

31 october 2013, Vicosa, Brazil

AGRO CAMPUS OUEST

Importância da fração mineral na ciência e tecnologia de produtos lácteos
Importance of the mineral fraction in the science and technology of dairy products

Frédéric Gaucheron

INRA, Agrocampus Ouest, UMR Science and Technology of Milk and Egg,
 65 rue de Saint Brieu, 35042, Rennes
 Email: frederic.gaucheron@rennes.inra.fr

FOOD AGRICULTURE ENVIRONMENT INRA

Generalities of Minerals

- Not synthesized by the human organism
- Present in low quantity
- Are not energetic sources
- Contribute to the good functions of the organism

Minerals

Macroelements :
 Ca, Mg, Na, K, P and Cl

Trace elements :
 Fe, Zn, Cu, Se.....

University of Vicosa, Brazil, 31 october to 2 November 2013 2

Main Macroelements in Milk

- ↪ Cations : Ca, Mg, Na & K
- ↪ Anions : P, Cl and citrate

University of Vicosa, Brazil, 31 october to 2 November 2013 3

Calcium and phosphate in milk and dairy products

FOOD AGRICULTURE ENVIRONMENT INRA

Calcium and Phosphorus in Milk

Trace elements

Chloride (~1200 mg/L)

Potassium (~1400 mg/L)

Sodium (~500 mg/L)

Magnesium (~120 mg/L)

Citrate (~1600 mg/L)

Phosphorus (~950 mg/L)

Calcium (~1200 mg/L)

University of Vicosa, Brazil, 31 october to 2 November 2013 5

The Salt Equilibria in Milk / different forms of calcium

Pi : Inorganic Phosphate

Aqueous Phase

CaCit⁻ [8 mM]

Cit³⁻ [1.6 mM]

HCl²⁻ [0.2 mM]

H⁺ [2 · 10⁻⁷ mM]

Ca²⁺ [2 mM]

↔

Micellar Phase

CaHPO₄ [0.6 mM]

HPO₄²⁻ [3 mM]

H₂PO₄⁻ [10 mM]

93 % caseins

- α₁-CN 36 %
- α₂-CN 10 %
- β-CN 34 %
- κ-CN 12 %

7 % of colloidal mineral complex phosphate, calcium, magnesium and citrate

Ca(HPO₄)_{0.7}(PO₄)_{0.2}·xH₂O

Nature, structure, role and intervention in the functionality ???

• Saturated in calcium phosphate (0.6 mM)

• All modifications of the physico-chemical characteristics of the soluble phase (pH, salt addition, pH_i...) → modifications of the salt equilibria between soluble and micellar phases

• Ionic strength : 0.08 M

University of Vicosa, Brazil, 31 october to 2 November 2013 6

Different Forms of Calcium in Dairy Products

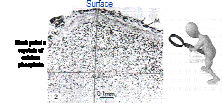
- The forms, locations and concentration of calcium are multiple and depend on the technologies used:
 - Free (ionic calcium \rightarrow Ca^{2+}) (milk, fermented milks, cheeses)
 - Associated to
 - casein molecules (milk, cheeses)
 - counter-ions (inorganic phosphate, citrate) to form soluble salts
 - α -lactalbumin, osteopontin,
 - free fatty acids (after lipolysis) to form soap
 - phosphopeptides (after proteolysis of casein)
 - Precipitates under
 - calcium phosphate (surface of Camembert)
 - calcium lactate (Cheddar)
 - calcium carbonate (Emmental)
- Important to consider the different forms of calcium which could be differently absorbed (effect of environment – lactose, phosphopeptide, others cations, presence of chelatants – Ca/P ratio, type of matrix, ...)



Different Forms of Phosphorus in Dairy Products

Its form, location and concentration depend on the considered product :

- Free but with different ionisation state depending on the pH value
 $(H_3PO_4 \leftrightarrow H_2PO_4^- \leftrightarrow HPO_4^{2-} \leftrightarrow PO_4^{3-})$ (milk, fermented milks, cheeses)
- Associated to casein micelles (milk, cheeses)
- Associated to calcium to form soluble salts and precipitates (surface of Mould cheeses)



Crystals in the surface of soft cheese (Coulommiers) observed by light microscopy / von Kossa coloration

- Associated with phosphopeptides (after proteolysis of casein)
- Associated to phospholipids, nucleic acids, sugar,
- Polyphosphates added for processed cheese



