

473  
2014

# Bulletin

of the International Dairy Federation

Compilation of some  
presentations from the  
**IDF World Dairy Summit 2013**  
Yokohama, Japan



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## Bulletin of the International Dairy Federation 473/2014

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# Compilation of some presentations from the IDF World Dairy Summit 2013 Yokohama, Japan

## C O N T E N T S

Foreword .....	1
1 Turkish dairy sector and traditional dairy products at a glance .....	3
2 Growth of pasteurized dairy products in Thailand .....	13
3 Metabolomics-based hydrophilic component profiling of cheeses using GC/MS .....	19
4 Variability of volatile organic compounds during the manufacture of Swiss-type cheese using selected ion flow tube mass spectrometry .....	29
5 Why children love milk? Milk companies and organizations together for school milk success, Polish experience .....	51
6 The history of milk in Japanese school lunches .....	55
7 The school milk scheme of the European Union .....	65
8 Development of the School Milk Programme in China .....	69
9 Preventive effects against infections by intake of yogurt fermented with <i>Lactobacillus delbrueckii ssp. bulgaricus</i> OLL1073R-1 .....	73
10 Milk consumption promotion in Chile: the PROMOLAC experience .....	79
11 Biodiversity in Japanese dairy systems: Floral diversification mechanisms in grazed grasslands .....	85
12 Revision of Codex Microbiological Criteria guideline .....	91
13 Feasibility and issues in the application of HACCP at the farm level in Japan .....	95

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# C ompilation of some presentations from the IDF World Dairy Summit 2013 Yokohama, Japan

## FOREWORD

The IDF World Dairy Summit brought together many stakeholders of the dairy chain and offered a unique platform for companies, academia and dairy leaders to share their knowledge and experience. It also provided an opportunity for people involved in the field to engage in a frank and open discussion about innovative research, the progress achieved and lessons learnt.

This Bulletin contains thirteen papers presented during the IDF World Dairy Summit in Yokohama, Japan from 28 October to 1 November 2013.

The conferences topics were Children and milk, Animal health and welfare, Dairy farming, Nutrition and health, Marketing, Environment and Food safety.

IDF would like to thank the authors whose contribution helped make the event in Yokohama memorable for the dairy sector and for the many

participants. Their contributions enable those who could not attend to learn about the new information presented at the IDF World Dairy Summit.

IDF wishes to express its sincere thanks to the IDF National Committee of Japan for organizing the 2013 edition of the IDF World Dairy Summit and for collecting these proceedings.

The next IDF World Dairy Summit will take place from 27-31 October in Tel Aviv, Israel. On behalf of IDF, I look forward to welcoming you all there.

Nico van Belzen, PhD  
Director General  
International Dairy Federation  
July 2014



# 1 Turkish dairy sector and traditional dairy products at a glance

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## 1. INTRODUCTION

Milk production in Turkey has increased over the last few years depending on holdings of milking animal and annual milk yield per cow. Total milk production in Turkey was 12.5 million tons in 2010, and increased dramatically to 13.6 million tons and 15.05 million tons in the following years and 17.4 million tons in 2013. In 2011, the number of milking cows was 5.3 million head. In 2012, liquid milk consumption per capita was estimated to be 33.1 kg/year.

Moreover, dairy foreign trade volume differs in both value and quantity. Dairy export value in 2010 was calculated to be 167 million USD and showed a significant increase in 2011 and reached 226 million USD. While there was a slight drop in dairy export value in 2012 (228.3 million USD), Turkey's dairy import value was 106 million USD in the same year.

Briefly, the Turkish dairy sector has been developed over the past years, swelling its production and consumption. However, one of the key issues in the Turkish dairy sector is stabilizing the raw milk prices. Increasing feed prices, which are the most important cost item in dairying, causes a rise in the total cost of production, resulting in a high raw milk price.

## 2. TURKISH TRADITIONAL DAIRY PRODUCTS

Turkish traditional dairy products are mostly made from sheep, goat and cow milk. While buffalo milk is hardly produced in Turkey, the milk is used

to manufacture butter, cream and some kinds of cheeses. Moreover, depending on the regional culture, the locally grown vegetables, spices and fruits are used as aroma.

Some of the traditional/regional dairy products are herby cheese, çeçil cheese, plait cheese, tongue cheese, aged skin bag cheese, çökelek, Mihalliç cheese, sack yogurt, ayran, creamy yogurt, butter made in a butter churn, buffalo milk cream, Maraş ice cream and Kurut.

## 3. TURKEY AT A GLANCE

### Republic of Turkey

- Population: 75.6 million
- Capital city: Ankara
- Most populous city: Istanbul
- Government: parliamentary democracy
- Official language: Turkish

Turkey, officially the Republic of Turkey, is a transcontinental country located mostly on Anatolia, in Western Asia and East Thrace in South-eastern Europe. Besides being a unitary parliamentary republic, Turkey is a democratic, secular, constitutional country with a diverse cultural heritage. As most of you possibly know, although Istanbul is the most popular and populous city in Turkey, Ankara is actually the capital.

## 4. LIVESTOCK HOLDINGS

Starting with livestock holdings in Turkey, both store cattle and dairy cow numbers in Turkey have been increasing since 2002. According to the

figures provided by Turkish Statistical Institute, there are more than 50 million head of animals in Turkey.

Compared with the previous year, while the cattle and buffalo numbers have increased by 12%, the increase rate for sheep and goat holdings has been calculated to be more than 10%.

### Livestock holdings

Year/Head	Cattle	Buffalo	Total bovine animals	Sheep	Goat	Total sheep and goat
2002	9 803 498	121 077	9 924 575	25 173 706	6 780 094	31 953 800
2003	9 788 201	113 356	9 901 458	25 431 539	6 771 675	32 203 214
2004	10 069 346	103 900	10 173 246	25 201 155	6 609 937	31 811 092
2005	10 526 440	104 965	10 631 405	25 304 325	6 517 464	31 821 789
2006	10 871 364	100 516	10 971 880	25 616 912	6 643 294	32 260 206
2007	11 036 753	84 705	11 121 458	25 462 293	6 286 358	31 748 651
2008	10 859 942	86 297	10 946 239	23 974 591	5 593 561	29 568 152
2009	10 723 958	87 207	10 811 165	21 749 508	5 128 285	26 877 793
2010	11 369 800	84 726	11 454 526	23 089 691	6 293 233	29 382 924
2011	12 386 337	97 632	12 483 969	25 031 565	7 277 953	32 309 518
2012	13 914 912	107 435	14 022 347	27 425 233	8 357 286	35 782 519

Total livestock in 2012 was 50 186 583 head

Source: Turkish Statistical Institute, 2013

## 5. MILKING ANIMALS

As for the milking animals, there are more than 5 million dairy cows and just 46 000 buffalo in Turkey. Similar to the figures for total animal holdings, dairy animal numbers have also been

developing year by year and thus, milk production, total dairy products and trade value relating to dairy has been increasing.

### Milking animals

Year/Head	Cow	Sheep	Goat	Buffalo	Total
2002	4 392 568	13 637 193	3 553 438	51 626	21 634 825
2003	5 040 362	12 477 217	3 126 656	57 378	20 701 613
2004	3 875 722	9 919 191	2 476 574	39 362	16 310 848
2005	3 998 097	10 166 091	2 426 993	38 205	16 629 386
2006	4 187 931	10 245 894	2 420 642	36 353	16 890 820
2007	4 229 440	10 109 987	2 263 630	30 460	16 633 517
2008	4 080 243	9 642 170	1 997 689	32 610	15 751 542
2009	4 133 148	9 407 866	1 830 814	32 361	15 404 189
2010	4 361 841	10 583 608	2 582 539	35 632	17 563 350
2011	4 761 142	11 561 143	3 033 111	40 218	19 395 614
2012	5 431 400	13 068 428	3 502 272	46 959	22 049 059

Population growth rate for milking cows between 2002 and 2012 was 23%

Source: Turkish Statistical Institute, 2013

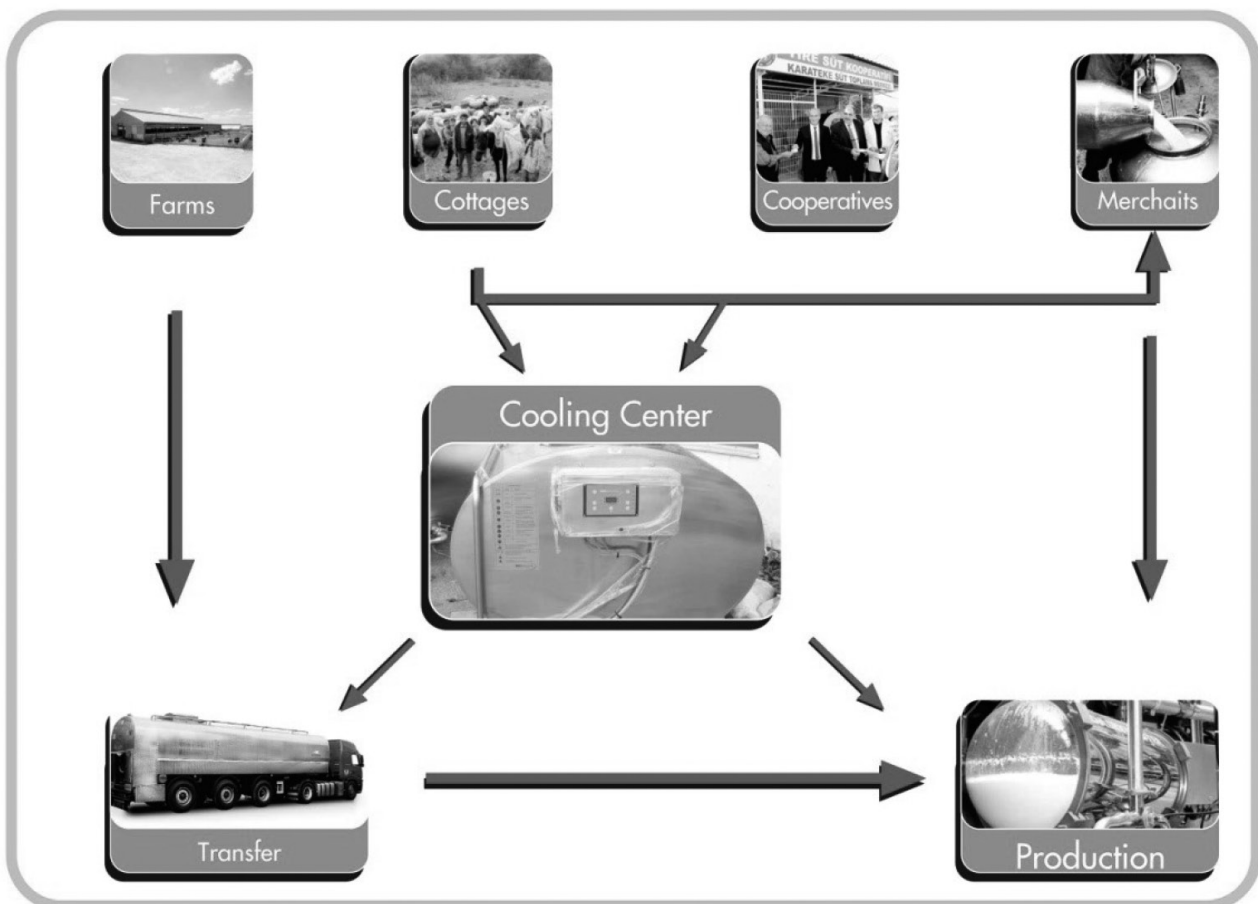


## 6. THE SUPPLY OF RAW MILK

We have many integrated dairy plants in Turkey. As you know, Turkey is a candidate country for EU and as part of this, the FVO mission came to Turkey and checked the dairy sector in our country. We intend to maintain food safety, not only in dairy sector but also in all the other sectors.

One of the important issues in Turkish dairy sector that needed to be dealt with is the milk delivery

rate for all species. According to the statistics, almost half of the total cow milk production is delivered to companies in Turkey. The rest of the quantity is processed by small dairy farms. Compared with 2011, cow milk delivery has increased monthly. Especially in the last few years, people mostly prefer to consume packaged dairy products due to revenue growth, high educational standards and increasing public awareness. This has caused a boost in milk delivery rates in Turkey.

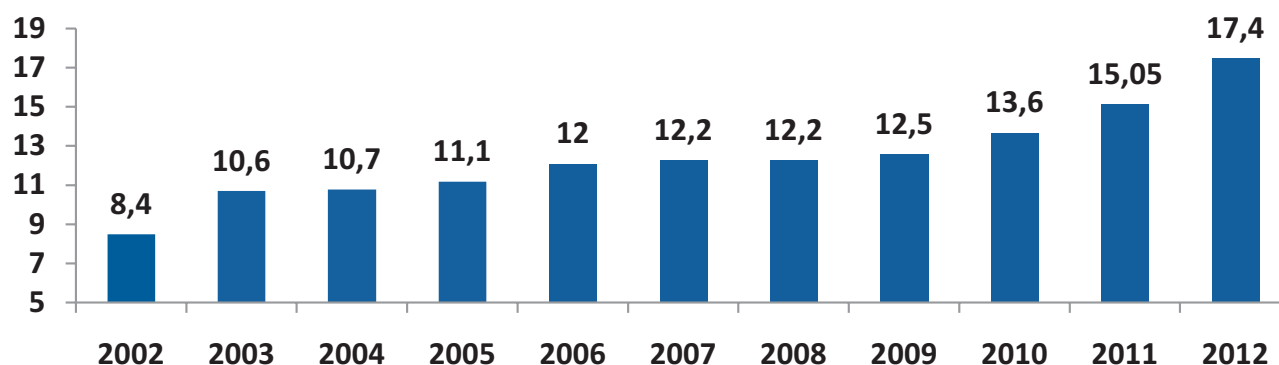


## 7. MILK PRODUCTION IN TURKEY

Milk production in Turkey has shown a significant development. Last year's total production was measured at 17.4 million tons, with a 15% increase compared with the previous year. But the main point shown in the figure below is that while the cow holdings in Turkey have increased by 23% in

the last decade, total milk production in Turkey has soared, more than doubling in the same time period. This demonstrates that milk yield per cow is becoming an increasingly important point for the farmers.

### Milk production in Turkey (million tons)

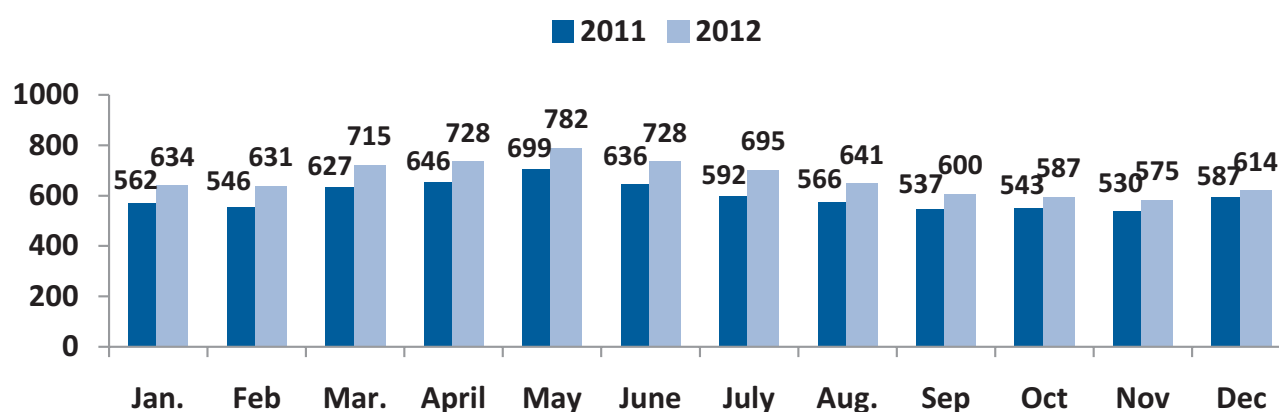


Source: Turkish Statistical Institute, 2013

## 8. COW MILK DELIVERY

As seen in the figure below, almost 8 million tons of milk was collected by the companies last year.

