Biological characterization of the estrous cycle in lactating Holstein cows

Y. Montanholi¹, S. Bourgon¹, A. Macdonald², P. Park², S. Lam², M. Kozak², B. Potvin², K. Colliver², L. Haas², A. Rocha², S. Miller²,³
Rationale & objective

"Map" biological responses to estrous state

Multi-stream assessment of estrous in dairy cows

Profitable Dairy Management

InterAg

Ontario

EAZI-BREED CIDR

AAC Funding Agri-Ideas

Agricultural Association Council
Rationale & objective

- Dairy cow fertility: increase in productivity

Pryce et al 2014
Rationale & objective

- **Dairy cow fertility: increase in productivity**

<table>
<thead>
<tr>
<th>Traits</th>
<th>High</th>
<th>Average</th>
</tr>
</thead>
<tbody>
<tr>
<td>Days to estrous</td>
<td>66</td>
<td>43</td>
</tr>
<tr>
<td>Ovulations before estrous</td>
<td>1.6</td>
<td>0.7</td>
</tr>
<tr>
<td>Days to conception</td>
<td>217</td>
<td>74</td>
</tr>
</tbody>
</table>

Harrison et al 1990
Rationale & objective

- Dairy cow farming: increased herd size

China constructs 100,000-cow dairy unit

The project will be the world’s largest dairy farm and will supply milk and cheese to Russia
Rationale & objective

- Milk composition
  - “Classical” composition
  - Freezing point
  - Fatty acids profile
  - Cortisol
  - Progesterone

- Standing vs. lying

- Fecal cort & P4

- Heart rate

- Vulva morphometry

- Body surface temperature
  - Infrared imaging
  - Contact temperature

Day of Cycle
Material and methods

Dairy cows: housing, feeding and biometrics

**Ingredients**

<table>
<thead>
<tr>
<th>Ingredients</th>
<th>% DM</th>
</tr>
</thead>
<tbody>
<tr>
<td>Wheat straw</td>
<td>30.80</td>
</tr>
<tr>
<td>Alfalfa haylage</td>
<td>11.60</td>
</tr>
<tr>
<td>Corn silage</td>
<td>34.90</td>
</tr>
<tr>
<td>Premix</td>
<td>22.70</td>
</tr>
</tbody>
</table>

**Performance**

<table>
<thead>
<tr>
<th>Performance</th>
<th>Value</th>
</tr>
</thead>
<tbody>
<tr>
<td>TMR feed intake (kg/d)</td>
<td>44.08</td>
</tr>
<tr>
<td>Milk yield (kg/d)</td>
<td>33.20</td>
</tr>
<tr>
<td>Crude fat content (%)</td>
<td>3.58</td>
</tr>
<tr>
<td>Crude protein (%)</td>
<td>2.96</td>
</tr>
<tr>
<td>Lactose content (%)</td>
<td>4.63</td>
</tr>
</tbody>
</table>

**Temperature Humidity Index**

July 15 to September 20
Material and methods

- Experimental design
Material and methods

- Estrous synchronization

Insert CIDR (progesterone)

-7

PGF2α

-1

Remove CIDR

0

Check for estrous

Estrous synchronization

CIDR (progesterone)

PGF2α

Remove CIDR

Check for estrous
Material and methods

Experimental timeline

Day - X  Day 1  Day 19  Day 22  Day 24

<table>
<thead>
<tr>
<th>Parameter</th>
<th>AM/PM</th>
<th>FULL</th>
<th>FULL</th>
<th>FULL</th>
<th>AM/PM</th>
</tr>
</thead>
<tbody>
<tr>
<td>Infrared imaging</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Surface temp.</td>
<td>FULL</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Heart rate</td>
<td></td>
<td>FULL</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Milk composition</td>
<td>AM/PM</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Vulva metrics</td>
<td>AM/PM</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>
Material and methods