The Importance of Calcium Phosphates

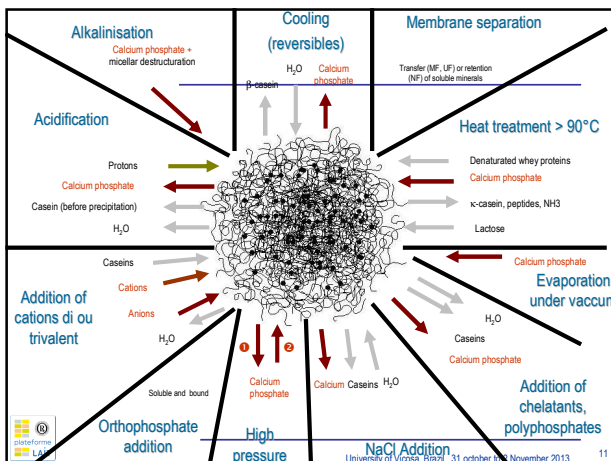
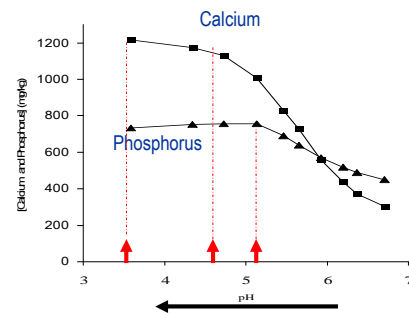
- Are partially responsible for physico-chemical properties of milk (structure of casein micelles, ...)
- Are modified by physico-chemical conditions (technologies) (pH, temperature, ionic environment, ...)
- Influence the technological properties of dairy products and processes of concentration and thermal treatment (heat stability, rennet and acid coagulations, rheological and textural characteristics, ...)



Very descriptive and several questions on these dairy calcium phosphates should be resolved



Calcium and Phosphorus Solubilizations during Milk Acidification



Concentrations of Calcium in Some Dairy Products

(mg / 100 g)

	Ca	Mg	Na	K	P	Zn	Fe	Cu	Se
LIQUID MILK	120								
Skim milk powder	1254	113	433	1448	112	3.86	0.435	0.1	0.13
UHT skimmed milk	113	20.6	11.9	173	88	0.41	0.05	0.007	0.04
Concentrated whole milk	264	25	11.9	297	213	0.19	0.15	0.05	1.6
Salted butter	24	2	570	24	24	0.16	0.02		1
Unsalted butter	24	2	11	24	24	0.02	0.02	0.003	1
Yoghurt with skimmed milk	127								2.2
Feta	489								16
Cheddar	729								11.9
Gruyere	763								11.6
Camembert	385								11.6
Provolone	750								11.6
Switzerland Emmental	791								11.6
Cottage cheese 1%	471								4
Parmesan	1194								10.9
Mozzarella	575	27	415	75	415	2.46	0.2	0.002	0.97
Hard cooked-cheeses	1095	20.9	482	105	780	2.22	0.21	0.02	0.2
Brie of Aownage	162	18.2	113.3	34.8	367	2.18	0.11	0.02	0.74
Fresh cheese 0%	116	11.7	42.1	124	122	0.49	0.02	0.02	1.9
Processed Cheese (25 % fat)	346	24	252	152	356	1.6	0.2	0.02	1.4
Fresh cheese (type Petit suisse 20 %)	117	101	124	94	122	0.04	0.02	0.04	1
Beaufort	695	30.5	639	117	777	0.14	0.04	0.02	1.23
Morbier	780	24	590	150	530	1.7	0.2	0.02	1
Comté	608	20.9	415	139	663	0.11	0.04	0.01	1
Roquefort	642	18	780	131	585	2.06	0.03	0.002	0.44



Concentrations of Phosphorus in Some Dairy Products (mg/100 g)