### Cow milk delivery (thousand tonnes)



Source: Turkish Statistical Institute

## 9. REGIONAL MILK PRODUCTION DENSITY

The provinces producing the most milk are Izmir, Balıkesir, Konya, Aydın, Çanakkale, Denizli and Burdur. While the Izmir, Balıkesir, Konya, Aydın, Çanakkale, Denizli and Burdur have more than 300 000 tonnes of milk production yearly, five

cities including Tekirdag are the second biggest producer cities.

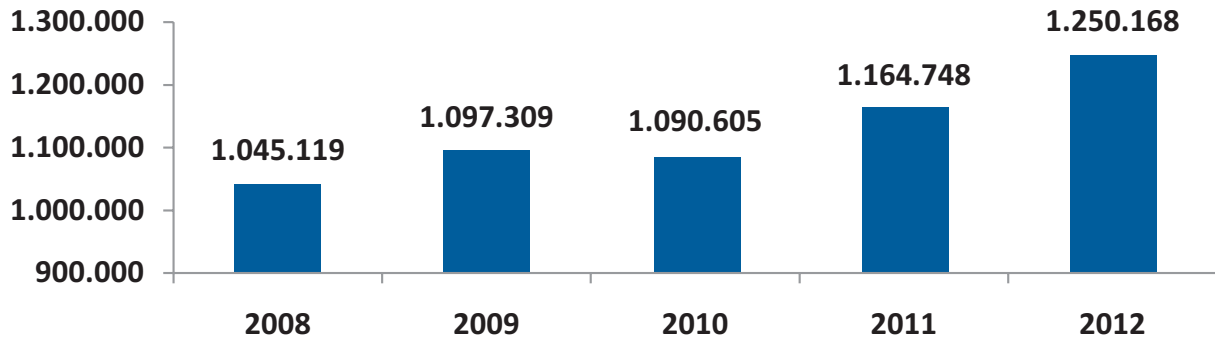
As for production of dairy products, Turkey is a unique country that has a wide diversity of dairy

products like ayran, yogurt and special kinds of cheeses. Due to the increasing milk consumption, liquid milk production growth shows a significant development. The figure shows that total liquid milk production in 2012 was 1 250 000 tons, which

is 7% more than the previous year. As you see, people living in Turkey generally prefer UHT milk to pasteurised milk, possibly because of its long life endurance.

## 10. LIQUID MILK PRODUCTION

Liquid milk production (tonnes)



Liquid milk production growth between 2011 and 2012 was 7.3%

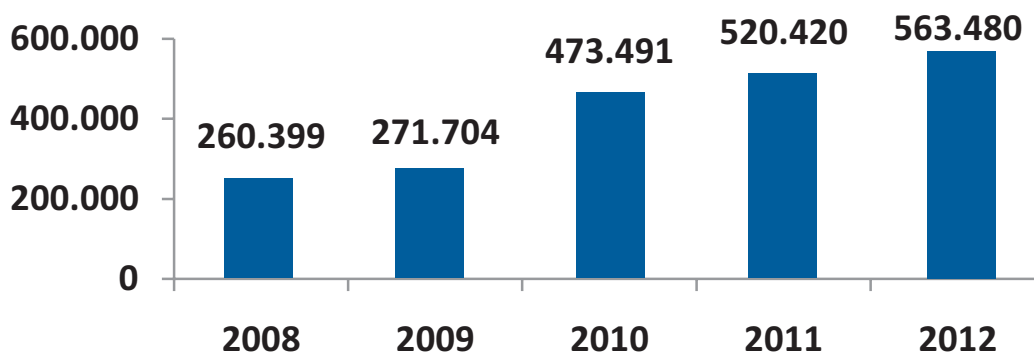
Source: Turkish Statistical Institute, 2013

Cheese is a special and unique dairy product for Turkish people, especially for breakfast. According to a survey carried out by a professor studying at Kafkas University, there are approximately 204 kinds of cheese in Turkey. This is evidently proof that Turkish people are keen on consuming cheese. That is the reason why our cheese

production has shown a slight increase for past 3 years. One of the main points regarding to cheese production in Turkey is that most kinds of cheese are made from cow milk. However, in rural areas, even in large cities like Istanbul and Ankara, cheeses made from goat or sheep milk are becoming increasingly popular.

## 11. CHEESE PRODUCTION

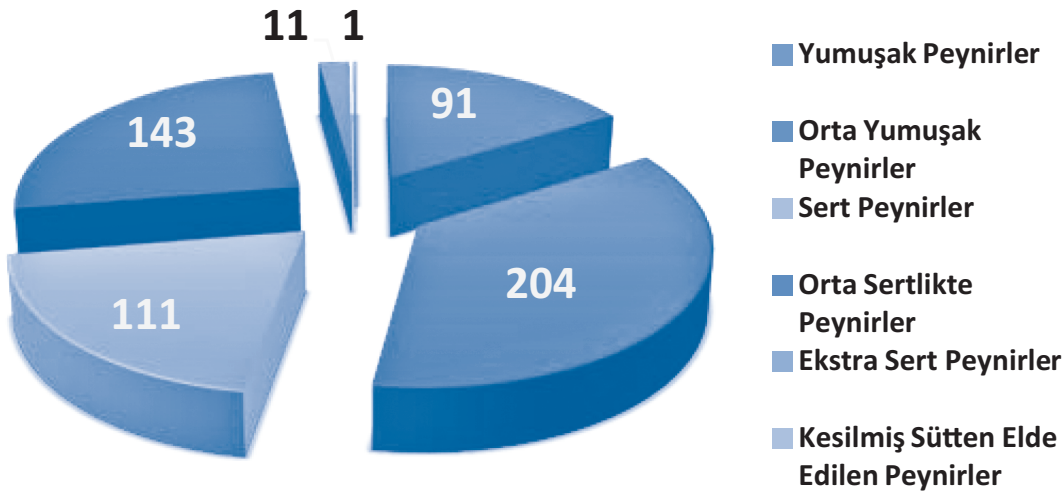
Cheese production (thousand tonnes)



96% of the total production is made from cow milk

Cheese production growth rate between 2012 and 2011 was 8.3%

## Cheese types



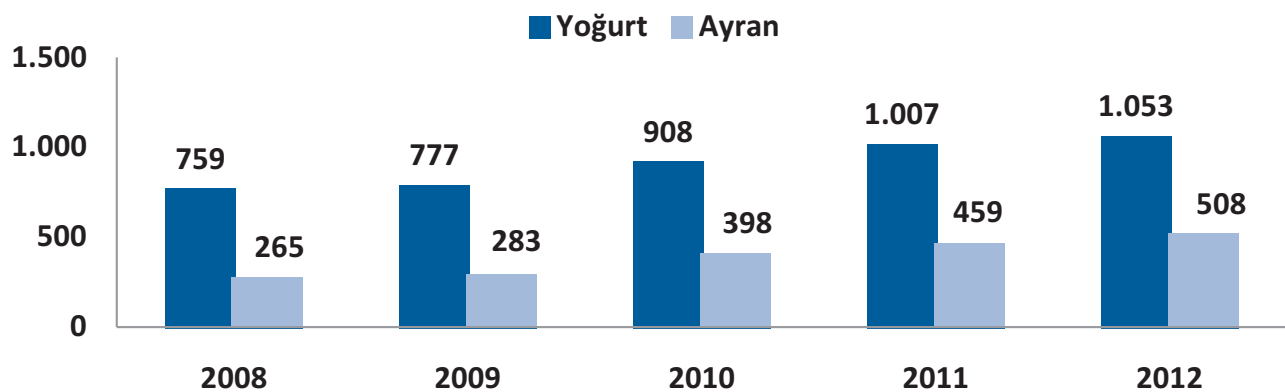
204 different kinds of cheese are produced

Source: Turkish Statistical Institute, 2013

Among all the dairy products produced in Turkey, yogurt is the second product regarding the production quantity. As traditional dairy products, ayran and yogurt are consumed in almost all parts of Turkey. As seen in the figure below, while yogurt production presented a modest increase in 2012, ayran production showed relatively high production increase. It should be noticed that ayran consumption rises in summer thanks

to it being a refreshing drink. What is quite an important development concerning ayran is that there is a collaborative study being carried out by the Food Agriculture and Livestock Ministry, National Milk Council and Codex Alimentarius on determining the concentration, salt content and standardized other ingredients and production methods.

## 12. YOGURT AND AYRAN PRODUCTION



Yogurt production rose 4.5% in 2012 to 1 053 657 tonnes  
Ayran production rose 10.7% in 2012 to 508 444 tonnes

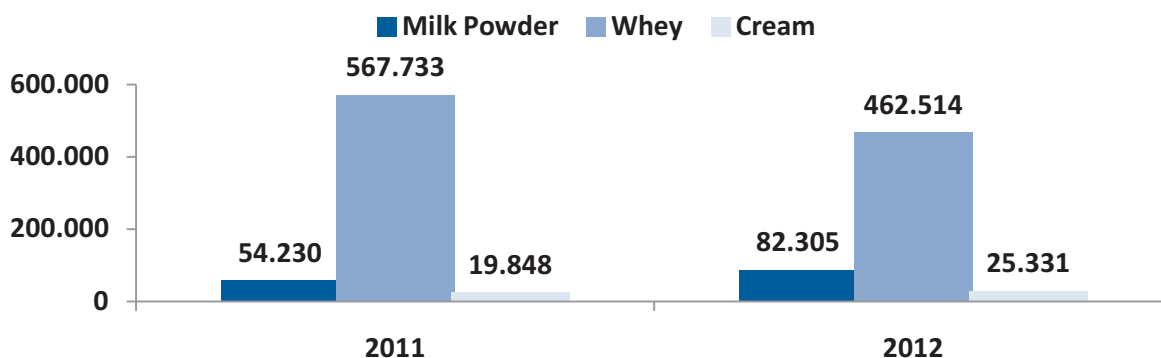
Source: Turkish Statistical Institute, 2013

### 13. MILK POWDER, WHEY PRODUCTS AND CREAM PRODUCTION IN TURKEY

Milk powder is an important dairy product in terms of intervention in the dairy market in Turkey. Within the scope of evaluation of surplus milk, milk powder producers are subsidized providing that the milk used by producing milk powder has to be bought at a reference price. According to the figures given, milk powder production showed a significant increase in 2012, apparently due to

the implementation of these subsidies. Similarly to milk powder production, cream production went up in 2012, while there was an considerable decline in whey production. Butter, which has a great effect on Turkish dairy foreign trade, demonstrates a significant production growth every year. Butter production increased by 15% in 2012, reaching 40 000 tonnes.

Milk powder, whey and cream production



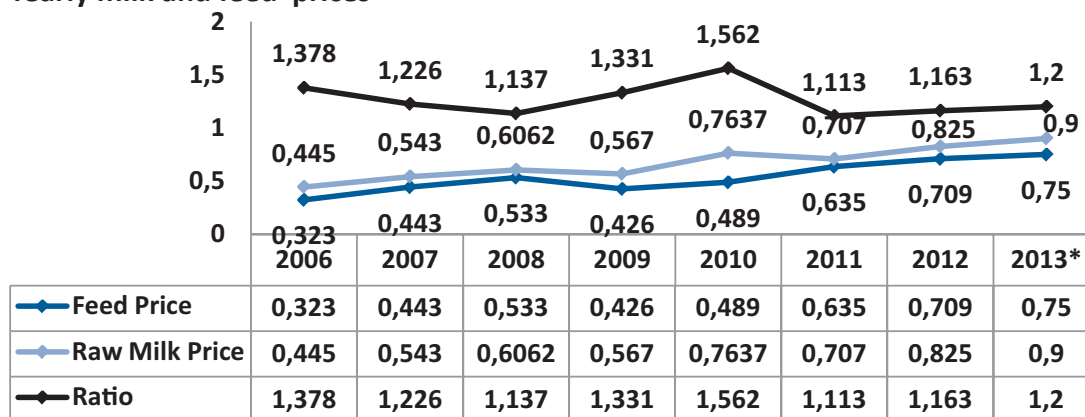
Whole milk powder production in 2012 was 35 119 tonnes (+81%)  
 Skimmed milk powder production in 2012 was 49 199 tonnes (+41%)  
 Butter production in 2012 was 40 139 tonnes (+15%)

Source: Turkish Statistical Institute, 2013

I am now going to move on to raw milk and feed price while giving some information on the milk-to-feed price ratio. Milk price in Turkey was previously determined by regional tenders, but for the last 2 years prices have been established for a particular period by negotiations between producer and processor representatives. As shown in table below, both milk and feed prices

have been swelling slightly every year. As all of you appreciate, feeding of animals constitutes 75% of the total cost of producing milk; it is quite evident that the most influential reason for this increase has been boosting of feed prices. One point that should be emphasized is that apart from those raw milk prices, farmers receive support payments such as raw milk, calf and feed subsidies.

Yearly milk and feed prices



Source: Turkish National Dairy Council, 2013

As for the farm structure in Turkey, most of the dairy farms in Turkey have between one and five cows. Because the farms that only have one cow are assumed to be a dairy farm, the percentage

of the farms having five or less cows is quite high. The thing needed to note is that the farms which have less than 10 cows constitute 80% of the total farms.

