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The value of reproductive tract scoring as a predictor of fertility and production outcomes in beef heifers

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ABSTRACT: In this study, 272 beef heifers were studied from just before their first breeding season (October 15, 2003), through their second breeding season, and until just after they had weaned their first calves in March, 2005. This study was performed concurrently with another study testing the economic effects of an estrous synchronization protocol using PG. Reproductive tract scoring (RTS) by rectal palpation was performed on the group of heifers 1 d before the onset of their first breeding season. The effect of RTS on several fertility and production outcomes was tested, and the association of RTS with the outcomes was compared with that of other input variables such as BW, age, BCS, and Kleiber ratio using multiple or univariable linear, logistic, or Cox regression. Area under the curve for receiver operating characteristic analysis was used to compare the ability of different input variables to predict pregnancy outcome. After adjustment for BW and age, RTS was positively associated with pregnancy rate to the 50-d AI season ($P < 0.01$), calf weaning weight ($r = 0.22$, $P < 0.01$), and pregnancy rate to the subsequent breeding season ($P < 0.01$), and negatively associated with days to calving ($r = 0.28$, $P < 0.01$). Reproductive tract scoring was a better predictor of fertility than was Kleiber ratio and similar in its prediction of calf weaning weight. It was concluded from this study that RTS is a predictor of heifer fertility, compares well with other traits used as a predictor of production outcomes, and is likely to be a good predictor of lifetime production of the cow.

Key words: age at puberty, beef heifer, Kleiber ratio, pregnancy rate, reproductive tract scoring, weaning weight

INTRODUCTION

In the past, conformation, BW, BCS, and calculated indices such as Kleiber ratio (KR; Kleiber, 1947; Scholtz and Roux, 1988) have been used to select heifers for breeding. However, selection based on age at puberty (AP) is desirable due to its correlation with fertility outcomes, and ultimately with lifetime production of the cow through repeated early calving dates (Anderson et al., 1991). Age at puberty in heifers is conveniently defined as the age at which a heifer displays visual signs of estrus for the first time (Pineda, 2003). Age at puberty is to some extent breed-determined and is a moderately heritable trait ($h^2 = 0.43$) with a favorable association with weaning weight and yearling weight of the offspring (Brinks, 1994).

Anderson et al. (1991) developed a standardized reproductive tract score (RTS) system to measure AP in heifers indirectly. This method involves palpation of the reproductive tract and ovarian structures per rectum and is scored from 1 to 5 (Table 1). Three possible applications of the RTS system have been recommended: first as a screening test to determine the pubertal status of heifers before the breeding season (Anderson et al., 1991), second as an indication of the nutritional requirements of heifers when sufficient time is allowed before the breeding season (Anderson et al., 1991), and third as a selection tool for AP (Pence and BreDahl, 1998; Pence et al., 2007). Reproductive tract scoring as a method of selection has been found to be correlated with AP, response to synchronization, and pregnancy rate to synchronized estrus, and has an estimated heritability of 0.32 (Anderson et al., 1991). Reproductive tract scoring is a repeatable (between and within veterinarian) and accurate measure of pubertal status (Rosenkrans and Hardin, 2003).
The objective of this study was to compare the usefulness of RTS as a predictor of fertility and production outcomes with other selection measures such as KR, BCS, BW, or age at the onset of the first breeding season.

MATERIALS AND METHODS

This study was performed under protocol number 36-5-620, as approved by the Animal Use and Care Committee of the University of Pretoria.

This was a prospective study performed simultaneously with a study to determine the economic effects of estrous synchronization using PG on 272 Bovelder heifers at Johannesburg Water’s Northern Farm (Holm, 2006; Holm et al., 2008).

