

## The effect of temperature on the hatching performance of ostrich chicks, and its implications for artificial incubation in forced draught wooden incubators

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Batches of 60–65 ostrich eggs were incubated in electronic incubators at 36.0, 36.5 and 37.3°C. The proportion of live chicks hatched relative to eggs set at 37.3°C (22/65 = 0.338) was impaired ( $p < 0.05$ ) relative to those incubated at 36.0 and 36.5°C (respectively 38/60 = 0.633 and 36/60 = 0.600). Shell deaths were higher ( $p < 0.05$ ) in fertile eggs incubated at 37.3°C than in those incubated at 36.0°C (28/50 = 0.560 vs. 14/52 = 0.269 respectively). Temperatures were recorded in three horizontal (front, middle and back) and three vertical (top, middle and bottom) positions in 10 forced draught wooden incubators. In general, a linear temperature gradient was found from the front to the back of these incubators. Temperatures recorded in the top tray of the incubators were higher ( $p < 0.05$ ) than those recorded in the bottom and middle trays. Temperatures recorded in the front and back of the bottom tray were higher ( $p < 0.05$ ) than those measured in corresponding positions of the middle tray. Mean temperatures of 37°C and higher were measured at the back of the top and bottom trays, as well as in the middle of the top tray. The hatching performance of ostrich chicks is likely to be impaired in these positions. It was assumed that younger embryos are less susceptible to excessive heat than older embryos. It was thus suggested that freshly laid eggs should routinely be placed in those positions where temperatures exceeded 37°C in forced draught wooden incubators.

Stelle van 60–65 volstruiseiers is in elektroniese broeimasjiene teen 36.0, 36.5 en 37.3°C gebroei. Die proporsie lewende kuikens uitgebroei relatief tot eiers gepak by 37.3°C (22/65 = 0.338) was laer ( $p < 0.05$ ) as by eiers wat by 36.0 en 36.5°C gebroei is (38/60 = 0.633 en 36/60 = 0.600 onderskeidelik). Dopvrektes was hoër ( $p < 0.05$ ) by vrugbare eiers wat by 37.3°C gebroei is as by eiers wat by 36.0°C gebroei is (28/50 = 0.560 en 14/52 = 0.269 onderskeidelik). Temperature in 10 houtbroeikaste is aangeteken in drie horisontale (voor, middel en agter) en drie vertikale (bo, middel en onder) posisies. Daar was, in die algemeen, 'n lineêre temperatuurgradiënt van die voorkant na die agterkant van die broeikaste. Temperature in die boonste laai van die broeikaste was hoër ( $p < 0.05$ ) as in die onderste en middelste laaie. Temperature in die voor- en agterkante van die onderste laai was hoër ( $p < 0.05$ ) as temperature in die ooreenstemmende posisies van die middelste laai. Gemiddelde temperature van 37°C en hoër is in die agterkant van die boonste en onderste laaie gemeet, sowel as in die middel van die boonste laai. Die uitbroeiprestasie van volstruiskuikens sal waarskynlik in hierdie posisies

benadeel word. Dit is aangeneem dat jonger embryos minder vatbaar sal wees vir oormatige hitte as ouer embryos. Dit word dus aanbeveel dat eiers wat vars gelê is, in die posisies in houtbroeikaste waar temperature 37°C oorskry, gepak moet word.

**Keywords:** Ostrich eggs, temperature, hatching success, artificial incubation

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## Introduction

The physiological requirements of the developing avian embryo which can be controlled in artificial incubators are the correct temperature, optimal humidity, correct gaseous environment, proper turning angle and egg orientation. Of these, temperature is the single most critical factor. The effect of temperature on hatchability of avian eggs has been the subject of several reviews (Barott, 1937; Orlov, 1962; Landauer, 1967; Lundy, 1969; Swart, 1988; Ar, 1996; Deeming, 1997). These reports indicate that temperature variation within incubators should be minimised. Barott (1937), for example, concluded that the optimum temperature for the best quality chicks and hatchability was 37.8°C for chicken eggs and that temperature variation should not exceed 0.3°C.