## 14. FARM STRUCTURE

Farm size (head)	Number of farms	Share in total (%)	Cow holdings (head)
1–5	811 778	58.74	2 063 726
6–9	293 799	21.25	2 095 781
10–25	198 117	14.35	2 767 188
26–49	62 858	4.54	1 812 749
50–100	11 681	0.84	866 621
101–199	2 798	0.20	388 490
200+	1 190	0.08	378 959
<b>Total</b>	<b>1 382 281</b>	<b>100</b>	<b>10 373 513</b>

80% of total farms have 1–9 cows

Source: Food Agriculture and Livestock Ministry, 2013

When it comes to the consumption figures, per capita milk consumption in Turkey is 230 kg milk equivalent. Compared with the year 2010, milk

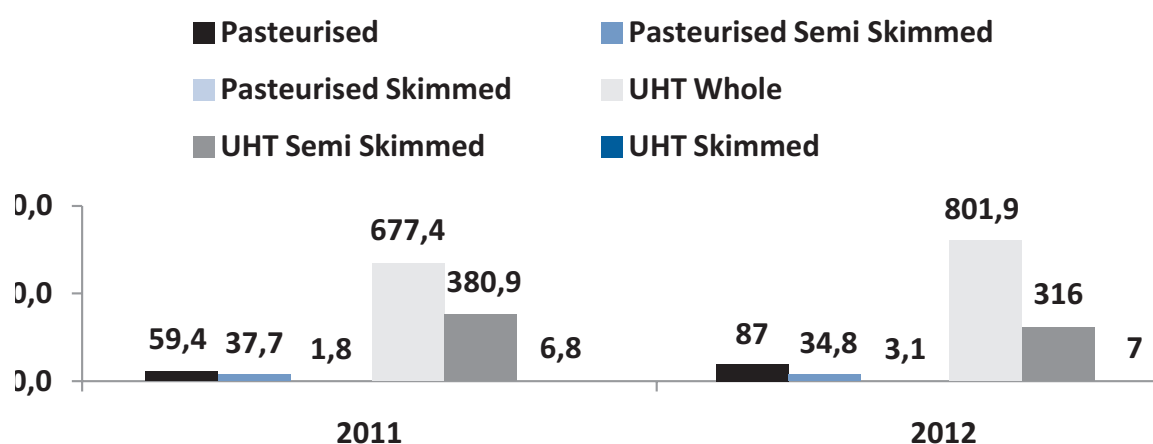
consumption figures increased by 30%. However, as far as I am concerned, calculation of milk consumption is not reliable enough because it is highly likely that same unit of milk could be calculated twice. In skimmed milk production for instance, 1.5% fat milk is sold; at the same time, the rest of the fat is sold as butter for instance. That is the reason why it is a little bit controversial in some cases.

Liquid milk consumption figures are determined using both packaged and farm use milk. Compare to the year 2010, liquid milk consumption increased about 30%. There is no doubt that the main effect of this increase is the school milk programme. With the cooperation of the Ministry of Health, the Ministry of Food Agriculture and Livestock, the Ministry of Education and the National Dairy Council, the School Milk Programme has been functioning for the past 2 years.

## 15. DAIRY CONSUMPTION

Per capita milk consumption in milk equivalent is 230 kg.

### Liquid milk consumption



Liquid milk consumption per capita is estimated at 33.1 kg

Source: Turkish Statistical Institute, 2013; Turkish National Dairy Council, 2013

The share of dairy products in the total export value of the 4<sup>th</sup> section in custom tariff is increasing year by year. The export of dairy products is becoming more important for Turkish foreign trade. Especially in 2011, there was a dramatic increase in dairy products. While total trade volume for dairy was around 168 million USD in 2010, it reached 225 million USD in 2011. Even though there was a slight decrease in total

trade volume, it is still not below 220 million USD.

The key export products for dairy are cheese and whey. While total cheese export volume was around 130 million USD, the export value for whey and whey products was about 20 million USD. As for the key markets for dairy, Turkish exporters are mostly doing business with the Middle East and North African countries.

## 16. FOREIGN TRADE

### 16.1. Exports

- Total dairy products export value: 225.3 million USD
- Key products for export: Cheese and whey powder

#### Key markets for dairy products

- Saudi Arabia
- Iraq
- Kuwait
- United Arab Emirates
- Azerbaijan
- Libya
- South Korea

(Source: Ministry of Economy)

The import value for dairy products was around 106 million USD last year. Butter is the key product

among all dairy products and generates more than half of the total dairy products value. The main suppliers for all products are Netherlands, New Zealand, Ireland, Switzerland and Italy.

### 16.2. Imports

Total dairy products import value: 105.9 million USD

Key products for import: Butter (15 000 tonnes for 2012)

#### Key suppliers for dairy imports

- Netherlands
- New Zealand
- Ireland
- Switzerland
- Italy

(Source: Ministry of Economy, 2013)

## 17. TRADITIONAL CHEESE PRODUCTION

Traditional cheese	Total numbers of units	Total production (tons/year)
White cheese	755	5 692 111
Processed cheese	158	95 981
Precipitated cheese	65	1 279
Dil cheese	143	24 171
Hellim cheese	13	1 037
Kaşar cheese	667	2 794 754
Village-type cheese	171	368 811
Moldy cheese	8	119.6
Lor cheese	506	387 388
Herby cheese (otlu)	19	571.9
Mesh cheese (örgü)	169	38 885
Fresh cheese	37	442.5
Tulum cheese (skin, overalls)	171	599 885
Fresh cheese (flavoured)	11	11 547

## 18. LOCAL CHEESE PRODUCTION

Local cheese	Total numbers of units	Total production (tons/year)
Wire cheese	755	5 692 111
Urfa cheese	158	95 981
Circassian cheese (Çerkez)	65	1 279
Abaza cheese	143	24 171
Mihallic cheese	13	1 037
Basket cheese (sepet)	667	2 794 754
Keş cheese	171	368 811
Ezine cheese	15	2 673
Yörük cheese (Tribal)	4	2 388
Kolot cheese (Golot)	7	90.6
Maraş parmesan cheese	8	323
Antep cheese	58	2 904
Other local cheeses	103	2786.4

## 19. FOREIGN-STYLE CHEESE PRODUCTION

Foreign-style cheese	Total numbers of units	Total production (tons/year)
Camembert cheese	1	0
Cheddar cheese	7	5 365
Edam cheese	5	11 927
Emmental cheese	3	3 336
Gouda cheese	3	12 702
Gruyere cheese	13	219
Parmesan cheese	2	10.6
Mozzarella cheese	18	4934.4

## 20. SOURCES

1. Ministry of Economy Report, 2013
2. Turkish Statistical Institute Statistics Report, 2013
3. Turkish National Dairy Council Report, 2013
4. Food Agriculture and Livestock Ministry Report, 2013



# 2 Growth of pasteurized dairy products in Thailand

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## ABSTRACT

As most of us in the industry may know, the majority of dairy product markets, especially milk, are stagnant or even in negative growth especially in the USA, UK and other developed countries. Many have asked why Thailand's milk market has grown consistently in the previous few years. In this paper I will try to share my understanding of the market, considering the following topics:

- **Supply side of milk and regulation:** to have a look at how the industry works in Thailand from the supply perspective .
- **Current milk market in Thailand:** to have a look where the demand of the market has been driven and where we aim to steer it towards.

## 1. SUPPLY SIDE OF MILK AND REGULATION

### 1.1. History of milk industry in Thailand

Milk in Thailand, like most south-east Asian countries; is not a cultural food. Milk was officially introduced in Thailand by HM King Rama IX in 1962 by promoting dairy farming in Thailand in cooperation with the Government of Denmark, forming the Thai-Danish Dairy Farm along with the Pilot Milk Production plant and setting-up of a milk cooperative program in Thailand. Today, the Thai-Denmark Dairy Farm is capable of producing 165 000 tons per year of UHT milk to feed Thailand's local market. With His Majesty Rama IX's support, milk consumption in Thailand has been quickly recognized. Thais consider it to be the utmost fortune for the Thai people that His Majesty the King has a keen interest in the Thai dairy industry.

### 1.2. Thailand milk supply chain from farm to consumer

Currently, domestic raw milk supply in Thailand is approximately 3095 tons per day. In which 61% is produced by cooperatives and the remaining 39% produced by Milk Collecting Centers and other sources combined. The Thai government supports a school milk program for students all the way through primary school. Therefore, 40% of the combined total domestic milk production has been allocated to the school milk program. The remaining 60% is for commercial use by the 10 major milk manufacturers. The 60% of supply allocated for commercial use is then mixed with imported milk (SMP and AMF) to ensure an adequate supply of milk for commercial use (see Table 1).

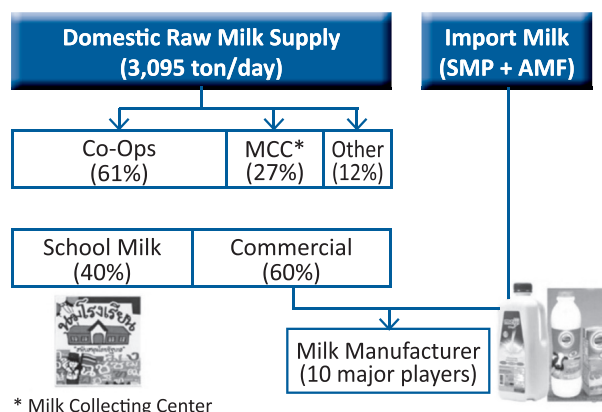
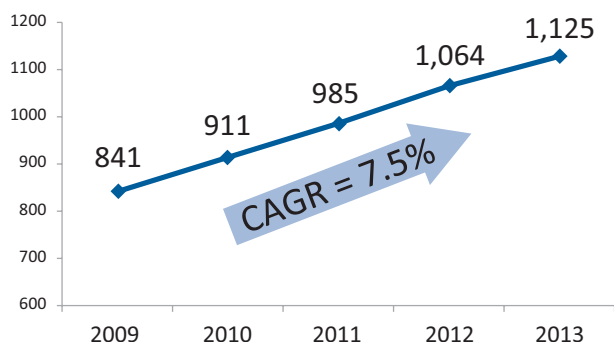


Table 1: Thailand's milk supply chain from farm to consumer

### 1.3. Raw milk supply and demand

Raw milk supply in Thailand has increased steadily at a CAGR rate of 7.5% from 2009 to 2013 (see Table 2). However, the sales growth of CP-Meiji over the same period was 16%. This means that CP-Meiji has to perform special projects to secure and ensure the quality of the raw milk supply.

**Raw Milk Production Volume ('000 ton)**



**Table 2:** Total Thailand raw milk supply trend from 2009 to 2013

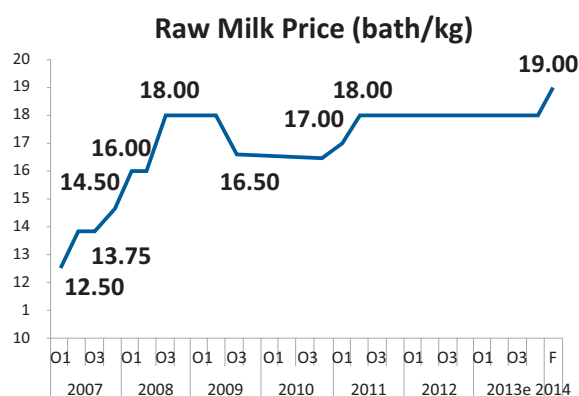
**1.4. Relationship between raw milk price and milk retail price**

Thai government views milk as an important nutritional product, therefore the government regulates both the raw milk price and milk retail price (see Table 3).

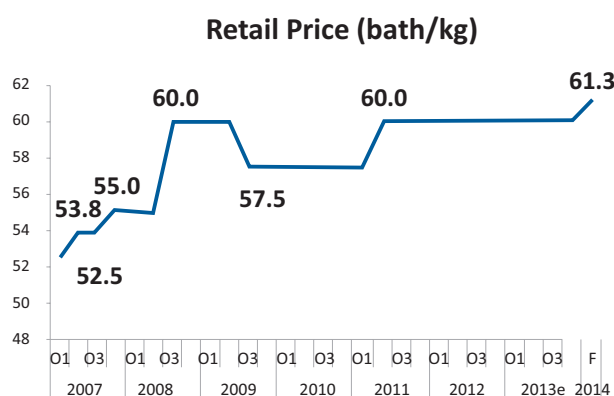
Because the Thai government intends to promote and support dairy farming for milk product manufacturing, they have set up a

regulated minimum price for raw milk, which all manufacturers purchasing raw milk from the dairy farmer have to follow. In the other hand, the retail price of milk products in Thailand is government regulated to promote milk consumption by Thai people. The Department of Internal Trade under the Ministry of Commerce regulates the maximum milk retail price.

**Generally, controlled retail price can be increased only when the raw milk price increases**



*Setup by Milk Board Committee and approve and control by Cabinet*



*Setup and control by Department of Internal Trade, Ministry of Commerce*

**Table 3:** Relationship between retail price and raw milk price

**1.5. Dairy farming development project**

Currently, there are 21 000 dairy farmers in Thailand, with 6 000 supplying milk to CP-Meiji daily. Some 70% of the 6000 dairy farm are small sized farms (10 milking cows or less). The average yield is 12.5 kg per cow per day, which has only increased slightly by a rate of 2.0% per year for the

past 5 years. There are 159 Milk Collecting Centers in Thailand, 26 of which supply to CP-Meiji daily. Most of the Milk Collecting Centers have already received GMP certification. In order to secure and ensure the quality of raw milk supply, CP-Meiji initiated a Joint Dairy Farming Development

Project, which is a collaboration between Milk Collecting Centers, dairy farmers, CPF (Charoen Phokphand Foods PCL) and CP-Meiji. The CPF Group is Thailand’s leading agro-industrial and food conglomerate. One of its businesses is a feed mill for cows and it is also a major shareholder of CP-Meiji. In order to produce the best quality product, CPF operates a high standard milking research facility capable of producing 25 kg of milk per cow per day. Current output is 20 tons per day and there are plans to double capacity within 2 years. CP-Meiji estimates its requirement of high quality raw material to be 325 tons per day in 2014, of which it will buy 50% from exclusive sources and 50% from shared sources. Therefore, CP-Meiji is under pressure to secure raw milk sources to sustain company growth. The objective of the project is to improve raw milk output from 13 to 15 kg per cow per day as well as improving

raw milk quality (see Table 4). The project includes the following measures:

1. **Milk hygiene:** Stakeholders of the project aim to improve product quality of cow milk through introduction of standard milk hygiene practices to be implemented at dairy farms.
2. **Milk feeding program:** Aiming to improve quality of output through a nutrition program for cows to ensure that optimum nutrition is fed to the cow according to its age and surrounding environment.
3. **Better breeding:** Through better breeding selection and breeding process, farmers are able to breed cows that have a higher chance of breeding success, are less prone to disease and provide better milk output in terms of both quality and volume.