LIQUID MILK	Ca	Mg	Na	K	P	Zn	Fa	Cu	Se
Skin milk powder	1250	120	100	1000	95	0.00	0.000	0.0	0.0
UHT skimmed milk	117	10	1.0	12	0.8	0.01	0.00	0.00	0.0
Concentrated whole milk	364	25	1.1	10	2.1	0.01	0.01	0.00	0.0
Salted butter	24	2	0.7	24	1	0.00	0.00	0.00	0
Unsalted butter	24	2	0.7	24	1	0.00	0.00	0.00	0
Yoghurt with skimmed milk	127	11	1	11	9	0.00	0.00	0.00	0
Feta	493	10	11.6	62	37	0.00	0.00	0.00	0
Cheddar	721	20	20	80	52	0.00	0.00	0.00	0
Gruyere	1011	20	20	80	52	0.00	0.00	0.00	0
Camembert	388	20	14	10	27	0.00	0.00	0.00	0
Provola	750	20	10	10	46	0.00	0.00	0.00	0
Switzerland Emmental	791	20	10	10	57	0.00	0.00	0.00	0
Cottage cheese 1%	61	1	1	1	134	0.00	0.00	0.00	0
Parmesan	1184	20	10	10	134	0.00	0.00	0.00	0
Mozzarella	373	21	1.0	10	41	0.00	0.00	0.00	0
Hard cooked cheeses	1050	43.8	100	100	600	0.00	0.00	0.00	0
Bleu d'Auvergne	583	18.1	11.0	10	30	0.00	0.00	0.00	0
Fresh cheese 0%	118	11.7	1.0	10	10	0.00	0.00	0.00	0
Processed Cheese (25 % fat)	346	34	10	10	10	0.00	0.00	0.00	0
Fresh cheese (type Petit suisse 20 %)	117	10	10	10	10	0.00	0.00	0.00	0
Beaufort	995	30.5	10	11	72	0.00	0.00	0.00	0
Morbier	760	20	10	10	50	0.00	0.00	0.00	0
Comté	969	18.9	1.0	10	10	0.00	0.00	0.00	0
Roquefort	643	10	10	10	30	0.00	0.00	0.00	0

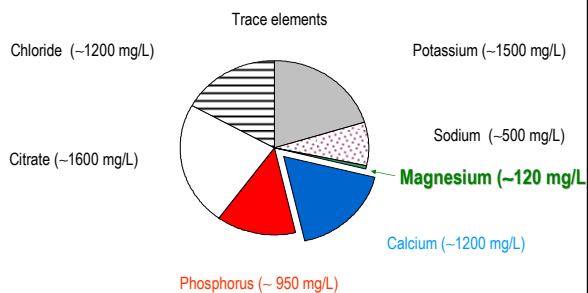
Milk and dairy products
☺
very good sources of phosphorus



Other macroelements : Mg, Na, K and Cl

FOOD
AGRICULTURE
ENVIRONMENT

Magnesium in Milk



Different Forms of Magnesium in Milk and Dairy Products

- In milk, magnesium is distributed between micellar (50 mg/L) and aqueous phase (70 mg/L)
- As described for calcium and phosphate, the chemical form, location and concentration of magnesium depend on the considered product :
 - Free
 - Associated to casein micelles
 - Associated to inorganic phosphate and citrate



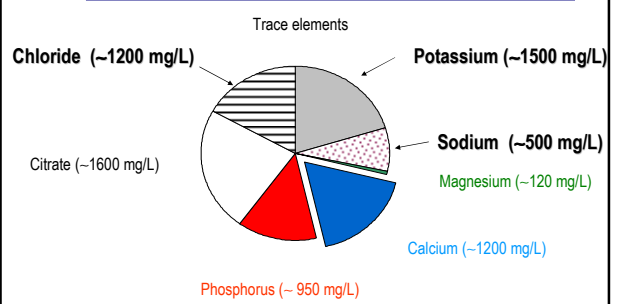
Concentrations of Magnesium in Some Dairy Products (mg/100 g)

LIQUID MILK	Ca	Mg	Na	K	P	Zn	Fa	Cu	Se
Skin milk powder	1250	120	100	1000	95	0.00	0.000	0.0	0.0
UHT skimmed milk	117	10	1.0	12	0.8	0.01	0.00	0.00	0.0
Concentrated whole milk	364	25	1.1	10	2.1	0.01	0.01	0.00	0.0
Salted butter	24	2	0.7	24	1	0.00	0.00	0.00	0
Unsalted butter	24	2	0.7	24	1	0.00	0.00	0.00	0
Yoghurt with skimmed milk	127	11	1	11	9	0.00	0.00	0.00	0
Feta	493	10	11.6	62	37	0.00	0.00	0.00	0
Cheddar	721	20	20	80	52	0.00	0.00	0.00	0
Gruyere	1011	20	20	80	52	0.00	0.00	0.00	0
Camembert	388	20	14	10	27	0.00	0.00	0.00	0
Provola	750	20	10	10	46	0.00	0.00	0.00	0
Switzerland Emmental	791	20	10	10	57	0.00	0.00	0.00	0
Cottage cheese 1%	61	1	1	1	134	0.00	0.00	0.00	0
Parmesan	1184	20	10	10	134	0.00	0.00	0.00	0
Mozzarella	373	21	1.0	10	41	0.00	0.00	0.00	0
Hard cooked cheeses	1050	43.8	100	100	600	0.00	0.00	0.00	0
Bleu d'Auvergne	583	18.1	11.0	10	30	0.00	0.00	0.00	0
Fresh cheese 0%	118	11.7	1.0	10	10	0.00	0.00	0.00	0
Processed Cheese (25 % fat)	346	34	10	10	10	0.00	0.00	0.00	0
Fresh cheese (type Petit suisse 20 %)	117	10	10	10	10	0.00	0.00	0.00	0
Beaufort	995	30.5	10	11	72	0.00	0.00	0.00	0
Morbier	760	20	10	10	50	0.00	0.00	0.00	0
Comté	969	18.9	1.0	10	10	0.00	0.00	0.00	0
Roquefort	643	10	10	10	30	0.00	0.00	0.00	0