- **Milk analysis:** progesterone and cortisol

- **Coat-A-Count Cortisol**
- **Coat-A-Count Progesterone**
Material and methods

- **Milk analysis**: composition and freezing point

<table>
<thead>
<tr>
<th></th>
<th>FAT</th>
<th>PROTEIN</th>
<th>UREA</th>
<th>LACTOSE</th>
<th>FREEZING POINT</th>
<th>SOMATIC CELL COUNT</th>
<th>Milkoscan FT6000</th>
<th>Fossomatic FC</th>
</tr>
</thead>
<tbody>
<tr>
<td>AM/PM</td>
<td>Milk</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

**Milk analysis**

- Composition:
  - Fat
  - Protein
  - Urea
  - Lactose

- Freezing point analysis:
  - Milkoscan FT6000
  - Fossomatic FC
Material and methods

- **Milk analysis:** Fatty-acid profile determination

J. Dairy Sci. 89:3690–3695

Estimating Fatty Acid Content in Cow Milk Using Mid-Infrared Spectrometry

H. Soyeurt,*†1,2 P. Dardenne,‡ F. Dehareng,‡ G. Lognay,*2 D. Veselko,§ M. Marlier,*2 C. Bertozzi,# P. Mayeres,*#2 and N. Gengler*||2

Nicolas Gengler
Clément Grelet
Amélie Vanlierde

Hélène Soyeurt
Frédéric Dehareng
Material and methods

- **Milk analysis**: Fatty-acid profile determination

- Trans FA
- C18:1 isomers
- Unsaturated
- Saturated
- Odd FA
- Omega 6
- Omega 3
- Iso & anteiso
- Long chain FA
- Mid chain FA
- Poly-unsaturated
- Small chain FA
- Mono-unsaturated
Material and methods

- Body surface temperature: thermochrons
Material and methods

- **Body surface:** Infrared imaging instrumentation

  - **Model A320**
    - Automated

  - **Model SC2000**
    - Handheld
Material and methods

- **Body surface**: Infrared imaging analyses

- **Right and left flanks**
- **Front and back foot**
- **Vulva**

**Maximum & Average temp**
Material and methods

Vulva morphometry
Material and methods

- Heart rate: Electrode-based heart rate

Model RS800 CX
Material and methods

- **Statistical analysis**

Repeated measures mixed models - THI covariate
Results and discussion

- **Milk analysis: progesterone**

- Estrus: >2.4 ng/mL (Roelofs et al 2006)

- Estrus cycle duration: 17 to 25 days
Results and discussion

**Milk analysis: cortisol (ng/mL)**

<table>
<thead>
<tr>
<th>State</th>
<th>Morning</th>
<th>Evening</th>
</tr>
</thead>
<tbody>
<tr>
<td>Non-estrous</td>
<td>1.32</td>
<td>1.09</td>
</tr>
<tr>
<td>Estrous</td>
<td>1.31</td>
<td>1.69</td>
</tr>
<tr>
<td><strong>P value</strong></td>
<td>0.94</td>
<td>0.02</td>
</tr>
</tbody>
</table>

- THI & cortisol $r = 0.23$ ($P = 0.03$)
- 86% of THI > 71 - evening
- Fecal cortisol metabolites: another dimension
Results and discussion

- **Milk analysis: cortisol**

Swanson et al. 1972
Results and discussion

**Milk analysis: composition & freezing point**

<table>
<thead>
<tr>
<th>Analyte</th>
<th>Estrous</th>
<th>Non-estrous</th>
<th>P value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Fat (%)</td>
<td>3.48</td>
<td>2.94</td>
<td>0.11</td>
</tr>
<tr>
<td>Lactose (%)</td>
<td>4.68</td>
<td>4.59</td>
<td>0.07</td>
</tr>
<tr>
<td>Protein (%)</td>
<td>2.82</td>
<td>2.95</td>
<td>0.04</td>
</tr>
<tr>
<td>Urea (mg/dL)</td>
<td>121</td>
<td>113</td>
<td>0.33</td>
</tr>
<tr>
<td>Somatic cell count (1000s/mL)</td>
<td>249</td>
<td>622</td>
<td>0.05</td>
</tr>
<tr>
<td>Freezing point (°C)</td>
<td>-0.560</td>
<td>-0.555</td>
<td>0.03</td>
</tr>
</tbody>
</table>