	Current	Objective
Raw Milk (kg/cow/day)	13	15
Standard Plate Count (SPC)	800,000	500,000
Somatic Cell Count (SCC)	700,000	500,000
Solid Non-Fat (SNF)	8.4	8.5

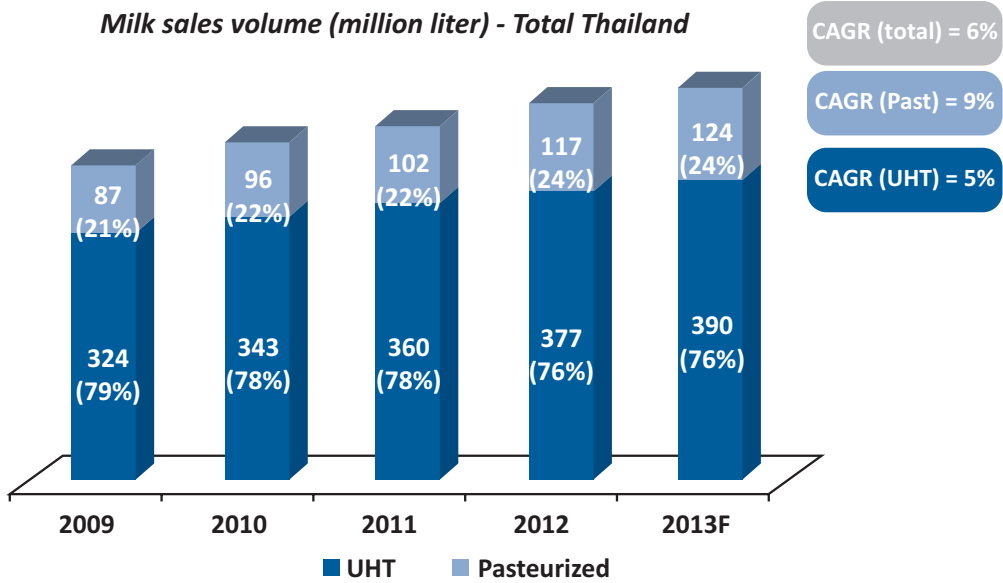
Table 4: Objectives of the Dairy Farming Development Project

## 2. CURRENT MILK MARKET IN THAILAND

### 2.1. Commercial milk product growth in the past 5 years

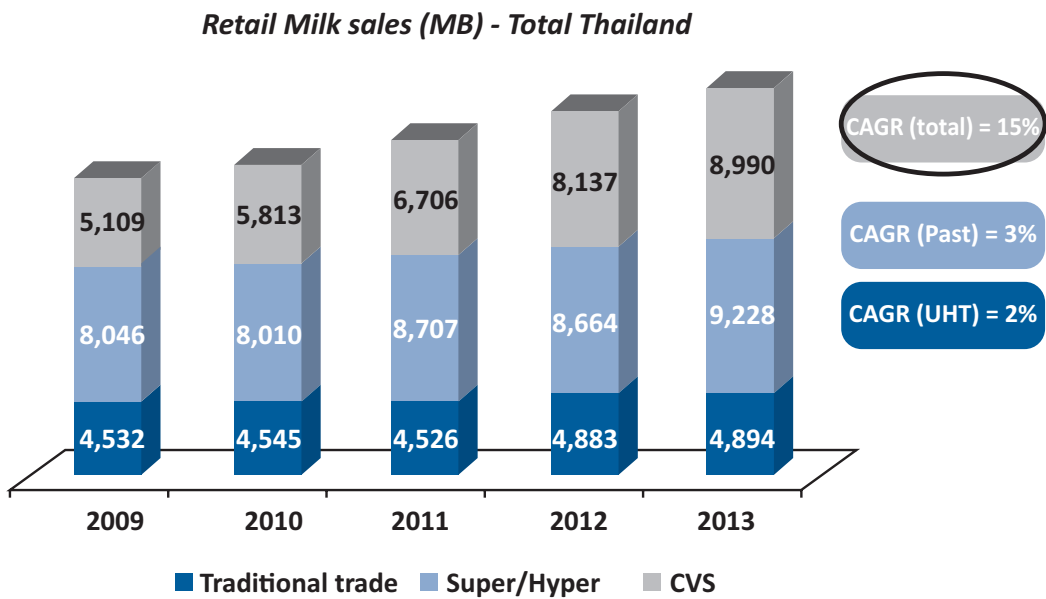
UHT is the main milk product in Thailand. However, the UHT market is slowly moving (CAGR 5%) towards pasteurized milk (CAGR 9%) (see Table 5). The market contribution of UHT product decreased continuously from 79% in 2009 to 76% in 2013. Each product type has a different product personality: pasteurized milk has a better taste than UHT milk but is an inconvenient product due to the requirement for temperature control from

factory to retailer and to the end consumer and has high logistic cost. On the other hand, UHT milk can serve the majority of the market through its lower price, longer shelf life and convenience of handling due to no requirement for refrigeration in the logistics process. The main reason for the mix shift is due to the growth of the convenience channel, which is able to facilitate distribution and temperature control infrastructure (see Table 6).



**Table 5: Commercial milk product growth in the past 5 years**  
 The Thai milk market is slowly shifting towards pasteurized milk

Source: Nielsen Retail Index



**Table 6. Retail milk sales in Thailand: by channel**  
 The convenience channel is the main growth driver for the Thai milk market  
 (Data includes pasteurized and UHT milk)

Source: Nielsen Retail Index

## 2.2. Current milk portfolio proposed to consumer

Currently, CP-Meiji is leader in the pasteurized milk market with 54% market share. The market is divided into two main segments:

1. **Mainstream segment:** managed through Core, Classic, and Seasonal flavours while focusing on price.
2. **Premium Segment:** managed by adding value through additional nutrition for the consumer. Two examples are the Meiji Gold Maxx line, which is premium milk enriched with 18 essential amino acids for brain and physical development of children, and Meiji Gold Love line, which is a complete care for women with nutritional benefits for blood, brain and bone.

## 2.3. CP-Meiji sales and distribution strategy

At CP-Meiji, we have always been pushing hard into the convenience channel due to our expertise and market leader position in the pasteurized milk segment, and as a result CP-Meiji grew by 18%, faster than convenience channel growth. The convenience channel is also a great place to create new product trials. Therefore, we used the convenience channel to create a trial of a small pack size (200 ml). If the product is successful, we can leverage into other channels more effectively.

## 2.4. “Quality is no compromise” policy at CP-Meiji

Aligning with the core values of both CP and Meiji, CP-Meiji will not compromise quality under any circumstances. We would like to highlight three main actions:

1. **Up-stream to down-stream reassurance:** We consider the quality of the supply chain distribution (logistics, warehouse management, infrastructure at retailer) before expanding into any new customer inside or outside Thailand. All this is to repay consumers for their trust in the CP-Meiji brand.

2. **100% product traceability:** Every bottle and carton of CP-Meiji milk is traceable via the database at CP-Meiji that tracks all information regarding the product. The product is monitored from the farm and throughout the whole supply chain process.
3. **100% quality control check:** At CP-Meiji, we do not do sample check, we check every batch of our product and record all information in our database. This ensures that every bottle is subject to the same highest standard.

## 2.5. Yoghurt trend

Lastly, I would like to share an interesting new emerging market segment in Thailand, the yoghurt segment, which grew by double digits every year for the previous 5 years (except in 2011 when Thailand was hit by a massive flood). CP-Meiji launched Bulgaria Yoghurt in August, 2013. Market feedback was overwhelming, despite the first launch being in only one key customer (7-Eleven). Meiji Bulgaria Yoghurt currently sits in the “Top 3 All Time Best Selling Item” in 7-Eleven Thailand.

## Yoghurt sales (MB) - Total Thailand

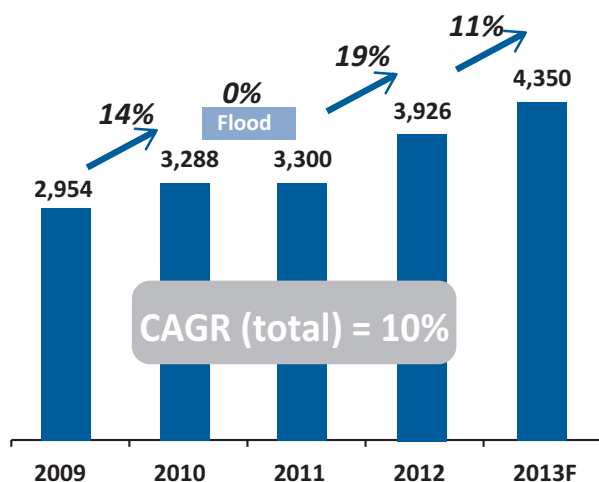


Table 7. Total Thailand yoghurt sales (Source: Nielsen Retail Index)

## Appendix

**meiji**  
 cp-meiji

	Value			% Growth	
	2011	2012	2013e	12/11	13/12
Milk production (ton) *	985,000	1,064,000	1,125,000	8.0%	5.7%
Milk delivered to dairies ***	591,000	638,400	675,000	8.0%	5.7%
The number of dairy farms **	20,458	20,486	21,023	0.1%	2.6%
The number of milking cows *	555,458	573,963	589,020	3.3%	2.6%
Milk consumption per capita ***	11.71	12.38	12.81	5.7%	3.4%
The number of dairy plants (Major Player) ***	10				
The distribution map of dairy farm areas *					
North					11%
North-East					20%
Central					68%
South					1%

Note : Milk Consumption

\* Dairy Products covers Milk, Soft Yoghurt, Cultured Yoghurt, excluding Soy milk

\*\*Data not include dairy product consumption via Food Services and Traditional Trade channel

Source :

\* Office of Agriculture Economics

\*\* Department of Livestock Development

\*\*\* CP-Meiji analysis based on data from Nielsen and National Statistics Office (NSO) Thailand

# 3 Metabolomics-based hydrophilic component profiling of cheeses using GC/MS

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## ABSTRACT

Exhaustive profiling of metabolites by metabolomics is a promising tool for qualitative evaluation of cheese. In the present study, gas chromatography/mass spectrometry (GC/MS) was used to analyze hydrophilic low molecular weight components in cheese. Partial least squares regression models were constructed to predict the relationship between the metabolite profile and two sensory attributes related to maturation, “rich flavor” and “sour flavor”. Compounds that played an important role in construction of each sensory prediction model were identified. Additionally, we developed a methodology combining metabolic profiling with multiple appropriate multivariate analyses, and verified it on an industrial scale for the ripening of Cheddar cheese. This method uses hydrophilic low molecular weight compound profiling by GC/MS and could be applied to determination of optimal conditions and quality monitoring for cheese ripening.

## 1. INTRODUCTION

Metabolomics is the comprehensive analytical study of all the metabolites formed in a given microorganism, plant, or animal. Using metabolomics, large numbers of metabolites can be analyzed to investigate the phenotype in high resolution and monitor metabolic fluctuations. Metabolomics is an effective post-genomic research tool that has been applied in many areas, including the study of human diseases, nutrition, drug discovery, and plant physiology [1-5]. Metabolomics is an extremely effective tool

for the analysis of biological metabolisms and for the analysis of food science processes. Therefore, its application to monitoring of the quality, processing, and safety of both raw materials and final products in food science has recently attracted attention [6-11].

Cheese is a popular milk product worldwide. It is made by adding rennet and lactic acid bacteria starter to milk, and then following a multi-step manufacturing process. During this process, and especially during ripening, biochemical changes that occur in the cheese produce a unique flavor, aroma and texture [12]. A number of factors, including the ingredients (added salts, enzymes and lactic acid bacteria), and the degradation and catabolism that occur during ripening, result in considerable diversity in the compounds present in the cheese and determine the “cheese quality”. It is difficult to manage cheese quality using conventional approaches that focus on one or several target compounds, and metabolomics is a promising tool for qualitative evaluation of the cheese.

Many studies have identified and quantified the odorous volatile compounds in cheese. Recent studies have focused on hydrophilic compounds that contribute to the taste of the cheese, such as the water-soluble extracts in Cheddar cheese [13], Gouda cheese [14], Swiss cheese [15,16], Comte [17] and goat’s milk cheese [18]. The application of metabolomics to hydrophilic compounds in cheese represents a major advance towards understanding cheese sensory quality in terms of the components of the metabolome. This study focused mainly on Cheddar cheese and Gouda



cheese, which are the most popular cheeses in Japan, and conducted metabolomics-based hydrophilic low molecular weight component profiling of these cheeses using GC/MS. Two applications of metabolic profiling of cheese, sensory predictive modeling of cheese [19] and monitoring of the ripening process of cheese [20], are described in this report.

## 2. SENSORY PREDICTIVE MODELING OF CHEESE

### 2.1. Relationship between the metabolome and flavor attributes in natural cheeses

Metabolic profiling of hydrophilic low molecular weight components in twelve natural cheeses, including six Cheddar cheeses and six Gouda cheeses, were conducted using GC/MS. Derivatization (methoxylation and silylation) of target analytes was conducted before GC/MS analysis. Quantitative descriptive analysis (QDA) [21] was performed to evaluate the cheese samples. Partial least squares (PLS) regression models were constructed to predict the relationship between the metabolite profile and two sensory attributes, “rich flavor” and “sour flavor”. The characteristics of these sensory attributes were “thick and rich, including umami taste and soy sauce-like flavor” for “rich flavor”, and “fundamental taste, typical of lactic and citric acids” for “sour flavor”. Example figures of predicted values versus measured values are shown for the “rich flavor” (Figure 1A) and “sour flavor” (Figure 1B), with the calibration set and the prediction set. The obtained sensory predictive model was interpreted using both diagnostic indices of model quality,  $R^2$  and  $Q^2$ , the root mean square error of estimation and the root mean square error of the prediction, respectively.  $R^2$  and  $Q^2$  denoted goodness of fit and predictability estimated by cross-validation, respectively. The root mean square error of the estimation is an index from internal validation that is used to evaluate the accuracy and precision of the model, while the root mean square error of the prediction is an index from external validation that is used to evaluate model robustness. The results from these diagnostic indices showed that a highly reliable model was obtained for the two sensory attributes “rich flavor” and “sour flavor”

by hydrophilic low molecular weight component analysis using GC/MS. Additionally, we successfully reconstructed the precise sensory predictive model with metabolic fingerprinting using gas chromatography with a flame ionization detector. This instrumentation is a relatively inexpensive instrument compared with GC/MS, is easy to maintain and operate, and is a valid alternative to GC/MS when metabolomics is applied in a practical setting as a novel quality evaluation tool for manufacturing processes or final products [22].



