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Statistical Considerations when using Hysteresis to Estimate Internal Heat Load in Dairy Cows

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Abstract

Water is often used to manage heat stress in dairy cattle. Sprinklers are often placed over the feed bunk or used while cattle are waiting to be milked, however in this experiment cattle were given control over water with a cow-activated shower. Previous studies have focused on how wetting can lower body temperature or reduce respiration rates. An alternative way to investigate this management practice is to examine internal heat loads. Internal heat load can be quantified by fitting a hysteresis loop to daily field data. The hysteresis loop is formed by a phase diagram of body temperature versus an environmental input. Internal heat load is the area inside the loop. The area can be estimated using a number of environmental measures. In this paper three environmental measures are considered: ambient air temperature, the temperature-humidity index and the heat-load index. The two stage harmonic least squares method is used to estimate internal heat load. Then a Bayesian MCMC model is used to predict internal heat load using the environmental inputs and test the effectiveness of allowing shower access on internal heat load reduction. Voluntary use of a shower reduces internal heat load and the strength of this effect increases with the degree of the heat challenge.

Keywords: Bayesian analysis, Energy dissipation, Farm animals, Heat Stress, Thermo-regulatory response.

1. Introduction

Numerous studies have found that heat stress in dairy cows can lead to problems ranging in severity from reduced milk production (Armstrong, D.V., 1994) to death (Hahn, G.L., 1989; Hahn, G.L. and Mader, T.L., 1997; Hubbard, K.G. et al., 1999) or issues with growth and reproduction (Forbes, J.M., 2007), (Fuquay, J.W., 1981), (Liu, F. et al., 2009). Most of these studies have focused on using body temperature (T_b) or panting levels as measures of heat stress. Elevated levels of T_b can signal problems for animals whose multitude of bodily functions depend on a stable T_b , but T_b is rarely the whole story as it does not capture the work done to maintain a stable T_b . As one of a variety of cooling mechanisms that cattle employ, respiration rate is often used as a proxy for the energy that an animal is expending to lower T_b , and it is a sensitive measure because it has considerable variability. (Hahn, G.L., A.M. Parkhurst, J.B. Gaughan, 1997) Internal heat load, however, is an alternative measure of heat stress that effectively reflects both energy use and other characteristics of heat tolerance.

The T_b displays hysteresis during a heat challenge, since T_b is dependent on the history of the system. Air temperature (T_a) is periodic and sinusoidal in nature and T_b is bivalued at a particular value of T_a . The same value of T_a can produce two possible values of T_b depending on its first derivative; a higher value of T_b when T_a is decreasing and a lower value of T_b when T_a is increasing. This is due to a lag in T_b response to changes in T_a . When the input, T_a , acts as a sinusoidal forcing function this system forms an elliptical hysteresis loop, whose area is an indication of energy transfer, referred to as internal heat load in the context of this study. Calculating the area of a hysteresis loop is an important topic in physics and other scientific branches as area is sometimes equal to the work done during a cycle (Brokate, M. and Sprekels,

J., 1996). The major benefit of internal heat load as a measure of heat stress is that it represents both absolute Tb and the effort put into reducing changes in Tb . The elliptical model also provides for the characterization of other dynamics occurring during the heat challenge. The model has five fundamental parameters and three other derived parameters in addition to internal heat load (lag, retention, and coercion) to be discussed below.

Yang and Parkhurst (2011) compared three parametric ellipse fitting methods, linear least squares, non-linear ellipse-specific least squares, and two stage simple harmonic least squares (harmonic2). They found that bootstrapped estimates reduced bias for all three methods and that the harmonic2 area estimates had the best coverage probability and least bias. However, their study focused on climate controlled experimental data while this study focuses on data collected in conditions with natural variation in weather. In an uncontrolled thermal challenge, the influence of the forcing function and the harmonics of the input are tempered by competing environmental effects, hence, robustness to deviations from the ellipse model is an important issue.

The major focus of this paper is on using internal heat load estimates to test the effectiveness of management strategies designed to reduce heat stress. Using water on cows is a commonly used technique to manage heat stress. Of U.S. dairies with over 500 cows, 62% use sprinklers or misters for heat abatement (USDA, 2010). Sprinklers are often placed over the feed bunk or used while cattle are waiting to be milked, giving them little choice over when to use this resource. In this experiment, cattle were given control over water with a cow-activated shower in order to

better understand how they would voluntarily use this resource. The specific objectives of this study are to: evaluate the usefulness of the elliptical model in fitting environmental data, assess the effect of the availability of showering on internal heat load, and determine whether the effect of the cow shower is dependent on the level of the heat challenge. If the shower treatment effect is significantly lower, there will be positive evidence for both the voluntary water use to reduce internal heat load and the ability of internal heat load estimates to test for treatment differences in future experiments where the environment is recorded but not regulated.

2. Materials and Methods

2.1. Experimental Design and Data

Data for this study comes from an experiment conducted by Legrand, A. et al. (2008) at UC-Davis to describe how and when dairy cattle voluntarily used an overhead water source and how use of this water affected behavioral and physiological indicators of heat stress. During the experiment, 12 cows had unlimited access to a weight activated shower while another control group of 12 cows were not given access to showers. Each trial was conducted for 5 days and a total of 6 trials were run during the summer months. For each trial, 4 of the 24 cows were placed in separate pens. Half of the pens had showers which remained fixed-in-place forming a split plot in time for each trial - without an error term for the whole plot (shower) error within trials. After 5 days all 4 cows were replaced with a new trial group of 4 cows that were given several days to become adjusted to their environment before the experiment recommenced. Internal T_b was measured every 5 minutes using a temperature logger inserted into the vaginal cavity. The T_{aw} was

measured every 5 to 10 minutes, and other environmental variables such as black globe temperature, humidity, wind speed, and wind direction were measured at the same time. Only half of each pen was covered from the elements. The showers were located in the uncovered portion 6m from the feed bunk. Cows could access the shower from all 4 sides.

