JWPR Journal of World's Poultry Research **2023, Scienceline Publication** J. World Poult. Res. 13(2): 191-198, June 25, 2023

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DOI: https://dx.doi.org/10.36380/jwpr.2023.21

Suitability of Inguinal and Axillary Sites for Temperature Measurement Using Digital Thermometers: A Comparison with Rectal Thermometry in Broiler Chickens

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Received: 14 March 2023 Accepted: 01 May 2023

ABSTRACT

Core body temperature is one of the physiological parameters that must be assessed during the monitoring of the thermic and or health status of broiler chickens. In this regard, cloacal thermometry is a standard method used for temperature measurement although it has many drawbacks. This study was conducted to explore the suitability of other anatomical sites for temperature measurement using a digital thermometer. This was a single-factor experiment that considered the anatomical site as the main factor with three levels (treatments), including cloacal (DTt_{cloacal}), axillary (DTt_{axillary}), and inguinal (DTt_{inguinal}) sites. Out of 84 broiler chickens, a total of 28 chickens were randomly selected for temperature measurement. The temperature was measured for each anatomical site, and the readings were analyzed using appropriate statistics. The cloacal site had the highest mean temperature ($41.40 \pm 0.17^{\circ}$ C), while the lowest mean value was observed for the axillary site $(41.12 \pm 0.19^{\circ}C)$. There was no significant difference between the mean cloacal and inguinal temperatures. The cloacal and inguinal temperature readings were significantly correlated. The results for the cloacal and inguinal temperature measurements revealed a non-significant bias. The agreement interval between these two methods was sufficiently lower than the maximum acceptable difference between the anatomical sites. Both cloacal and inguinal temperature measurements had similar median points. The results indicated an underestimation of the temperature readings for the axillary site compared to those of the other sites. In conclusion, this study has revealed that the application of a digital thermometer using the inguinal site gives temperature readings that are similar to those of the conventional cloacal method.

Keywords: Axillary site, Broiler chicken, Cloacal site, Digital thermometer, Inguinal site, Temperature

INTRODUCTION

Chicken remains one of the fastest growing, most prolific, and thus most numerous livestock species worldwide (Paputungan et al., 2020; Lawal and Hanotte, 2021). Its adaptability to diverse environmental conditions and potential for breeding improvement provide a unique resource for addressing the persistent food security challenge (Lawal and Hanotte, 2021; Nawaz et al., 2021). Despite their adaptation, the realization of the full productive and reproductive potential of chicken has been undermined by many factors, including diseases, heat stress, and inter alia (Paputungan et al., 2020; Ahmed et al., 2022). This is particularly the case among the chickens that evolved to suit the temperate regions, and they are exotic to the sub-Saharan region (Omodewu and Tiamiyu, 2021). For example, broiler chickens (broilers) are highly susceptible to heat stress and diseases compared to their indigenous counterparts (Omodewu and Tiamiyu, 2021; Ahmed et al., 2022). That notwithstanding, this breed is widely preferred for rearing because of its productive and meat quality characteristics (Chang, 2007; Ahmed et al., 2022). Given this, the broiler industry must improve its production methods, particularly in the areas of health, environment, and animal welfare (Chang, 2007).

Among the crucial requirements, there is a need to implement timeous on-farm strategies to minimize the resulting undue economic losses attributed to the negative effects of heat stress and diseases, among others (Nawaz et al., 2021; Ahmed et al., 2022). These primarily undermine the productive and reproductive performance in terms of egg and meat production, as well as the welfare and mortality rate of broiler chickens (Goel, 2020; Kim et al., 2021). Consequently, one strategy needed to minimize losses is the timely diagnosis of disease or febrile conditions in chickens through routine clinical examination (Sellier et al., 2014; He et al., 2022). The initial part of the clinical examination can employ many physiological parameters, such as body temperature, heart respiratory rate. serum biochemistry. rate. and reproductive performance (Abdisa, 2017; Omodewu and Tiamiyu, 2021). Under pathological or heat stress conditions, the temperature will change due to infection or emergence response (He et al., 2022). For example, some poultry diseases, such as avian influenza, fowl typhoid, and leg conditions, like bamboo foot, increase temperature. Accordingly, a timeous detection of this temperature increment facilitates disease diagnosis and prompt farm-decision making (He et al., 2022). Other than the health aspect, body temperature-taking is useful in ascertaining the thermoregulatory and reproductive status of broiler chickens (Sellier et al., 2014). However, the measurement of temperature is often bypassed due to the potential risks, such as stress and injury, to the chicken or handlers (Anderson et al., 2019).