Egg temperatures under natural incubation conditions, however, may vary considerably between species, suggesting some flexibility in temperature requirements (Romanoff, 1934; Huggins, 1941; Martin & Insko, 1935; Wilson *et al.*, 1979; Kühn *et al.*, 1982). The mean incubation temperature for ostrich eggs under natural conditions was reported to be approximately 35°C for the outer shell surface and 36°C for the nest air (Swart & Rahn, 1988).

Early research on temperature effects on incubation involved exposing developing embryos to temperatures above that considered optimal (Alsop, 1919; Orlov, 1962; Landauer, 1967; Lundy, 1969). Lundy (1969) concluded that the effect of high temperature on hatchability increased with increased temperature. Landauer (1967) concluded that the upper temperature limit for normal development was closer to the optimum temperature than the lower limit for normal growth. This suggested a greater sensitivity of the embryo to temperatures increased above optimum levels than to temperatures below optimum levels.

No research has been done so far with regard to the exposure of ostrich embryos to high temperatures. The effect of increased temperatures on the hatchability of ostrich chicks was therefore investigated. It is estimated that more than 70% of all ostrich eggs in the Klein Karoo area are incubated in forced-draught wooden incubators (B.F.Visser, Klein Karoo Agricultural Development Centre, P.O. Box 313, Oudtshoorn 6620, South Africa, pers. comm.). The temperature fluctuations in such forced-draught wooden incubators have also not been studied adequately. This study was undertaken to determine whether heat distribution was evenly spread within such incubators.

## Material and methods

The study on the effect of increased temperatures on hatchability was conducted in the incubation facilities of the Oudtshoorn experimental farm. Facilities and procedures for the collection and storing of eggs were described in the literature (Van Schalkwyk *et al.*, 1996; Van Schalkwyk, 1998). Three 75 egg capacity La National® electronic incubators were used with settings at 36°C, 36.5°C and 37.3°C. The relative humidity was kept at a constant 28% throughout. The number of eggs set per incubator ranged from 60 to 65.

Ten 200-egg forced-draught wooden incubators of the same design and manufacturer were monitored on five commercial farms. Air circulation and heat distribution in these incubators (height 2 m and width 1 m) are maintained by a paddle fan with air intakes on either side of the incubator, 50 cm from the bottom. The corresponding outlets are 50 cm from the top. Heat generation is by two

elements situated on the floor of the incubator. The thermostat probe is situated halfway between the middle and the top of the incubator. Temperature setting is usually done according to a hand-operated thermometer hanging in the middle against the door window.

In this study, temperatures were recorded with an electronic Henna thermometer in the top, middle and bottom trays of each incubator. Within each tray these recordings were done just behind the observation window, in the middle and at the back of the tray. The temperature measurements were taken individually after a period of 20 min was allowed for the equilibration of temperatures within incubators. The thermometer was placed in the incubator in each case, and the temperature was recorded through a closed glass window in the incubator door.

### Statistical analysis

Numbers of infertile eggs and chicks hatched were expressed as proportions of eggs set. Dead-in-shell eggs and eggs with malpositioned embryos (assessed according to the descriptions of Deeming, 1995) were correspondingly expressed as proportions of fertile eggs.  $\chi^2$  procedures were used to compare these proportions (Siegel, 1956), utilizing the Bonferroni correction to account for the fact that three comparisons were made.

The analysis of the temperatures recorded in each of the 10 wooden incubators was complicated by the fact that measurements were made in nine different locations within each incubator. The covariation arising from repeated measurements in the same incubator was accounted for by fitting the following mixed, linear model to the data (Harvey, 1990):

$$y_{ijkl} = \mu + B_i + h_j + v_k + h_j v_k + e_{ijkl}$$

where

$y_{ijkl}$  = an individual temperature recording,

$\mu$  = the overall mean,

$B_i$  = the random effect of the  $i$ 'th incubator,

$h_j$  = the fixed effect of the  $j$ 'th horizontal position ( $j$  = front, middle or back),

$v_k$  = the fixed effect of the  $k$ 'th vertical position ( $k$  = top, middle or bottom),

$h_j v_k$  = the two-factor interaction between horizontal and vertical position,

$e_{ijkl}$  = the random error term, used to test the other effects for significance.

