Effect of milking interval on alveolar and cisternal compartments in the udder of dairy sheep
(S.01, #6, p. 3)

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Dairy sheep industry (FAOstat, 2004):
- World: 220 M of dairy sheep producing 8.2 Mt of milk
- Europe: 15% dairy sheep producing 35% milk (2.9 Mt)
- Mainly located in the Mediterranean countries
- Oriented to dairy products (cheese & yogurt)
Introduction (2/2)

- **Structural constraints**
  - Increasing flock sizes (150 to 10,000 ewes)
  - Low unitary milk yield (0.2 to 2.0 L/milking)
  - Milking (2X daily) represents >50% working time in sheep dairy farms
  - High milking costs (0.1 to 0.3 €; ×100 dairy cow costs)

- **Biological features:**
  - Larger cisternal compartment (40-80%) than dairy cows
  - Large “effective storage volume” (~3 L)

\[
V = \pi r^2 h = 9,425 \text{ cm}^3
\]

- Gland:Milk = 1:1
- Cisternal (70%) = 3.2 L
- Alveolar (30%) = 1.4 L
Objectives

To study the effect of milking interval on:

- Udder compartments (alveolar & cisternal)
- Milk yield
- Milk composition
- Indicators of mammary epithelial cell tight junctions opening (plasma and milk)

In 2 dairy ewe breeds at same lactation stage and under similar management conditions:

- Manchega (medium yield, high composition)
- Lacaune (high yield, medium composition)
Material and Methods (1/3)

24 dairy ewes at mid lactation (DIM, 70 ± 3) with healthy udders (SCC, 64 ± 48 ×10³ cells/mL; <5 cfu/mL), penned and fed indoors (INRAtion v. 3.3):

- **Manchega** (n = 12; 70 kg BW; milk, **1.11 ± 0.09 L/d**)
- **Lacaune** (n = 12; 76 kg BW; milk, **2.32 ± 0.11 L/d**)

**Milking:** 2×12 parallel stalls milking parlor (Westfalia-Surge; 42 kPa, 120 p/min and 50%)

- **Regular milkings (2X): 14-10 h** (8 am and 6 pm).
- **Experimental milkings:** 6 groups of 4 ewes according to a crossover design with 6 milking intervals (4, 8, 12, 16, 20, and **24 h**) at random, replicated at wk 11 and 16.

<table>
<thead>
<tr>
<th>Group</th>
<th>Ewes, n</th>
<th>Treatment order</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>1</td>
</tr>
<tr>
<td>1</td>
<td>4</td>
<td>24 h</td>
</tr>
<tr>
<td>2</td>
<td>4</td>
<td>4 h</td>
</tr>
<tr>
<td>3</td>
<td>4</td>
<td>16 h</td>
</tr>
<tr>
<td>4</td>
<td>4</td>
<td>20 h</td>
</tr>
<tr>
<td>5</td>
<td>4</td>
<td>8 h</td>
</tr>
<tr>
<td>6</td>
<td>4</td>
<td>12 h</td>
</tr>
</tbody>
</table>
Milk compartments:
- **Residual milk removal (0 h):** milking after i.v. injection of oxytocin (2 IU/ewe; Veterin Lobulor, Lab. Andreu, Barcelona).
- **Cisternal area (4, 8... and 24 h):** B-ultrasonography (Ultra Scan 900, Ami Medical Alliance, Montreal; Ruberte et al. (1994) after i.v. injection of an oxytocin blocking agent (0.8 mg/ewe; Tractocile, Ferrin lab., Madrid; Rovai et al., 2008).
- **Cisternal milk (4, 8... and 24 h):** milking under Tractocile effects (<18 min; Wellnitz et al., 1999).
- **Alveolar milk (4, 8... and 24 h):** milking after i.v. injection of oxytocin (2 IU/ewe).

Milk composition: NIRA (InfraAlyzer-450, Bran+Luebbe, Nordersted) for total solids, fat, protein, true protein, casein, and lactose.
- **Cisternal milk (4, 8... and 24 h)
- **Alveolar milk (4, 8... and 24 h)

SCC (Somatic Cells Count): automatic cell counter (Fossomatic 5000, Foss Electric, Hillerød) calibrated for sheep milk.
Blood sampling and analyses: at each milking interval from the jugular vein using heparinized tubes. Refrigerated (4°C) and centrifuged (490 × g, 15 min) for plasma (−20°C).