The age of the heifers at the start of the breeding season ranged from 364 to 486 d (median 431), and their BW ranged from 261 to 407 kg (median 316). Two days before the onset of the AI season (d −1), all heifers were weighed, body condition scored using a 5-point scale, and reproductive tract scored (Anderson et al., 1991; Table 1). Kleiber ratio was calculated as growth rate per metabolic BW (ADG/end BW0.75), using birth weight as the beginning BW, and the BW on d −1 as the end BW. To avoid potential bias caused by synchronization, heifers were ranked by RTS first and BW second, and then block randomized in pairs to the synchronized or the unsynchronized group (Holm et al., 2008). As a result of this, each RTS category contained exactly 50% synchronized and 50% unsynchronized heifers.

Frozen semen of 11 different bulls was allocated to heifers according to normal farm practice. Farm management and other staff were blinded to categories (synchronization and RTS), and heifers were managed as 1 group. The AI season started on October 15, 2003 (d 1) and lasted for 50 d. Detection of estrus was done by visual observation and marking during each night, and a scratch-off adhesive estrus detection aid was used additionally for the first estrus of each heifer (Estrotect, AgHold, Howick, South Africa). Upon detection of standing estrus, heifers were inseminated once per day at 0900 h by 1 experienced inseminator. After a window period of 5 d, there was a period of 42 d natural breeding with bulls, using a multisire system with a heifer:bull ratio of 34:1.

Day of the AI season and semen batch were recorded for all inseminations during the breeding season. A veterinarian palpated the heifers via rectum to determine pregnancy status 90 d after the removal of bulls. Pregnancy status was confirmed by calving date. Abortions, dystocia, birth date, birth weight, sex of calf, calf mortality, cow mortality, and weaning weight were subsequently recorded. The subsequent breeding season started on November 1, 2004, consisting of a 50-d AI period followed 14 d later by a 42-d bull breeding period. No estrous synchronization was used, but similar records were collected during the subsequent breeding season. All calves were weaned on the same day (March

<table>
<thead>
<tr>
<th>RTS</th>
<th>Number</th>
<th>Age, d (mean; 95% CI)</th>
<th>BW, kg (mean, 95% CI)</th>
<th>BCS (1 to 5 scale) (mean, 95% CI)</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>16</td>
<td>420&lt;sup&gt;a&lt;/sup&gt;</td>
<td>309&lt;sup&gt;b&lt;/sup&gt;</td>
<td>3.8&lt;sup&gt;b&lt;/sup&gt;</td>
</tr>
<tr>
<td>2</td>
<td>70</td>
<td>423&lt;sup&gt;a&lt;/sup&gt;</td>
<td>309&lt;sup&gt;a&lt;/sup&gt;</td>
<td>3.6–4.0</td>
</tr>
<tr>
<td>3</td>
<td>81</td>
<td>432&lt;sup&gt;a&lt;/sup&gt;</td>
<td>313&lt;sup&gt;a&lt;/sup&gt;</td>
<td>3.6–3.8</td>
</tr>
<tr>
<td>4</td>
<td>74</td>
<td>434&lt;sup&gt;c&lt;/sup&gt;</td>
<td>320&lt;sup&gt;b&lt;/sup&gt;</td>
<td>3.8&lt;sup&gt;b&lt;/sup&gt;</td>
</tr>
<tr>
<td>5</td>
<td>30</td>
<td>439&lt;sup&gt;a&lt;/sup&gt;</td>
<td>318&lt;sup&gt;a&lt;/sup&gt;</td>
<td>3.9&lt;sup&gt;b&lt;/sup&gt;</td>
</tr>
</tbody>
</table>

<sup>a</sup>Values within columns with no superscripts in common differ significantly (P < 0.05).
<sup>b</sup>Ci = confidence interval.
Table 3. Effects of prebreeding age, BW, and BCS on reproductive tract score (RTS; multiple regression)

<table>
<thead>
<tr>
<th>Variable</th>
<th>b</th>
<th>SE</th>
<th>95% CI</th>
<th>P-value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Age</td>
<td>0.013</td>
<td>0.003</td>
<td>0.006, 0.019</td>
<td>&lt;0.01</td>
</tr>
<tr>
<td>BW</td>
<td>0.002</td>
<td>0.003</td>
<td>-0.003, 0.007</td>
<td>0.40</td>
</tr>
<tr>
<td>BCS</td>
<td>0.219</td>
<td>0.195</td>
<td>-0.163, 0.601</td>
<td>0.26</td>
</tr>
</tbody>
</table>

1CI = confidence interval.