Table 1. Pen Design for a Single Trial. The water trough was inside the barn while showers and the feed bunk were located outside.

South-most Pen 1	Pen 2	Pen 3	North-most Pen 4
Barn			
Water Trough	Shared Water Trough		Water Trough
Outside Area			
Control	Shower	Control	Shower
Feed Bunk			

Two heat indices were calculated from the environmental data: the thermal heat index (*THI*) and the Heat Load Index (*HLI*) reported in Igono, M. et al. (1992), and Gaughan, J.B. et al. (2008) respectively. These indices are alternative ways to quantify the size of the heat challenge in addition to T_a .

Table 2: Components and Formulas for Heat Indices

Heat Index	Components	Formula
Thermal Heat Index (THI)	T_a , Relative Humidity (RH)	$(1.8 \times T + 32) - [(0.55 - 0.0055 \times RH) \times (1.8 \times T - 26)]$
Heat Load Index (HLI)	Black Globe Temperature (BGT), Wind Speed (WS), Relative Humidity (RH)	IFBGT > 25, $8.62 + (0.38 \times RH) + (1.55 \times BGT) + \exp(-WS + 2.4) - 0.5 \times WS$ Else, $10.66 + (0.28 \times RH) + (1.3 \times BGT) - WS$

2.2. Models

2.2.a. Two-Stage Simple Harmonic Least Squares

The two-stage simple harmonic least squares method based on the work of Lapshin (1995) is used to model elliptical hysteresis. The input T_a and output T_b are estimated consecutively.

$$\begin{pmatrix} T_a(t) \\ T_b(t) \end{pmatrix} = \begin{pmatrix} b_a \cos(2\pi\omega t + \phi_a) + c_a + \varepsilon_a \\ b_b \cos(2\pi\omega t + \phi_a) - a \sin(2\pi\omega t + \phi_a) + c_b + \varepsilon_b \end{pmatrix} \quad (\text{eq1})$$

In the two-stage simple harmonic least squares method (eq1), five fundamental parameters, the center coordinates (c_a , c_b), saturation points (b_a , b_b), and retention (a) along with three derived parameters *area*, *lag*, and coercion (C) can be estimated, Figure 1. The phase angle of the input is ϕ_a , time is t , and the frequency, ω , is the reciprocal of the period. (Yang F., Parkhurst A.M., 2011). The saturation point occurs where T_a reaches its highest value. Retention a , is perhaps the most clinically important ellipse parameter with the exception of internal heat load. Retention is defined as the amount of heat left in the body after a heat challenge has receded to its mean. It is the length of the T_b axis from the centroid to the point where it intersects with the perimeter of the ellipse. If there is no hysteresis, retention equals 0. The derived parameter, area can then be calculated as

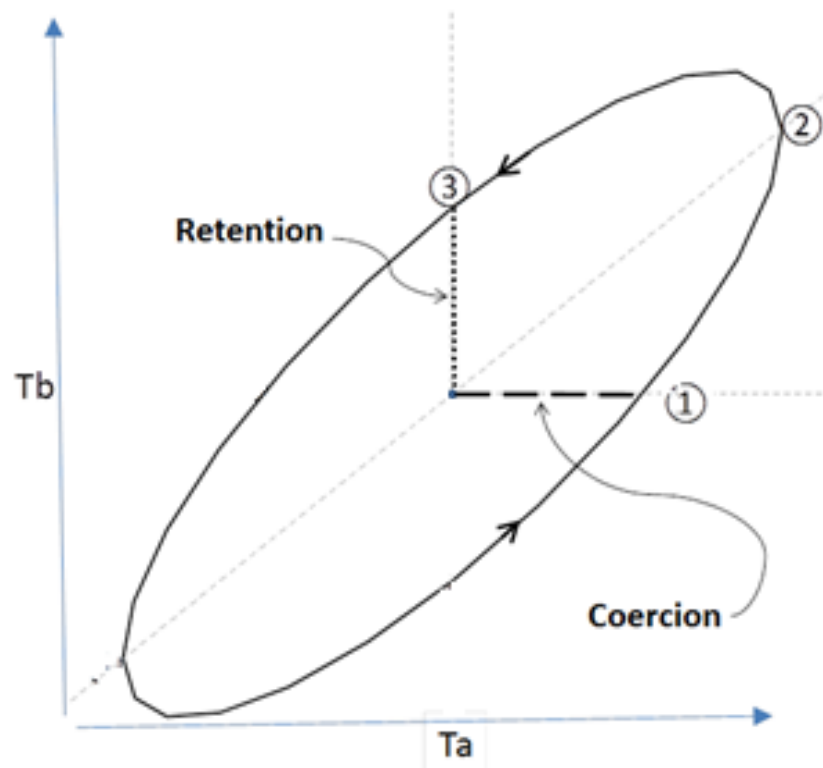
$$\text{area} = \pi a b_a \quad (\text{eq2})$$

In addition, two other derived parameter, lag and coercion C , are calculated as

$$\text{lag} = \arctan(a / b_b) \quad (\text{eq3})$$

$$C = b_a \sin(\text{lag}) \quad (\text{eq4})$$

Figure 1. Ellipse Parameters. Coercion is the distance from the center to point 1, the saturation point is at 2, and retention is the distance from the center to point 3. Internal heat load is the *area* within the ellipse and *lag* is the time it takes to get from point 2 to the maximum value of T_b . T_a and T_b are ambient temperature and body temperature.