FAT = Lactose

SCC Freezing point

Protein
Milk analysis: Mid-infrared spectra for fat acids

<table>
<thead>
<tr>
<th>Fat acids (g/L)</th>
<th>Estrous</th>
<th>Non-estrous</th>
<th>P value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Mono-unsaturated</td>
<td>1.14</td>
<td>0.92</td>
<td>0.03</td>
</tr>
<tr>
<td>Poly-unsaturated</td>
<td>0.14</td>
<td>0.12</td>
<td>0.08</td>
</tr>
<tr>
<td>Unsaturated</td>
<td>1.29</td>
<td>1.04</td>
<td>0.03</td>
</tr>
<tr>
<td>Long chain</td>
<td>1.68</td>
<td>1.33</td>
<td>0.02</td>
</tr>
<tr>
<td>Omega 3</td>
<td>0.025</td>
<td>0.020</td>
<td>0.09</td>
</tr>
<tr>
<td>Omega 6</td>
<td>0.091</td>
<td>0.079</td>
<td>0.09</td>
</tr>
<tr>
<td>Trans</td>
<td>0.16</td>
<td>0.13</td>
<td>0.04</td>
</tr>
<tr>
<td>C18:1 isomers</td>
<td>1.02</td>
<td>0.81</td>
<td>0.02</td>
</tr>
</tbody>
</table>
Results and discussion

- **Body surface temperature:** Contact device

---

![Graph of milk progesterone levels during the estrous cycle](image)

- **Proestrus**
- **Non-estrous**
- **Estrous**

<table>
<thead>
<tr>
<th>Days of the estrous cycle</th>
<th>Milk progesterone (ng/mL)</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>0</td>
</tr>
<tr>
<td>6</td>
<td>12</td>
</tr>
<tr>
<td>11</td>
<td>24</td>
</tr>
<tr>
<td>16</td>
<td>48</td>
</tr>
<tr>
<td>21</td>
<td>60</td>
</tr>
</tbody>
</table>

- **Morning**
- **Evening**
Results and discussion

- **Body surface temperature**: Contact device

Differing letters within body location are different.
Results and discussion

- **Body surface temperature**: Contact device

Marrone et al 1976
Results and discussion

**Body surface temperature**: Infrared imaging

<table>
<thead>
<tr>
<th>Location</th>
<th>Estrous</th>
<th>Non-estrous</th>
<th>P value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Foot</td>
<td>31.24</td>
<td>30.48</td>
<td>0.08</td>
</tr>
<tr>
<td>Flank</td>
<td>34.70</td>
<td>33.39</td>
<td>0.03</td>
</tr>
<tr>
<td>Vulva</td>
<td>34.41</td>
<td>33.90</td>
<td>0.11</td>
</tr>
</tbody>
</table>

Hurnik et al 1985
Results and discussion

- Vulva morphometry

Halachmi et al 2008
Results and discussion

**Heart rate**

<table>
<thead>
<tr>
<th>Condition</th>
<th>Heart Rate (BPM)</th>
<th>P Value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Non-estrus</td>
<td>86.02</td>
<td>0.01</td>
</tr>
<tr>
<td>Estrus</td>
<td>87.64</td>
<td></td>
</tr>
</tbody>
</table>

≠ ~2,300 beats/day
Remarks

- Possibilities for inline milk analysis
- Possibilities for body surface temperature monitoring

Technologies based on multi-proxies and multi-proxies with application for $$$ / ENVIRO relevant traits.
Financial & technical support

Canwest DHI
Profitable Dairy Management

Gemboux Agro-Bio Tech
Université de Liège

Ontario

InterAg
Delivering innovative technologies

AAC
Funding Agri-Ideas