

1 **Vertical back movement of cows during locomotion: Detecting lameness with a simple**
2 **image processing technique**

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6 **SUPPLEMENTARY FILE**

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8 **Supplementary materials and methods**

9 The images were processed in three main steps:

10 i: Image pre-processing; ii: Image processing; iii: Decision.

11

12 *i. Image pre-processing:* The images were processed by using software (Matlab R2018a,
13 MathWorks Inc). In this step, stored images (Figure 1a) were prepared for calculations and
14 decision process with the effort of clear, low-sized “black & white” (B&W) images with a
15 minimum number of image errors. Firstly, the colored (RGB–Red-Green-Blue) images were
16 clipped to get the desired region of interest (ROI, back arch of cows) (Figure 1b). Images were
17 converted to 1080×1920×1 size, including only the red dimension, due to easy image extraction
18 in front of the red background (Figure 1c). This 1080x1920x1-sized image was filtered using
19 a 5x5 median filter to obtain a smoother image (Figure 1d). Then, the images were converted
20 to a complete B&W image for the ROI extraction (Figure 1e; the cow is black, and the
21 background is white). Another search was done on images to detect and correct the noise and
22 pixel errors by finding the small and distinct white areas. Thus, the distinct white dots that
23 artifacts may cause in the image were also converted to black to obtain a complete black cow
24 image (Figure 1f).

25

26 *ii. Image Processing/Analysis:* Each complete black cow image was processed to obtain a
27 clear back posture image of the cows. Therefore, to identify the ROI (back arch of cows), the
28 head (Figure 2a, L_h) and tail (Figure 2a, L_t) parts were ignored in each image. Thus, the
29 problems that arise from the movement of the head and tail are eliminated and the highest point
30 of the cow’s back was achieved (Figure 2b, L_b).

31

32 This kind of black and white image is being stored in 2D matrices including 0’s (black) and
33 1’s (white) only (Figure 3). Therefore, the highest point of the cow’s back arch was calculated
34 in the computer by a simple counting process of zeros in each column.

35

36 The number of black pixels was calculated by counting the number of 0's in each column of
37 each matrix. This number also corresponds to the height of each column as well. Then, the
38 maximum of each column gives the maximum height of the overall image (cow back arch).
39 This number was stored in memory and the same procedure was repeated until the end of the
40 same cow's movement was finished. For example, if we had N images of the same cow while
41 it was walking, N-many numbers, which represent the maximum height of each image, were
42 recorded in memory. These N-recorded data were stored for use in the decision step of ALDS.

43

44 *iii. Decision:* In the previous steps, the image was processed and the highest points in each
45 image were stored. This procedure was applied to all cows and a data matrix is generated as

46 follows:

$$\mathbf{C} = \begin{bmatrix} f_{1,1} & \cdots & f_{1,N_1} \\ \vdots & \ddots & \vdots \\ f_{M,1} & \cdots & f_{M,N_M} \end{bmatrix}$$

47

48 where M is the total number of cows (in our case M=63), N_i is the number of images for the i^{th}
49 cow. Each row of this matrix was allocated for data of different cows as:

50
$$\mathbf{C}_i = [f_{i,1}, f_{i,2}, \dots, f_{i,N_i}]; i = 1, 2, \dots, M$$

51

52 If there was no cow inside the image, the maximum height (or the maximum number of black
53 pixels) tended to be zero; and if a cow was visible in the image, the height was increased and
54 changed from image to image. Similarly, when the cow was not invisible in the image, the
55 height decreased and reached zero as expected. An example plot of stored \mathbf{C}_i 's for the same
56 cow with LS=5 is given in Figure 4a.

57

58 The mean value of these curves depends on the cow's size (height) and the length of the
59 recorded data (N_i 's). Hence, the data had to be normalized for a correct decision-making
60 procedure. In this step, thresholding and normalization were applied as well.