After the initial parameter estimates are made, T_a and T_b residuals are collected and used to bootstrap the harmonic2 ellipse estimates. Harmonic2 ellipses with bootstrapping were used in this study since the bootstrapped parameter estimates with a bias adjustment are more accurate than the original estimates and bootstrapping leads to an estimate of the standard error for area (Yang F., Parkhurst A.M., 2011). A total of 120 ellipses were fit for each combination of the 4 pens, 5 days and 6 trials.

2.2.b. Linear Mixed Effects Models

Linear mixed effect models are fit separately to the 4th trial and the full data set. First 4 models for trial 4 are considered. As later tests showed that trial 4 results are somewhat extraordinary among the 6 trials these single trial models should not be considered representative. The first model, (eq5a), considers day a random effect with a treatment interaction.

$$Area_{ijk} = \mu + b_1 * treatment_i + random(day)_j + b_3 * treatment_i * random(day)_j + random(cow)_j + e_{ijk} \quad (eq5a)$$

Where μ is the overall mean, $treatment_i$ is the i th treatment level of the dummy variable representing shower access and both day and cow are normally distributed random effects. The ε_{ijk} are i.i.d. $N(0, \sigma^2)$. The next modification, (eq5b), checks for the interaction between treatment and day. In this model, day is considered to have both a fixed and random component,

$$Area_{ijk} = \mu + b_1 * treatment_i + b_2 * treatment_i * day_j + random(day)_j + random(cow)_j + e_{ij} \quad (eq5b)$$

The next modification, (eq5c), further reduces the model by focusing on the multiplicative effects of treatment and day by removing the fixed effect for treatment and the random component for day.

$$Area_{ijk} = \mu + b_1 * treatment_i * day_j + random(cow)_j + e_{ijk} \quad (eq5c)$$

The final modification, (eq5d) checks for additive effects of treatment and day.

$$(eq5d)$$

$$Area_{ijk} = \mu + b_1 * treatment_i + b_2 * day_j + random(cow)_j + e_{ijk}$$

The model for the full dataset includes environmental covariates and an additional treatment effect for pens sharing a water trough.

$$area_{ijk} = \mu + b_1 * treatment_k + b_2 * HLI_i + b_3 * treatment_k * HLI_i + b_4 * sharewater + b_5 * HLI^2 + b_6 * temp.range + b_7 * temp.range * treatment + random(cow)_j + random(day)_i + treatment_k * random(day)_i + e_{ijk} \quad (eq6)$$

In all cases, reduced models are chosen on the basis of smaller AIC. The R-package, lmer, (R Development Core Team, 2011) is used to obtain parameter estimates.

2.2.c. Weighted Residuals

A weighting scheme is introduced to account for the uncertainty of the harmonic2 area estimates.

The absolute values of the model residuals are fit using

$$|residual|_{ijk} = \mu + b_1 * boot.error_{ijk} + b_2 * treatment_k * HLI_i + b_3 * treatment_k + b_4 * HLI_i + e_{ijk} \quad (eq7)$$

and the reciprocal of the squared predicted absolute residual is then used as a system of weights within the original model.

2.2.d. Bayesian Model

Weighted linear models assume that the weight matrix is known exactly, which is not true when variance is estimated using a regression model as in the case above. Using a Bayesian Markov Chain Monte Carlo model (MCMC) eliminates this problem by modeling variance along with area. Bayesian models also allow for more intuitive interpretation of results using posterior

probability distributions and work well with hierarchical data.

The final model was obtained as the one with the lowest deviance information criterion (DIC). The final Bayesian internal heat load model given below does not contain variables for wind speed or whether cows shared water troughs.

$$Area_{ijk} = \mu + b_1 * trt_k + b_2 * HLI_i + b_3 * Ta.range_i + b_4 * trt_k * HLI_i + b_5 * trt_k * Ta.range_i + cow_j + day_i + trt_k * day_i + e_{ijk} \quad (eq.8)$$

The log of the variance of e_{ijk} is in turn modeled via (9), denoted as Bayesian Residual Model

$$\log(\text{var}(e_{ijk})) = \text{intercept} + \text{bootstrap.error}_{cd} \quad (eq.9)$$

In the area model (eq. 8) each of the independent effects (such as treatment) receives a non-informative normal prior centered at zero with a variance of 10,000. The group effects for cow and day come from a normal distribution with mean 0 and variance σ^2 , where σ^2 itself has a uniform prior distribution from 0 to 10000. Using data from Yang F. et.al.(2010), the intercept is given a normal prior with a mean of 15.99 and a variance of 100. This is the only informative prior used, and it is given a large variance because it comes from a study of Hereford steers in an environmentally controlled setting. Mean *HLI* and *Ta* range are both centered to facilitate comparisons and improve convergence of the MCMC procedure. The mean of *HLI* is 69 and the mean of *Ta* range is 20.

