The science of cheese manufacture and ripening

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The dairy industry in Ireland

- Market capitalisation €7.7bn
- Kerry Group.
  - Export 80% of production
  - Exports worth €2.7bn (10th biggest dairy exporter)
  - 136,000 tonnes of butter
  - 180,000 tonnes of cheese (mainly Cheddar)

- Ingredients:
  - 5.4bn litres of milk
  - 1.1m dairy cows

- Irish owned multinationals:
  - 60% of production

- Exports worth €2.7bn

- 136,000 tonnes of butter

- 180,000 tonnes of cheese (mainly Cheddar)

- Ingredients:
  - 5.4bn litres of milk
  - 1.1m dairy cows

The Irish problem: seasonally-dominated milk production pattern

- Seasonal pattern reflects historical Irish farming practices
- Favours production of long-life dairy products
- Hampers diversification (key industry goal)
- Most cheese factories closed over the winter
Overview of dairy chemistry (of relevance to cheese)

Mammals
- Mamma (Latin, breast). Characteristic: secretion of milk
- Three subclasses:
  - Monotremes
  - Metatheria (Marsupials)
  - Eutheria
  - About 99% of all species
  - Placental mammal; young develop in utero
  - Nourished from maternal blood via placenta
  - Born at different stages of development
  - Mammary glands located externally, end in a teat
  - Number varies from two (probably most common, e.g., human, goat, sheep, mare, etc) to 14-16 (pig)
  - Each gland anatomically separate (produces milk of different composition)
  - Evolved from sebaceous glands which secreted an oily substance to coat hair

Udder issues...
MAMMARY GLAND

• Essentially an assembly line:
  • Absorbs water, glucose, amino acids, monoglycerides and fatty acids and inorganic salts from blood across basal membrane
  • In gland, converted and assembled into lactose, proteins and lipids
  • Exported through apical membrane into lumen

The basic components of milk
- Water: 87.3% Cow - Lactose (sugar): 4.8% Cow
- Fat: 3.7% Cow - Protein: 3.4% Cow

The basic function of milk
- To provide everything a newborn mammal needs until weaning...
- Unsurprising that different species' milk has very different composition which varies during lactation...

Variability of milk
- Milk is a highly variable biological fluid
- Why? Nutritional requirements of neonate vary with stage of development and between species
- Variation in milk composition caused by:
  - Species
  - Individuality of the animal (genetics)
  - Breed (genetics)
  - Age of the animal
  - Health of the animal (mastitis and other diseases)
  - Nutritional status
  - Interval between milkings
  - Stage of lactation, etc.
Lactose: milk sugar

- Disaccharide made from two simple sugars: galactose and glucose

- Two forms: α, β, with very different properties

Technological problems with lactose

- Low solubility
- Difficult to crystallize
- Large crystals – sandiness
- Mutarotation characteristics
- Hygroscopicity
- Low sweetness
- Adsorption of flavours and pigments
  [may be exploited to advantage in foods]
WHY DO MAMMALS SYNTHESISE LACTOSE?

Ready source of energy
Fat and lactose principal sources of energy in milk

Energy in cows’ milk
~ 50% fat
~ 30% lactose
~ 20% protein

Energy from sugars – readily assimilable

Applications of lactose

Humanized infant formulae
demineralized whey powder or lactose

Instantizing/free-flowing agent in foods
agglomeration due to lactose crystallization

Confectionery industry
improves functionality of shortening
anti-caking agent at high relative humidity
certain types of icing
Maillard browning, if desired
accentuates other flavours (chocolate)

Flavour adsorbent
flavour volatiles

Bulking agent in tablets

Lactose malabsorption (“intolerance”)…
- Dull climate in northern Europe…
- Milk source of clean “water” in hot countries?
### Table 3.1: Fat content of milk from various species (g l⁻¹)