Fig. 1 A. Sensory predictive model for “rich flavor”

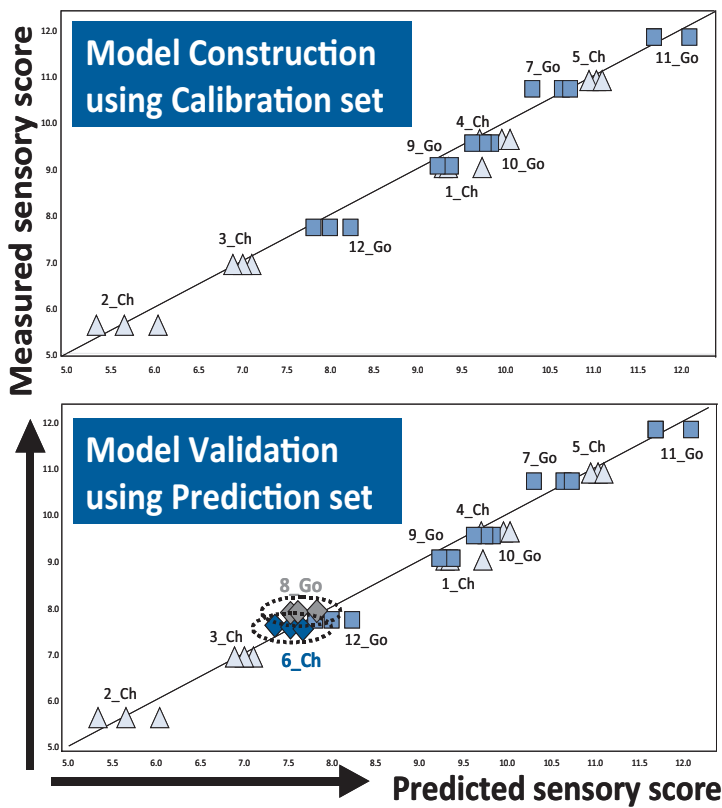


Fig. 1 B : Sensory predictive model for “sour flavor”

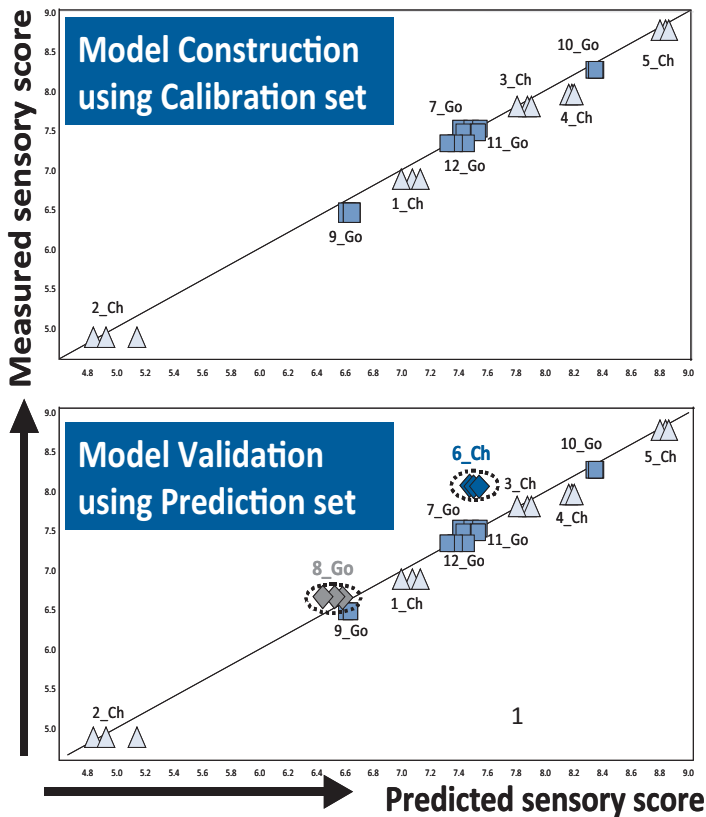


Figure 1: Relationships between predicted and measured QDA scores based on the PLS model for (A) “rich flavor” and (B) “sour flavor”. Calibration set: five Cheddar cheeses (triangles, 1\_Ch, 2\_Ch, 3\_Ch, 4\_Ch, and 5\_Ch) and five Gouda cheeses (squares, 7\_Go, 9\_Go, 10\_Go, 11\_Go, and 12\_Go). Prediction set (diamonds): Cheddar cheese (6\_Ch) and Gouda cheese (8\_Go)

## 2.2. Analysis of flavor components in cheese

The metabolites that contributed significantly to the PLS prediction model for sensory evaluation were identified from variable importance for the projection values. This is an index computed from the influence of every *x*-term (metabolite) on a *y*-variable (sensory attribute) in the model. The important compounds in the sensory prediction model were identified as 12 amino acids and lactose for the “rich flavor”, and 4-aminobutyric

acid, ornithine, succinic acid, lactic acid, proline and lactose for the “sour flavor”. For the “rich flavor”, umami-tasting amino acids, bitter-tasting amino acids, and sweet-tasting amino acids were all found, and both positive and negative contributors to the “sour flavor” were detected. The coexistence of these compounds provides complexity and a sensation of ripeness to the cheese flavor. From these results, we can determine how metabolites contribute to specific flavors, and those metabolites that act as markers of cheese flavor for design and management of cheese quality.

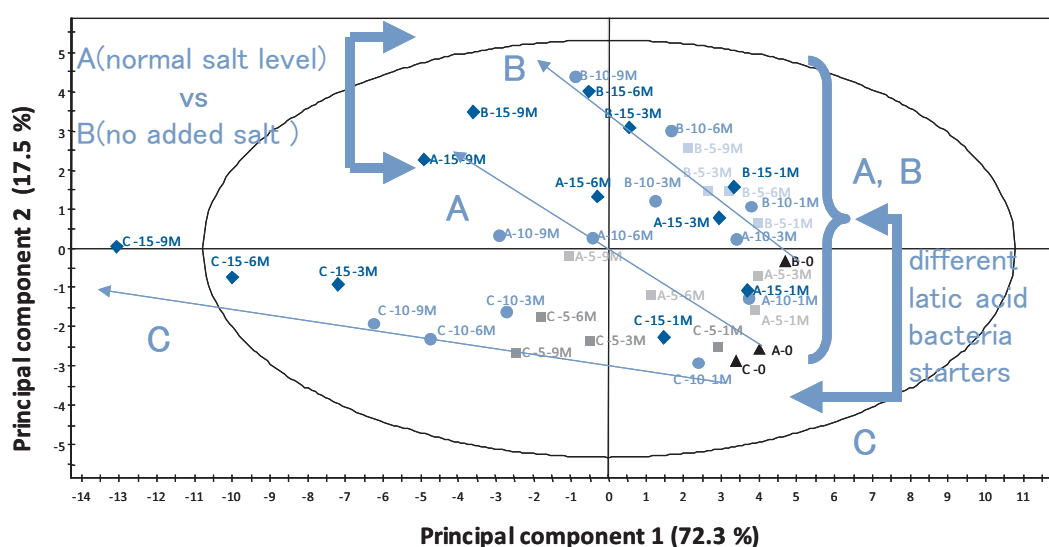
## 3. MONITORING THE RIPENING PROCESS OF CHEESE

### 3.1. Metabolic profiling of three types of cheddar cheese

Three types of Cheddar cheese were manufactured by common protocols [23] and coded as A, B, and C. Cheeses A and B were produced using the same lactic acid bacteria starter, but while cheese A contained added salt, cheese B did not. Cheese C contained added salt and was produced using a different lactic acid bacteria starter to cheeses A and B. Thirteen samples, consisting of one sample taken before ripening and 12 samples taken after ripening under different temperature and time conditions, were collected for each cheese (A, B, and C). The cheeses were ripened at 5, 10, or 15°C, and for 1, 3, 6, or 9 months. All samples were analyzed using GC-MS, and 190 peaks, including amino

acids, sugars, organic acids, and inorganic acids, were observed. Among these, 55 compounds were identified or partially identified (compound class identified).

The data from these 55 peaks were summarized and assessed with PCA. The PCA results (score plot) of these data are shown in Figure 2. As shown by the arrows in Figure 2, for all the cheese types (A, B, and C) the metabolic profile changed and diverged as maturation proceeded. Therefore, changes in the levels of different compounds present in cheese will vary depending on the manufacturing conditions, including the ingredients used.

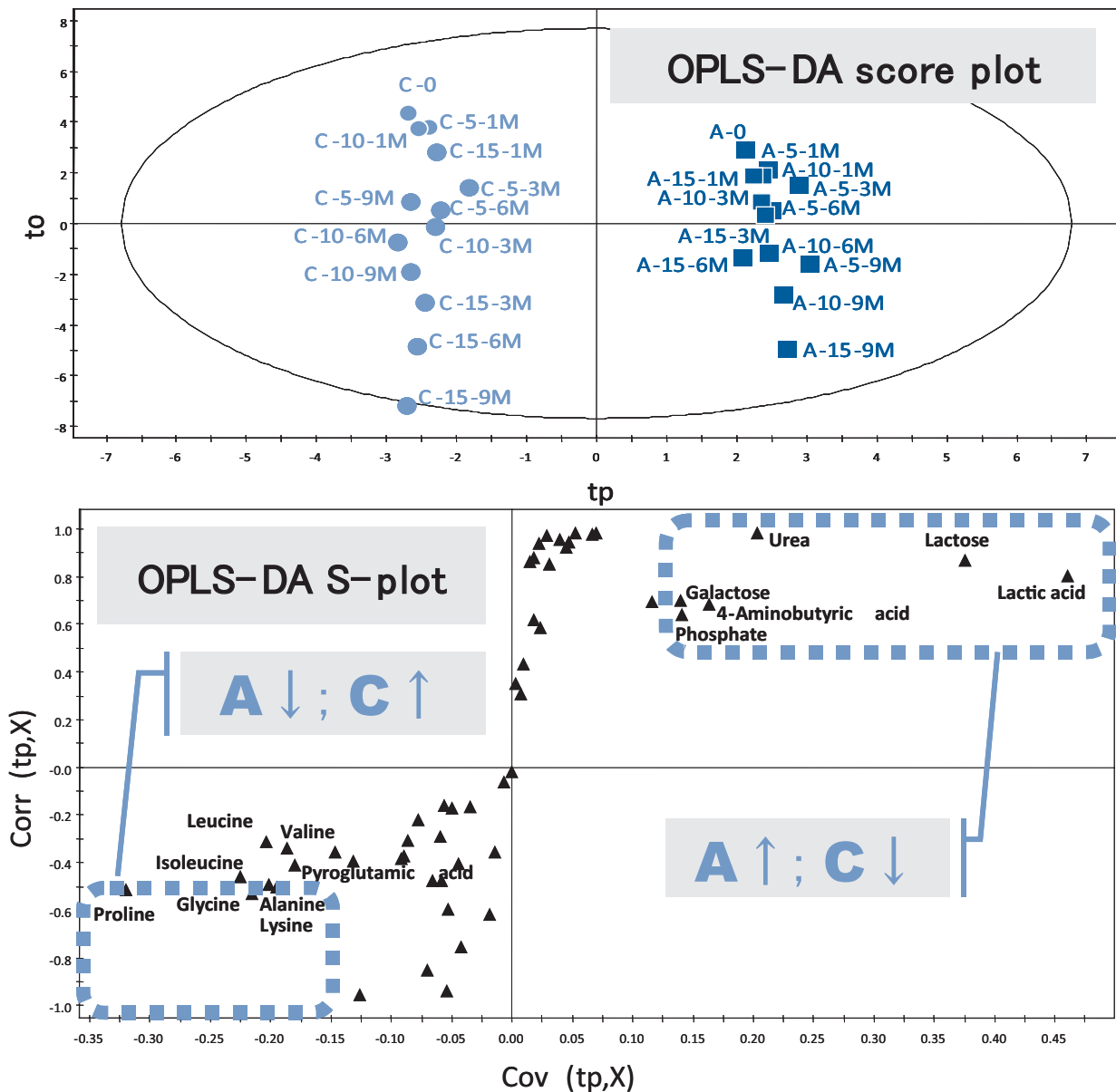


**Figure 2:** Principal components analysis score plots for all samples based on the GC/MS metabolome. Measurements A-0, B-0, and C-0 were taken before ripening samples of cheeses A, B, and C, respectively. Ripening conditions are provided in the sample name; for example, A-5-1M means cheese A ripened at 5°C for 1 month

### 3.2. Characteristic metabolites in the cheese types

Differences among cheeses A, B, and C were analyzed by statistical discriminant analysis using orthogonal PLS discriminant analysis (OPLS-DA) [24]. The effect of the kind of lactic acid bacteria starter on the metabolic profile was investigated by comparing cheeses A and C, which both contained added salt but were produced with different lactic acid bacteria starters. Figure 3 shows the score plot and S-plot of OPLS-DA for this comparison. Lactic acid, lactose, urea,

4-aminobutyric acid, galactose, and phosphate were found in greater amounts in cheese A than cheese C. Proline, glycine, alanine, and lysine were found in greater amounts in cheese C than in cheese A. The combination of metabolic profiling and OPLS-DA provided quantitative analysis of the ripening process by identifying characteristic compounds and how the levels of these changed during maturation.

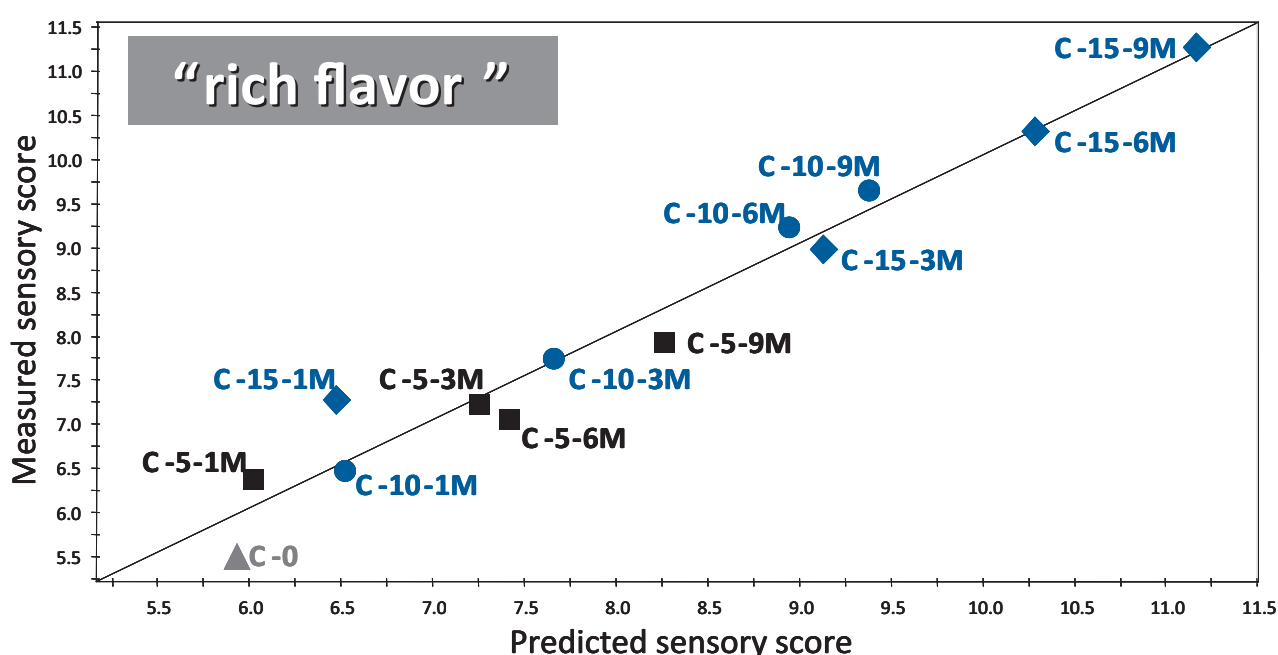


**Figure 3:** Score plots and S-plots for orthogonal partial least squares-discriminant analysis models. Cheddar cheese sample A versus Cheddar cheese sample C. A-0 and C-0 are pre-matured samples of cheeses A and C, respectively. Ripening conditions are provided in the sample name; for example, C-5-1M means cheese C ripened at 5°C for 1 month. The *dotted squares* in the S-plot show the threshold zone, representing beyond |0.5| in the vertical axis, and beyond |0.15| in the horizontal axis. *to* identical orthogonal score (the intraclass variation); *tp* predictive score (the interclass variation); *Corr* correlation; *Cov* covariate

### 3.3. Relationship between the metabolic profile and sensory attributes

The relationship between changes in the metabolic profile and the sensory characteristics of the cheese was evaluated to assign a sensory evaluation score to an index for determining ripening conditions and monitoring the ripening process. The relationship between the ripening conditions and sensory scores was investigated by sensory predictive modeling using the metabolic profiles over time for cheese C. Figure 4 shows the predicted sensory score from metabolome analysis plotted against the measured sensory score for

the “rich flavor”. This model showed an excellent fit. In the case of metabolome changing with ripening, the relationship between the metabolite profile and sensory attributes was modeled. When the cheese ripening process is monitored using this model, cheese quality is evaluated in terms of both the metabolite profile and sensory score by predicting the intensity of the “rich flavor”. This procedure can be conducted during quality inspection of the manufacturing process and of the finished products.