There are many anatomical sites, including cloaca, tympanic membrane, axilla, eyes, wattles, forehead, inter alia, which have been studied and or used for temperature measurement in chickens (Anderson et al., 2019; Kim et al., 2021). Among these sites, the rectum or cloaca remains the conventional site that is widely used for measuring temperature in many species, including chicken (Pourjafar et al., 2012; Aluwong et al., 2017). Generally, the cloacal or rectal temperatures are closer (about 2°C) to the core body temperature compared to those of the other body sites, particularly the peripheral (shell) parts (Anderson et al., 2019). Similarly, many temperature sensors, such as mercury and digital thermometers, temperature loggers, and infrared devices, have been innovated or used; however, the mercury and digital thermometer types remain the standard measuring devices (Anderson et al., 2019; Bloch et al., 2020). Mercury and digital thermometers are applied per cloacal, and their readings are usually close to the core body temperature (Anderson et al., 2019).

Although mercury and digital thermometers are widely favored for their reliable accuracy, these devices can be a source of transmissible diseases or lead to rectal injury to the chicken (Pourjafar et al., 2012; Anderson et al., 2019; Kim et al., 2021). This is more so when these

devices are applied per cloacal by an unskilled handler. Less invasive evaluation of body temperature will result in a complete physical examination and provide a more accurate and appropriate clinical decision about the chicken's status (Anderson et al., 2019). Thus, it is necessary to identify and validate the less-risky anatomical sites in chickens, whose temperatures are closely in agreement with the cloacal or the core body temperature. Currently, digital thermometers are the most common temperature devices that are utilized in veterinary clinics; hence they are readily available for veterinary use (Abdisa, 2017; Kahng and Brundage, 2019). Additionally, they are time-saving, accurate, and user-friendly to clinicians (Anderson et al., 2019; Kahng and Brundage, 2019). However, some clinicians and many poultry farmers can easily injure or propagate infections among the chicken, when this device thermometer particularly is inappropriately applied per cloaca. Hence, this study was undertaken to assess the suitability of inguinal and axillary sites for temperature measurement using a digital thermometer in broiler chickens.

MATERIALS AND METHODS

Ethical approval

This study was conducted with approval (No. 1595-2021) by the institutional committee on animal research, University of Zambia, Lusaka, Zambia. Furthermore, the feeding, housing, handling of the chickens, and experimentation were carried out in compliance with the guide for the care and use of agricultural animals in research and teaching (ASAS, 2020).

Study location

This study was conducted at the Poultry farm owned by the Department of Animal Science, School of Agricultural Sciences, University of Zambia, Lusaka, Zambia, in January 2023. Zambia is located at latitude S 14° 20' 0" and longitude E 28° 30' 0" (GeoNames geographical database Google Earth-January, 2023). Furthermore, this country lies in the tropics, specifically in the Southern-African region.

Experimental chicken and management

A total of 100 broiler chickens (one-day-old) of the strain Cobb 500 were procured for this study. The average weight of these chickens at the time of procurement was 37.30 g. These chickens were raised in the poultry unit that belonged to the Department of Animal Science. In terms of their management, all the chickens were

vaccinated against gumboro, Newcastle, and marek's diseases, following an established vaccination schedule at the field station. A 17 light and 7 dark photoperiods was subjected to the chicken, with the lighting provided from 05:00 to 22:00 hours. The ambient temperature was maintained at about 32°C, using infrared lamps, for 21 days. Physical observation and ambient temperature measurement with liquid-in-glass thermometers (Easy-Read[®], Thomas Scientific, New Jersey, USA) were employed to monitor heat stress among the chicken. The ventilation in the poultry house was provided using a wire mesh, of which the control of temperature and humidity levels was not comparable with an automated system. With regard to feeding, three different diets were used, including broiler starter, broiler grower, and broiler finisher. The type of diet provided was dependent on the age of the broiler chicken. Additionally, all the feeds were sourced from Entrust stock feed Limited, Lusaka, Zambia. The stated (provided) nutrition information for the feeds (diets) used is presented in Table 1. All the diets were fortified with premix containing a range of vitamins, minerals, and amino acids at an undisclosed rate. In this case, the effect of individual components of the premix on the physiology or performance of the chicken may not be pinpointed. Nevertheless, all the chickens were allowed to feed ad libitum, with the feeders routinely refilled at 10:00 and 15:00 hours. Similarly, these chickens had unrestricted access to clean water for drinking. About 10% mortality rate was recorded; at the time of the experiment, a total of 84 chickens were physically healthy and available for the trial.