Milk and dairy products
☺
good sources of magnesium



Sodium, Potassium and Chloride in Milk



Sodium, Potassium and Chloride in Milk and Dairy Products

- Monovalent ions (Na⁺, K⁺, Cl⁻)
- Sodium, potassium and chloride are mainly in the aqueous phase of milk and dairy products
- Mainly free or weakly associated to ions of opposite charge
- In dairy products, the concentration of NaCl is increased by salting or brining (cheeses)
 - Contribute to :
 - Draining
 - Organoleptic properties of the cheeses
 - Selection of microorganisms and enzyme activities during ripening
 - Important variations depending on the type of the cheeses



Concentrations of Na and K in Some Dairy Products

mg/100g

	Ca	Mg	Na	K	P	Zn	Fe	Cu	Se
LIQUID MILK	126	112	59	150	85	26	0.23	0.2	0.5
Skim milk powder	1254	1112	420	1449	960	31.98	0.243	0.2	10.5
URT skimmed milk	1121	1018	414	1291	863.4	30.41	0.232	0.202	10.8
Concentrated whole milk	284	251	114	287	213	6.93	0.35	0.32	11.8
Salted butter	24	2	114	26	24	0.105	0.02	0.02	1
Unsalted butter	24	2	11	24	24	0.105	0.02	0.02	1
Yoghurt with skimmed milk	123	111	15	172	84	28.44	0.11	0.051	2.2
Feta	663	114	1118	62	337				
Cheddar	721	23	621	98	512				
Gruyere	1071	38	328	81	600				
Camembert	188	33	842	187	347				
Provolone	758	23	875	138	488				
Switzerland Emmentaler	791	38	152	77	387				
Cottage cheese 1%	61	1	117.8	66	131				
Parmesan	1184	24	171	52	854				
Mozarella	875	21	415	75	412				
Hard cooked cheeses	955	21.8	405	103	665				
Bleu d'Auvergne	583	17.1	224	84.8	391				
Fresh cheese 8%	118	11.7	42.1	124	182				
Processed Cheese (25% fat)	348	12	282	152	185				
Fresh cheese (type Petik salisse 26%)	117	10	75	98	123	0.18	0.2	0.011	0
Baufort	395	20.5	628	117	728	0.18	0.21	0.028	0.22
Morbier	160	30	100	100	520	7	0.01	0.02	0
Comté	899	21.8	412	118	661	2.1	0.23	0.1	7
Roquefort	662	21	175	81	582	20.16	0.23	0.051	11.6

For cheeses :

- Addition of NaCl during salting
- Low amount due to removal of K during draining



Prediction of the Salt Equilibria in Dairy Formula



Original model predicting salt equilibria



Calculations based on the combination of anions and cations according to their reciprocal affinity and considering pH, ionic strength, and solubility of calcium phosphate.

The calculations provided the concentrations of all species (free and complex) and their distribution between micellar and aqueous phases

Very interesting tool for preparation of various dairy formulations containing different concentrations of minerals and caseins



Interests and Use of the Software Milk Salts

- Good correlations between experimental and calculated values
- Several hundred combinations of dairy formula taking into account concentrations in proteins, minerals and pH can be calculated
- Facilitates the understanding of salt equilibria and could explain relationship between ionic environment and milk protein functionalities
- Helps the manufacturers in their decisions regarding modifications of the ionic environment for the development of dairy products (enriched milks, cheeses, yoghurts, etc)
- Replaces laboratory experiments and save money and time.