<table>
<thead>
<tr>
<th>Species</th>
<th>Fat content</th>
<th>Species</th>
<th>Fat content</th>
</tr>
</thead>
<tbody>
<tr>
<td>Cow</td>
<td>13–47</td>
<td>Mammal</td>
<td>71</td>
</tr>
<tr>
<td>Buffalo</td>
<td>47</td>
<td>Rabbit</td>
<td>183</td>
</tr>
<tr>
<td>Sheep</td>
<td>40–99</td>
<td>Guinea-pig</td>
<td>39</td>
</tr>
<tr>
<td>Goat</td>
<td>41–45</td>
<td>Snowshoe hare</td>
<td>71</td>
</tr>
<tr>
<td>Musk-ox</td>
<td>109</td>
<td>Musk</td>
<td>134</td>
</tr>
<tr>
<td>Dall-sheep</td>
<td>12–206</td>
<td>Musk</td>
<td>134</td>
</tr>
<tr>
<td>Moose</td>
<td>18–105</td>
<td>Chinchilla</td>
<td>117</td>
</tr>
<tr>
<td>Antelope</td>
<td>93</td>
<td>Rat</td>
<td>103</td>
</tr>
<tr>
<td>Elephant</td>
<td>85–100</td>
<td>Red kangaroo</td>
<td>9–128</td>
</tr>
<tr>
<td>Human</td>
<td>38</td>
<td>Delphin</td>
<td>62–320</td>
</tr>
<tr>
<td>Horse</td>
<td>19–30</td>
<td>Manatee</td>
<td>35–225</td>
</tr>
<tr>
<td>Monkeys</td>
<td>10–51</td>
<td>Pygmy sperm whale</td>
<td>133</td>
</tr>
<tr>
<td>Lemurs</td>
<td>8–33</td>
<td>Harp seal</td>
<td>580–532</td>
</tr>
<tr>
<td>Pig</td>
<td>88</td>
<td>Bear (four species)</td>
<td>108–151</td>
</tr>
</tbody>
</table>


### Lipid class

<table>
<thead>
<tr>
<th>Lipid class</th>
<th>% of total lipid (w/w)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Triacylglycerols</td>
<td>97.5</td>
</tr>
<tr>
<td>Diacylglycerols</td>
<td>0.36</td>
</tr>
<tr>
<td>Monoacylglycerols</td>
<td>0.027</td>
</tr>
<tr>
<td>Cholesterol</td>
<td>0.31</td>
</tr>
<tr>
<td>Cholesteryl esters</td>
<td>Trace</td>
</tr>
<tr>
<td>Free fatty acids</td>
<td>0.027</td>
</tr>
<tr>
<td>Phospholipids</td>
<td>0.6</td>
</tr>
</tbody>
</table>