 Table 1. Chemical nutritional characteristics of feed(s)

 provided to broiler chickens at 55 days of age

| Nutrition composition | Starter (1-12 D) | Grower (13-23 D) | Finisher (24-55 D) |
|--------------------------------|---------------------|---------------------|-----------------------|
| Crude protein (min %) | 20.0 | 19.0 | 19.0 |
| Moisture content (max %) | 12.0 | 12.0 | 12.0 |
| Crude fiber (max %) | 5.0 | 5.0 | 5.0 |
| Phosphorus (available %) | 0.6 | 0.5 | 0.4 |
| Calcium (max %) | 1.0 | 1.2 | 0.85 |
| Lysine (min %) | 1.23 | 1.0 | 1.04 |
| Total methionine (min %) | 0.5 | 0.4 | 0.39 |
| Metabolizable energy (kcal/kg) | 2900 | 3100 | 3200 |

D: Days; min: Minimum; max: Maximum; %: Percentage

Experiment design

This was a cross-sectional study that employed a single-factor experimental design. The study considered anatomical site as the main factor, which had three-factor levels (as treatments), including inguinal, axillary, and

cloacal locations of the chicken. The measurement of temperature was done when the chickens were 55 days of age, with an average weight of 2.80-3.21 kg. A total of 28 physically-healthy chickens were randomly selected for temperature measurement. For each chicken, temperature measurements, namely DTt_{cloacal}, DTt_{axillary}, and DTt_{inguinal}, for inguinal, axillary, and cloacal sites, respectively, were conducted, and DTt_{cloacal} was considered as a control. The sampling strategy was based on the previous procedure (Anderson et al., 2019). The order for the measurement of sites was determined by means of a simple random assignment, which used folded papers bearing the names of each site. Additionally, the study followed all the precautions necessary to minimize the potential effects of psychogenic fever or hyperthermia. Temperature measurement was conducted in the morning (8:00-11:45 hours) and before feeding the chicken. A functional veterinary digital thermometer (DT; GB Kruuse digital thermometer, New Taipei City, Taiwan) was used to measure the temperature. The measuring range and the resolution of the DT were 30.0-43.9°C and 0.1°C, respectively.

The measurement of temperature

Before the measurement of temperature, a chicken was physically restrained (Nash, 1976). The procedure for the DT application was based on the manufacturer's directions, with some additions depending on the site. With the chicken restrained, the DTt_{cloacal} was obtained by inserting a sterile DT into the cloaca (approximately 2 cm), gently and at a slight angle dorsally, towards the cloacal wall. The device was left in position for 20-60 seconds until the degree sign stopped flashing, and an alarm went off before the DTt_{cloacal} readings were recorded (°C). The DT was properly disinfected using isopropyl alcohol before temperature-taking for each chicken to prevent cross-contamination and/or disease transmission. A similar procedure was followed to obtain the DTt_{axillary} readings. Here, the DT probe was snugly placed between the chicken's breast and biceps, approaching from the cranial aspect and aiming dorsally toward the shoulder joint. Similarly, the DTt_{inguinal} was measured by snugly positioning the DT probe between the chicken's breast and thigh, aiming dorsally/deep into the hip joint. A double temperature measurement was performed for each site, and the average of the two measurements was recorded as a single datum. Furthermore, the same researchers took the temperature to minimize human errors and undue stress on the chicken. During the experiment, the temperature and

humidity ranged from 26.8 to 27°C and 72 to 75%, respectively.

