pull time. BW at placement time decreased with longer durations between the pull and placement times. However, BW at 7 d after placement was greater in the 24 h holding time group than in shorter (6 h) or longer (48, 60, and 72 h) preplacement holding time groups. In the present study,

a greater number of chicks (19,200) were raised, and it was clearly demonstrated that mortality, as a direct indicator of flock health and welfare, was not affected by preplacement holding times of 60 h or less under optimum conditions, but when the preplacement holding time was extended to 72 h, mortality at placement and 7 d of age after placement was increased compared to the other holding time groups. In addition, compared to the control (no gel), feeding hydrogel during the preplacement holding period had no beneficial effect on BW gain and cumulative mortality during the first week of the growing period.

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