61

62 Normalization was applied for each cow separately as $\mathbf{D}_i = \alpha_i \mathbf{C}_i$ ($i = 1, 2, \dots, M$) where the α_i
63 is the normalization coefficient for i^{th} cow calculated as:

64
$$\alpha_i = \frac{\sqrt{\mathbf{C}_i \mathbf{C}_i^T}}{N_i}; i = 1, 2, \dots, M$$

65

66 where ‘T’ represents vector-transpose operation, M is the total number of cows, N_i is the
 67 number of images for the i^{th} cow. Using recorded \mathbf{C}_i 's, α_i was calculated for the i^{th} cow as:

$$68 \quad \alpha_i = \frac{\sqrt{\mathbf{C}_i \mathbf{C}_i^T}}{N_i} = \frac{\left([f_{i,1}, f_{i,2}, \dots, f_{i,N_i}] \begin{bmatrix} f_{i,1} \\ f_{i,2} \\ \vdots \\ f_{i,N_i} \end{bmatrix} \right)^{1/2}}{N_i} = \frac{\sqrt{\sum_{n=0}^{N_i} (f_{i,n})^2}}{N_i}; i = 1, 2, \dots, M$$

69

70 Hence, after normalization, the new data is:

$$71 \quad \mathbf{D}_i = \frac{\sqrt{\sum_{n=0}^{N_i} (f_{i,n})^2}}{N_i} \mathbf{C}_i = \alpha_i [f_{i,1}, f_{i,2}, \dots, f_{i,N_i}]; (i = 1, 2, \dots, M)$$

72 Then thresholding was applied to eliminate non-desired regions. A threshold level (β_i) was
 73 chosen separately for each cow using mean and standard deviation (std) values of \mathbf{D}_i as:

$$74 \quad \beta_i = \text{mean}(\mathbf{D}_i) - \frac{\text{std}(\mathbf{D}_i)}{2}; i = 1, 2, \dots, M$$

75

76 \mathbf{D}_i was a $1 \times N_i$ size vector hence, it includes N_i data. By using threshold value (β_i) a new vector
 77 ($\widehat{\mathbf{D}}_i$) was arranged by deleting some data in \mathbf{D}_i ($\alpha_i f_{i,n}; i = 1, 2, \dots, M$ and $n = 1, 2, \dots, N_i$) using
 78 the algorithm as:

- 79 1. Take $\mathbf{D}_i = \alpha_i [f_{i,1}, f_{i,2}, \dots, f_{i,N_i}]$
- 80 2. Take $n=1, m=1$
- 81 3. If $\alpha_i f_{i,n} \geq \beta_i$
 - 82 a. $g_{i,k} = f_{i,n}$
 - 83 b. $n=n+1, k=k+1$
- 84 4. Else
 - 85 a. $n=n+1$
- 86 5. If $n \leq N_i$, go to Step 3
- 87 6. Else
 - 88 a. $K_i = k$; (final length of the new vector)
 - 89 b. New vector $\widehat{\mathbf{D}}_i$ (size “ $1 \times K_i$ ”) is ready
 - 90 c. Stop

91

92 The effect of this procedure can be seen in Figure 4b. After thresholding, a new vector (g) was
93 generated from f values as $\widehat{\mathbf{D}}_i = [g_{i,1}, g_{i,2}, \dots, g_{i,K_i}]$; ($i = 1, 2, \dots, M$), ($K_i \leq N_i$). Using the
94 new vector $\widehat{\mathbf{D}}_i$ the locomotion score (LS) for i^{th} cow was determined by the following decision
95 algorithm:

- 96 1. Take $\widehat{\mathbf{D}}_i = [g_{i,1}, g_{i,2}, \dots, g_{i,K_i}]$
- 97 2. Calculate the mean of $\widehat{\mathbf{D}}_i \leftrightarrow \mu_i$
- 98 3. Calculate the standard deviation of $\widehat{\mathbf{D}}_i \leftrightarrow \sigma_i$
- 99 4. Calculate lameness factor θ_i as $\leftrightarrow \theta_i = \left(\frac{\sigma_i}{\mu_i}\right) * 100$
- 100 5. Decide the new computerized automatic lameness score (ALDS)
 - 101 a. If $\theta_i < \rho_1 \rightarrow ALDS = 1$ (normal)
 - 102 b. If $\rho_2 > \theta_i \geq \rho_1 \rightarrow ALDS = 2$
 - 103 c. If $\rho_3 > \theta_i \geq \rho_2 \rightarrow ALDS = 3$
 - 104 d. If $\rho_4 > \theta_i \geq \rho_3 \rightarrow ALDS = 4$
 - 105 e. If $\theta_i \geq \rho_4 \rightarrow ALDS = 5$ (severely lame)
- 106 6. Stop

107

108 In step 4, the calculated lameness factor (θ_i) is very small; hence it is scaled by 100 to convert
109 it into a more interpretable value. Here, $\rho_1, \rho_2, \rho_3, \rho_4$ are threshold values for the LS procedure
110 and must be chosen carefully for each farm. As explained in section 2.2, 12 cows were reserved
111 to determine these threshold values for the calibration of the proposed system.

112

113 Initially, colored images were clipped to obtain the region of interest (ROI), which in this case
114 is the back arch of cows. Images were converted to a single-channel black and white format
115 (1080×1920×1) to facilitate easy extraction against a red background. A 5×5 median filter was
116 applied to smooth the images. Further processing involved the removal of noise and pixel errors
117 by identifying and correcting small white areas, ensuring clear black cow images. Each black
118 cow image underwent processing to extract a clear back posture. The head and tail parts of the
119 cow were ignored in each image to eliminate potential movement artifacts, enabling the
120 identification of the highest point of the cow's back. The black and white images were stored
121 in 2D matrices comprising only 0s (black) and 1s (white). The height of the cow's back arch
122 was determined by counting the number of black pixels in each column, with the maximum
123 height recorded for each image. These recorded heights were stored for subsequent analysis in

124 the decision-making step. The processed images and recorded heights were used to generate a
125 data matrix for all cows. Each cow's data was normalized to account for differences in size and
126 recorded data length. Thresholding was applied using mean and standard deviation values to
127 eliminate non-desired regions and create a new vector. This new vector represented the
128 processed data after thresholding. Using this vector, the locomotion score (LS) for each cow
129 was determined based on a decision algorithm. The calculated lameness factor was scaled by
130 100 to ensure interpretability. Threshold values for the LS procedure (ρ_1 , ρ_2 , ρ_3 , ρ_4) were
131 carefully chosen for the farm and used to assign the new computerized automatic lameness
132 score (ALDS).

133

134 **Supplementary Figure S1:**

135 The walking path at the exit of the milking parlor.

136

137 **Supplementary Figure S2:**

138 Camera and walking path configuration.

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140 **Supplementary Figure S3:**