**Figure 4:** Relationships between predicted and measured quantitative descriptive sensory analysis (QDA) scores for “rich flavor” based on the PLS regression model for cheese C samples. C-0 is a pre-matured sample of cheese C. Ripening conditions are provided in the sample name; for example, C-5-1M means cheese C ripened at 5°C for 1 month

### 3.4. Selection of marker compounds for monitoring the cheese ripening process

The PCA for metabolome data from cheese C during ripening was performed to select marker compounds for monitoring cheese ripening. The score plot and loading plot are shown in Figure 5. The PCA loading plot (Figure 5b) shows that the amino acids proline, leucine, valine, isoleucine, pyroglutamic acid, alanine, glutamic acid, glycine, lysine, tyrosine, serine, phenylalanine, methionine, aspartic acid, and ornithine contributed to the positive value on the PC1 axis. Increases in the peak intensities of proline and ornithine, which

were two of the marker candidate compounds, during ripening at different temperatures are shown in Figure 6. Peak intensities (y-axis) increased as ripening progressed. Additionally, a higher ripening temperature increased the peak intensities for ripening periods of the same duration. From these results, the compounds, mainly the amino acids mentioned above, were identified as relevant markers for monitoring the ripening process.

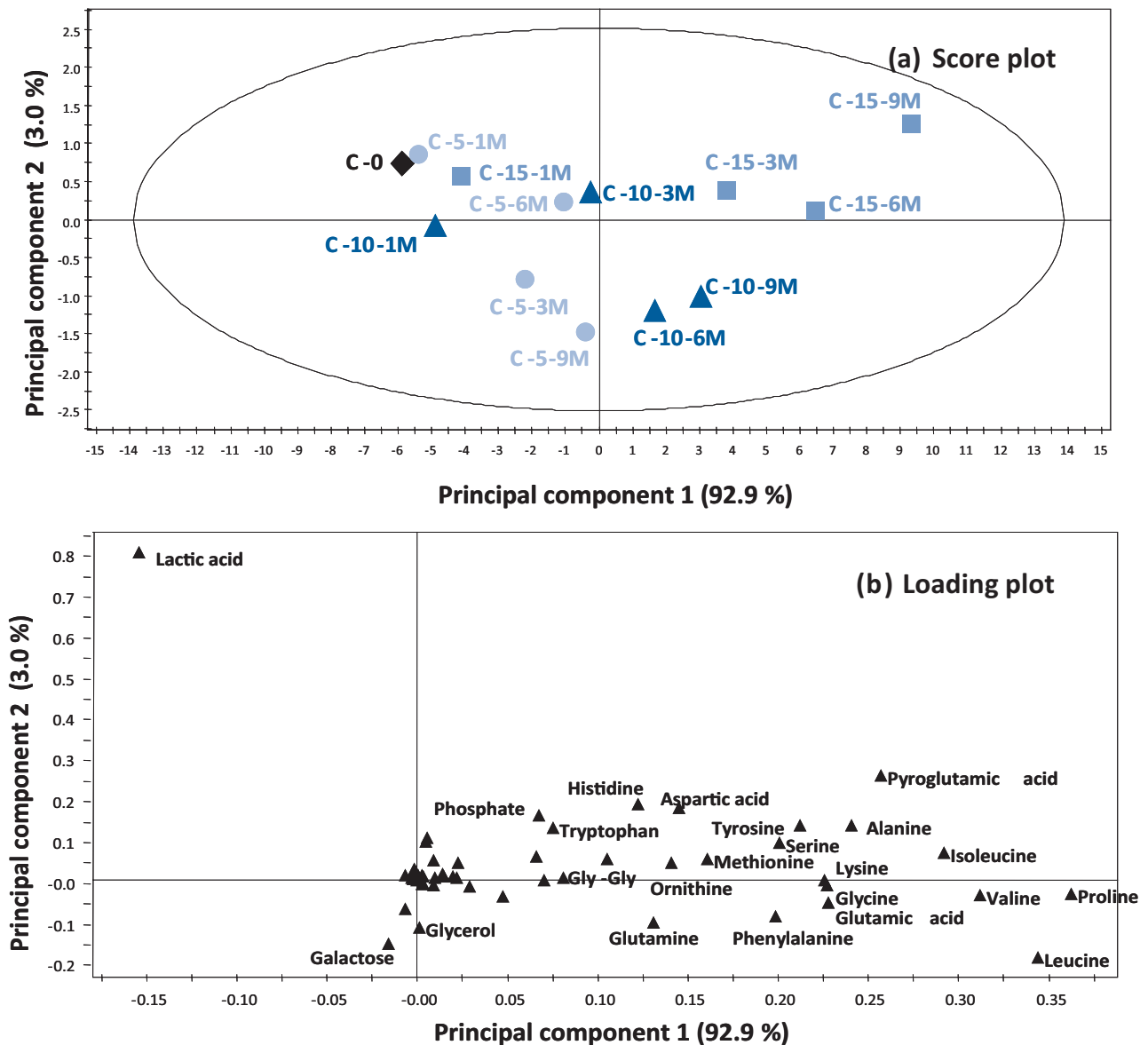


Figure 5: Principal components analysis plots based on the GC-MS metabolome. (a) Score plot and (b) loading plot for Cheddar cheese sample C. C-0M is a pre-matured sample of cheese C. Ripening conditions are provided in the sample name; for example, C-5-1M means cheese C ripened at 5°C for 1 month

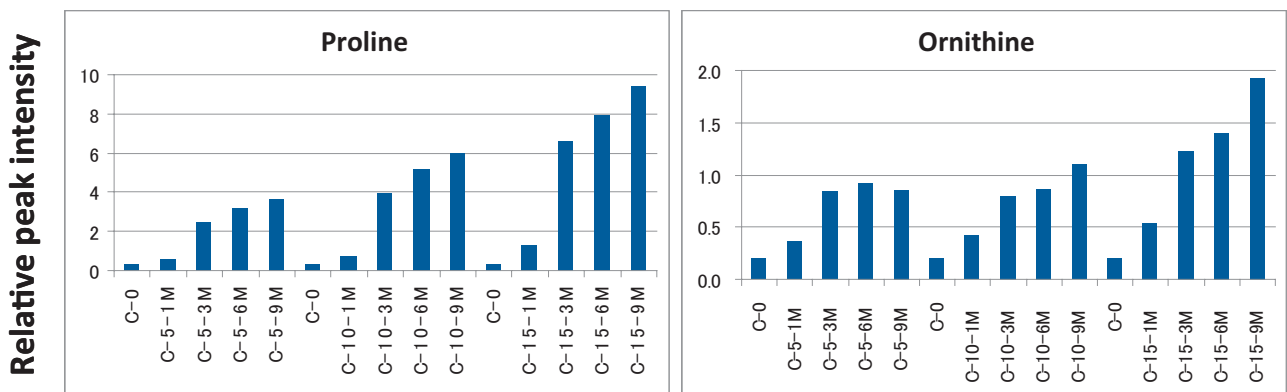


Figure 6: Changes in the peak intensities of two marker candidates, proline and ornithine, in the ripening process. C-0 is a pre-matured sample of cheese C. Ripening conditions are provided in the sample name; for example, C-5-1M means cheese C ripened at 5°C for 1 month

## 4. CONCLUSION

Metabolomic studies of hydrophilic low molecular weight components in natural cheeses were conducted using GC/MS. We obtained good prediction models for “rich flavor” and “sour flavor” sensory characteristics. The compounds that contributed to the prediction from the model were extracted. We then developed a method that combined metabolic profiling with multiple appropriate multivariate analyses, and verified it on the industrial scale for the ripening of Cheddar cheese. This method allows practical application of the profiling hydrophilic low molecular weight compounds by GC/MS to determination of optimal conditions for cheese ripening and monitoring of cheese quality.

As an extension of this work, an extremely valuable database of metabolomic analysis data with R&D and/or manufacturing data will be constructed and utilized. This will allow us to conduct numerical simulations in the R&D and manufacturing of cheese, which reduce the cost and time required for R&D and improve the accuracy of manufacturing in reaching quality targets. It is expected that metabolomics will be applied to other food products in the future.

## 5. ACKNOWLEDGEMENTS

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# 4 Variability of volatile organic compounds during the manufacture of Swiss-type cheese using selected ion flow tube mass spectrometry

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## ABSTRACT

The determination of the volatile organic compound (VOC) profiles in Swiss cheese samples from four different stages of manufacture was achieved using static headspace sampling with selected ion flow tube-mass spectrometry (SIFT-MS). Ten cheese samples from two different batches of Swiss cheese manufactured from a single factory were obtained. The stages included: out-of-press, end of pre-cool, end of warm room and at time of cutting. Multivariate stage-level classification using SIMCA showed significant discrimination of VOC profiles among the four manufacturing stages. This suggests a varied VOC profile/behavior at each stage of manufacture. The VOC profile of samples from the end of warm room stage was highly discriminatory, relative to other stages. Key discriminating VOCs and the highest VOC concentration at each stage were identified. Significant variability of VOC profiles among the five vats within the same make-date and stage of manufacture was also observed. There was an increased inhomogeneity in VOC profiles between vats towards the final stages of manufacture. Variabilities were most probably related to varied fermentation activities, changes in microflora, biochemical factors, and physical and environmental conditions. These findings provide in-depth knowledge on the biochemistry and microbiology associated with VOC production. These would be beneficial, especially for effective/efficient control in bringing consistency in texture, flavor and aroma of Swiss cheese, and could

potentially shed light towards understanding and potentially minimizing split defect formation in Swiss cheese.

**Keywords:** Volatile organic compound, VOC, Swiss cheese, SIFT-MS

## 1. INTRODUCTION

The flavor and quality perception of cheese is very highly dependent on microbial fermentation of milk constituents and cheese body. The aroma and flavor development in cheeses, as the ripening process progresses, involve a very complex blend and balance of microbiological and biochemical changes to the curd, leading to the characteristic flavor and texture of a particular variety (Langsrud and Reinbold 1973; McSweeney 2004).

The flavor of Swiss cheese is widely described as being nutty, mild, and slightly sweet, with its flavor becoming more aromatic as the cheese ages (Fröhlich-Wyder and Bachmann 2004; Steffen and others 1993). The procedures for the manufacture of Swiss cheese, relative to the traditional Emmentaler, have evolved over the years. These include treatment of milk, extent of mechanization, microorganisms used, cooking temperature, weight, shape, ripening time and shelf life (Steffen and others 1993). Because of these reasons, although not exclusively, the texture, flavor and aroma of the resulting Swiss cheese products could possibly differ for different cheese manufacturers. Previous work has shown

significant differences in Swiss cheese samples obtained at the time of packaging from different factories (Taylor and others 2013; Castada and others 2013).

On a molecular perspective, the flavor and aroma in cheese can be characterized using its volatile organic compound (VOC) composition. VOC analyses have been proven as an effective tool in the elucidation of flavor profiles, which provide valuable information on the organoleptic qualities of food products (Harper and others 2011; Xu and Barringer 2010a, b; Fleming-Jones and Smith 2003; Spanel and Smith 1999). However, one of the challenges in the analysis of VOCs is their isolation, identification and subsequent quantitation. The main reasons for this difficulty include the elusive character of VOCs, their presence in minute amounts and their very unstable nature (Harper and others 2011; Grab 2004).

The use of selected ion flow tube mass spectrometry (SIFT-MS) has proven to be useful in the analysis of headspace (H/S) volatile compounds in various research studies across different disciplines (Taylor and others 2013; Wampler and Barringer 2012; Harper and others 2011; Ozcan and Barringer 2011; Azcarate and Barringer 2010; Xu and Barringer 2010a,b; Xu and Barringer 2009; Spanel and Smith 2007; Allardyce and others 2006; Smith and Spanel 2005; Wilson and others 2001; Wilson and others 2002; Spanel and Smith 1999; Spanel and Smith 1996).

SIFT-MS has been described in detail elsewhere (Spanel and Smith 1996; Smith and Spanel 1999; Smith and Spanel 2005). Briefly, SIFT-MS utilizes a soft chemical ionization technique to instantly quantify compounds in whole air samples. Such technique produces a relatively cleaner mass spectrum, thereby minimizing interferences and conflicting ion product issues. The reagent ions are produced when water vapors are drawn through a microwave discharge generating a mixture of ions. These ions are focused via a series of lenses in an upstream quadrupole which are filtered by their mass-to-charge ratio, resulting in a pure ion beam containing only selected precursor ions ( $\text{H}_3\text{O}^+$ ,  $\text{NO}^+$  and  $\text{O}_2^+$ ). This ion beam is injected into the flow-tube through a venturi along with helium as a carrier gas where the sample mixture is subsequently introduced. Controlled chemical reactions occur between the sample compounds

and the selected precursor ions within the flow-tube, where fixed reactions result in the formation of product ions, which are then focused into a second quadrupole. The downstream quadrupole selects individual product ions, based on their mass-to-charge ratio, and directs them toward the detector. The detector/particle multiplier counts the product ions to create a mass spectrum. By interpreting the mass spectrum, the sample composition is determined and each compound is quantified (Syft Technologies 2011; Smith and Spanel 1999).

The main objective of this research was to determine the VOC profile of Swiss cheese samples from each major stage of manufacture using SIFT-MS. Subsequently, the key discriminating VOCs at each stage was determined and the behavior of individual VOCs in each stage was monitored. Such behavior can be related to the biochemical origin of each VOC. Through this approach, a collection of VOC profiles could be made that can be used as a reference for (i) monitoring VOCs in Swiss cheese making; (ii) calculating odor activity values (OAVs) for building aroma profiles; (iii) fundamental understanding of the chemistry and microbiology of VOC production in cheese; (iv) studying the biochemical/biophysical behavior of VOCs to explain and control the flavor/aroma heterogeneity in Swiss cheese.

## 2. METHODOLOGY

### 2.1. Cheese sampling

The Swiss cheese samples used in this study were provided by a cheese manufacturing company in the United States. Two batches of cheese samples were obtained from each of the four stages of manufacture namely: out-of-press, end of pre-cool, end of warm room and at time of cutting and packaging. In each batch, five cheese samples from five different vats were obtained, giving a total of ten cheese samples per stage of manufacture.

### 2.2. Sample preparation and headspace VOC analysis

Homogeneous representative sub-sampling of Swiss cheese for succeeding headspace VOC

analysis was ensured via coning and quartering of the shredded cheese sample. If not immediately utilized, shredded cheese samples were vacuum packed and stored at freezing temperatures ( $-20\pm 1^{\circ}\text{C}$ ). Five grams of shredded sample were weighed into a 500 ml Schott bottle and capped with septum-lined screw cap. Headspace equilibration was allowed by incubating the bottle in a  $40^{\circ}\text{C}$  ( $\pm 1^{\circ}\text{C}$ ) water bath for 1 h. Immediately after incubation, headspace sampling was done by inserting the SIFT-MS passivated sampling needle into the septum of the bottle. A second, open-ended needle was also inserted into the septum to allow pressure stabilization during sampling. An empty bottle was scanned between samples to act as blank and to zero the instrument before running the next sample.