29, 2005), and the experiment was terminated on April 1, 2005.

Days to first AI was defined as the day of the breeding season on which a heifer was inseminated for the first time. Days to calving was defined in a similar fashion, and the first day of the calving season was defined as the day when the first calf was born. When a heifer did not achieve the specified status by the end of the time period a maximum value was given to that heifer (e.g., 50 in the case of days to first AI), but these values were censored for the purpose of Cox regression.

Effects of age, BW, and BCS on RTS were assessed using multiple linear regression. The various outcomes (days to first AI, pregnancy rates, days to calving, and calf weaning weight) were then compared between categories of RTS. Proportions were compared using the Fisher exact test, and means and medians were compared using ANOVA with the Tukey-Kramer multiple comparison test and Kruskal-Wallis 1-way ANOVA, respectively. The effects of RTS on the outcomes, adjusted for BW, BCS, and age, were then estimated using Cox regression for days to AI and days to calving, logistic regression for pregnancy rates, and multiple linear regression for weaning weight. The usefulness of each of the predictor variables (age, BW, BCS, KR, and RTS), when used alone to predict the outcomes, were compared using the \( R^2 \) statistic for linear regression models (weaning weight), the pseudo \( R^2 \) values for Cox regression models (days to AI and days to calving), and the area under the curve (AUC) of the receiver operating characteristic (ROC) curve for binary outcomes (pregnancy). Areas under the ROC curves were compared using the algorithm of DeLong et al. (1988). Statistical analyses were done using NCSS 2004 (NCSS, Kaysville, UT), Epicalc 2000 (http://www.brixtonhealth.com/epicalc.html), and Stata 10.1 (StataCorp, College Station, TX).

RESULTS

The summary of the 5 RTS categories on d −1 (Table 2) shows that heifers with RTS 1 and 2 were younger than those with RTS 3, 4, and 5, whereas heifers with RTS 3 were younger than those with RTS 5. It further shows that heifers with RTS 2 and 3 were lighter than those with RTS 4, and heifers with RTS 2 and 3 had lower BCS than those with RTS 4 and 5.

Using simple linear regression, age, BW, and BCS before the onset of the breeding season were each associated with RTS \( (P = 0.03, P < 0.01, \text{and } P < 0.01, \text{respectively}) \). However, in a multiple regression model of RTS using age, BW, and BCS as predictors, only prebreeding age was independently associated with RTS \( (P < 0.01; \text{Table 3}) \).

In a multiple regression model, age, BW, and BCS were all associated with prebreeding KR \( (P < 0.01) \), whereas RTS was not independently associated with KR \( (P = 0.76; \text{data not shown}) \).

The univariable effects of prebreeding RTS on pregnancy rates, days to first AI, days to calving, and weaning weight of the calves are summarized in Table 4. Using logistic regression, RTS and BW before the onset of breeding showed positive univariable associations with pregnancy rate after the first breeding season \( (P < 0.01 \text{ and } P = 0.04, \text{respectively}) \), whereas age, BCS, and KR did not \( (P = 0.07, P = 0.17, \text{and } P = 0.28, \text{respectively}) \). Associations between predictor variables measured before the first breeding season and pregnancy rate after the second breeding season were significant for RTS \( (P = 0.02) \), BW \( (P = 0.01) \), and BCS \( (P = 0.03) \), but were not for age \( (P = 0.10) \) and KR \( (P = 0.73) \).

