Milk quality essentials for dairy manufacturers

Hilton Deeth
(h.deeth@uq.edu.au)

NCDE/Dairy Australia Webinar 10 February 2016
Introduction

To ensure processed dairy products (milk, cheese, etc) are of high quality, we need to start with a good quality raw milk and treat the finished products properly.

Main elements of milk quality are:

• Chemical aspects (composition, lipolysis, light exposure)
• Somatic cell count/mastitis
• Bacteria and their enzymes
• Sensory characteristics – smell, taste
• Presence/absence of contaminants
Chemical aspects
Chemical aspects

Milk composition (approximate)

- Lactose 5%
- Fat 4%
- Protein 3%
  - includes enzymes – phosphatase, lipase, protease (plasmin)
- Minerals ~1%
- Vitamins, carotene (yellow colour)
- Chemical contaminants – inhibitory substances, taints

Many factors affect milk composition – hence it is impossible to give precise compositional data
Chemical aspects - factors affecting milk composition

Stage of lactation

- colostrum (day 1 - day 4 after calving) is very different from mature milk

<table>
<thead>
<tr>
<th></th>
<th>Colostrum (day 1)</th>
<th>Milk (day 6)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Total solids (%)</td>
<td>23.9</td>
<td>12.9</td>
</tr>
<tr>
<td>Protein (total) (%)</td>
<td>14.0</td>
<td>3.1</td>
</tr>
<tr>
<td>• Casein (%)</td>
<td>4.8</td>
<td>2.5</td>
</tr>
<tr>
<td>• Immunoglobulins (%)</td>
<td>6.0</td>
<td>0.09</td>
</tr>
<tr>
<td>Fat (%)</td>
<td>6.7</td>
<td>4.0</td>
</tr>
<tr>
<td>Ash (minerals) (%)</td>
<td>1.1</td>
<td>0.7</td>
</tr>
<tr>
<td>Vitamin B12 (mg/L)</td>
<td>49</td>
<td>6</td>
</tr>
</tbody>
</table>

- Colostrum up to 5-days should not be included in bulk milk
- Colostrum has low clotting temperature and causes fouling in heat exchangers
Chemical aspects - factors affecting milk composition 2

- Late-lactation milk is different from early and mid-lactation milk
  - Higher protein concentration
  - Fat concentration does not change but free fatty acid level is higher due to fat breakdown (lipolysis)
  - Higher somatic cell count
  - Higher plasmin (protease) activity
  - Higher non-protein nitrogen level due to plasmin activity in udder
  - Longer clotting time
    - Leads to high moisture cheese
  - Reduces stability of milk powders

Stage of milking

- Fat content lowest in first & highest in last portion
Animal species and breed

- Composition of milk from different animal species and different breeds varies considerably.
## Composition of milk of different animals

<table>
<thead>
<tr>
<th>Species</th>
<th>Total solids %</th>
<th>Fat %</th>
<th>Protein %</th>
<th>Lactose %</th>
<th>Ash %</th>
</tr>
</thead>
<tbody>
<tr>
<td>Cow</td>
<td>12.7</td>
<td>3.7</td>
<td>3.4</td>
<td>4.8</td>
<td>0.7</td>
</tr>
<tr>
<td>Goat</td>
<td>12.3</td>
<td>3.9</td>
<td>3.2</td>
<td>4.5</td>
<td>0.8</td>
</tr>
<tr>
<td>Camel</td>
<td>15.0</td>
<td>5.4</td>
<td>3.8</td>
<td>5.2</td>
<td>0.7</td>
</tr>
<tr>
<td>Buffalo</td>
<td>16.8</td>
<td>7.4</td>
<td>3.8</td>
<td>4.8</td>
<td>0.8</td>
</tr>
<tr>
<td>Sheep</td>
<td>19.3</td>
<td>7.4</td>
<td>4.5</td>
<td>4.8</td>
<td>1.0</td>
</tr>
<tr>
<td>Yak</td>
<td>19.3</td>
<td>7.9</td>
<td>5.3</td>
<td>5.2</td>
<td>1.0</td>
</tr>
<tr>
<td>Horse</td>
<td>11.2</td>
<td>1.9</td>
<td>2.5</td>
<td>6.2</td>
<td>0.5</td>
</tr>
<tr>
<td>Human</td>
<td>12.2</td>
<td>3.8</td>
<td>1.0</td>
<td>7.0</td>
<td>0.2</td>
</tr>
<tr>
<td>Blue whale</td>
<td>55.0</td>
<td>40.9</td>
<td>11.9</td>
<td>1.3</td>
<td>1.4</td>
</tr>
</tbody>
</table>
## Composition of milk of various breeds of cows