### Component fatty acids (% w/v) in the milk fat of some species.

<table>
<thead>
<tr>
<th>Acid</th>
<th>Cow</th>
<th>Goat</th>
<th>Sheep</th>
<th>Pig</th>
<th>Human</th>
<th>Lion</th>
<th>Horse</th>
</tr>
</thead>
<tbody>
<tr>
<td>C₈</td>
<td>2.8</td>
<td>3.0</td>
<td>4.0</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>0.4</td>
</tr>
<tr>
<td>C₁₀</td>
<td>2.3</td>
<td>2.5</td>
<td>2.1</td>
<td>-</td>
<td>0.3</td>
<td>-</td>
<td>0.9</td>
</tr>
<tr>
<td>C₁₂</td>
<td>1.1</td>
<td>2.8</td>
<td>2.0</td>
<td>1.3</td>
<td>-</td>
<td>-</td>
<td>2.6</td>
</tr>
<tr>
<td>C₁₄</td>
<td>3.0</td>
<td>10.0</td>
<td>6.0</td>
<td>-</td>
<td>1.6</td>
<td>0.7</td>
<td>2.5</td>
</tr>
<tr>
<td>C₁₆</td>
<td>2.9</td>
<td>6.0</td>
<td>2.8</td>
<td>-</td>
<td>6.9</td>
<td>0</td>
<td>5.6</td>
</tr>
<tr>
<td>C₁₈</td>
<td>9.0</td>
<td>12.3</td>
<td>5.3</td>
<td>1.5</td>
<td>8.5</td>
<td>4.3</td>
<td>7.0</td>
</tr>
<tr>
<td>C₂₀</td>
<td>24.0</td>
<td>27.9</td>
<td>17.5</td>
<td>26.9</td>
<td>20.9</td>
<td>26.4</td>
<td>16.1</td>
</tr>
<tr>
<td>C₂₂</td>
<td>13.2</td>
<td>6.0</td>
<td>15.6</td>
<td>6.5</td>
<td>7.3</td>
<td>2.1</td>
<td>2.9</td>
</tr>
<tr>
<td>Odd Carbon</td>
<td>3.7</td>
<td>0.9</td>
<td>1.5</td>
<td>0.8</td>
<td>2.7</td>
<td>?</td>
<td>tr</td>
</tr>
<tr>
<td>Branched</td>
<td>1.5</td>
<td>-</td>
<td>-</td>
<td>1.2</td>
<td>-</td>
<td>?</td>
<td>-</td>
</tr>
<tr>
<td>Monounsaturated</td>
<td>33.3</td>
<td>25.1</td>
<td>38.6</td>
<td>45.0</td>
<td>39.5</td>
<td>42.7</td>
<td>29.9</td>
</tr>
<tr>
<td>Polyunsaturated</td>
<td>3.8</td>
<td>3.8</td>
<td>5.6</td>
<td>18.8</td>
<td>10.1</td>
<td>23.8</td>
<td>28.8</td>
</tr>
<tr>
<td>Trans</td>
<td>5.0</td>
<td>-</td>
<td>12.1</td>
<td>-</td>
<td>1.4</td>
<td>?</td>
<td>-</td>
</tr>
<tr>
<td>Conjugated</td>
<td>1.1</td>
<td>2.7</td>
<td>-</td>
<td>-</td>
<td>?</td>
<td>-</td>
<td>-</td>
</tr>
</tbody>
</table>
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Notable Features of Fatty Acid Profile of Milk Fat

- High level of C<sub>4:0</sub>
  - only natural lipid containing this FA
- Moderate levels of C<sub>6:0</sub> to C<sub>10:0</sub>
  - rare in lipids
- Short- and medium-chain FAs highly flavoured
- Low level of polyunsaturated fatty acids (nutritional)
  - High level of CLA (very desirable)
- Low level of trans PUFAs
  - these are undesirable

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Fatty Acid Distribution in Milk Lipids

- C<sub>4:0</sub> to C<sub>10:0</sub> totally or mainly at sn-3 position
- C<sub>12:0</sub> to C<sub>16:0</sub> mainly at sn-2 position
- C<sub>18:0</sub> at sn-1 and sn-2
- C<sub>18:1</sub> at sn-1 and sn-3

![Diagram of fatty acid distribution]

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**Fig. 50. Diagrammatic Representation of the Events which Accompany the Secretion of Fat Globules from the Secretory Cells of a Lactating Bovine.**

Fat droplets and protein particles are designated by F and P, respectively.
Milk proteins: caseins and whey proteins

Recognised about 120 years ago that there are two main families of proteins in milk

Skim milk
Add acid

pH 4.6

Soluble
Whey (or serum)
Proteins (~20%)
α-Lactalbumin
β-Lactoglobulin
Bovine serum albumin
Immunoglobulins
Enzymes
And many more...

Insoluble
CASEINS (~80%)
Latin: caseus = cheese
αs1-, αs2-, β-, κ-casein

Industrial products
(Cheese)
Acid casein
Sodium caseinate

Ratio of caseins to whey proteins vary

Cow, goat, sheep, buffalo
~80:20 casein:whey protein

Equine (mare)
~50:50 casein:whey protein

Human
~50:50 casein:whey protein

Controlled destabilisation of caseins

- Fast acidification: isoelectric precipitation of caseins to make acid casein
- Slow quiescent bacterial acidification: yogurt, acid-coagulated cheeses
- Rennet coagulation: rennet casein, most cheeses
- However, must avoid in manufacture of cream liqueurs
Caseins and whey proteins

• Whey proteins do not precipitate from milk at pH 4.6; casein precipitate. **Operational definition of casein.**
• Caseins are acted upon by chymosin (an enzyme in preparations called rennet used in manufacture of most cheeses), causing a slight change resulting in coagulation of caseins in presence of Ca$^{2+}$. Whey proteins are not coagulated by chymosin.
• Caseins very stable to heating; whey proteins are quite heat labile (denature easily).