For the variance model (eq. 9) the intercept receives a normal prior with mean 3.2 and variance

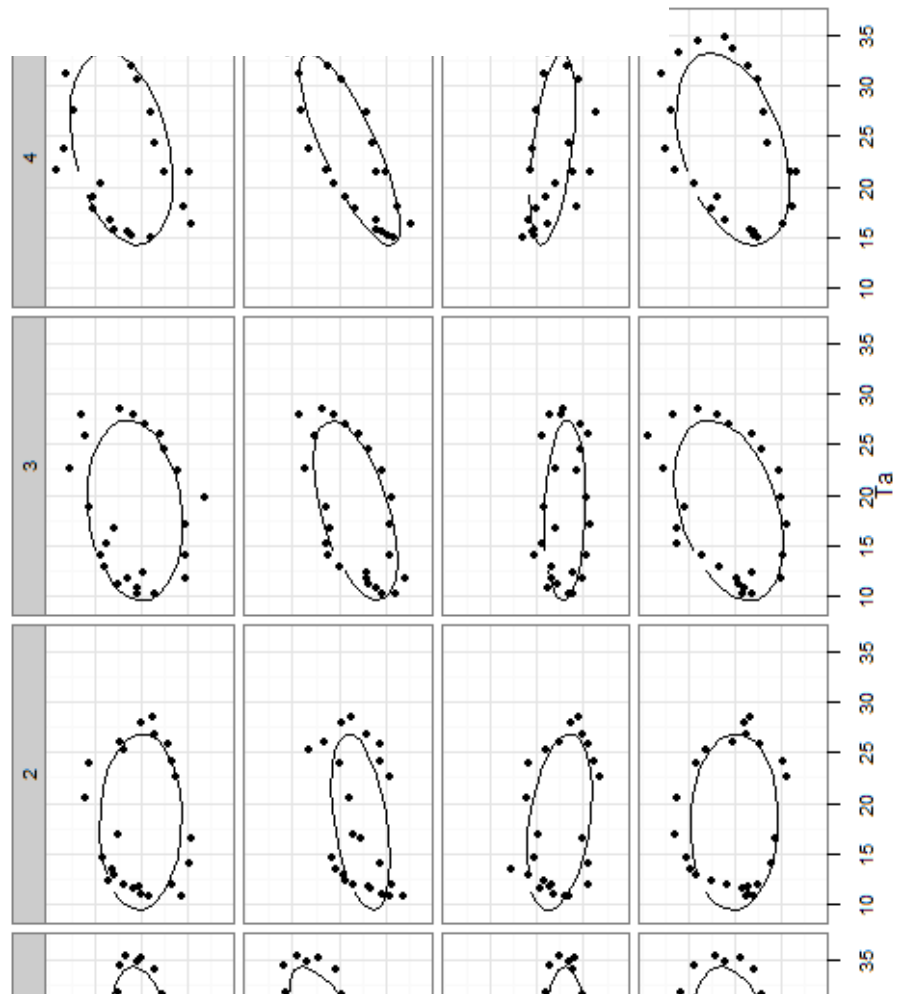
20. Since $e^{3.2}=24.5$ the expected value of the area variance is 24.5. The prior on bootstrap error is also normal with a mean of 0 and a variance of 1,000. The program is run in Winbugs (Lunn D.J. et. al., 2000) using 3 chains and 11000 iterations, of which only the last 15,000 are retained after a burn in period and thinning.

3. Results and Discussion

3.a. Bootstrapped Two-Stage Simple Harmonic Model

Figure 2 shows bootstrapped harmonic2 ellipses for every day from trial 4. These ellipse areas are those used in the single trial models. Some ellipses, such as cow 2098 day 5 fit quite well, whereas others exhibit serious deviations from the ellipse, e.g. cow 2103 day 2. The bootstrap standard error for area can be used to provide an estimate for the size of these deviations.

Figure 2: Example of Bootstrapped Harmonic2 Ellipses for 4 Cows over 5 Days in Trial 4.



3.b.Linear Models

3.b.1.Single Trial Linear Mixed Effects Model Results

To illustrate the fitting of single trial models (eq 5.a-d) to internal heat loads, trial 4 is used as an illustration. Table 3 shows that the multiplicative model with random cow effects (eq.5c) has the best fit. The treatment*day interaction is statistically significant with a p-value of 0.002. This means that the effect of treatment is not constant across days, within the small sample of trial 4 alone.

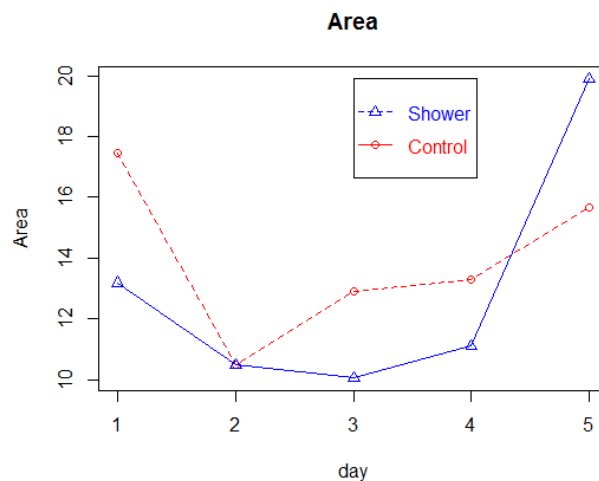
Table 3: Results from fitting Single Trial Models (eq5.a-e) to Heat Load Areas from Trial 4.(n=

20: 4 Cows and 5 Days)

eq	Model	Cows.d.	Days.d.	Residuals.d.	AIC	BIC	logLik
5.a	Crossed re C,day	3.51	2.39	1.72	118	124	-51.8
5.b	Trt Crossed re C, day	3.51	1.41	1.72	108	121	-41.1
5.c	Trt*Day re Cow	3.51	-	1.72	101	113	-38.5
5.d	Trt + Day re Cow	3.44	-	2.37	110	118	-47.0

This significant interaction is further revealed in the Treatment by Day interaction plot, Figure 3. On days 2 and 5 cows with access to a shower do not seem to have lower heat loads, although it is unlikely that this is due to more than random variation. On day 2 heat load is relatively small for all cows suggesting that there was little reason to engage in showering. Day 5, however, is a drastically different situation as heat load increases with showering.