- **Lactose**: enzymatic assay (Lactose/D-Galactose UV-method; Boehringer Mannheim/R-Biopharm, Darmstadt).
- **Na and K**: Inductively coupled plasma atomic emission spectroscopy (Chemical Analysis Service, UAB).

Statistical Analyses: PROCMIXED of SAS v.8.2. and differences between LSM were determined with the PDIFF test of SAS.
Results (1/7): Milk accumulation in the ewe’s udder according to milking interval and breed

- **Manchega**
  
  \[ y = 37.5 \times x \]
  
  \( R^2 = 0.98 \)
  
- **Lacaune**
  
  \[ y = 86.6 \times x \]
  
  \( R^2 = 0.99 \)

**Notes:**
- a, b, ..., f \( P < 0.05 \)
Results (2/7): Cisternal area according to milking interval and ewe breed

![Graph showing cisternal area in Manchega and Lacaune breeds over different milking intervals with significance levels indicated.](image-url)
Results (3/7): Milk fat content in the ewe’s udder according to milking interval and breed

Manchega

Lacaune

Cisternal  Alveolar  Total

Fat (%)

Milking interval (h)

a, b, ... f  $P < 0.05$
Results (4/7): Milk total protein content in the ewe’s udder according to milking interval and breed.

- **Cisternal**: Shows a steady increase in protein content over the milking intervals.
- **Alveolar**: Demonstrates a significant rise in protein content, especially noticeable at longer intervals.
- **Total**: Reflects a combined trend from both Cisternal and Alveolar, indicating overall protein changes.

**Species Comparison**:
- **Manchega**: Exhibits a consistent pattern with slight variations across intervals.
- **Lacaune**: Displays a more pronounced change, particularly evident at intervals greater than 16 hours.

**Statistical Significance**: 
- **a, b** indicate a p-value of less than 0.05, suggesting significant differences between the groups.
Results (5/7): Milk lactose content in the ewe’s udder according to milking interval and breed

- **Manchega**
  - **Cisternal**
  - **Alveolar**
  - **Total**

- **Lacaune**
  - **Cisternal**
  - **Alveolar**
  - **Total**

\[ a, b \] \( P < 0.05 \)
Results (6/7): Milk Na and K content in the ewe’s udder according to milking interval and breed

<table>
<thead>
<tr>
<th>Milking Interval (h)</th>
<th>Na or K (g/L)</th>
<th>Na/K Ratio</th>
</tr>
</thead>
<tbody>
<tr>
<td>4</td>
<td>0.5</td>
<td></td>
</tr>
<tr>
<td>8</td>
<td>0.7</td>
<td>0.2</td>
</tr>
<tr>
<td>12</td>
<td>0.9</td>
<td>0.3</td>
</tr>
<tr>
<td>16</td>
<td>1.2</td>
<td>0.4</td>
</tr>
<tr>
<td>20</td>
<td>1.5</td>
<td>0.5</td>
</tr>
<tr>
<td>24</td>
<td>1.8</td>
<td>0.6</td>
</tr>
</tbody>
</table>

- **Manchega**
- **Lacaune**

\[ a, b, c \quad P < 0.05 \]
Results (7/7): Lactose content in plasma of dairy ewes according to milking interval and breed

Plasma lactose (µmol/L)

Milking interval (h)

Manchega

Lacaune

a, b P < 0.05
Conclusions

- Both dairy breeds stored large amounts of milk within the cisterns and were able to tolerate extended milking intervals during short-term.
- Milk fat content decreased when milking interval was extended, but milk protein steadied.
- Ultrasonography was useful for evaluating cistern size and identifying large-cisterned ewes.
- High-yielding ewes presented greater cisternal compartment suggesting easier adaptation to extended milking intervals.
- Tight junction disruption differed according to breed, being earlier in Manchega (small cisterns) than in Lacaune ewes (large cisterns).
- Dairy ewes may be able to support 20 h of udder filling without negative effects on milk yield or milk composition, making possible the use of once-a-day or occasional (weekend) milking omission schedules.
Thanks for your attention!