The intraclass correlation derived from the between incubator variance component could be used to estimate the repeatability of temperature recorded within incubators.

### Results

The proportion of fertile eggs was independent of incubation temperature (Table 1). The proportion of live chicks hatched expressed relative to eggs set was lower ( $p < 0.05$ ) in eggs incubated at 37.3°C than in those incubated at lower temperatures. Expressed relative to fertile eggs, shell deaths were higher ( $p < 0.05$ ) in eggs incubated at 37.3°C than in those incubated at 36.0°C. There was a suggestion ( $p < 0.10$ ) for the incidence of malpositioned embryos to increase with increased incubation temperature, but significance could not be demonstrated.

The model fitted to the temperatures recorded in different positions in the wooden incubators accounted for 85.2% of the overall variation. A small, albeit significant ( $p = 0.042$ ), portion of this variation was associated with the covariation of the same incubator being measured repeatedly. The intraclass correlation (repeatability  $\pm$  SE) derived from the variance components was small ( $0.11 \pm 0.10$ ). The measurements were thus assumed to be sufficiently uncorrelated to validate analysis of

**Table 1** The hatching performance of ostrich eggs subjected to incubation at different temperatures

Parameter	Temperature			$\chi^2*$
	36°C	36.5°C	37.3°C	
Number of eggs set	60	60	65	
Number of fertile eggs	52	53	50	
Proportions relative to eggs set				
Fertile eggs	0.867	0.833	0.769	3.53
Chicks hatched	0.633 <sup>a</sup>	0.600 <sup>a</sup>	0.338 <sup>b</sup>	13.21
Proportions relative to fertile eggs				
Shelldeaths	0.269 <sup>a</sup>	0.321 <sup>a,b</sup>	0.560 <sup>b</sup>	10.37
Malpositions	0.231	0.283	0.440	5.57

\* Critical  $\chi^2$  ( $p = 0.05$ ) with 2 degrees of freedom = 5.99

<sup>a,b</sup> Proportions with different superscripts differ ( $p < 0.05$ ) in rows

variance procedures. Despite being significant, the interaction between horizontal and vertical position only accounted for 2.4% of the temperature variation, compared to 48.2% for horizontal position and 30.8% for vertical position.

There was a linear gradient from the front to the back of the incubators, successive means for the front, middle and back differing ( $p \leq 0.05$ ) in the top (36.6, 37.0 and 37.7°C respectively; SE = 0.07) and middle (36.0, 36.4 and 36.8°C respectively; SE = 0.07) (Figure 1). In the bottom, the mean temperatures recorded in the front (36.3°C) and middle (36.5°C) differed ( $p \leq 0.05$ ) from that recorded in the back (37.0°C). The mean temperature recorded in the top of the incubators was higher ( $p \leq 0.05$ ) than that recorded in the middle and bottom. In the back of the incubators, the mean temperature recorded at the bottom (37.0°C) was higher ( $p \leq 0.05$ ) than that recorded in the middle (36.8°C).

## Discussion and conclusions

Elevated incubator temperatures in the present study resulted in an increase in embryonic mortality and abnormalities in ostrich embryos. These results are consistent with previous studies on a variety of avian species. As incubation temperature was raised above the optimum level for maximum hatchability the mortality and the number of crippled and deformed chicks increased (Barott, 1937; Romanoff *et al.*, 1938). Leighton *et al.* (1964) found that heart and kidney enlargement and heart failure was the result of 11- and 12-day old embryos being exposed to 42.5°C for 1 week. An embryonic mortality of 50% or more resulted from exposure of 16-day broiler embryos to 43.3°C for 9 h. Romanoff *et al.* (1938) suggested, however, that mortality could be appreciably reduced by a slightly higher temperature of 0.25°C above optimum during the early part and a slightly lower temperature of 0.2°C below optimum during the latter part of incubation. These findings were in agreement with later reports (Romijn & Lokhorst, 1955; Tullett, 1990; Wilson, 1991), and presumably related to temperatures experienced by embryos during natural incubation. It was demonstrated by Tazawa *et al.* (1989) that the metabolic rate of early chicken embryos increased by approximately 10% more for every 1°C increase in incubator temperature than that of late embryos. This resulted in increased oxygen consumption at elevated incubation temperatures during early incubation. In the later stages of incubation, oxygen consumption was independent of incubation