Data analysis

Data were analyzed in the Statistical Package for Social Scientists (SPSS[®] IBM 26 version, USA) using descriptive statistics, including the selected central tendency, dispersion, and distribution measures. The temperatures taken from different anatomical sites were analyzed by a One-way ANOVA test using the General Linear Model, a univariate analysis procedure. The following model was used to determine the main effect of the treatment factor;

$$Y_{ij} = \mu + A_i + e_{ij}$$

Where, Y is the dependent variable representing a value of the measured parameter, u signifies the overall mean, A_i refers to the effect of the treatment factor (site) with three levels (i = cloacal, inguinal, and axillary sites), e_{ij} defines the random error term. Tukey's HSD (post-hoc) test was employed to obtain the mean treatment pair(s), where significant differences existed. The correlation between the sites (by DTt readings) was determined using a Pearson's correlation test. In all cases, significance was taken at a level of p < 0.05. The Bland-Altman plot was employed to analyze the agreement between the two temperature measurements that exhibited the highest correlation coefficient.

RESULTS

Mean temperature readings for different anatomical sites

The mean temperature readings taken by a digital thermometer (DTt), including $DTt_{cloacal}$, $DTt_{axillary}$, and $DTt_{inguinal}$, for the different anatomical sites, namely cloacal, axillary, and inguinal, respectively, are presented in Table 2. The mean $DTt_{cloacal}$ was the highest among the observed treatments, while the mean $DTt_{axillary}$ was the lowest. Analysis of variance revealed a main effect of the anatomical site of broiler chickens on the mean DTt readings, F(2, 81) = 12.75, p < 0.05, $\eta_p^2 = 0.339$. The post hoc analyses (Tukey's HSD test) indicated that the mean $DTt_{cloacal}$ was higher than that of the $DTt_{axillary}$ (p < 0.05), but did not differ significantly from the mean $DTt_{inguinal}$ (p > 0.05). Additionally, the mean $DTt_{axillary}$ (p < 0.05).

| Table 2 | 2. The | mean | temperature | readings | from | different |
|---------|----------|----------|---------------|------------|---------|-----------|
| anatomi | cal site | es of bi | oiler chicken | s at 55 da | ys of a | age |

| Variable | DTt readings | | |
|-----------------|--------------------|------------------------------------------------|--|
| Anatomical site | Mean ± SD (°C) | Difference from DTt _{cloacal} (°C) | |
| Cloacal | 41.40 ± 0.17^{a} | - | |
| Axillary | 41.12 ± 0.19^{b} | 0.28 | |
| Inguinal | 41.39 ± 0.19^a | 0.01 | |

DTt: Temperature readings by a digital thermometer, SD: Standard deviation, °C: Degrees Celsius. ^{a,b}Different superscript letters within the same column indicate a significant difference (p < 0.05).

Correlation between temperature readings and different sites

Results from multiple correlation analyses of the temperature readings, including $DTt_{cloacal}$, $DTt_{axillary}$, and $DTt_{inguinal}$, are presented in Table 3. The $DTt_{cloacal}$ had a relationship with both the $DTt_{axillary}$ and $DTt_{inguinal}$ readings. The highest correlation was observed between the $DTt_{cloacal}$ and $DTt_{inguinal}$ (r(28) = 0.90, p < 0.05). Among the treatments, the correlation between the $DTt_{cloacal}$ and the $DTt_{axillary}$ readings was the lowest (r(28) = 0.758, p < 0.05).

 Table 3. Correlation between temperature readings taken at different anatomical sites of broiler chickens at 55 days of age

| | DTt _{cloacal} | DTt _{axillary} | D Tt _{inguinal} |
|---------------------------------|------------------------|--------------------------------|---------------------------------|
| DTt _{cloacal} | 1 | | |
| DTt _{axillary} | 0.758** | 1 | |
| D Tt _{inguinal} | 0.900** | 0.823** | 1 |

DTt: Temperature readings by a digital thermometer, correlation coefficient 0.00-0.10: Negligible, 0.10-0.39: Weak, 0.4-0.69: Moderate, 0.7-0.89: Strong, 0.9-1.0: Very strong correlation, Correlation coefficient with an asterisk (**): statistical significance at p < 0.05

Comparison between the cloacal and inguinal temperature measurements

The Bland-Altman plot of the temperature readings (Figure 1) presented a relationship between the $DTt_{cloacal}$ and $DTt_{inguinal}$ measurement methods. This plot displays the points that were within the 95% confidence interval (CI) of the distributed data; the mean of $DTt_{cloacal}$ and $DTt_{inguinal}$ and the difference between the two measurements were used to generate the plot. The results revealed that, with a 95% CI, many paired DTt readings were not significantly different (p > 0.05).