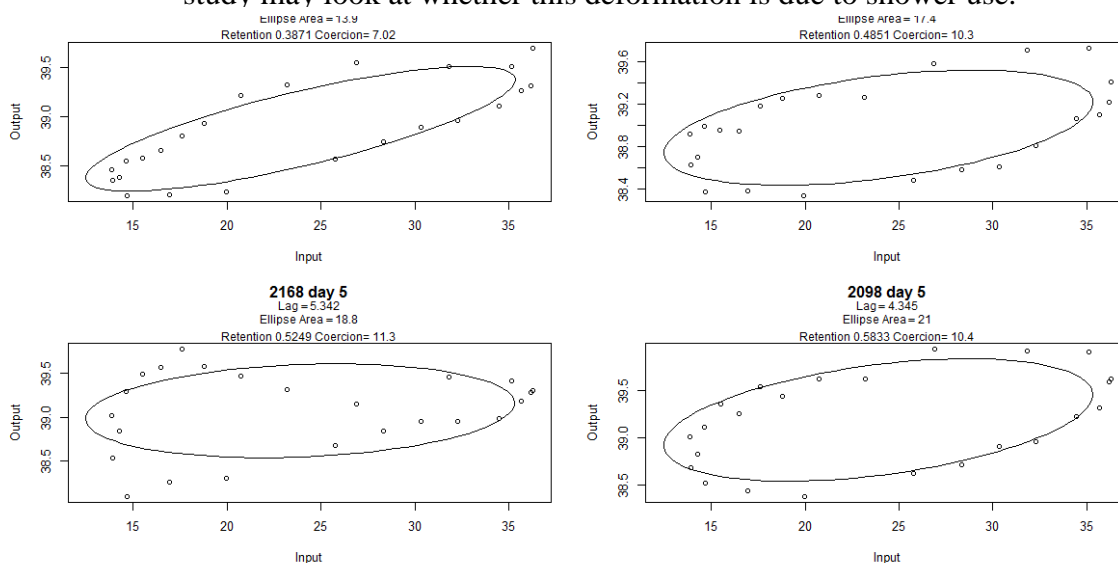
Figure 3: Trial 4 Treatment by Day Interaction Plot using average area for two cows.



The four ellipses formed on day 5 are enlarged in Figure 4. Both showered cows (bottom row) have areas larger than those of the two control cows (top row) and all four cows appear to have ellipses with a mean *T_b* around 39° C. In addition, body temperatures for cow 2168 do not seem

to follow a sinusoidal pattern. It may be that this lack of fit is at least partially responsible for the positive effect of showering, and it will be necessary to account for unequal variances in the full model.

Figure 4: Bootstrapped Harmonic2 Ellipses for Trial 4 day 5. Upper row is control cows, lower row is showered cows. Notice how 2168 day 5 does not look like an ellipse. A future study may look at whether this deformation is due to shower use.



It is also possible to measure the area of hysteresis loops that use *HLI* as an input. As *Tb* is more dependent on *HLI* than *Ta* there is a theoretical basis for switching inputs in this way. The *THI* is less interesting as a possible input because it will be shown later that it is not a very good predictor of heat load. Figure 5 shows how *HLI* levels at night are radically different from those during the day. The plot appears to be vertically disconnected between day and night. This is likely due to solar radiation. In contrast a plot of *Ta* over time, Figure 6, indicates no such discrepancy. This makes it difficult to use *HLI* instead of *Ta* as the input for internal heat load estimation.

Figure 5: *HLI* Time Series Separated by Trial appears to be constructed of two separate sinusoids

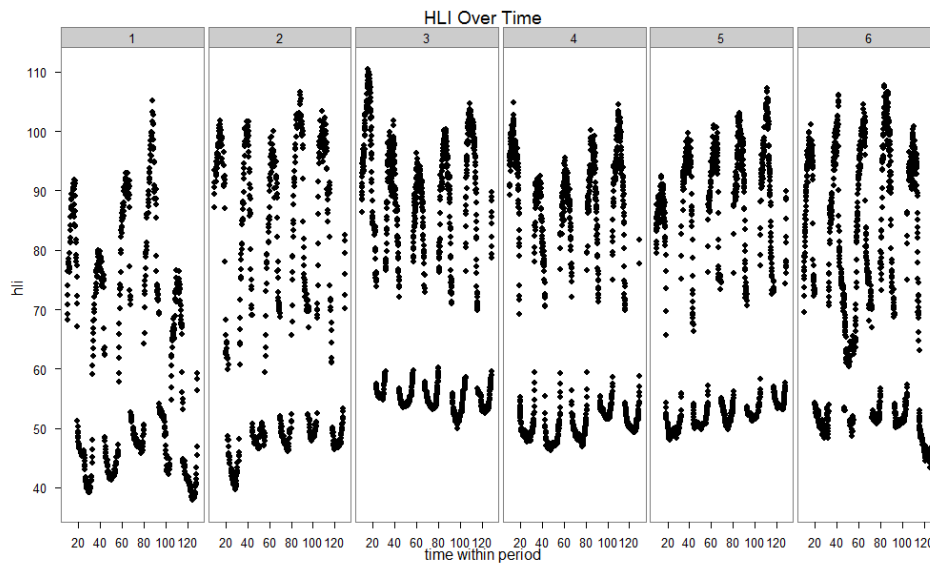
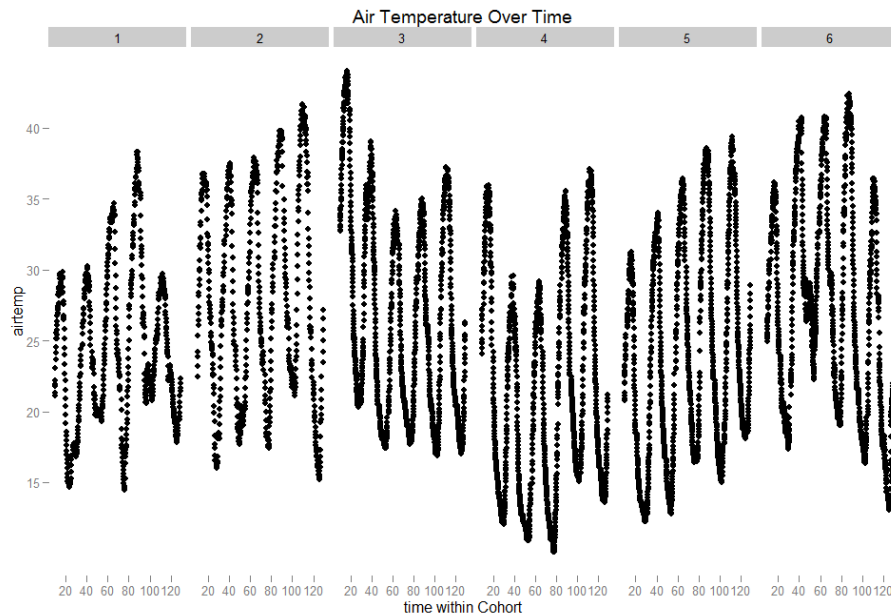