### 2.3. Instrumentation

A Voice200™ SIFT-MS (Syft Technologies Ltd., Christchurch, New Zealand) was used for the instantaneous identification and quantification of VOCs in the sample headspace down to the low parts-per-trillion (ppt) level. The V200 is capable of detecting VOCs with characteristic boiling point (BP) of less than  $392^{\circ}\text{F}$  ( $200^{\circ}\text{C}$ ) and molar mass range of 10–300 Da. It is sensitive down to a unit mass resolution throughout the mass range and with a detection limit of 50 ppt by volume (pptv) above its measuring range and linearity detection of 100 pptv level to 0.1% level. (Syft Technologies 2011). The V200 was operated using selected ion mode (SIM) in quantifying the specific compounds of interest. SIM scan limits the number of masses counted per scan, thus allowing longer detection time per mass and delivers higher quantitative precision than the full mass scan (FMS) mode. FMS mode was also run during the analysis for spectral referencing and assists in the identification of masses in the headspace sampling, especially in cases of resolving product ion conflicts. Table 1 lists the 28 compounds used in the SIM scan method. These compounds, in general, are considered as high impact compounds based on their concentration being higher than their threshold value (Taylor and others 2013; Castada and others 2013). This table also lists the reaction products, rate coefficients for each reagent ion and potential interferences with their corresponding resolutions.

### 2.4. Statistical analysis

Data fitting and analysis of least square (LS) means of the VOC concentrations were carried out using the PROC MIXED of Statistical Analysis System (SAS, v9.3) (Cary, NC: SAS® Institute Inc.). The randomized complete blocked design (RCBD) model included the fixed effects of factories (3 degrees of freedom, DF) and segment (2 DF) and the random effects of cheese, cheese-x-factory and residual error. The differentiation of split and non-split Swiss cheese samples using VOCs was performed using soft independent modeling of class analogy (SIMCA) by Pirouette®, v3.11 (Bothell, WA: Infometrix Inc). SIMCA is a comprehensive chemometric modeling software with a supervised classification technique that builds a distinct confidence region around each class after applying principal component analysis (PCA) (Lavine 2006). PCA is a visualization tool providing a technique to reduce the dimensionality of multivariate data sets. The concept of variation is central to PCA where it finds linear combinations of the original independent variables, which account for maximal amounts of variation (Multivariate data...2008).

No	Compound	Reagent	Reaction rate, k (cm <sup>3</sup> /s)	Branching ratio (%)	Mass-to-charge ratio (m/z)	Product	Conflict and resolution
1	(E)-2-nonenal	H <sub>3</sub> O <sup>+</sup>	4.8x10 <sup>-9</sup>	100	141	C <sub>9</sub> H <sub>17</sub> O <sup>+</sup>	None
		H <sub>3</sub> O <sup>+</sup>			159	C <sub>9</sub> H <sub>17</sub> O <sup>+</sup> • H <sub>2</sub> O	None
		NO <sup>+</sup>	3.8x10 <sup>-9</sup>	80	139	C <sub>9</sub> H <sub>15</sub> O <sup>+</sup>	None
		NO <sup>+</sup>			20	C <sub>9</sub> H <sub>15</sub> O <sup>+</sup> • NO <sup>+</sup>	None
		O <sub>2</sub> <sup>+</sup>	3.7x10 <sup>-9</sup>	18	83	C <sub>5</sub> H <sub>7</sub> O <sup>+</sup>	None
					84	C <sub>5</sub> H <sub>8</sub> O <sup>+</sup>	None
					96	C <sub>7</sub> H <sub>12</sub> <sup>+</sup>	None
2	2,3-butanedione	H <sub>3</sub> O <sup>+</sup>	1.7x10 <sup>-9</sup>	100	87	C <sub>4</sub> H <sub>7</sub> O <sub>2</sub> <sup>+</sup>	*Potential conflict with 3-methylbutanal; 87 m/z for 3-methylbutanal is not scanned (has lower BR of 70%)
		NO <sup>+</sup>	1.3x10 <sup>-9</sup>	35	43	C <sub>2</sub> H <sub>3</sub> O <sup>+</sup>	None
		NO <sup>+</sup>		65	86	C <sub>4</sub> H <sub>6</sub> O <sub>2</sub> <sup>+</sup>	None
		H <sub>3</sub> O <sup>+</sup>	3.6x10 <sup>-9</sup>	30	65	C <sub>5</sub> H <sub>6</sub> <sup>+</sup>	None
3	2-methylpropanal	O <sub>2</sub> <sup>+</sup>	3.0x10 <sup>-9</sup>	70	72	C <sub>4</sub> H <sub>8</sub> O <sup>+</sup>	*Potential conflict with butanal; 72 m/z for butanal is not calculated (has lower BR of 35%)
4	3-methylbutanal	NO <sup>+</sup>	3.0x10 <sup>-9</sup>	100	85	C <sub>5</sub> H <sub>9</sub> O <sup>+</sup>	*Potential conflict with 3-methylbutanoic acid; 85 m/z for 3-methylbutanoic acid is not calculated (has lower BR of 30%)
		H <sub>3</sub> O <sup>+</sup>				3.6x10 <sup>-9</sup>	30
5	3-methylbutanoic acid	H <sub>3</sub> O <sup>+</sup>	3.0x10 <sup>-9</sup>	95	103	C <sub>5</sub> H <sub>11</sub> O <sub>2</sub> <sup>+</sup>	None
		NO <sup>+</sup>	2.5x10 <sup>-9</sup>	70	132	C <sub>5</sub> H <sub>10</sub> O <sub>2</sub> <sup>+</sup> • NO <sup>+</sup>	None
6	3-methylindole	H <sub>3</sub> O <sup>+</sup>	3.3x10 <sup>-9</sup>	100	132	C <sub>9</sub> H <sub>10</sub> N <sup>+</sup>	None
		NO <sup>+</sup>	2.5x10 <sup>-9</sup>	100	131	C <sub>9</sub> H <sub>10</sub> N <sup>+</sup>	None
		O <sub>2</sub> <sup>+</sup>	2.4x10 <sup>-9</sup>	20	130	C <sub>9</sub> H <sub>10</sub> N <sup>+</sup>	None
				75	131	C <sub>9</sub> H <sub>10</sub> N <sup>+</sup>	None
7	Acetic acid	H <sub>3</sub> O <sup>+</sup>	2.6x10 <sup>-9</sup>	100	61	CH <sub>3</sub> COOH <sub>2</sub> <sup>+</sup>	None
					79	CH <sub>3</sub> COOH <sub>2</sub> <sup>+</sup> • H <sub>2</sub> O	None
					97	CH <sub>3</sub> COOH <sub>2</sub> <sup>+</sup> • 2H <sub>2</sub> O	None
8	Ammonia	H <sub>3</sub> O <sup>+</sup>	2.6x10 <sup>-9</sup>	100	18	NH <sub>4</sub> <sup>+</sup>	None
					36	NH <sub>4</sub> <sup>+</sup> • H <sub>2</sub> O	None
		O <sub>2</sub> <sup>+</sup>	2.6x10 <sup>-9</sup>	100	17	NH <sub>3</sub> <sup>+</sup>	None

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No	Compound	Reagent	Reaction rate, k (cm <sup>3</sup> /s)	Branching ratio (%)	Mass-to-charge ratio (m/z)	Product	Conflict and resolution
9	butanal	NO <sup>+</sup>	3.5x10 <sup>-9</sup>	100	71	C <sub>4</sub> H <sub>7</sub> O <sup>+</sup>	*Potential conflict with 2-methylpropanal; 71 m/z is not calculated for 2-methylpropanal (has lower reaction rate of 3.1x10 <sup>-9</sup> )
							*Potential conflict with butanoic acid; 71 m/z is not calculated for butanoic acid (has lower reaction rate of 1.9x10 <sup>-9</sup> )
							*Potential conflict with ethyl butanoate; 71 m/z is not calculated for butanoic acid (has lower reaction rate of 1.9x10 <sup>-9</sup> )
10	butanoic acid	H <sub>3</sub> O <sup>+</sup>	2.9x10 <sup>-9</sup>	90	89	C <sub>3</sub> H <sub>7</sub> COOH <sub>2</sub> <sup>+</sup>	*Potential conflict with methionol; 89 m/z for methionol is not calculated (has lower BR of 30%)
					107	C <sub>3</sub> H <sub>7</sub> COOH <sub>2</sub> <sup>+</sup> • 2H <sub>2</sub> O	None
		NO <sup>+</sup>	1.9x10 <sup>-9</sup>	50	118	NO <sup>+</sup> • C <sub>3</sub> H <sub>7</sub> COOH	None
11	diethyl sulfide	O <sub>2</sub> <sup>+</sup>	2.5x10 <sup>-9</sup>	30	75	C <sub>3</sub> H <sub>7</sub> S <sup>+</sup>	None
			2.5x10 <sup>-9</sup>	35	90	C <sub>4</sub> H <sub>10</sub> S <sup>+</sup>	None
12	dimethyl disulfide	NO <sup>+</sup>	2.4x10 <sup>-9</sup>	100	94	(CH <sub>3</sub> ) <sub>2</sub> S <sub>2</sub> <sup>+</sup>	None
		O <sub>2</sub> <sup>+</sup>	2.3x10 <sup>-9</sup>	80	94	(CH <sub>3</sub> ) <sub>2</sub> S <sub>2</sub> <sup>+</sup>	None
13	dimethyl sulfide	H <sub>3</sub> O <sup>+</sup>	2.5x10 <sup>-9</sup>	100	63	(CH <sub>3</sub> ) <sub>2</sub> S.H <sup>+</sup>	None
		NO <sup>+</sup>	2.2x10 <sup>-9</sup>	100	62	(CH <sub>3</sub> ) <sub>2</sub> S <sup>+</sup>	None
		O <sub>2</sub> <sup>+</sup>	2.2x10 <sup>-9</sup>	25	47	CH <sub>3</sub> S <sup>+</sup>	None
14	dimethyl trisulfide	H <sub>3</sub> O <sup>+</sup>	2.8x10 <sup>-9</sup>	100	127	C <sub>2</sub> H <sub>6</sub> S <sub>3</sub> H <sup>+</sup>	None
		NO <sup>+</sup>	1.9x10 <sup>-9</sup>	100	126	C <sub>2</sub> H <sub>6</sub> S <sub>3</sub> <sup>+</sup>	None
		O <sub>2</sub> <sup>+</sup>	2.2x10 <sup>-9</sup>	15	111	CH <sub>3</sub> S <sub>3</sub> <sup>+</sup>	None
		O <sub>2</sub> <sup>+</sup>	2.2x10 <sup>-9</sup>	45	126	C <sub>2</sub> H <sub>6</sub> S <sub>3</sub> <sup>+</sup>	None
15	ethanol	H <sub>3</sub> O <sup>+</sup>	2.7x10 <sup>-9</sup>	100	47	C <sub>2</sub> H <sub>7</sub> O <sup>+</sup>	None
			2.7x10 <sup>-9</sup>		65	C <sub>2</sub> H <sub>7</sub> O <sup>+</sup> • H <sub>2</sub> O	None
			2.7x10 <sup>-9</sup>		83	C <sub>2</sub> H <sub>7</sub> O <sup>+</sup> • (H <sub>2</sub> O) <sub>2</sub>	None
		NO <sup>+</sup>	1.2x10 <sup>-9</sup>	100	45	C <sub>2</sub> H <sub>5</sub> O <sup>+</sup>	None
			1.2x10 <sup>-9</sup>		63	C <sub>2</sub> H <sub>5</sub> O <sup>+</sup> • H <sub>2</sub> O	None
			1.2x10 <sup>-9</sup>		81	C <sub>2</sub> H <sub>5</sub> O <sup>+</sup> • (H <sub>2</sub> O) <sub>2</sub>	None
		O <sub>2</sub> <sup>+</sup>	2.3x10 <sup>-9</sup>	75	45	C <sub>2</sub> H <sub>5</sub> O <sup>+</sup>	None
			2.3x10 <sup>-9</sup>		63	C <sub>2</sub> H <sub>5</sub> O <sup>+</sup> • H <sub>2</sub> O	None
			2.3x10 <sup>-9</sup>		81	C <sub>2</sub> H <sub>5</sub> O <sup>+</sup> • 2H <sub>2</sub> O	None

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No	Compound	Reagent	Reaction rate, k (cm <sup>3</sup> /s)	Branching ratio (%)	Mass-to-charge ratio (m/z)	Product	Conflict and resolution
16	ethyl butanoate	H <sub>3</sub> O <sup>+</sup>	3.0x10 <sup>-9</sup>	80	117	C <sub>6</sub> H <sub>12</sub> O <sub>2</sub> • H <sup>+</sup>	None
					135	C <sub>6</sub> H <sub>13</sub> O <sub>2</sub> <sup>+</sup> • H <sub>2</sub> O	None
		NO <sup>+</sup>	2.4x10 <sup>-9</sup>	30	146	C <sub>6</sub> H <sub>12</sub> O <sub>2</sub> <sup>+</sup> • NO <sup>+</sup>	None
17	ethyl hexanoate	H <sub>3</sub> O <sup>+</sup>	3.0x10 <sup>-9</sup>	100	145	C <sub>8</sub> H <sub>16</sub> O <sub>2</sub> • H <sup>+</sup>	*Potential conflict with dimethyl trisulfide; 145 m/z for dimethyl trisulfide is not calculated (has lower reaction rate of 2.8x10 <sup>-9</sup> and a secondary product)
		H <sub>3</sub> O <sup>+</sup>	3.0x10 <sup>-9</sup>		163	C <sub>8</sub> H <sub>16</sub> O <sub>2</sub> • H <sup>+</sup> • H <sub>2</sub> O	None
		NO <sup>+</sup>	2.5x10 <sup>-9</sup>	95	174	C <sub>8</sub> H <sub>16</sub> O <sub>2</sub> <sup>+</sup> • NO <sup>+</sup>	None
18	ethyl methyl sulfide	H <sub>3</sub> O <sup>+</sup>	2.4x10 <sup>-9</sup>	100	77	CH <sub>3</sub> SHC <sub>2</sub> H <sub>5</sub> <sup>+</sup>	None
19	furaneol	H <sub>3</sub> O <sup>+</sup>	4.0x10 <sup>-9</sup>	100	129	C <sub>6</sub> H <sub>8</sub> O <sub>3</sub> • H <sup>+</sup>	None
			4.0x10 <sup>-9</sup>		147	C <sub>6</sub> H <sub>8</sub> O <sub>3</sub> • H <sub>3</sub> O <sup>+</sup>	None
		NO <sup>+</sup>	2.5x10 <sup>-9</sup>	95	128	C <sub>6</sub> H <sub>8</sub> O <sub>3</sub> <sup>+</sup>	None
		O <sub>2</sub> <sup>+</sup>	3.0x10 <sup>-9</sup>	100	128	C <sub>6</sub> H <sub>8</sub> O <sub>3</sub> <sup>+</sup>	None
20	γ - decalactone	H <sub>3</sub> O <sup>+</sup>	3.0x10 <sup>-9</sup>	100	171	C <sub>10</sub> H <sub>18</sub> O <sub>2</sub> • H <sup>+</sup>	None
		NO <sup>+</sup>	2.5x10 <sup>-9</sup>	100	200	C <sub>10</sub> H <sub>18</sub> O <sub>2</sub> • NO <sup>+</sup>	None
21	homofuraneol	H <sub>3</sub> O <sup>+</sup>	3.0x10 <sup>-9</sup>	100	143	C <sub>7</sub> H <sub>10</sub> O <sub>3</sub> • H <sup>+</sup>	None
		H <sub>3</sub> O <sup>+</sup>	3.0x10 <sup>-9</sup>		161	C <sub>7</sub> H <sub>10</sub> O <sub>3</sub> • H <sup>+</sup> • H <sub>2</sub> O	None
		NO <sup>+</sup>	2.5x10 <sup>-9</sup>	100	142	C <sub>7</sub> H <sub>10</sub> O <sub>3</sub> <sup>+</sup>	None
		O <sub>2</sub> <sup>+</sup>	2.5x10 <sup>-9</sup>	100	142	C <sub>7</sub> H <sub>10</sub> O <sub>3</sub> <sup>+</sup>	None
22	hydrogen sulfide	H <sub>3</sub> O <sup>+</sup>	1.6x10 <sup>-9</sup>	100	35	H <sub>3</sub> S <sup>+</sup>	None
			1.6x10 <sup>-9</sup>		53	H <sub>3</sub> S <sup>+</sup> • H <sub>2</sub> O	None
		O <sub>2</sub> <sup>+</sup>	1.4x10 <sup>-9</sup>	100	34	H <sub>2</sub> S <sup>+</sup>	None
23	methional	O <sub>2</sub> <sup>+</sup>	2.5x10 <sup>-9</sup>	75	104	C <sub>4</sub> H <sub>8</sub> OS <sup>+</sup>	None
24	methionol	NO <sup>+</sup>	2.5x10 <sup>-9</sup>	100	106	C <sub>4</sub> H <sub>10</sub> OS <sup>+</sup>	None
		O <sub>2</sub> <sup>+</sup>	2.5x10 <sup>-9</sup>	30	89	C <sub>4</sub> H <sub>9</sub> S <sup>+</sup>	None
			2.5x10 <sup>-9</sup>	40	106	C <sub>4</sub> H <sub>10</sub> OS <sup>+</sup>	None
25	methyl mercaptan	H <sub>3</sub> O <sup>+</sup>	1.8x10 <sup>-9</sup>	100	49	CH <sub>4</sub> S.H <sup>+</sup>	None
			1.8x10 <sup>-9</sup>		67	CH <sub>4</sub> S.H <sup>+</sup> • H <sub>2</sub> O	None
26	phenylacetaldehyde	H <sub>3</sub> O <sup>+</sup>	3.0x10 <sup>-9</sup>	100	121	C <sub>8</sub> H <sub>8</sub> O • H <sup>+</sup>	None
		H <sub>3</sub> O <sup>+</sup>	3.0x10 <sup>-9</sup>		157	C <sub>8</sub> H <sub>8</sub> O • H <sup>+</sup> • 2H <sub>2</sub> O <sup>+</sup>	None
		NO <sup>+</sup>	2.5x10 <sup>-9</sup>	15	91	C <sub>7</sub> H <sub>7</sub> <sup>+</sup>	None
		NO <sup>+</sup>	2.5x10 <sup>-9</sup>	60	120	C <sub>8</sub> H <sub>8</sub> O <sup>+</sup>	None