Figure 1. Bland-Altman plot visualizing the agreement between $DTt_{cloacal}$ and the $DTt_{inguinal}$ readings obtained from broiler chickens in Zambia



Figure 2. The Box-and-Whisker plot demonstrating the relative distribution of temperature readings for each anatomical site

The distribution of temperature readings for each anatomical site

The pictorial presentation of the relative dispersion and distribution of the DTt readings for each anatomical site, including the cloaca, axilla, and inguinal, is shown in Figure 2. The DTt_{inguinal} range was generally larger than that of the DTt_{cloacal}. The DTt_{cloacal} and DTt_{inguinal} generally had similar medians, with most DTt readings tightly clustered around the median. Generally, the analysis showed an underestimation of the values (DTt_{axillary} readings) for the axillary site compared to those of the other sites. There was considerable skewing to the lower temperature limits for the case of DTt_{axillary} readings.

DISCUSSION

Cloacal temperature taking has been conventionally regarded as the "standard/gold method" for core body temperature assessment in chickens because of its physiological accuracy and repeatability (Anderson et al., 2019; Reis et al., 2022). Nevertheless, some drawbacks associated with cloacal temperature taking, such as injuries and disease transmission among chickens, have recently compelled researchers to search for less invasive and safer temperature sensors (Anderson et al., 2019; Kim et al., 2021). Temperature measurement is often bypassed for fear of the potential risks associated with cloacal thermometry (Anderson et al., 2019). So far, many advanced temperature sensors have been studied and or used, including infrared thermometers, infrared cameras, inter alia (Sellier et al., 2014; Bloch et al., 2020). However, many of these are expensive for an average chicken farmer or generally not user-friendly, and some are not as accurate as the case is for cloacal-based devices (Cândido et al., 2018; Bloch et al., 2020). Cognizant of this, the current study found a non-invasive temperature measurement method or site whose readings agree with the conventional cloacal temperature.

Furthermore, the observed main effect of the measurement site on the mean temperature value supports an earlier study that confirmed the dependency of a thermometer's readings on the point of skin used (Abioja et al., 2019; Reis et al., 2022). In addition, the absence of a significant difference between the cloacal and inguinal temperatures indicated the proximity of the latter to the core body temperature in broiler chickens. This disagrees with the earlier study findings that reported significantly lower temperatures of most skin points, compared with the cloaca (Abioja et al., 2019; Reis et al., 2022). This may be

attributed to the type of thermometer used and the proximity of the inguinal site to the liver and other innards that are metabolically very active. Additionally, the disposition of the thigh and breast parts that make up the inguinal site, as well as the feathers around this site, perhaps minimized the excessive heat loss (Saeed et al., 2019).

With regard to the axillary site, the current study findings varied from the previous study that showed similar mean cloacal and axillary temperatures (Abioja et al., 2019). This disparity may be attributed to the type of thermometers used. Nevertheless, the current mean temperature values for both the axillary and inguinal sites were within the reported normal chicken body temperature range of 40.6-43.0°C (Reece, 2009). This may be attributed to the fact that these sites are mostly kept covered, given their anatomical predisposition, with less heat loss to the environment. On the other hand, the similarity between the observed temperatures and the reported normal range points to the fact that the production performance of the study chicken was not critically affected by disease or environmental conditions, which agrees with the findings of an earlier study conducted in Brazil (Reis et al., 2022).

The correlation between the cloacal and inguinal temperature in the current study revealed the potential use of the latter for core body temperature assessment using digital thermometers. From a previous report, the cloacal temperature was found to correlate well with the core body temperature (Candido et al., 2020). The observed positive and very strong correlation between the cloacal and inguinal temperatures qualifies the inguinal site for the digital thermometer application in broiler chickens. Given the observed non-significant bias between the cloacal and inguinal temperature measurements (p > 0.05), the current results of the Bland-Altman analysis confirmed an agreement between these two temperature measurement methods. Moreover, the agreement interval is sufficiently narrow, compared to the maximum acceptable difference $(\pm 0.2^{\circ}C)$ for temperatures between two anatomical sites (Fulbrook, 1993). On the other hand, although the current correlation between the cloacal and axillary temperatures was significantly strong (p < 0.05), which finding is consistent with the previous reports, their observed mean difference renders the axillary site inappropriate for the digital thermometer application (Abioja et al., 2019).