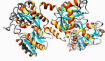
Trace elements in milk and dairy products : Fe, Zn, Cu and Se



FOOD
AGRICULTURE
ENVIRONMENT



Iron in Milk and Dairy Products

- Different states of ionisation (Fe^{2+} or Fe^{3+})
- In milk (associated to casein, citrate, lactoferrine) 
- Results depend on the analytical methods used for the determination
- Milk and dairy products are considered as very poor sources of iron
⇒ low contributions of dairy products to total iron intake



Iron Content of Some Dairy Products mg/100g

	Ca	Mg	K	P	Zn	Fe	Ca	Fe
LIQUID MILK	120	10	100	100	95	0.25	0.25	0.25
Skin milk powder	1254	115	525	1085	300	0.35	0.35	0.35
URT skimméd milk	113	10.8	114	100	88.8	0.27	0.27	0.27
Concentrated whole milk	384	25	114	201	113	0.33	0.33	0.33
Salted butter	24	0	578	54	24	0.08	0.08	0.08
Unsalted butter	24	0	11	12	24	0.07	0.07	0.07
Yoghurt with skimméd milk								
Feta						0.001	0.001	0.001
Cheddar						0.001	0.001	0.001
Gruyere						0.001	0.001	0.001
Camembert						0.001	0.001	0.001
Provolaone						0.001	0.001	0.001
Switzerland Emmental						0.001	0.001	0.001
Cottage cheese 1%						0.001	0.001	0.001
Parmesan						0.001	0.001	0.001
Mozzarella						0.001	0.001	0.001
Hard cooked cheeses	1650	15.8	605	110	690	0.2	0.2	0.2
Bleu d'Auvergne	900	13.4	112.9	64.8	301	0.46	0.46	0.46
Fresh cheese 9%	100	11.7	100	100	100	0.10	0.10	0.10
Processed Cheese (25% fat)	340	24	252	112	300	0.1	0.1	0.1
Fresh cheese (type Petit suisse 20%)	117	10	24	98	120	0.48	0.48	0.48
Beaufort	995	20.5	338	111	730	0.13	0.13	0.13
Morbier	780	30	300	100	520	0.1	0.1	0.1
Comé	900	48.9	412	116	664	0.1	0.1	0.1
Roquefort	662	10	1000	81	382	0.08	0.08	0.08

Milk and dairy products
⊗
very poor sources of iron



Zinc and Copper in Milk

Zinc :

- Divalent cation
- Milk concentration ~ 3-4 mg/L
- Locations :
 - mainly associated (95%) to casein micelles
 - secondly associated to citrate molecules in the aqueous phase

Copper :

- Milk concentration ~ 0.1 mg/L
- Milk and dairy products : poor sources of copper ⇒ low contribution to the intake of copper
- Locations :
 - 8% to whey proteins
 - 44% to casein
 - 47% to low molecular weight fraction (citrate)



Zinc and Copper in Some Dairy Products mg/100 g

	Ca	Mg	K	P	Zn	Fe	Ca	Fe
LIQUID MILK	120	10	100	100	95	0.25	0.25	0.25
Skin milk powder	1254	115	525	1085	300	0.35	0.35	0.35
URT skimméd milk	113	10.8	114	100	88.8	0.27	0.27	0.27
Concentrated whole milk	384	25	114	201	113	0.33	0.33	0.33
Salted butter	24	0	578	54	24	0.08	0.08	0.08
Unsalted butter	24	0	11	12	24	0.07	0.07	0.07
Yoghurt with skimméd milk								
Feta						0.001	0.001	0.001
Cheddar						0.001	0.001	0.001
Gruyere						0.001	0.001	0.001
Camembert						0.001	0.001	0.001
Provolaone						0.001	0.001	0.001
Switzerland Emmental						0.001	0.001	0.001
Cottage cheese 1%						0.001	0.001	0.001
Parmesan						0.001	0.001	0.001
Mozzarella						0.001	0.001	0.001
Hard cooked cheeses	1650	15.8	605	110	690	0.2	0.2	0.2
Bleu d'Auvergne	900	13.4	112.9	64.8	301	0.46	0.46	0.46
Fresh cheese 9%	100	11.7	100	100	100	0.10	0.10	0.10
Processed Cheese (25% fat)	340	24	252	112	300	0.1	0.1	0.1
Fresh cheese (type Petit suisse 20%)	117	10	24	98	120	0.48	0.48	0.48
Beaufort	995	20.5	338	111	730	0.13	0.13	0.13
Morbier	780	30	300	100	520	0.1	0.1	0.1
Comé	900	48.9	412	116	664	0.1	0.1	0.1
Roquefort	662	10	1000	81	382	0.08	0.08	0.08