Caseins are phosphoproteins (contain PO$_4$ attached to Ser residues). Negatively charged; can bind Ca$^{2+}$.

• Phosphate bound to casein is called organic phosphate (also phosphate associated with caseins in form of salts: colloidal calcium phosphate; see later)

Caseins low in sulphur-containing amino acids (Met, Cys). Caseins contain ~0.8% S; whey proteins relatively rich (~1.7%). Very little Cys in caseins.

• Whey proteins contain relatively more Cys. Responsible for some changes that occur to whey proteins on heating...

• When milk is heated, β-lactoglobulin (a whey protein) is denatured and its SH group is exposed. Can interact with κ-casein via a –S-S– bond. As κ-casein is involved in rennet coagulation of milk, it is very difficult to make cheese from high heat treated milk!
Caseins

- Typical concentrations of $\alpha_s^1$-CN, $\alpha_s^2$-CN, $\beta$-CN and $\kappa$-CN in bovine milk are 12-15, 3-4, 9-11 and 2-4 g L$^{-1}$
- $\alpha_s^1$-CN, $\alpha_s^2$-CN, $\beta$-CN are the calcium-sensitive caseins; $\kappa$-CN is calcium-insensitive
- Caseins account for ~75-80% of protein in bovine milk (ratio of casein:whey protein known as the “casein number”; important technological parameter)

<table>
<thead>
<tr>
<th>Casein</th>
<th>Approx Amount</th>
<th>Approx. Ratio</th>
</tr>
</thead>
<tbody>
<tr>
<td>$\alpha_s^1$-CN</td>
<td>37%</td>
<td>4</td>
</tr>
<tr>
<td>$\alpha_s^2$-CN</td>
<td>10%</td>
<td>1</td>
</tr>
<tr>
<td>$\beta$-CN</td>
<td>35%</td>
<td>4</td>
</tr>
<tr>
<td>$\kappa$-CN</td>
<td>12%</td>
<td>1</td>
</tr>
</tbody>
</table>

Properties of the caseins

- Little 2º, 3º structure. Caseins thus cannot be denatured in normal sense. High level of Pro one reason for this disordered structure
- Open, flexible structure. Very susceptible to proteolysis (one of their natural functions is to be digested!); also important in cheese during ripening.
- Very heat stable.
- Very strong tendency to self-associate in solution. If you dissolve sodium caseinate in water, most molecules present as aggregates of 250-500 kDa (10-20 molecules). Association mainly due to hydrophobic bonding.

Casein association

- All caseins associate with themselves and other caseins
- For example:
  - at 4°C, $\beta$-casein exists as monomers (MW 25 kDa); as temperature increases, forms thread-like chains of about 20 units at 8.5°C; larger aggregates at higher temperatures, depending on [protein]
  - $\alpha_s$-Casein forms tetramers (MW 113 kDa); degree of polymerisation increases with temperature and [protein]
  - $\kappa$-Casein present largely as ~S-S- linked dimers (unless reduced)
- Casein interaction in the presence of Ca$^{2+}$ leads to the formation of casein micelles
The casein micelle…
Or, why milk is white!

Colloids
- A colloid (colloidal dispersion, colloidal solution, sol)
  - A very finely-divided solid
  - Large molecules (e.g., proteins, polysaccharides, nucleic acids, synthetic polymers)
  - Aggregates of small molecules (association/accidental colloids, micelles)

Typical characteristics:
- Non-diffusable through semi-permeable membrane (non-dialysable)
- Can scatter light (translucent, cloudy, white)
- Include:
  - Emulsions (liquid/liquid)
  - Foams and aerosols (gas/liquid)
  - Solids (solid/liquid)