Univariable Cox regression analyses of days to first AI and of days to calving showed negative associations with prebreeding RTS and BCS \( (P < 0.01) \), but no association with prebreeding BW, age, or KR. Univariable linear regression of calf weaning weight showed associations with prebreeding RTS \( (P < 0.01) \), age \( (P = 0.04) \), and KR \( (P = 0.05) \), but not with BCS \( (P = 0.37) \) or BW \( (P = 0.65) \). Calves of heifers with RTS 1 or 2 \( (n = 33) \) had a mean weaning weight of 186.7 kg \( (95\% \text{ confidence interval: 176.0 to 197.4 kg}) \), differing from calves of heifers with RTS 3, 4, or 5 \( (n = 102) \) with a mean weaning weight of 210.1 kg \( (95\% \text{ confidence interval: 203.8 to 216.4 kg}) \).
A summary of the multivariable (logistic, Cox, and linear) regression models for the various outcomes using prebreeding RTS, age, and BW as predictor variables is presented in Table 5. There were consistently significant associations between RTS and the outcomes ($P < 0.01$), but not for age or BW. When days to calving was added as a predictor variable to the logistic regression model for pregnancy to the subsequent AI season, days to calving was associated with the outcome ($P < 0.01$), but RTS was not ($P = 0.09$). When an interaction term between synchronization group and RTS was included as a predictor variable in these models, the interaction term was not associated with any of the outcomes ($P > 0.2$).

In Table 6, the usefulness of each prebreeding variable when used on its own for predicting various economically important outcomes are compared. For each outcome, RTS was a better predictor (explained more of the variation in the outcome) than any of the 4 other prebreeding variables.

The AUC of the ROC curve for RTS (0.67) was greater than that for age, BCS, KR ($P < 0.01$), and BW ($P = 0.02$), but did not differ from the AUC for age (0.57; $P = 0.14$).

**DISCUSSION**

Previous studies have clarified the basic principles of the onset of puberty (Day et al., 1984, 1987; Foster, 1994). Puberty in cattle occurs when a certain level of somatic development (critical BW) is reached, causing the prepubertal negative feedback of estradiol on the pituitary gland, hypothalamus, or both, to be terminated, which leads to the first ovulation. Environmental factors affecting the onset of puberty in heifers include nutrition, seasonal effects, climate, and biostimulation (Pineda, 2003). Figure 1 summarizes the factors affecting AP and also the pathways through which AP influences production outcomes.

In the present study, RTS was associated with age, BW, and BCS before the first breeding season, but it seems that, in this group of heifers, RTS was associated more strongly with age than with BW of the heifer. This is in contrast with an older theory that a heifer needs to reach a specific level of somatic development (BW) for the onset of puberty to be induced (Day et al., 1984, 1987; Foster, 1994) and may indicate that there is

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### Table 5. Multivariable associations of prebreeding reproductive tract score (RTS), BW, and age with some important production and reproduction outcomes in beef heifers

<table>
<thead>
<tr>
<th>Predictor variable</th>
<th>Pregnancy after the first AI season</th>
<th>Pregnancy after the subsequent AI season</th>
<th>Days to first AI</th>
<th>Days to calving</th>
<th>Calf weaning weight</th>
</tr>
</thead>
<tbody>
<tr>
<td>RTS</td>
<td>1.78 (1.64)</td>
<td>1.04—1.32</td>
<td>1.25 (1.14—1.44)</td>
<td>1.14—1.84</td>
<td></td>
</tr>
<tr>
<td>BW</td>
<td>1.01 (1.00)</td>
<td>0.99—1.00</td>
<td>0.84 (0.73)</td>
<td>0.73</td>
<td></td>
</tr>
<tr>
<td>Age</td>
<td>1.00 (0.98—1.01)</td>
<td>0.99—1.02</td>
<td>0.31 (0.24)</td>
<td>0.24</td>
<td></td>
</tr>
</tbody>
</table>

1Area under the curve for receiver operating characteristic analysis.
2Pseudo-$R^2$-value for univariable Cox regression.
3$R^2$-value for univariable linear regression.
4RTS = reproductive tract score.