The observed similar median points and close distribution of readings within the lower and upper quartiles for both cloacal and inguinal temperatures attest to observations on their means and correlation coefficient.

To cite this paper: Abigaba R and Sianangama PC (2023). Suitability of Inguinal and Axillary Sites for Temperature Measurement Using Digital Thermometers: A Comparison with Rectal Thermometry in Broiler Chickens. J. World Poult. Res., 13(2): 191-198. DOI: https://dx.doi.org/10.36380/jwpr.2023.21

These observations largely indicate the potential and reliability of the inguinal temperature measurement method for broiler chickens' thermal status monitoring. Moreover, inguinal thermometry is devoid of the reported rectal thermometry drawbacks, namely invasiveness, disease transmission, and potential temperature variations attributed to fecal masses, muscle tone, digestion, inter alia (Pourjafar et al., 2012; Kim et al., 2021). On the other hand, the findings of the current study on the axillary temperature agree with the report of a recent study which indicated lower skin temperature readings compared to cloacal temperature (Reis et al., 2022).

The current findings reveal the potential of inguinal thermometry for monitoring the temperature or health of broiler chickens, more so among smallholder farmers. Moreover, early identification of the health status of chickens contributes to the timeous decision-making in smallholder or commercial poultry farms (Ahmed et al., 2022; Reis et al., 2022). Temperature estimation is used as a physiological marker for chickens' thermic status during disease, reproductive status, and heat stress assessments (He et al., 2022; Reis et al., 2022). For example, disease infections in chickens cause abnormal temperature, avian influenza, fowl typhoid, inter alia, and fever (He et al., 2022). Hence, this clinical symptom helps in the evaluation or identification of the sick chicken. It is noteworthy that changes in environmental or farm temperature can also influence the body temperature of broiler chickens (Ahmed et al., 2022). The high temperature increases susceptibility to diseases and compromises the productivity of the chicken (Ahmed et al., 2022). The application of inguinal thermometry, which is generally safer and more user-friendly than the cloacal temperature measurement method (Anderson et al., 2019), will likely benefit the poultry subsector. Nevertheless, whether the inguinal thermometry gives similar results among younger broilers or other chicken breeds remains unclear.

CONCLUSION

A timeous body temperature taking in chickens, including broilers, is crucial as part of their initial clinical examination to confirm disease status, heat stress, and productive and/or reproductive status. This study has revealed that applying a digital thermometer using the inguinal site gives as accurate temperature readings as the case with the conventional cloacal method/site. Hence the inguinal temperature is recommended, as a reliable physiological marker, for the body temperature assessment in broiler chickens. The inguinal site is non-invasive and user-friendly to clinicians and smallholder chicken farmers. The limitations of the current study included the use of one breed, small sample size, and one age (adult) group of the chicken. Thus, future studies intended to replicate the current research must consider these factors for the generalization of the findings.

DECLARATION

Acknowledgments

All authors acknowledge the support from all Field Station Staff and the Department of Animal Science. The authors also applaud Dr. Oswin Chibinga and Dr. Benson H. Chishala, the Head of the Department of Animal Science and Dean School of Agricultural Sciences, respectively, for their administrative and technical support.

Funding

This study received no external funding.

Authors' contribution<mark>s</mark>

Pharaoh C. Sianangama designed and supervised the study and reviewed the manuscript, and Rubaijaniza Abigaba conceived and designed the study, collected and analyzed data, and wrote the manuscript. Both authors approved the final manuscript for publication.

Conflict of interests

The authors declare no conflict of interest.

Ethical consideration

The authors declare that this manuscript is original and has not been submitted elsewhere for possible publication. The authors also declare that the data used/presented in this manuscript has not been fabricated.

Consent to publish

Both authors informed their consent before the study conduction.

Availability of data and materials

The authors will provide data of the present study in case of reasonable request.

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