<table>
<thead>
<tr>
<th>Breed</th>
<th>Fat</th>
<th>Protein</th>
<th>Lactose</th>
</tr>
</thead>
<tbody>
<tr>
<td>Holstein/Friesian</td>
<td>3.5</td>
<td>3.1</td>
<td>4.9</td>
</tr>
<tr>
<td>Ayrshire</td>
<td>4.1</td>
<td>3.6</td>
<td>4.7</td>
</tr>
<tr>
<td>Brown Swiss</td>
<td>4.0</td>
<td>3.6</td>
<td>5.0</td>
</tr>
<tr>
<td>Guernsey</td>
<td>5.0</td>
<td>3.8</td>
<td>4.9</td>
</tr>
<tr>
<td>Jersey</td>
<td>5.5</td>
<td>3.9</td>
<td>4.9</td>
</tr>
</tbody>
</table>
Chemical aspects - lipolysis

Lipolysis or fat hydrolysis

- In raw milk caused by **natural milk enzyme, lipase**
- Milk lipase is killed by pasteurisation
- Forms free fatty acids in raw milk which can cause rancid, unclean off-flavours in milk, butter, cheese
- Also reduces foaming capacity of milk – not good for cappuccino coffee
- Can happen at farm when milk is cooled
  - especially in milk from cows in late lactation – also from cows on poor quality feed
- Can happen at farm or factory if milk is pumped or agitated, with foaming, or if homogenised (pasteurised) milk is mixed with raw milk
**Titratable acidity and pH**

- The acidity of milk increases and pH decreases with bacterial growth
- Normal titratable acidity is ~ 0.11-0.12% lactic acid
- Normal pH is ~6.7
Chemical aspects – effect of light

- Exposure of milk (raw or processed) to light causes production of unpleasant odour and flavour
- All milk should be protected from light wherever possible
- This is a major reason for opaque and coloured plastic bottles and sleeves on bottles
- Problems can arise when milk is held in a display cabinet with fluorescent lighting
  - Example: flavour defects were detected in 2% pasteurised milk in clear plastic bottle at 6°C under fluorescent light in 15-30 min by trained tasters and in 54 min to 2 hours by untrained consumers
  - 50% of milk remains in display cabinets for ≥ 8 hours
- Paperboard cartons with aluminium layer provides excellent light protection
Chemical composition standards

• Raw milk standards set by companies

• For example:
  • fat > 3.2%
  • protein > 3.2%
  • acidity < 0.16% lactic acid
  • pH > 6.6
  • Freezing point should be ~-0.540°H; -0.525°H is international limit – higher values (less negative) indicate added water.
## Chinese standard for raw milk