Casein micelle
- ~95% of casein in milk exists as large colloidal aggregates known as casein micelles
- Micelles are ~84% protein, ~6% low molecular weight salts (minerals) known as colloidal calcium phosphate
- Highly hydrated; micelles contain ~4 g H₂O g⁻¹; only ~15% of water bound to protein, rest simply occluded within spongy micelle structure
- Generally spherical; diameters 50-500 nm (average ~120 nm)
- Average mass 10¹⁸ Da (many small particles but represent only a small proportion of volume or mass)
- Very closely packed in milk (average of only two micelle diameters apart); very large surface area. About 10⁻¹⁴⁻¹⁰¹⁶ micelles per ml milk
- Scatter light! That’s why milk is white
### Average Characteristics of Casein Micelles

<table>
<thead>
<tr>
<th>Characteristic</th>
<th>Value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Composition (dry weight)</td>
<td>94% protein, 6% salts (colloidal calcium phosphate)</td>
</tr>
<tr>
<td>Shape</td>
<td>spherical</td>
</tr>
<tr>
<td>Diameter</td>
<td>130-160 nm (range 50-600)</td>
</tr>
<tr>
<td>Molecular mass (hydrated)</td>
<td>1.3 x 10^9 Daltons</td>
</tr>
<tr>
<td>Molecular mass (dehydrated)</td>
<td>5 x 10^8 Daltons</td>
</tr>
<tr>
<td>Number of peptide chains (MW: 23,000)</td>
<td>~ 2 x 10^4</td>
</tr>
<tr>
<td>Number of micelles/ml milk</td>
<td>10^14 - 10^16</td>
</tr>
<tr>
<td>Mean free distance</td>
<td>240 nm</td>
</tr>
</tbody>
</table>

### Controlled destabilisation of casein micelles

- Fast acidification: isoelectric precipitation of caseins to make acid casein
- Slow quiescent bacterial acidification: yogurt, acid-coagulated cheeses
- Rennet coagulation: rennet casein, most cheeses
- However, must avoid in manufacture of cream liqueurs

### Casein micelle stability

- Stable to many processing operations (except obviously those designed to destabilise micelles!)
- Stable to high temperatures (requires heating at 140°C for 15-20 min at normal pH of milk to coagulate; even then coagulation not due to denaturation but to other changes to milk, particularly ↓ pH)
- Stable to compaction unless ultracentrifuged
- Stable to normal commercial homogenisation
- Stable at high [Ca^{2+}] (at least 200 mM up to 50°C)
Casein micelle stability

- Aggregate and precipitate at isoelectric point of the caseins (pH 4.6)
- Retain some structure even if much colloidal calcium phosphate removed (up to 70%). More CCP removal results in disintegration of the micelle
- Destabilised by action of enzymes in rennet (see later)
- Unstable in ~40% ethanol at pH 6.7 (and lower [ethanol] as ↓pH)
- Destabilised by freezing (cryodestabilisation) due to ↓pH and ↑[Ca²⁺] in the unfrozen part as milk freezes

Structure of the casein micelle

- Various models of the casein micelle have been proposed. Still much debate!
- All models must account for the following facts about the casein micelle...

Structure of the casein micelle

1. κ-Casein must be able to stabilise the other caseins
2. κ-CN content of micelles is inversely proportional to size (i.e., small micelles contain more κ-casein)
3. κ-Casein must be easily hydrolysed by chymosin during rennet coagulation (large molecules)
4. When milk heated, β-lactoglobulin interacts with κ-casein via –S–S– bonds
5. Micelles have a porous structure; protein only occupies ~25% of total volume
6. Removal of CCP results in disintegration of micelle into particles of mass ~3 x 10⁶ Da
Structure of the casein micelle

7. Micelles can be dispersed (dissociated) by urea and SDS, suggesting involvement of hydrogen bonds and hydrophobic interactions
8. Micelles can be destabilised by alcohols, acetone suggesting important role for electrostatic interactions
9. At low temperatures, β-casein dissociates from the micelle (10-50% of β-casein non-micellar at 4°C)
10. Electron microscopy suggests interior of micelle is not uniformly electron dense
11. Micelles have surface (zeta) potential of -20 mV at pH 6.7

Submicellar model

Micelles are composed of submicelles (of mass of ca. 10^6 Da and diameter of 10-15 nm), linked together by CCP, giving the micelle an open, porous structure. κ-CN rich submicelles are found more commonly at the surface of the micelle. This gives the micelle a κ-CN rich surface layer with protruding “hairs” formed from the C-terminal region of the protein (but with some of the other caseins at the surface also). κ-CN deficient submicelles are at the centre of the micelle.

