Reticulo-rumen temperature as a predictor of estrus in dairy Gir heifers

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Summary

Reticulo-rumen sensor data were used for estrus prediction in Gir heifers. Temperature-sensing reticular boluses were orally administered to 60 heifers, and the ruminal temperature (RRT) and animal activity were recorded every 10 minutes. The cows were synchronized using a hormonal protocol. Logistic regression with binary distribution was employed in the analysis to determine the response variables: Yi = 1 for presence, or Yi = 0 for the absence of estrus; using the GLM function of the R software. Two models were adjusted for data analysis: Model 1 considered only the effect of RRT as a dependent variable, while Model 2 included the effects of RRT and animal activity. The AIC inferred that Model 2 was the best fit regarding the data. The area under the ROC curve was used as an indicator of the model's ability to discriminate between non-occurrence (0) and occurrence (1) of estrus. Regarding the probability of correct estrus identification, Y2 retained a value of 0.64 and Y1 of 0.62. These values are considered low and evidence that the correct identification capability of estrus is reduced. In other words, the use of other approaches to determine estrus occurrence is required in order to improve model calibration and/or develop more robust methodologies.

Keywords: estrus, livestock precision farming, reproduction, ruminal boluses, zebu cattle.

Introduction

The Gir breed is an important genetic resource for milk production in the tropics. It is widely crossbred with Holstein cattle given its resistance to adverse conditions in the tropical environment. The animals of this breed are sexually late and have short and often nocturnal cycles, hindering the identification of estrus in commercial herds (Pires *et al.*, 2003).

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Precision technologies, such as temperature sensors, can assist in estrus identification. The body temperature increases in 0.4ºC during the estrus of cows (Cooper-Prado *et al*., 2011; Adams *et al*., 2013). Temperature-sensing reticulo-rumen boluses can be employed to continuously monitor body temperature variations of animals during time, aiding in the detection of estrus (Sieves *et al*., 2004), calving (Costa Jr. *et al*., 2016), and other physiological events. The adoption of precision technologies enables to phenotype traits related to the sexual precocity of zebu females and to take measurements that are associated with postpartum reproductive activities, providing significant information for the selection of these traits in zebu cattle.

Most of the studies that utilized ruminal sensors to predict estrus were conducted using taurine breeds, which exhibit distinct estrus behavior when compared to zebu cattle (Pires *et al*., 2003). Therefore, the prediction equations developed for taurine breeds may result in different performance values than zebu cattle, leading to low efficiency in the detection of estrus in these animals.

The objective of the present study was to utilize the data obtained from reticulo-rumen sensing for the prediction of estrus in Gir breed heifers.

Material and methods

Temperature-sensing reticular boluses (TX-1442, Smaxtec Animal Care, Austria) were orally administered to cows using a custom bolus gun in the ruminal region of 60 Gir dairy heifers. The sensors retain a temperature measurement range of 0°C to 50°C and an activity index of 0 to 100%. The reticulo-rumen temperature variation (RRT) and animal activity were recorded every 10 minutes.

The RRT data were collected using a telemetry system, through an antenna, with a reading range of 30 meters. The antenna was allocated in the pen where it gathered the RRT data from the animals while they were handled. The readings were sent via radio signal and accessed using the Smaxtec Messenger online software (Smaxtec Animal Care, Austria).

After a 45-day adaptation period, the heifers were synchronized regarding estrus using the following hormonal protocol: D0 - Application of the Intravaginal Progestogen implant (P4) + benzoate (BE); D7 - Application of Prostaglandin (PGF); D9 - Application of Cypionate of Estradiol (CP) + Equine Chorionic Gonadotropin (ECG) + removal of the implant; D11 - Realization of Artificial Insemination + use of the Gonadotropin-releasing Hormone (GnRH).

Behavioural observations were made every 10 minutes during 48 hours, starting two hours after the removal of the Intravaginal Progestogen implant. The sexual behaviour related to estrus consisted of mounting acceptance. Ultrasound imaging was used to identify animals that ovulated after 60 hours following implant removal. A total of 59 heifers ovulated.

During the statistical analyses, all temperature data inferior to 37.72°C were discarded, given the low temperature is associated with water consumption (Bewley *et al*., 2008). A total of 19,660 records regarding stored information on reticulo-rumen temperature and activity were used.

In order to evaluate the optimal model for prediction capability, the animals were divided into five groups (four groups of 12 cows, and one with 11) at random. The analyses were repeated five times and, in each round, a different group was considered as a validation population. Data analysis relied on the use of a logistic regression model with binary distribution to determine the response variables (Yi = 1 for presence, or Yi

= 0 for the absence of estrus), using the GLM function of the R software. Two models were adjusted for data analysis: Model 1 included only the RRT effect as a dependent variable, while Model 2 involved the effects of RRT and animal activity.

Additionally, the response variable (presence or absence of estrus) was defined by two distinct forms (Y1 and Y2). In Y1, the presence of estrus (1) was considered during the interval between 32 and 45 hours after the removal of the progesterone implant, assuming that zebu females have an average estrus duration of 13 hours (Pires *et al.*, 2003). In turn, in Y2, the presence of estrus was associated with the period from the first to the last accepted mounting, based on behavioural observations. Models 1 and 2 were compared trough Akaike's information criteria (AIC) with Y1 or Y2 as variable responses. The best model was used to determine the occurrence of estrus in the validation population, and the prediction accuracy was calculated as the correct number of predictions considering the total number. The ROCR package of the R software was employed to construct ROC curves, which are used to evaluate the model's ability to predict estrus. When the area under the curve is closer to 1.0, the probability of the occurrence of true-positive results is greater.