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### Table 6. Univariable predictive ability of 5 prebreeding variables for some important production and reproduction outcomes in beef heifers

<table>
<thead>
<tr>
<th>Predictor variable</th>
<th>Pregnancy after the first AI season</th>
<th>Pregnancy after the subsequent AI season</th>
<th>Days to first AI</th>
<th>Days to calving</th>
<th>Calf weaning weight</th>
</tr>
</thead>
<tbody>
<tr>
<td>RTS</td>
<td>0.67 (0.66)</td>
<td>0.99—1.02</td>
<td>0.31 (0.24)</td>
<td>0.24</td>
<td></td>
</tr>
<tr>
<td>BCS</td>
<td>0.56 (0.53)</td>
<td>0.99—1.02</td>
<td>0.31 (0.24)</td>
<td>0.24</td>
<td></td>
</tr>
<tr>
<td>BW</td>
<td>0.58 (0.53)</td>
<td>0.99—1.02</td>
<td>0.31 (0.24)</td>
<td>0.24</td>
<td></td>
</tr>
<tr>
<td>Age</td>
<td>0.54 (0.57)</td>
<td>0.99—1.02</td>
<td>0.31 (0.24)</td>
<td>0.24</td>
<td></td>
</tr>
<tr>
<td>Kleiber ratio</td>
<td>0.51 (0.42)</td>
<td>0.99—1.02</td>
<td>0.31 (0.24)</td>
<td>0.24</td>
<td></td>
</tr>
</tbody>
</table>
an age-related induction of puberty that is not related to the BW of the heifer, in agreement with Yelich et al. (1995) and Pence et al. (2007). On the other hand, it may also indicate some variation in the critical BW of individual heifers that needed to be achieved to induce puberty, meaning that there was some scope for selection for AP in this population. Reproductive tract score was not associated with prebreeding KR in this study as was shown using multiple regression, whereas age, BW, and BCS all contributed to the variation in KR.

Reproductive tract score was associated with all important fertility and production outcomes in this study, which is in agreement with previous studies (Anderson et al., 1991; Pence and BreDahl, 1998; Pence et al., 2007). In general, heifers with RTS 1 and 2 had longer days to first AI and days to calving and decreased pregnancy rates and calf weaning weights compared with those with RTS 4 and 5. After adjustment for BW and age, RTS showed a significant association with all the outcomes, and these associations were not confounded or modified by synchronization group in this study. These results, along with other objective measures, indicate that variation in RTS accounted for more of the variation in the fertility and production outcomes than did variation in BW, age, or KR. This indicates that RTS represents a measure of the true genetic variation

**Figure 1.** Diagram illustrating factors affecting age at puberty (AP) and the pathways through which AP may influence production outcome.
of AP within the population, which is in agreement with Pence et al. (2007).

In this study, RTS and BCS, which are the more subjective measurements (compared with BW and age), explained more of the variation in the fertility and production outcomes than did the objective measurements. This supports the findings of Rosenkrans and Hardin (2003), that RTS has good accuracy despite less favorable repeatability. The subjectivity of RTS is not only caused by the less favorable repeatability, but also by the complexity of the scoring system. It is our experience that many heifers do not fit a particular RTS score exactly, and it is for the operator to decide which of the measures to give the most weight (Holm, 2006). More research is needed to clarify which of the different measures of the RTS system gives the best prediction of reproduction outcome; this may improve the accuracy of RTS. Ultrasound may also improve the repeatability of RTS.

Comparing RTS with Other Predictors of Heifer Performance

Reproductive tract score showed a consistently stronger association with fertility and production outcomes than did KR (Table 6). This is evidence that RTS can be used as a primary selection tool for heifers before the onset of breeding without any detrimental effect on production. The association of RTS with weaning weight of the offspring was mostly indirectly through its effect on days to calving. This was shown by the multiple regression model for weaning weight of the offspring: RTS was associated with weaning weight of the offspring, but this association was not significant when days to calving was added to the model as a predictor.