Explains many properties of micelles
Problem! Submicelles with different levels of κ-casein have never been found!

Other models

• Submicellar model explains many important properties of the caseins
• Never enjoyed unanimous support! Mainly because model suggests there should be two types of submicelles; those rich in κ-casein and those poor in κ-casein. These have not been found!
Another model……

Holt model (1992). The micelles is a tangled web of flexible casein molecules forming a gel-like structure in which nanocrystals of CCP are an integral feature.

The C-terminal regions of κ-CN molecules protrude from the surface, forming a "hairy" layer.

Dalgleish model

Major feature is the porosity of the model

Charge on a protein varies with pH

Isoionic point (pI) = pH at which protein has no net charge (it remains charged, but the number of positive charges equals the number of negative charges)

Isoelectric point = pH at which a protein does not move in an electric field, because they have no net charge
Casein Micelles: Stability

Due to:
- Zeta (surface) potential, -20 mV
- Steric stabilization

If we can overcome this effect, we can get casein micelles to form a gel:
- solid phase
- as a result, milk coagulates (liquid → solid)

Two ways to do this:
1. Cut off the protective ‘hairs’ – use a proteolytic enzyme (rennet)
2. Neutralise the charge – bring pH down to 4.6 (isoelectric point of casein)

Either results in………..

But gels (rennet v. acid) have different properties!

Acid gelation of milk

Courtesy of Dr Mark Auty, Teagasc, Moorepark
Milk proteins: caseins and whey proteins

Recognised about 120 years ago that there are two main families of proteins in milk.

Skim milk

Add acid

pH 4.6

Soluble
Whey (or serum)
Proteins (~20%)

Insoluble
CASEINS (~80%)
Latin: caseus = cheese

Acid whey also contains casein-derived peptides (proteose peptones) and rennet whey also contains GMP from casein.

Preparation of whey proteins

1. Protein fraction soluble at pH 4.6
2. Protein fraction soluble after rennet coagulation of the caseins
3. Soluble in saturated NaCl
4. In supernatant after ultracentrifugation of milk
5. By gel permeation chromatography

Wheys prepared by all (except 5) will contain lactose and salts. Slight differences between wheys obtained by these methods.

Whey processing

- In the past seen as troublesome...
  - Disposal down effluent treatment plant
  - Spread on land
  - Feed to pigs
- Now a very valuable starting point for production of food ingredients:
  "Cheese is the by-product of whey manufacture!"
Whey processing

- In the past seen as troublesome...
  - Disposal down effluent treatment plant
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  - Feed to pigs

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  "Cheese is the by-product of whey manufacture!"

- Whey powder
  - Demineralised whey powder

- Whey protein concentrate
Whey processing

• In the past seen as troublesome...
  Disposal down effluent treatment plant
  Spread on land
  Feed to pigs
• Now a very valuable starting point for production of food ingredients
  “Cheese is the by-product of whey manufacture!”
• Whey powder
  Demineralised whey powder
  Whey protein concentrate
  Whey protein isolate

Lactose

Fermented to ethanol
Whey processing
- In the past seen as troublesome...
  Disposal down effluent treatment plant
  Spread on land
  Feed to pigs
- Now a very valuable starting point for production of food ingredients
  “Cheese is the by-product of whey manufacture!”
- Whey powder
- Demineralised whey powder
- Whey protein concentrate
- Whey protein isolate
- Lactose
- Fermented to ethanol
  Isolation of individual whey proteins

Milk salts
- The salts system of milk can be considered as a two-phase system consisting of casein-bound colloidal calcium phosphate in quasi-equilibrium with an aqueous solution of soluble salts