Figure 6: *Ta* Time Series Separated by Trial appears to be a single sinusoid.



3.c. Full Model

In the less important no weights case (eq. 6), the 2 interaction terms $\text{treatment} * HLI$ and $\text{treatment} * Ta.\text{range}$ are jointly statistically insignificant with a p-value of 0.17. These two variables are tested together as they suffer from multicollinearity. HLI and treatment by themselves are also statistically insignificant at the $\alpha=0.05$ level with p-values of 0.23 and 0.075 respectively, although the difference between 0.075 and 0.05 is small. $Ta.\text{range}$ on the other hand is statistically significant with a p-value of less than 0.001. The use of weights is then considered. The residual model (eq. 7) is fit using residuals from the model that includes the interaction terms, and the bootstrap error coefficient has a p-value of less than 0.001. Neither HLI nor treatment is a statistically significant predictor of the residuals absolute value with p-values

greater than 0.25 in both cases. The treatment*day interaction term is removed from the weighted and unweighted models as it increases AIC by 4 points.

Table 4 shows the results of the weighted model using *HLI* and *Ta.range* as covariates. The treatment**HLI* and treatment**Ta.range* interactions have a joint p-value of 0.015 although due to multicollinearity they are both statistically insignificant when tested separately with p-values above 0.1. Additional models found the effects of shared trough and HLI^2 to be statistically non-significant.

Table 4: Results of Weighted Linear Mixed Effects Model. (n=120, : 24 Cows, 30 Days)

Random Effects	Standard Deviation
Day	0.98
Cow	2.80
Residual	1.51

Fixed Effects	Estimate	Standard Error
Intercept	-9.97	4.64
Showering Effect	8.54	4.89
<i>HLI</i>	0.11	0.08
Showering* <i>HLI</i>	-0.10	0.08
<i>Ta.range</i>	0.78	0.12
Showering* <i>Ta.range</i>	-0.20	0.13

As the estimates here are similar to those for the Bayesian model we will hold off on interpreting them for now.

3.d. Bayesian Results

The final Bayesian heat load model (eq. 8), selected to have the lowest DIC does not contain variables for wind speed or shared troughs. The treatment by day effect is also eliminated as the DIC with this effect included is 495.2, which is higher than the DIC of 494.0 with it removed. A model which includes period has an almost identical DIC of 493.9, and although the decision was made to report the model without period for reasons of simplicity, it should be noted that including period does not change the estimates and standard errors for the treatment effect or its interactions. Posterior distributions for the parameters of interest were then obtained, as can be seen in Figures 7 and 8.

Figure 7: Markov Chains Showing Convergence of Posterior Distribution for Treatment.

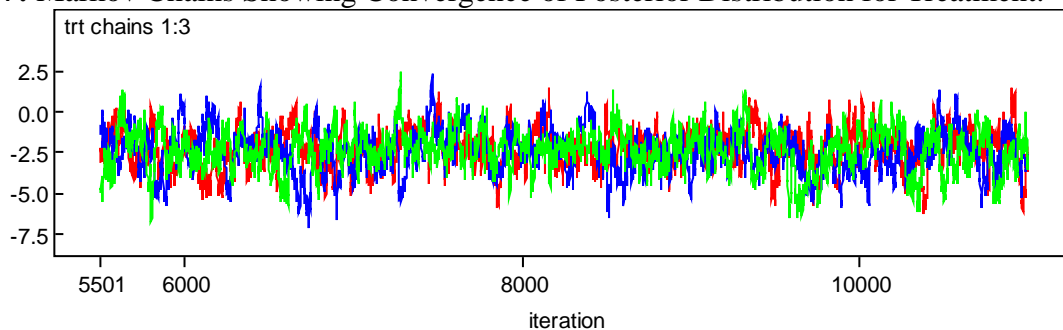
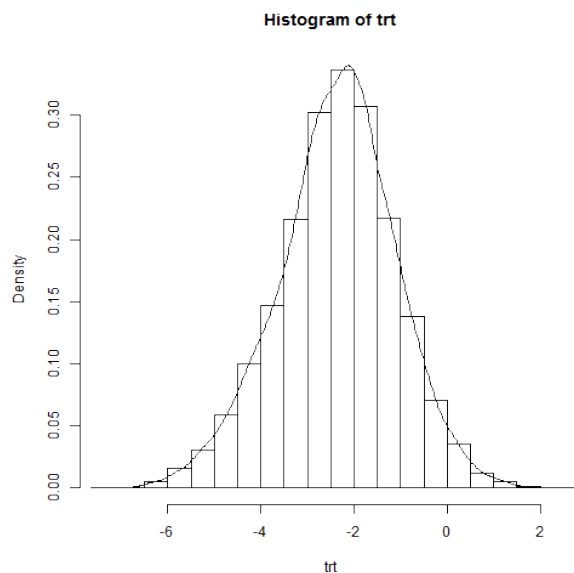


Figure 8: Posterior Distribution for Treatment Effect on Internal Heat Load at Mean Value of HLI.