Results and discussion

The observations showed that estrus occurred between 30 and 48 hours after implant removal. Diskin *et al.* (2002) stated that the synchronization, combining progesterone and PGF, led to the occurrence of a considerable number of cows in estrus between 36 and 60 hours after the removal of the progesterone implant. The earlier observation of the receptivity period may be attributed to the application of ECG at the time of implant removal, which may have anticipated the occurrence of estrus (Baruselli *et al.*, 2008).

The AIC values indicated that Model 2, which associated the ruminal temperatures with the activity of the animals, provided better fit than Model 1 (only RRT) regarding Y1 and Y2 (Table 1). The inclusion of activity in the model improved the adjustment. Some authors obtained similar results, in which increased activity of the animals was observed during the estrus when compared to other stages of the reproductive cycle (Lovendahl & Chagunda, 2010).

Yoshioka *et al.* (2010) observed a positive correlation between increased activity with estrus and ovulation in beef cattle. When using body temperature sensors, previous studies showed that the ruminal temperature of the animals increased in 0.3°C to 0.7°C during the estrus (Bailey *et al.*, 2009; Cooper-Prado *et al.*, 2011; Adams *et al.*, 2013). These results corroborate with the present study, in which the model that considered the two variables (temperature and activity) exhibited optimal fitness.

Table 1. AIC values employing training populations and estrus period or mounting acceptance as response variables, considering reticulo-rumen temperature (RRT) or reticulo-rumen temperature+activity (RRT+A) as independent variables.

Training population	Estrus Period Model 1 RRT	Estrus Period Model 2 RRT+A	Mounting acceptance Model 1 RRT	Mounting acceptance Model 2 RRT+A
1	17251	17014	11686	11469
2	17162	16951	10823	10737
3	17552	17265	11087	10862
4	16849	16564	10821	10655
5	17685	17428	11676	11521
Mean	17300	17044[1]	11219	11049*

[1]Best values according to the AIC.

Considering Model 2 individually, the validation population data was used to test the prediction ability of the model. The area under the ROC curve (AUC) is an indicator of the model's ability to discriminate between the non-occurrence (0) and occurrence (1) of the event (Table 2). An AUC that was higher than 0.50 indicated that the discrimination was not at random. Thus, regarding Y1, the probability of correctly identifying the estrus (true-positives) was 0.62 on average, while the identification probability of false-positives was 0.38. For Y2, the probability of correctly identifying the estrus was slightly improved (0.64 on average), and the probability of false-positives was of 0.36. These values are considered small and demonstrate how the correct identification capability of estrus remains reduced. However, the better definition of the response variable, such as the use of hormonal dosages for the determination of estrus occurrence, is believed to result in an enhanced calibration of the model for the prediction of estrus in zebu cattle.

Table 2 - Parameter estimates and standard errors (between brackets), prediction accuracy, and area under the ROC curve of the estrus period (Y1) or mounting acceptance (Y2) traits regarding Model 2.

Training population	Estimates*			Accuracy	Area under ROC curve
	Intercept	RRT	Activity		
		Estrus Period (Y1)			
1	-11.00 (1.66)	0.24 (0.04)	0.045 (0.003)	0.77	0.65
2	-15.26 (1.72)	0.35 (0.04)	0.042 (0.003)	0.78	0.64
3	-17.09 (1.70)	0.39 (0.04)	0.050 (0.003)	0.73	0.60
4	-13.28 (1.71)	0.29 (0.04)	0.049 (0.003)	0.73	0.62
5	-16.10 (1.66)	0.39 (0.04)	0.0450 (0.003)	0.76	0.58
		Mounting Acceptance (Y2)			
1	-29.25 (2.17)	0.68 (0.05)	0.053 (0.003)	0.92	0.64
2	-29.68 (2.36)	0.69 (0.06)	0.036 (0.004)	0.87	0.72
3	-43.40 (2.316)	1.03 (0.06)	0.058 (0.004)	0.86	0.52
4	-29.70 (2.29)	0.68 (0.06)	0.049 (0.004)	0.86	0.68
5	-31.65 (2.19)	0.74 (0.05)	0.045 (0.003)	0.89	0.66

**P* values < 2e-16.

The accuracies for Y1 and Y2 were 0.75 and 0.88 in average, respectively (Table 2). The lower accuracy of Y1 can be attributed to the subjectivity of the variable since Y1 definition was based on the expected range in time according to the hormonal protocol, and Y2 was based on the observed behaviour. The artificial induction of luteal regression in reproductive protocols, with the use of prostaglandin, can result in lower proportions of synchronization than expected (< 65%), and the estrus can occur in up to 130 hours following treatment (Viana *et al.*, 1997). On the other hand, mounting acceptance is the leading indicator of estrus in zebu cows (Lamothe-Zavaletha *et al.*, 1991).

The model that used the information regarding ruminal temperature and animal activity displayed improved fitness.

The probability of the detection of estrus (true-positives) was similar for both response variables. However, the values indicated the occurrence of errors in estrus detection. Therefore, it is necessary to use other approaches to improve the calibration of the model or to use more robust methodologies.

Acknowledgements

The authors thank to Epamig, CNPq, CAPES, and FAPESP (2015/241743) for financial support

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