Milk salts
- The salts of milk are mainly the phosphates, citrates, chlorides, sulphates, carbonates and bicarbonates of sodium, potassium, calcium and magnesium
- Approximately 20 other elements are found in milk in trace amounts including Cu, Fe, Si, Zn and I
- There is no lactate in freshly-drawn milk but it may be produced in milk by bacterial fermentation of lactose
- The ash content of milk remains relatively constant at 0.7-0.8% but the relative concentrations of various ions can vary considerably. It is important to remember that “ash” (produced following heating a sample at 500-500°C in a muffle furnace) is not the same as the salts of milk
Indigenous enzymes

- First enzyme in milk (lactoperoxidase) identified in early 1880s
- Perhaps 70 indigenous enzymes in milk
  ~20 well characterised
- Most have no obvious physiological role in milk
- Arising from four principal sources:
  Blood (via defective mammary cell membranes)
  Cytoplasm of mammary cell
  Milk fat globule membrane (and thus Golgi membranes)
  Somatic cells

Technological significance:
- Deterioration
  Lipoprotein lipase, plasmin, somatic cell proteinases
- Indices of thermal history of milk
  Alkaline phosphatase, Lactoperoxidase, Catalase
- Indices of mastitic infection
  N'-Acetylglucosaminidase, Catalase, Acid phosphatase
- Antimicrobial activity
  Lactoperoxidase, Xanthine oxidoreductase, Lysozyme
- Digestion
  Bile-salts stimulated lipase (human milk)
Cheese manufacture

Why make cheese…?

- Originally a form of food preservation
- Preservation action due to...
  1. **DEHYDRATION.** Cheese is a medium moisture food (30-50%), stability inversely related to moisture. $a_w$ usually > 0.95
  2. **ACID.** Medium acid food (~pH 5 for many varieties)
  3. **ANAEROBIC CONDITIONS**
  4. **NaCl.** Depending on variety, 0.7-10%. What is important is salt-in-moisture (2-15%)

Why make cheese…?

- Originally, the greater the stability, the better.
- Stability not now primary objective
- Consistency, high quality, good flavour, texture important
- **BALANCED RIPENING**
How many cheeses in the world…?

- About 800 varieties/variants; 400 in France?
- Sandine & Elliker (1970) suggest >1000
- Walter & Hargrove (1972) described 400, listed names of another 400
  But concluded probably only 18 distinct types of natural cheese
- Burkhalter (1981) classified 510 varieties
- > 500 listed by International Dairy Federation
- Jim Path, 1400 varieties ([www.cdr.wisc.edu](http://www.cdr.wisc.edu))

- Numerous minor, locally
  cheese.

The truth is that we probably don’t know how many cheeses are in the world!

Classification of cheese

- Traditional classification based on moisture (“hard”, “semi-hard”, “soft”…)
- We can agree that most varieties in one of two categories…

1. RENNET COAGULATED (most varieties)
2. ACID COAGULATED (e.g., Cottage cheese, Quarg, Cream cheese…)

Cheese Classification
- Few large groups?
- Many very similar "varieties"?

Paleontology
- Few large groups?
- Many very similar species?

"Lumpers" Few, heterogeneous taxa
"Splitters" Many, overlapping taxa
On what criteria do we classify cheese…?

- No completely right or wrong answer to this question; depends on objectives, background, personal choice...
- Some factors of more importance to some people...

Scientific/technological criteria

- Cheese texture/moisture
- Method of coagulation
- Ripening/technological parameters
- Microflora
- Include products other than natural cheese
- Type of milk

Marketing/business criteria

- Geographical region
- Tradition
- PDO status
- Brand identity

Classification based on ripening indices

- Davis (1965) suggested this should be possible "within a few years"; still not possible to do so reliably 44 years later!
- Problems:
  - Cheese ripening is dynamic: cheeses can differ greatly with age
  - Considerable variation within each variety
- However, some interesting developments...