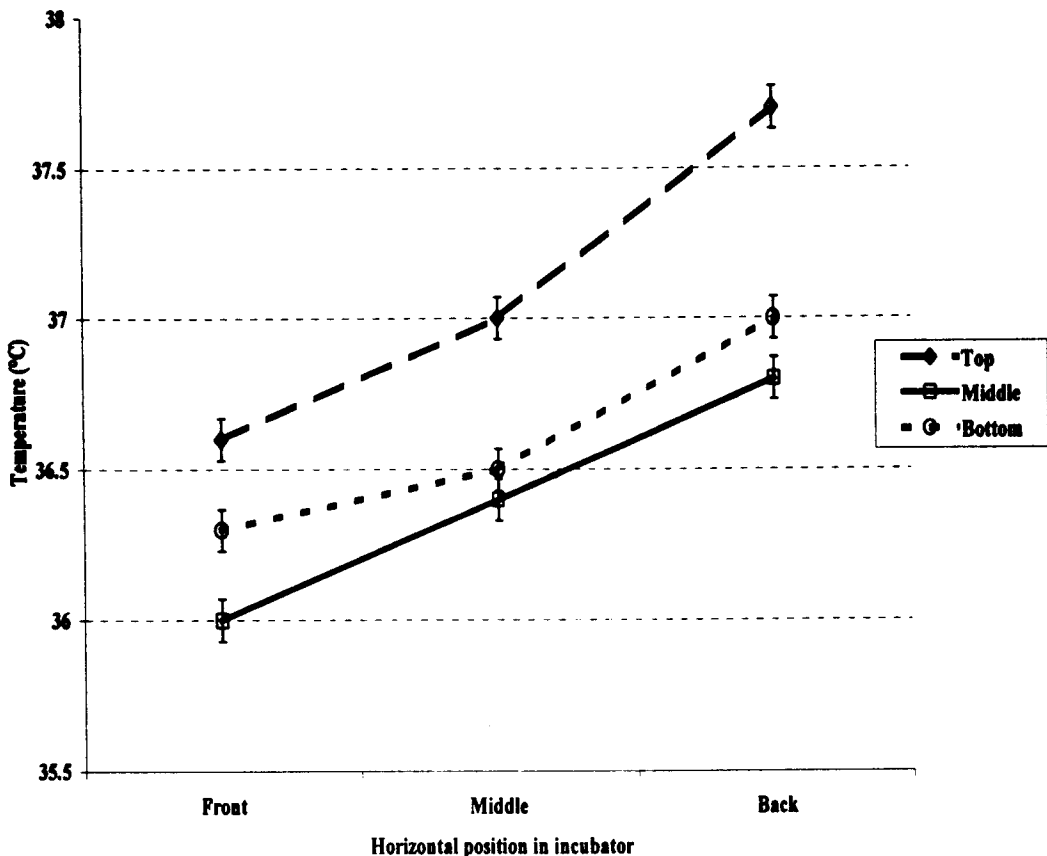
temperature. These results accorded with subsequent reports in the literature (Meir & Ar, 1990; Deeming & Ferguson, 1991). Hoyt *et al.* (1978) and Reiner & Dzapó (1995) studied the respiratory gas exchange and heat production in ostrich eggs incubated at 35°C and 36.3° respectively. In the former study, the increase in metabolic rate (as reflected by oxygen consumption) started after approximately 28 days of incubation, compared to approximately 23 days in the latter study. The rate of oxygen consumption per kg of egg was similar from 35 days of incubation onwards in both studies.

Under natural incubation conditions, the young embryo floats just below the brood patch, and enjoys a warmer temperature than the average (Ar, 1996). Overheating of embryos is a key problem in commercial incubation of any species of bird because metabolic heat production by the embryo during the second half of development can raise the egg temperature as much as 2°C above the incubator temperature (Deeming, 1997). Ar (1996) suggested that the overheating of the embryo in constant temperature incubators can be circumvented in modern single stage incubators (mostly used by commercial chicken and turkey hatcheries) by the changing of the incubator temperature as incubation progresses to offset changes in embryonic heat production. Decuypere & Michels (1992) and Ar (1996) suggested that the tolerance of the early embryo to increased incubation temperatures may be used in future to accelerate early development in artificial incubation, without subjecting the late embryo to heat stress. Deeming *et al.* (1993) implemented the principle of a descending incubator temperature during the incubation of ostrich eggs. No significant improvement in hatchability was observed, possibly because of using inappropriate control eggs.

