

The effect of lower egg storage temperature and length of storage period

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This study investigated the effect of lower storage temperature on the albumen quality, the blastoderm development, and the hatchability of short and long stored broiler hatching eggs.

In this study, freshly collected hatching eggs from a commercial flock of Ross 308 at 57 weeks of age were stored for two days at 0, 4, 8 and 16°C (control) in four identical temperature controlled rooms.

After the first two days, the eggs from all groups were also stored for two days or 12 days at 16°C before incubation. Therefore, total egg storage periods were either four days or 14 days. The albumen quality (height and pH) and developmental stages of the blastoderm before and after the first two days of the storage were determined.

There were eight sub-treatment groups comprising four storage temperatures (ST) groups × 2 storage periods (SP). There were 10 replicates (150 eggs per tray) per ST × SP sub-treatments, with a total of 12,000 eggs were set in a single stage incubator in a commercial hatchery.

All unhatched eggs were opened and examined macroscopically to determine fertility or embryonic mortality (early dead [0-7 days], middle [8-17 days], late dead [18-21 days plus pipped eggs]) to calculate the percentage hatchability of fertile eggs.

In this study, as expected, the eggs stored for 14 days had a significantly

lower hatchability of fertile eggs owing to increase the percentage of early embryonic mortality, contaminated eggs and the second grade chicks than the short-period stored (four days) eggs ($P < 0.05$).

There was a significant difference in the albumen height and pH for the storage temperature groups ($P < 0.01$). Albumen height was higher at 0°C compare to fresh, 4, 8° and 16°C (control) groups whereas the albumen pH increased at 16°C compare to the other storage temperature groups at two days of the storage period.

There was no significant difference in the embryonic development due to the storage temperature. For the fertile hatchability, a significant interaction was noted between ST and SP, which showed that the ST had no effect in four days stored eggs, whereas 0°C group had a significantly lower fertile hatchability compare to the other ST (4, 8 and 16°C) groups when the storage period was extended (14 days).

It can be concluded that lower storage temperature (between 8 and 0°C) improved the albumen quality compared to control temperature (16°C).

Furthermore, hatchability was affected negatively only stored at 0°C during the first two days of 14 days storage in eggs from the older flock.

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Research and implications of eggshell temperature, CO₂ and O₂ during incubation

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In artificial incubation, hatchability and hatchling quality can be influenced by animal related factors, such as breeder age and by environmental factors, such as temperature and gas concentrations. In this presentation, focus will be put on eggshell temperature and on CO₂ and O₂ concentrations, during several phases of incubation.

To optimise eggshell temperature, the incubator temperature needs to be adjusted continuously to cope with changes in metabolic heat production by the embryo and by evaporative heat loss through the eggshell.

Metabolic heat production is in turn related to oxygen availability, which is related to the eggshell conductance.

Suboptimal incubator temperature in early incubation has been associated with retarded embryonic development (particularly at too low temperatures) and teratological effects, expressed in a higher percentage of malformed embryos (particularly at too high temperatures).

Suboptimal incubation temperature in late incubation has been associated with delayed hatching and lower robustness in later life (low temperature) and higher embryonic mortality, poor hatchling quality and higher risk on metabolic disorders in later life (high temperature).

Based on numerous studies the current advice for optimal eggshell

temperature is 37.5 to 38.0°C throughout incubation. CO₂ concentrations has been adjusted during different stages of incubation. Concentrations up to 4% or even higher have been used, but quite often adjustments in CO₂ concentrations are confounded with changes in relative humidity, temperature and O₂ concentration.

Recently, we have demonstrated that effects of CO₂ concentration up to 0.8% after day 8 of incubation had limited effects on embryonic development and hatchling quality appear as long as the other factors were maintained constant.

Higher CO₂ concentrations during late incubation might force hatching, with risks of premature hatching and poorer hatchling quality. Oxygen concentration is particularly of interest in relation to incubation on high altitude.

With lower oxygen pressure, O₂ availability for the embryo is reduced.

To maintain the balance between embryonic metabolism and oxygen availability, it has been suggested to reduce the eggshell temperature, because otherwise the risk of overheating in late incubation is considerable.

It can be concluded that incubation is a sensitive combination of temperature, CO₂ and O₂ and the hatchery manager can strongly influence the hatchability and hatchling quality by playing with these factors.

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Monitoring cooling during incubation of geese eggs

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Monitoring the eggshell temperature in the setter, with different measuring tools at critical points is widely used in chicken incubation, however the literature on the use of the same tools in incubation of goose eggs is limited.

One of the most critical points during incubation of goose eggs is cooling and spraying the eggs outside the setters during the second half of incubation, until 84.2-86.0°F temperature is achieved at the apex.

The aims of this field measurement were to check the applicability of Tinytag loggers, Braun thermometer and Flir thermal camera to monitor eggshell temperature of geese eggs, and to identify the most informative location on the trolley to use as reference point during cooling, preferring the side eggs as they are more accessible.

The eggs were measured on the 15th and 22nd day of incubation, at the beginning and at the end of cooling with all devices from 24 individual locations. Egg temperature was measured at the equator of the goose eggs, as common practice in chicken eggs.

However, the target temperature of cooling goose eggs during incubation has to be determined at the apex, so with Flir both equator and apex temperature were recorded.