Milk and dairy products
⊗ good sources of zinc
⊗ very poor sources of copper



Selenium in Milk and Dairy Products

- Concentration in milk : 30 µg/l
- Concentration in dairy products : poor knowledge
- Location : mainly in the skim milk
- Association with protein fraction (casein and whey proteins)



Biological and physiological roles of the minerals present in milk and dairy products



General Roles of Minerals (7-9 g/l of milk)

- Structural functions (bone) (Ca, P, Mg)
- Homeostasis (Na, K, Cl, Ca, P)
- Muscular contraction (Na, K, Cl, Ca)
- Blood coagulation (Ca)
- Structure of cellular membranes
- Enzymatic systems (Cu, Fe, Zn, Se,...) and activation of enzyme (Ca, Mn, Fe, Mg, Cu, Zn,...)
- Complexes hormones/trace elements (Zn and I)



Recommended Daily Allowance (RDA) of Some Micronutrients of Milk for Human

600 ml of milk



720 mg of Ca	65 mg of Mg	0.1 mg of Fe	2.4 mg of Zn	0.3 mg of Cu	20 µg of Se
↓	↓	↓	↓	↓	↓
~80 %	~16 %	~2.5 %	~20 %	~5 %	~33 %
of the RDA	of the RDA	of the RDA	of the RDA	of the RDA	of the RDA



The dairy products : unique sources of minerals



- Micronutrients are various in terms of :
 - Concentration (milk to cheeses)
 - Forms, locations and interactions
- They contributes significantly to our state of health
- For calcium and phosphate, the data concerning their impact on the technofunctionalities of dairy products is not clear
- To understand the biological effects of these mineral, it is necessary to know the concentration but also to integrate the chemical form, their associations with others compounds (ions, proteins)



Perspectives : Some Actual Questions on Minerals

- What are the role and structure of calcium phosphate in casein micelles ?
- Do exist relationships between qualitative/quantitative mineralisation and technological functionalities
- What are the evolutions of these micronutrients during the manufacture of dairy products?
- What are the effects of external compounds present in the food on the absorption of these micronutrients?
- Is it nutritionally interesting to enrich dairy products in micronutrients?



2ND INTERNATIONAL SYMPOSIUM ON MINERALS & DAIRY PRODUCTS

FROM MILK MINERAL SPECIATION TO NUTRACEUTICALS



26-28 February 2014
Auckland, New Zealand

WWW.MADP2014.COM

*Provide information on the latest scientific developments in different areas (chemistry, biochemistry, physics, dairy science, food technology, engineering, health and nutrition) related to milk minerals in dairy-based systems.

*Discuss industrial problems associated with milk minerals (e.g. heat stability, processed cheese).

*Propose new strategies in research and development sectors.

*Provide information on the health and nutritional aspects of fortified milks and nutraceuticals.

*Report on new physical and chemical methods to quantify mineral

Four sessions

- Fundamental aspects
- Nutrition and health
- Industrial applications
- General session



1ST INTERNATIONAL MEETING ON



Milk
Vector of
development

May 21-23, 2014 • Rennes, France

ORGANISED BY AGRENIUM (INRA, CIRAD, AGROCAMPUS OUEST) AND CORFILAC



 **Milk: Vector of development**

In industrialised countries as in developing countries, regardless of climate and the species domesticated, milk and dairy products often have an important place in daily life and represent an undeniable vector of local development and food safety. The diversity of milk production and processing practices throughout the world is considerable and has invaluable heritage value. Besides playing an unquestioned role in nutrition, these products often play a key role in local economies and regional development, with multiple development models due to local constraints and opportunities, all within a context of increasing globalisation.

The objective of this conference is to exchange experiences of the diversity of observed milk-sector developments, of technical and sanitary issues, assemble the initiatives and networks involved, measure the importance of women in this development, and better identify the role, when present, of public policies. To jointly improve coordination of activities related to milk and its associated products, particularly joint research projects, training activities, or advisory activities, is the main aim of this meeting.

Public targeted: Research and technical institutes, agronomic or veterinary institutions of higher education, NGOs and associations/foundations, chambers of agriculture, political leaders in the development field, private companies, etc.

  
<http://colloque.inra.fr/lait2014>
 University of Viçosa, Brazil, 31 October to 2 November 2013 27


Thank you for your attention

&

Professor Antonio





  
 University of Viçosa, Brazil, 31 October to 2 November 2013 38