MALDI-ToF MS
"Lumpers"
Few, heterogeneous groups

"Splitters"
Many, similar "varieties"

Fox et al. (2000, 2003)

- Initial classification (Fox, 1993) based on method of coagulation
- Three "superfamilies"
  Rennet, acid, heat/acid coagulated
- Rennet coagulated family subdivided into relatively homogeneous groups based on characteristic ripening agent(s) or technology
- Other cheeses/derived products listed separately

Acid-Curd Cheeses
- High moisture (70-80%)
- Consumed fresh
- Short shelf-life
- Important food ingredients
University College Cork

**CHEESE**

- HEAT/ACID COAGULATED (e.g., Ricotta)

- Concentration/crystallization
  - Minor group of Norwegian "cheeses" made from whey

**CHEESE ANALOGUES**

- NATURAL CHEESE
- ENZYME-MODIFIED CHEESE
- DRIED CHEESES
- HEAT/ACID COAGULATED (e.g., Ricotta)

**PROCESSED CHEESE**

- Most varieties of cheese may be processed

- **Cottage, Cream, Quarg**
- **Ricotta**

**Internal Bacterially-ripened Mould-ripened Surface-ripened**

- Pasta-filata Varieties
- High-salt Varieties

- **Surface Mould** (usually *P. camemberti*)
- **Internal Mould** (*P. roqueforti*)

- **Brie**
- **Camembert**
- **Roquefort**
- **Danablu**
- **Stilton**
- **Havarti**
- **Limburger**
- **Munster**
- **Port du Salut**
- **Trappist**
- **Taleggio**
- **Tilsit**
- **Domiati**
- **Feta**
- **Mozzarella**
- **Kashkaval**
- **Provolone**

- **Swiss-type** (Lactate metabolism by *Propionibacterium* spp.)
- **Dutch-type** (Eyes caused by citrate metabolism)

- **Edam**
- **Gouda**
- **Emmental**
- **Gruyère**

- **Extra-hard**
  - **Grana Padano**
  - **Parmesan**
  - **Asiago**
  - **Sbrinz**

- **Hard**
  - **Cheddar**
  - **Cheshire**
  - **Graviera**
  - **Ras**

- **Semi-hard**
  - **Caerphilly**
  - **Mahon**
  - **Monterey Jack**

- **M**
  - **a**
  - **m**
  - **s**
  - **d**
  - **a**
  - **m**
  - **s**

**CONCENTRATION/CRYSTALLIZATION**

- **Mysost**

**Cheeses with eyes**

- **Swiss-type**
- **Dutch-type**

**PROCESSED CHEESE**

- Most varieties of cheese may be processed

**DRIED CHEESES**

- Concentration/crystallization
  - Minor group of Norwegian "cheeses" made from whey
Processed cheese produced by comminuting, melting and emulsifying into a smooth homogeneous blend one or more natural cheeses and optional ingredients (e.g., stabilizers, milk protein powders, flavours, colours, preservatives) using heat and mechanical shear and usually emulsifying salts.

Processed cheese
• Long maturation (usu. > 2 yr), hard, grainy texture
  • May be consumed young (when softer) or mature (often grated)
  • Semi-skim milk, high cooking temperature, loss of moisture during ripening (all leading to hard texture)

• Very heterogeneous group
  • Cheddar and ‘British Territorial’ varieties (Cheshire, Derby, Gloucester, Leicester) and others...
Cheeses with “eyes”

- Mechanical openings caused by incomplete fusion of curd pieces (defect in many varieties)
- “Eyes” result from gas-producing microorganisms. Shiny interior, more regular size, shape
- Swiss-type cheeses: CO₂ produced from lactate by Propionibacterium freudenreichii
- Dutch-type cheeses: gas produced by citrate fermentation by Cit.+ lactococci or Leuconostoc sp.
Hybrid “Blue Brie”

- White mould (P. camemberti) on surface, blue veins (P. roqueforti) in centre
- Add spores of P. roqueforti to milk, heat treat cheese to kill spores at surface, spray P. camemberti on surface, pierce cheese to allow oxygen in. P. roqueforti grows in piercings.
Smear-ripened cheeses (soft)

- Smear formation
  - pH 5.3 after manufacture
  - Growth of oenococcus (e.g., O. lactis) and organisms involved in pH reductions to 4.6

Surface ~pH 5.3 after manufacture

- Yeast growth (e.g., D. hansenii and Geotrichum candidum)
- pH increases to >6.0

- Gram-positive bacterial flora
  - Very complex ecosystem
  - Micrococceae
  - Brevibacterium sp.
  - Arthrobacter sp.
  - Corynebacterium sp.

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