Tables 5 and 6 show summary statistics for these parameter estimates: the mean, standard deviation, and the probability that the parameter is less than zero. The probability that allowing access to a shower negatively affects heat load at the mean values of HLI and *Ta* range is approximately 97%. This showering effect has a mean of -2.4 and increases in absolute size with *HLI* and *Ta* range. Thus, the interactions of treatment with both *HLI* and *Ta* range are significant.

Table 5: Statistics from Bayesian Area Model Posterior Distributions. $P(x < 0)$ is the posterior probability that the variable of interest is less than 0.

Parameter	mean	s.d.	$P(x < 0)$
Intercept	13.1	0.9	0.00
treatment* <i>HLI</i>	-0.1	0.1	0.90
Treatment	-2.4	1.3	0.97

<i>HLI</i>	0.1	0.1	0.09
<i>Ta</i> range	0.8	0.1	0.00
<i>Ta</i> .range*treatment	-0.2	0.1	0.96
sigma.cow	9.8	3.5	
sigma.day	1.6	0.8	

Table 6: Statistics from Bayesian Variance Model. $P(x < 0)$ is the posterior probability that the variable of interest is less than 0.

Parameter	mean	s.d.	$P(x < 0)$
Intercept	0.9	0.2	0.00
bootstrap standard error	1.5	0.4	0.00

Figure 9 shows how well trend lines based on *HLI*, *Ta* range and treatment group fit the data. It reveals how the control (solid line) and shower (dashed line) internal heat loads vary depending on *HLI* and *Ta* range. Figure 10 focuses on the reduction in head load for the shower cows by showing the size of the predicted reduction in head load associated with showering at the 30 combinations of *HLI* and *Ta* range present in the data. These results are consistent with Legrand et al.(2011) that use of showers increases with *Ta* and this seems to be the most likely explanation for the treatment by day interaction found in trial 4.

Figure 9: HLI vs. Observed (symbols/color) and Predicted Area (lines). Treatments are Control (dot with solid line) and Shower (triangle with dashed line). The size of the symbol indicates the Size of the T_a Range. Predicted internal heat load increases with air temperature and HLI for both treatment groups.

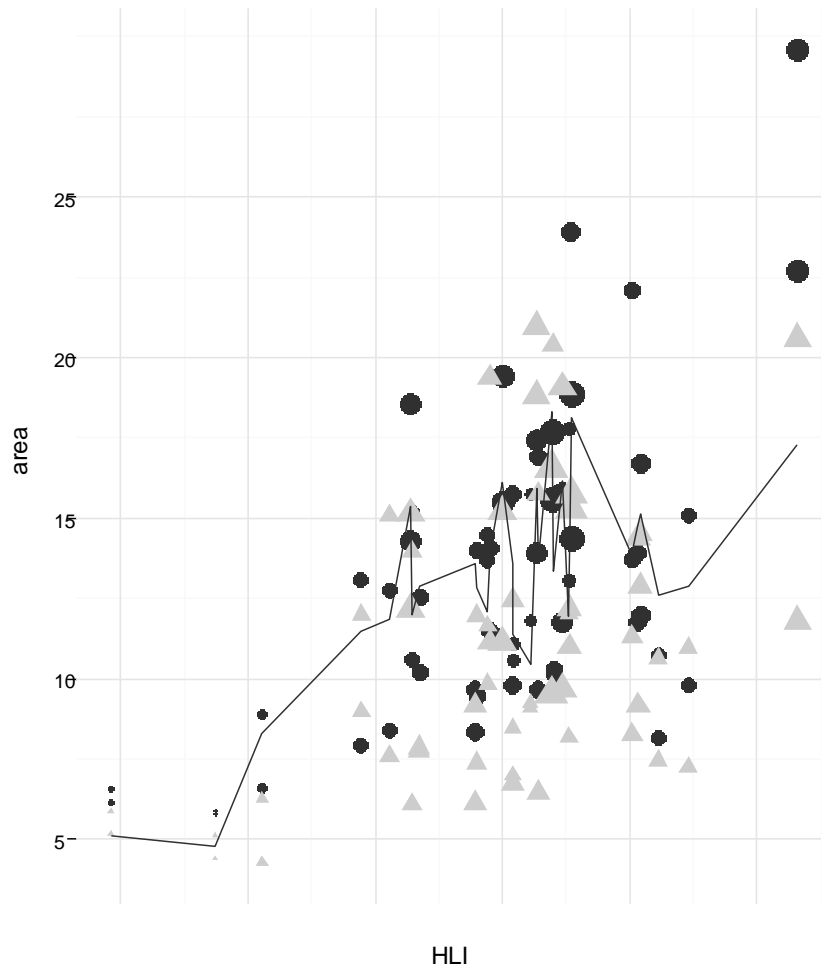
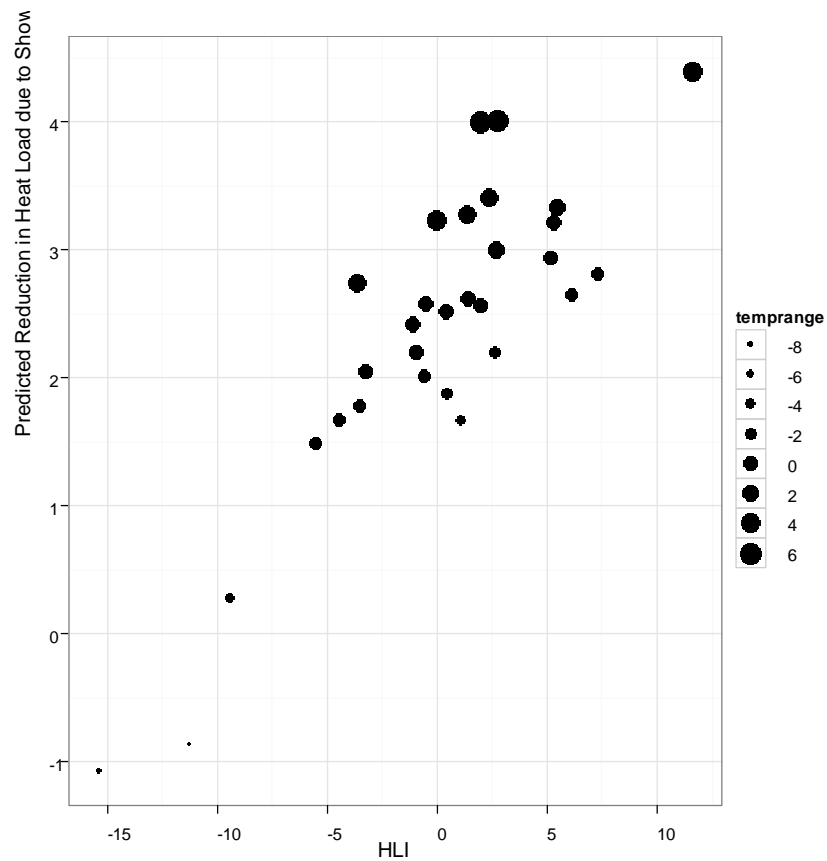