If RTS had been used as a selection criterion in this group of heifers before breeding, using RTS 2 as the cut-off point, thereby selecting the best 94% of heifers, the pregnancy rate to the 50-d AI season would not have increased (56 vs. 58%, \( P = 0.79 \)). Using RTS 3 as the cut-off point, thus selecting the best 68% of heifers, would have resulted in an increase in pregnancy rate to the 50-d AI season from 56 to 64% \( (P = 0.10) \). Although impractical because of the proportion of heifers (62%) that would have needed to be culled, using RTS 4 as cut-off would have resulted in an increase in pregnancy rate from 56 to 73% \( (P < 0.01) \). It seems that, in this group of heifers, it would have been most sensible to use RTS 3 as cut-off for selection. Of course, this will not always be the case because it depends on the timing of RTS and the proportion of heifers in the group that have reached puberty by that time. If the best 68% of heifers in this group were selected using KR, it would not have increased pregnancy rate to the 50-d AI season \( (56 \text{ vs. } 57\%, \ P = 0.96) \). The superiority of RTS as a selection tool for fertility is well demonstrated by this, despite the fact that d −1 was probably not the best time to use RTS as selection tool in this group of heifers. Although speculative, it is possible that scoring heifers 1 or 2 mo earlier may have resulted in stronger associations with the outcome. More research is needed to determine the best time to do RTS on yearling heifers as a selection tool for fertility.

Receiver-operating characteristic analysis is a useful tool to compare the predictive ability of RTS and other measures on pregnancy outcome, although the idea of RTS is not simply to predict pregnancy outcome, but rather as a selection tool for fertility. The AUC of the ROC curve provides a summary of the overall ability of a diagnostic test or predictor variable to correctly classify or predict a binary outcome. In this study, the AUC can be interpreted as the probability that a randomly chosen pregnant (to the 50-d AI period) heifer had a greater prebreeding RTS than a randomly chosen nonpregnant heifer. It is clear that, although RTS was nowhere near perfect \( (\text{AUC} = 1) \), it was better than that of any of the other measures. In contrast, the AUC for BCS, age, and KR were not different from 0.5, indicating no predictive ability.

Long-term Benefits of Using RTS as a Selection Tool

Selecting for RTS leads to a reduction in days to calving (Table 4), which allows heifers more time to recover from the stress of calving and to become prepared for the next breeding season. First calf cows are known to be the group under most pressure to conceive again in the subsequent breeding season, due to the fact that they are still growing and also nursing a calf, which puts tremendous pressure on their energy and protein metabolism, to the detriment of fertility (Chenoweth and Sanderson, 2001). Reproductive tract score was shown in this study to influence not only the immediate calving season, but also the pregnancy rate to the subsequent breeding season. It was shown in this study that the association of prebreeding RTS with the pregnancy rate to the second breeding season was not direct, but was confounded by the association between RTS and days to calving during this first calving season. The proportion of heifers with RTS 4 and 5 that remained in the herd until their second breeding season was 77% \( (80 \text{ of } 104) \), whereas that proportion for heifers with RTS 1 to 3 was 54% \( (90 \text{ of } 167) \), demonstrating an increased survival of heifers with greater RTS.

In addition to this, among the heifers that were retained until their second breeding season, there was a strong association between RTS before first breeding season and pregnancy outcome of the second breeding season, most likely due to the effect of RTS on days to calving. The effect of days to calving on pregnancy rate of the subsequent breeding season is well known (Chenoweth and Sanderson, 2001) and was also shown using these data (Holm, 2006). It can be seen here that not only should the direct benefit of using RTS as selection tool for heifers be taken into account, but also...
the effect that selection using RTS will have during the subsequent breeding seasons, and therefore on lifetime production of the cows.

Due to its ease of measurement, good heritability, and association with feed conversion ratio (Nkrumah et al., 2004), KR has been used as an important selection tool for replacement heifers. Results from the present study indicate that selecting for RTS will not select against production measures such as KR, due to their poor association with each other. However, RTS is primarily an indicator of AP and could be used in addition to production variables, such as KR in a selection policy.

In conclusion, RTS before the onset of the breeding season is a predictor of heifer reproductive performance, even after adjustment for age, BW, and BCS. It is a better predictor of fertility than other traits commonly used (BW, BCS, and KR), compares well with these traits in predicting production outcomes, and is likely to be a predictor of lifetime production of the cow.

LITERATURE CITED