**Table 2 Chemical and physical requirements**

<table>
<thead>
<tr>
<th>Item</th>
<th>Criteria</th>
<th>Test method</th>
</tr>
</thead>
<tbody>
<tr>
<td>Freezing Point $^{a,b}/(°C)$</td>
<td>-0.500 $\sim$ -0.560</td>
<td>GB 5413.38</td>
</tr>
<tr>
<td>Specific gravity $(20°C/4°C) \geq$</td>
<td>1.027</td>
<td>GB 5413.33</td>
</tr>
<tr>
<td>Protein $(g/100g) \geq$</td>
<td>2.8</td>
<td>GB 5009.5</td>
</tr>
<tr>
<td>Fat $(g/100g) \geq$</td>
<td>3.1</td>
<td>GB 5413.3</td>
</tr>
<tr>
<td>Impurity $(mg/kg) \leq$</td>
<td>4.0</td>
<td>GB 5413.30</td>
</tr>
<tr>
<td>Milk Solids-not-fat $(g/100g) \geq$</td>
<td>8.1</td>
<td>GB 5413.39</td>
</tr>
<tr>
<td>Acidity $(^oT)$</td>
<td></td>
<td>GB 5413.34</td>
</tr>
<tr>
<td>Bovine Milk $^b$</td>
<td>12-18</td>
<td></td>
</tr>
<tr>
<td>Ovine Milk</td>
<td>6-13</td>
<td></td>
</tr>
</tbody>
</table>

$^a$ Inspection after 3 hours of milking  
$^b$ only applies to Holstein cows
Somatic cells
Somatic cells

“Somatic cells” literally means “body cells”

- Mostly white blood cells – technically polymorphonuclear leukocytes or neutrophils
  - reaction to mastitic infection – Streptococci, Staphlococci, coliforms

Some cells are epithelial cells shed from inside of the udder when an infection occurs

Size: 7-15 μm in diameter – much larger than bacteria
Somatic cell counts (SCC)

Problems associated with mastitic milk depend on the bulk SCC (BSCC)

< 150,000/mL Excellent

150,000-250,000/mL Good - below 250,000 is the level for premium payment in most Australian dairy companies; some use 200,000/mL

250,000-400,000/mL Moderate mastitis and cell count control

400,000/mL Limit in Australia, NZ, EU, Canada

>400,000/mL “Not fit for human consumption” (EU)

750,000/mL Legal limit for grade A milk in US

1,000,000 Legal limit in Brazil
Why are we concerned about somatic cells?

Producers lose money through reduced production and treatment costs, and are inconvenienced by having to keep milk separate from bulk supply.

Milk with high somatic cell count (SCC) has changed composition (less casein & whey proteins, more blood proteins, reduced lactose, higher salt levels, increased pH to >7).

Bacteria causing mastitis add to bacterial load but are destroyed by pasteurisation.

Reduced quality and yield of products made from milk with high somatic cell counts (> 250,000/mL).
**Effect of mastitic milk on products**

Somatic cells contain proteases and lipases

Proteases are quite heat-stable and cause bitterness and age gelation in UHT milk.

- the higher the BSCC, the shorter the time to appearance of bitter flavour and gelation in UHT milk

Mastitic milk has higher free fatty acid (FFA) levels than normal milk, due to:

- lipases in somatic cells; and
- incomplete synthesis of triglycerides from FFA pool
Significance of mastitic milk in cheesemaking

Reduced cheese yield:
• Mastitic milk contains more plasmin which degrades protein and causes more protein to be lost in the whey.
• Reduced casein directly affects cheese yield.
• Poor curd formation (longer flocculation time, slower rate of curd firming, and reduced maximum firmness) contributes to yield loss as fines.

Reduced cheese quality:
• Decreased curd strength due to high whey proteins, low caseins, high pH and altered calcium-phosphate-caseinate balance.
• Higher moisture cheese due to impaired curd syneresis.
• Soft, less elastic, sticky and grainy cheese texture.
• Increased flavour intensity, usually with off flavours.
Bacteria and their enzymes
Bacteria in raw milk

All raw milk contains bacteria

Bacteria are < 1 μm in diameter and 2-8 μm long

(1 μm = 0.001 mm = 0.00004 inch)

Have different shapes: some spherical, some rod shaped

Bacteria are the reason for heat treatment of milk

Bacteria produce enzymes which cause milk to spoil

Bacteria come from:

• The cow - udder & teat surface (mastitis)
• The environment – air, water, soil (dust, mud)
• Equipment
• Humans
Growth of bacteria