141 Diagram illustrating the summary of the study's materials and methods.

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143

144 **Supplementary Figure S1:**

145



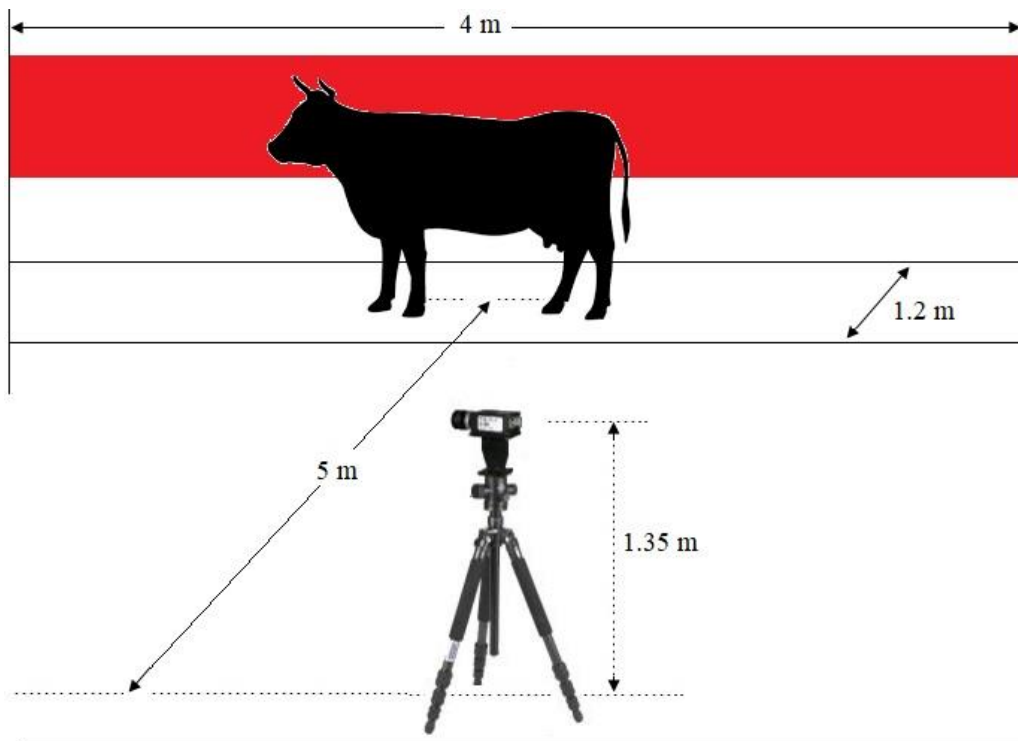
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149 **Supplementary Figure S2:**

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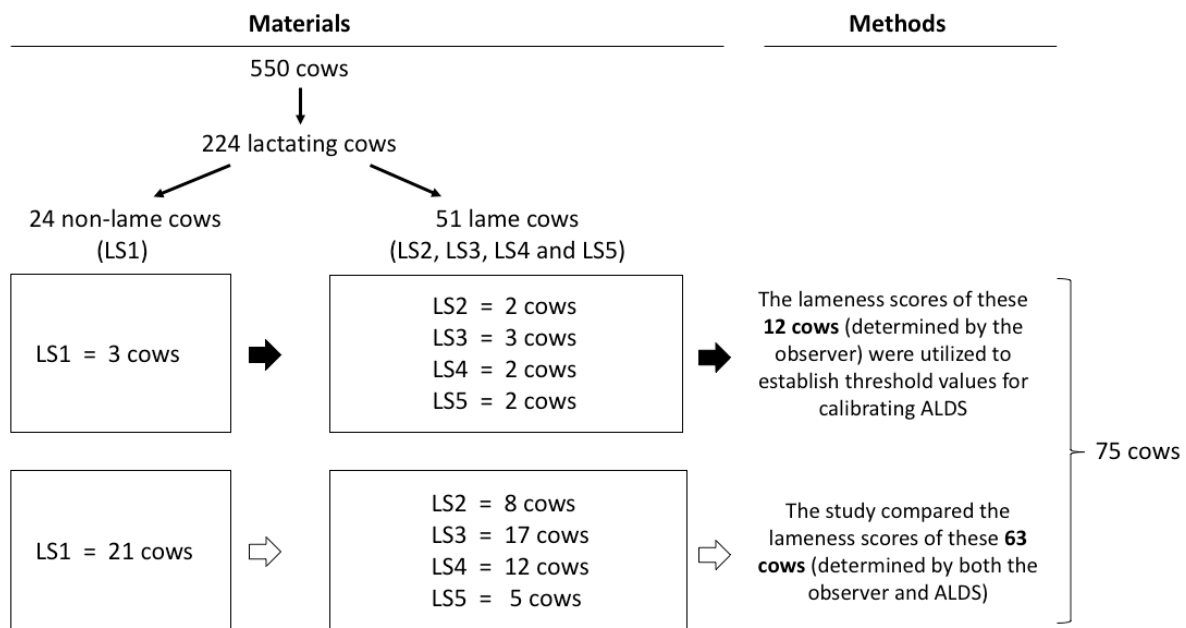
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154 **Supplementary Figure S3:**

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