Four of the ostrich chicks hatched at 37.3°C in the present study had eye abnormalities. Nilsen (1968) suggested that eye abnormalities were caused by circulation disturbances when chicken eggs were exposed to temperatures of between 40 and 42.2°C during the first six days of incubation. Ande & Wilson (1981) observed that chicken poults hatched after severe heat stress were not only weaker than contemporaries incubated normally, but that there was also an increased incidence of clubbed wry down and the exhibition of an unsteady gait. Barott (1937) found that chicks incubated at 38.9°C were smaller, less lively with more abnormalities than chicks incubated at 37.3 to 37.8°C. French (1994) found that the hatching success of turkey eggs incubated at 38.5°C was impaired relative to eggs incubated at 37.5°C. Overheated embryos had a mortality peak between 15 and 20 days and an increased mortality after 24 days of incubation. Overheated eggs were characterised by a high incidence of embryos with heads in the small end, excess albumen, ruptured yolk sacs, oedematous heads, eye cataracts and swallow down plumules. Ar (1996) suggested that heat conductance in and around the egg is an important factor determining embryonic temperature. An insulating air layer occurs around the egg in still air incubators. Because of this, the capability of these incubators to get rid of excess heat is of utmost importance. The magnitude of egg temperature changes in relation to incubator temperature is markedly reduced in forced-draught incubators, where the excess embryonic heat is removed by constant air circulation (Sotherland *et al.*, 1987). This is in contrast to natural incubation, where the brood patch temperature regulates egg temperature in chickens (Turner, 1991) and ostriches (Swart & Rahn, 1988). Measurements made throughout natural incubation indicated that egg temperature approaches that of the brood patch toward the end of natural incubation (Drent, 1970; Swart & Rahn, 1988; Handrich, 1989). Ar (1996) suggested that the chorio-allantoic blood circulation plays an important role in the dispersion of heat away from the brood patch, as well as dealing with the additional heat output generated by increased metabolic action of the older embryo. Both the brood patch and egg temperature at the end of incubation may thus serve to provide an indication of the comfort zone of the developing embryo (Ar, 1996). Embryonic temperature was estimated at 36 to 37°C, resulting in the recommendation to set incubator temperature at 36.5°C. This suggestion is supported by the present

results, where a decreased hatchability was only obtained when ostrich eggs were heated to a temperature exceeding 37°C.

The temperature gradient in the forced-draught wooden incubators in our study was probably due to ineffective heat distribution, as affected by the design and speed of the paddle fans. In practice, these incubators operated partly on the principles of still-air incubators. The increased temperatures in the top of the incubators can thus probably be related to natural convection, as the heat-generating elements are situated in the bottom of the incubators. The proximity to these elements probably resulted in the suggestion of higher temperatures recorded in the bottom of the incubators compared to those recorded in the middle (Figure 1). It is important to note that temperatures of 37°C and higher were recorded in three positions. Hatchability of artificially incubated ostrich eggs may be impaired at these temperatures. It is suggested that ostrich farmers should set older eggs in those parts of the incubator that do not exceed 36.5°C when small wooden incubators are used. If the temperature sensitivity displayed by ostrich embryos corresponds to those reported for chickens and turkeys, the setting of older eggs at extreme temperatures of 37°C and higher will potentially result in late embryonic deaths and an increased incidence of abnormalities. Younger eggs can possibly



**Figure 1** The interaction between vertical position (top tray or bottom tray) and horizontal position (front, middle or back) for temperatures recorded in 10 forced draught wooden incubators. Vertical bars on the line represent standard errors.

tolerate higher temperatures better, and should be set in the trays at the top and bottom, where higher temperatures were recorded. Further research is required to elucidate the effect of high temperatures on newly set ostrich eggs.

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