The analysis of the database shows

a strong (>0.70) and significant ($P<0.05$) correlation between:

● Braun measurement at different locations after cooling and apex measurements.

● Flir measurements of eggs on different locations on the tray and on the trolley, both measured at the equator and apex of the eggs.

● Side egg apex measurements and temperature measured on the eggs in the middle of the tray at the beginning and the end of cooling at any location on the trolley.

Further analysis of the Flir data shows a strong (0.91) and significant ($P<0.001$) linear correlation between the measurements at the equator and at the apex of the egg. Linear regressions ($R^2=0.8142$) were determined using apex and equator measurements as dependent:

● Apex °F = 0.9609x Equator °F + 2.5437 and Equator °F = 0.8474x Apex °F + 15.771.

It can be concluded that the side eggs on the setter trolleys are suitable to monitor the cooling process if we measure them with Flir thermal camera either at the equator or apex, or use Braun at the equator and convert the value to the apex target using the equations above.

For Tinytag this is only valid to the top tray, but it can be caused by the tape we used to fix the sensor, so a re-measurement is required. ■

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Sexual sorting of chicken embryos using the HINTW gene in allantoic fluid

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The poultry industry is undergoing a revolution to end the male-chick culling, pushing for better solutions to meet its requirements: work with all eggs' colours, >98.5% accuracy, low disturbance and price, sexing before day seven, and >20,000 eggs/hour. Currently, the most accurate technique relies on PCR using allantoic fluid (AF) from day nine, despite being essential to apply sexing methods before embryo pain perception (day seven).

To date, PCR techniques applied

before day nine have focused only on tissue or blood cells, which is highly invasive and impacts hatchability. In our work, we established a PCR protocol based on a female-specific gene in the W-chromosome, HINTW, a highly conserved gene.

We incubated 80 ISA brown eggs (37.7°C, 50% RH) and extracted 100 µL of AF from day 6-9. Subsequently, purified genomic DNA was amplified with newly designed HINTW-specific primers and DMRT-1

gene primers as control.

First, the melting temperature confirmed the specificity of amplified sequences from female embryos by comparison with the HINTW template (82°C), which was 1.5°C higher in male samples.

Next, based on the cycle threshold, the HINTW sequence seemed to be amplified in 100% of the female and 94% of the male samples, the latter possibly related to cells from the mother hen or sequences with similarities at the Z-chromosome.

Notably, the difference between the mean cycle threshold of females

(mean ± standard deviation = 25.3±2.78) and males (mean ± standard deviation = 35±1.8) was statistically significant ($p<0.0001$).

A linear fitting of the cycle thresholds revealed a 100% sexing accuracy using AF samples before day seven, whereas the gene is amplified before cycle 31 in females.

As such, the newly developed assay allows minimally invasive sorting of chicken embryos as early as day six. Future work will focus on improving sampling strategy and decreasing assay time below the current 60 minutes. ■

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Effect of lighting regimes in automatic setters and hatchers on ostrich hatching performance

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The environment in which an embryo develops can have a lasting effect on well-being throughout its life. Traditionally, ostrich eggs are incubated in complete darkness and are only exposed to light when the incubator is opened. Exposing embryos to light during incubation benefitted hatchability, chick quality and hatching time in other poultry species.

The effect of light exposure during incubation on ostrich eggs was investigated with material collected from the pair-bred ostrich flock on the Oudtshoorn Research farm, South Africa.

A total of 4,042 eggs were set in electronic setters to investigate the effect of different lighting regimes (no light vs. light for 24 hours a day) cool white LED lighting with a colour temperature of 6,500 K (900lm) in setters and hatchers in a completely randomised design.

Of these eggs, 3,815 were transferred to hatchers at 35 days of incubation, where they were randomly assigned to a two (lighting regime in the setter) x two (lighting regime in the hatcher) factorial design. Lighting regimes in the setter reduced ($P<0.01$) early embryonic mortality (0.051±0.007 in eggs supplied with light vs. 0.082±0.007 in eggs incubated in darkness) and total embryonic mortality (0.264±0.015 in eggs supplied with light vs. 0.319±0.015 in eggs

incubated in darkness). When only eggs transferred to the hatcher were considered, a similar tendency was observed for late embryonic mortality (0.202±0.013 in eggs supplied with light vs. 0.223±0.013 in eggs incubated in darkness; $P=0.09$).

In these eggs, embryonic mortality was unaffected by lighting regime in the hatcher ($P>0.20$) as well as the interaction between incubation regime in the hatcher and the setter ($P>0.20$).

Evaporative water loss to 21 and 35 days of incubation was unaffected by the lighting regime ($P>0.05$). In contrast to the results for water loss, eggs subjected to continuous lighting during incubation hatched earlier than those incubated in complete darkness (respectively 42.51±0.04 vs. 42.73±0.04 days; $P<0.01$).

Expressed relative to a 24-hour day, these results imply that eggs subjected to lighting hatched, on average, 5.3 hours earlier than those incubated in darkness.

These results suggested that provision of continuous lighting during incubation benefitted embryonic survival in ostrich chicks.

The biology underlying this result is not explicitly known, but further studies where alternative lighting regimes are considered may lead to a better understanding of the mechanisms that may be involved. ■

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