Figure 10: The predicted reduction in heat load due to showering is greater at higher levels of mean *HLI* and *Ta* range. The size of the bubbles reflects size of *Ta* range. $\text{range.Reduction} = 2.5 + 0.1 * \text{HLI} + 0.8 * \text{Ta.range}$



The *HLI* daily mean together with *Ta* range are statistically significant predictors of the heat load experienced by a dairy cow over the course of a day. The *Ta* range may only be statistically significant for heat load because it is circularly related to the amplitude of *Ta* which plays a direct part in the ellipse estimation, and whether it has a practical effect on heat stress or is just a statistical artifact of the way heat load is estimated is an open question. Initial results of a regression on retention, a measure of heat stress that is orthogonal to the ellipses x axis, suggest that it does not. It is important to note that after the inclusion of mean *HLI* in the model, neither *HLI* range nor mean *Ta* lead to a reduction in the DIC. If the range of the heat challenge during the day truly has an effect on heat stress this effect should be more visible from *HLI* range than *Ta* range, as *HLI* itself has a stronger relationship with internal heat load than *Ta*.

Other models that replace the *HLI* terms with mean *THI* or mean *Ta* result in a DIC that is actually greater than that from simply omitting the *HLI* terms as can be seen in Table 7.

Table 7: Deviance Information Criteria (DIC) for models using various environmental inputs

Model	DIC
<i>HLI</i>	494.0
<i>THI</i>	497.4
<i>Ta</i>	497.5
No environmental input outside of <i>Ta</i> range	494.4

3.e. Comparison to Previous Results

The introduction of the heat load model provides evidence for a difference in heat stress between the two treatment groups, control and shower, that enhances results based on summary statistics. Legrand et al. (2011) reported a mean *Tb* for the 2 groups that was roughly identical at 38.9° C (p=0.568). Moreover they reported that t-tests on the daily minimum, maximum, and amplitude

of Tb were also statistically non-significant with p-values greater than 0.3. However when Tb was at its peak (18:00 to 21:00), threehourly mean Tb differences were detected ($p < 0.05$).

4. Conclusion

Internal heat load provides a quantifiable measure of heat stress over the course of a day that has the potential to provide more information than either raw Tb or daily statistical summaries. Harmonic2 ellipses fit to data from dairy cows housed outdoors were used to detect a statistically significant reduction in internal heatload associated with access to a cow-controlled shower. This reduction increases with the size of the heat challenge as measured by the Ta range and HLL , suggesting that dairy cows increase their shower usage on hotter days (as shown by Legrand et al., 2011) and that this extra shower use is effective in reducing heat load. Information on shower use by day would be needed to test the causal reason for this interaction effect. The HLL is more closely related to heat load, than either Ta or THI , possibly because it contains information about wind speed and solar radiation.

Bootstrapped two-stage harmonic least squares area estimates come bundled with a bootstrap standard error. This standard error can be used to place greater importance on area estimates that have greater precision using either 1) weighted least squares or 2) a Bayesian analysis that models variance along with area. Future users of bootstrapped internal heat load estimates should use one of these methods to correctly specify variance matrices of area prediction models when the assumption of controlled sinusoidal input is tenuous.

Internal heat load reduces a day's worth of information into a single number, and internal heat load provides a measure of heat stress that is compelling theoretically and useful clinically. In the case illustrated here Bayesian analysis of heat load in conjunction with *HLI* and *Ta* range shows that shower use reduces heat load and that this treatment effect increases with the size of the heat challenge.

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