Bacteria can only “grow” by cell division

1 → 2,   2 → 4,  
4 → 8,   8 → 16,.....
### Types of bacteria

#### Spoilage and pathogenic
- Spoilage bacteria cause milk to “go off”; pathogens cause disease
- Pasteurisation [72°C/15 s or 65°C/15 min] designed to kill pathogen bacteria (and most spoilers)
- Thermisation [65°C/15 s] kills many spoilage bacteria but does not kill all pathogens – designed to increase shelf-life of raw milk

#### Sporeforming and non-sporeforming
- Most bacteria in raw milk do not form spores and are easily killed
- A spore is a dormant [non-growing] state which forms when growth conditions are unfavourable
- Spores are very heat-resistant –
  - not affected by pasteurisation
  - killed by UHT
- Spores turn into a vegetative form can “grow”
Types of bacteria (cont)

• Thermoduric – not killed by pasteurisation
  • Include spores and some non-spore-formers

• Types classified by growth temperature:
  • Mesophiles (grow best around room temperature)
  • Psychrotrophs (will grow at low temperatures although prefer to grow around room temperature)
  • Thermophiles (will only grow >~50°C)

• Lactic acid bacteria:
  • major type of bacteria in raw milk as it comes from the udder
  • grow at room temperature, i.e., they are mesophilic
  • cannot grow at low temperature
  • used to make cheese, yoghurt, etc
Milk bacterial counts

Several types of bacterial count:

- TPC (total plate count)/APC (aerobic plate count)/SPC (standard plate count)
- PC (psychrotroph count)
- thermoduric count/LPC (laboratory pasteurised count)
- spore count (psychrotrophic, mesophilic, thermophilic)
- coliform and \textit{E. coli}
- \textit{Salmonella}
- \textit{Listeria} count
- Stapylococci
- Yeasts and moulds
How is a bacterial count measured?

By a machine such as a Bactoscan or manually

Manual total plate count (TPC) outline:

- Milk sample is diluted, say 1 in 100
- A small amount, say 0.1 mL is put onto a nutritious agar gel in a Petri dish or plate
Doing a TPC manually (cont)

- Plates are incubated at say 30°C for 2-3 days to allow individual bacterial cells to grow (i.e. by dividing)
- the bacterial cells clump together to form a colony like a small bead
- All colonies on a plate are counted
- Plate opposite has 95 colonies

[Note: plates usually have colonies of a variety of shapes, sizes and colours]
Still doing a manual TPC

Working out the count from the plate:

If we count 95 colonies on the plate, that is 950 per 1 mL of diluted sample (we used 0.1 mL) and we have diluted it 100 times; therefore count is 950 x 100 or 95,000 colony forming units (cfu)/mL or $9.5 \times 10^4$ cfu/mL

- good milk has 1,000-10,000 cfu/mL
- poor quality milk has > 100,000 cfu/mL
- very poor quality milk has >1,000,000 cfu/mL
- spoiled milk generally has > 10,000,000 cfu/mL
Counts other than TPC

Other bacterial counts require:

• Some pre-treatment, e.g., heating for thermodurics and spore counts; or

• Special microbiological media. e.g., coliforms; or

• Special incubation conditions, e.g., psychrotrophic bacteria counts at 7°C
Non-manual methods

Virtually all bacteriological counts are now done by machines

A common one is the BactoScan

- Uses flow cytometry
- All milk components other than bacteria are broken down or made soluble
- Bacterial cells are labelled with fluorescent dye
- Individual cells are counted as they pass through a capillary tube
BactoScan 2

Analyses up to 200 samples/h

Measures total count

Counts may be higher than manual TPCs

• Counts individual cells while plate counts count colonies which may grow from clumps of bacteria
## Good, bad and ugly counts

**A good milk**

<table>
<thead>
<tr>
<th>Parameter</th>
<th>Count</th>
</tr>
</thead>
<tbody>
<tr>
<td>SPC</td>
<td>&lt; 10,000 cfu/mL</td>
</tr>
<tr>
<td>Thermoduric</td>
<td>&lt; 200,000 cfu/mL</td>
</tr>
<tr>
<td>Coliforms</td>
<td>&lt; 50 cfu/mL</td>
</tr>
<tr>
<td>SCC</td>
<td>&lt; 200,000/mL</td>
</tr>
<tr>
<td>Spores</td>
<td>&lt; 100 cfu/mL</td>
</tr>
</tbody>
</table>

Reinemannnn, D. J. (2011)


---

**Figure 1. Diagnostic Chart for Bulk Tank Bacteria Counts.**
Some microbiological standards for raw milk

<table>
<thead>
<tr>
<th>Country</th>
<th>TPC (cfu/mL)</th>
</tr>
</thead>
<tbody>
<tr>
<td>USA</td>
<td>100,000 (Individual farm),</td>
</tr>
<tr>
<td></td>
<td>300,000 (comingled)</td>
</tr>
<tr>
<td>UK</td>
<td>100,000</td>
</tr>
<tr>
<td>China</td>
<td>2,000,000</td>
</tr>
</tbody>
</table>
Why do we worry about high counts?

- Problems with high pathogenic bacterial counts, e.g. coliforms/\textit{E. coli} or \textit{Salmonella} are obvious
  - If present in processed milk (pasteurised, ESL, UHT) can cause illness - result in product recalls

- Problems with high TPC can be less obvious
  - If really high, the milk will be spoiled 😞
  - If high but not spoiled, it may contain enzymes formed by the bacteria
  - Enzymes are formed when TPC is > about 100,000 cfu/mL
  - Main problem enzymes are proteases and lipases
Problems with bacterial enzymes formed in raw milk

- Most have high heat stability - not destroyed by ESL or UHT heating
- The proteases cause bitterness in ESL and UHT milk, and cheese; also cause gelation during storage of UHT milk
- Lipases cause rancid off-flavours in milk and cheese, and also reduce the foaming capacity of milk
What about the quality of pasteurised or ESL milks?

- Somatic cells are no longer an issue
- Bacteria are still a problem
- Pathogenic bacteria can only be present if they enter the milk after heat treatment
  - Hence cleanliness of filling process is most important
- Post processing contaminants, e.g., *Pseudomonas*, are the main spoilage bacteria
- If spoilage bacteria are prevented from entering the milk after the heat treatment, pasteurised milk will keep for several weeks
# Chinese pasteurised milk microbiological standard

## Table 3 Limit of microorganism

<table>
<thead>
<tr>
<th>Items</th>
<th>Sampling $^a$ and limit (CFU/g or CFU/ml)</th>
<th>Analytical methods</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>$n$</td>
<td>$c$</td>
</tr>
<tr>
<td>TPC</td>
<td>5</td>
<td>2</td>
</tr>
<tr>
<td>Coliform</td>
<td>5</td>
<td>2</td>
</tr>
<tr>
<td>Staphylococcus aureus</td>
<td>5</td>
<td>0</td>
</tr>
<tr>
<td>Salmonella</td>
<td>5</td>
<td>0</td>
</tr>
</tbody>
</table>

$^a$ Analysis and treatment of samples apply to GB4789.1 and GB4789.18.
What about the bacteriological quality of UHT milks

- UHT milk should be “commercially sterile”, i.e., no bacteria will grow during ambient storage
- How do we tell if it is “commercially sterile”? Can be stored at 30°C for 14 days and then analysed for TPC
  - Count should be zero but < 100/mL is OK
- Rapid methods are now used
  - Most common are based on ATP detection
  - Only ATP from living bacteria are detected
  - Based on bioluminescence
    - ATP reacts with luciferin/luciferase (from fireflies) to produce bioluminescence
  - Several commercial instruments, e.g., from Celsis, Promicol, 3M, Charm.
Factors affecting quality of raw milk

On farm and in factory

- **Time and temperature**
- Maintenance of equipment
- Cleaning and sanitation practices

Other on-farm factors

- Weather/environmental conditions – dust, mud
- Animal health/husbandry
- Inclusion of abnormal milk – mastitic, colostrum
- Feed (silage, green feed, protein content)
- **stage of lactation/season**
Raw milk quality is important as it affects the quality of all processed milk and milk products made from it

(you can’t make a silk purse out of a sow’s ear)
Thank you for your attention