**Table 1.** Selected ion mode (SIM) method development. List of compounds, reagent ion, rate coefficients, product ions and potential interferences used in the V200™ SIM scan method



### 3. RESULTS AND DISCUSSION

#### 3.1. Stage-level VOC profile variability

The effect of the four different stages of manufacture on VOCs was evaluated for each of the 30 compounds for the two batches of cheeses. Each stage could be differentiated from each other stage for both Batch A and Batch B (Figure 1). In this section, the significant discrimination of VOC profiles among the different stages of manufacture, as well as the key compounds for the discrimination process will be discussed.

Figure 1A shows the VOC profile class projections simulations as well as the interclass distances (ICD) between the stages using SIMCA for the two batches of Swiss cheese samples. The class projections provide visual evaluation of the degree of class separation among the four stages of manufacture while the interclass distances

quantify the separation between classes. Three-factor PCA was performed during the SIMCA processing and the coordinates of the ellipse, which are based on the standard deviations of the scores in each principal component (PC) direction for each class, are projected into these three-factor PCA spaces. These bounding ellipses form a confidence interval for the distribution of these classes. As can be seen from these figures, separation of the four stages is significant, based on their ICDs, which are all greater than 4. It is generally accepted that an ICD of 3 and greater indicates statistically significant separation (with 95% confidence) among classes being differentiated (Subramaniam and others 2011; Kvalheim and Karstang 1992). It can also be observed that the VOC profile at the end-of-warm room stage has a higher level of discrimination compared to all other stages.

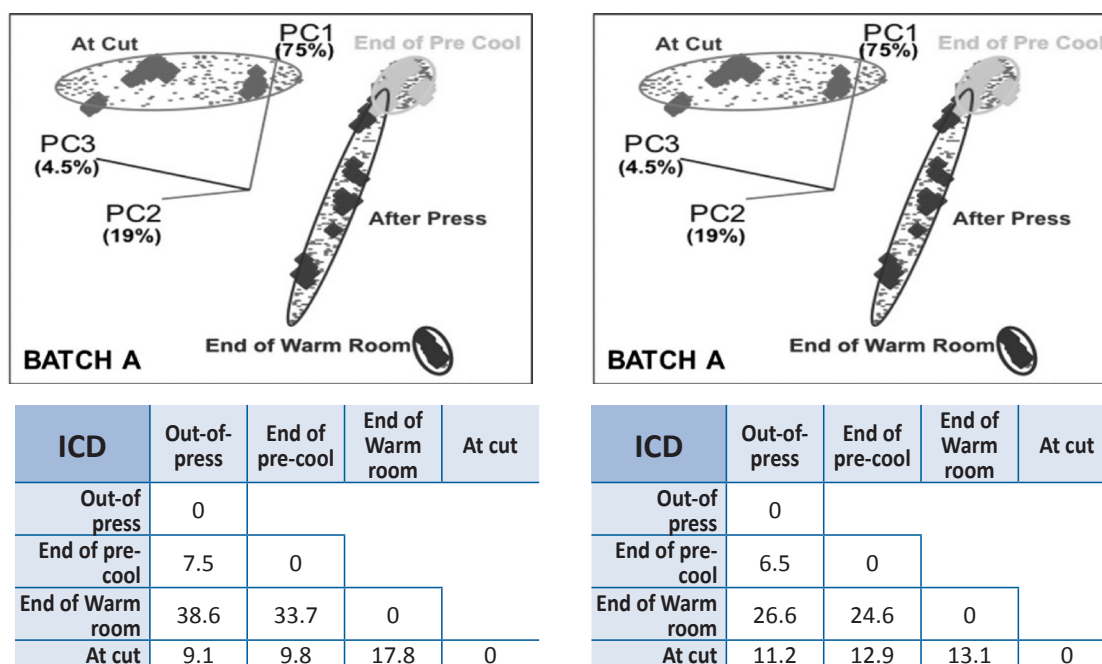


Figure 1A. SIMCA class projections and ICDs of the VOC profiles from the four stages of cheese manufacturing

The differentiation between the cheeses could be related to the discriminating power of the different compounds based on the SIMCA analyses, as shown in Table 2. The data in the table shows the hierarchy of the each VOC based on their discriminating power in each batch (A and B) of cheese samples. The discriminating power provides important information on which

variables are best at discriminating between the classes. The discriminating power provides an indication of how much a variable discriminates between “correct” and “incorrect” classification by comparing the average residual variance of each class fit to all other classes and the residual variance of all classes fit to each class (Multivariate data...2008).

Batch A - Compound	Discriminating Power	Batch B - Compound	Discriminating Power
Methional	27 391.8	Propanoic acid	8663.7
Propanoic acid	3272.6	Ethanol	732.9
Acetic acid	1388.1	Methional	523.8
Butanal	312.3	Butanoic acid	473.7
Phenylacetaldehyde	311.3	Dimethyl sulfide	272.3
Ethanol	245.5	Acetic acid	168.9
3-Methylbutanoic acid	178.7	Butanal	150.1
2-Methylpropanal	147.2	Phenylacetaldehyde	139.5
Ethyl methyl sulfide	108.0	Methionol	119.5
Ethyl butanoate	95.3	Diethyl sulfide	91.4
Dimethyl disulfide	91.8	(E)-2-nonenal	74.7
Tetramethylpyrazine	89.2	3-Methylbutanal	74.5
Dimethyl sulfide	82.3	Ethyl butanoate	68.7
(E)-2-nonenal	74.4	Tetramethylpyrazine	63.7
Methionol	62.3	Ethyl methyl sulfide	51.2
Methyl mercaptan	54.0	2,3-Butanedione	43.3
3-Methylbutanal	53.0	3-Methylindole	42.7
2,3-Butanedione	52.8	2-Methylpropanal	35.9
Butanoic acid	42.8	Ethyl hexanoate	33.0
$\gamma$ -decalactone	37.1	3-Methylbutanoic acid	31.7
Diethyl sulfide 33.9; Furaneol 23.6; Dimethyl trisulfide 22.3; Ammonia 21.1; Ethyl hexanoate 12.1; Homofuraneol 11.1; Hydrogen sulfide 9.3; 3-Methylindole 8.6		Dimethyl disulfide 29.0; $\gamma$ -decalactone 27.3; Methyl mercaptan 26.0; Furaneol 23.0; Ammonia 13.4; Homofuraneol 11.2; Dimethyl trisulfide 10.9; Hydrogen sulfide 2.8	

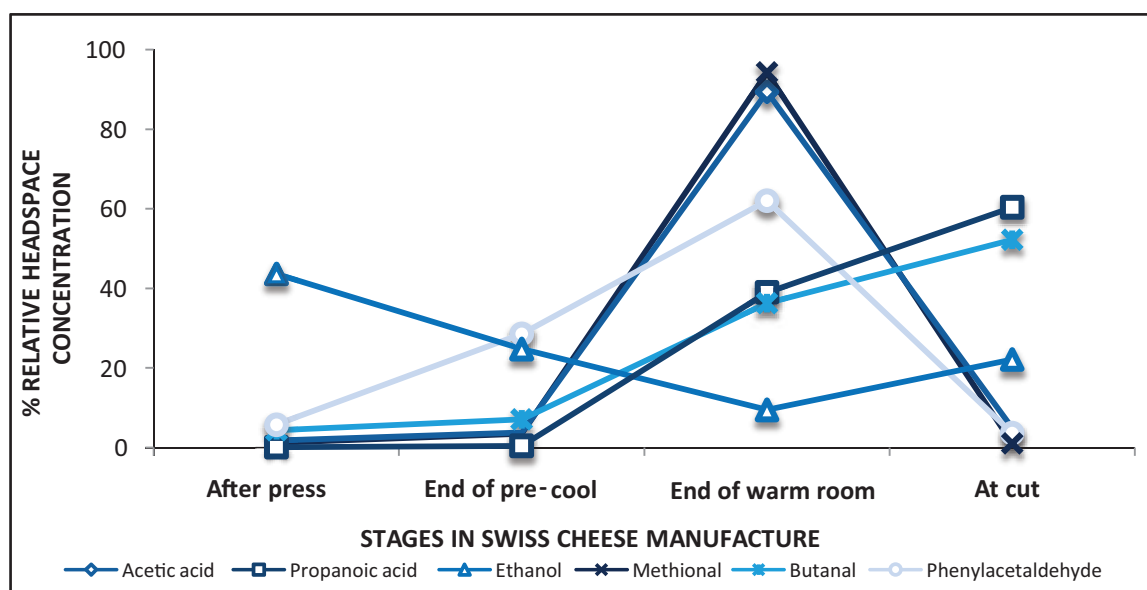
**Table 2.** Discriminating power of individual VOCs from SIMCA analysis

Based on the results, six VOCs (methional, propanoic acid, acetic acid, ethanol, butanal and phenylacetaldehyde), out of the 28 VOCs analyzed, are in the top ten highly discriminating key compounds in both batches of cheese samples. These six VOCs are key variables in discriminating the four stages of manufacture and served as basis for the classification and therefore, separation of the four stages.

The percentage relative headspace concentrations of these six key discriminating VOCs in each stage of manufacture are presented in Figure 1B. In this figure, the relative variability of each VOC

is distinct at each stage, as well as the profile of individual VOC throughout the four stages of manufacture. Most of these VOCs have their concentration maxima towards the latter stages of manufacture (i.e., ripening of the cheese). Acetic acid, methional and phenylacetaldehyde are highest during the warm room storage of the cheese; propanoic acid and butanal have their maximum concentration at the time of cutting; and ethanol is highest during pressing. The profile behaviors of these compounds along with other compounds are discussed more thoroughly in the next section (“Evaluation of the stage-level concentration profile of individual VOCs”).





**Figure 1B.** % Relative headspace concentration of the six key discriminating VOCs based on their discriminating power (simulated in SIMCA) between the four stages of Swiss cheese manufacture

### 3.2. Evaluation of the stage-level concentration profile of individual VOCs

Different compounds showed different profiles at different stages of the process, as shown in Figures 2A -2H. With a few exceptions, the profiles of the compounds were similar for the cheeses in both Batch A and Batch B. The exceptions were ethyl butanoate, ethyl hexanoate, phenylacetaldehyde, 3-methylindole, butanoic acid and some of the sulfur compounds. The variabilities are due to the differences in the level of concentration between

the two batches of cheeses for a particular stage or stages of manufacture. Table 3 shows which compounds are highest (average of the two batches of cheese) at each stage of manufacture. Few compounds had their highest concentration during pressing, most during pre-cool, some during the warm room and at the time of cutting. Identification of these compounds is important in being able to relate them back to their biochemical and microbiological origin, which could provide insights on the mechanism and activities occurring in each stage of manufacture.

STAGES OF SWISS CHEESE MANUFACTURE			
Out-of-press	End of pre-cool	End of warm room	At cut/Packaging
(E)-2-nonenal	3-Methylindole	3-Methylindole	2-Methylpropanal
Ethanol	2,3-Butanedione	Acetic acid	3-Methylbutanal
Methyl mercaptan	3-Methylbutanoic acid	Dimethyl disulfide	Ammonia
	Butanoic acid	Methional	Butanal
	Diethyl sulfide	Phenylacetaldehyde	Furaneol
	Dimethyl sulfide	Tetramethylpyrazine	Hydrogen sulfide
	Dimethyl trisulfide		Homofuraneol
	Ethyl butanoate		Propanoic acid
	Ethyl hexanoate		
	Ethyl methyl sulfide		
	$\gamma$ -decalactone		
	Methionol		

**Table 3.** VOCs with highest concentration detected in each stage of manufacture

The following section describes, in detail, the changes in the levels of concentration of each VOC in relation to their biochemical pathway of origin and some information on their organoleptic

qualities in cheese. Each VOC will be discussed according to their functional group as categorized and presented in Table 4.

Functional Group	Compound	Functional Group	Compound	
Alcohol	Ethanol	Sulfur compounds	Methyl mercaptan	
Aldehydes	(E)-2-Nonenal		Ethyl methyl sulfide	
	2-Methylpropanal		Dimethyl sulfide	
	Butanal		Dimethyl disulfide	
	2-Methylbutanal		Dimethyl trisulfide	
Aromatic - Aldehyde	3-Methylindole		Diethyl sulfide	
	Phenylacetaldehyde		Methional	
Ketones	2, 3-Butanedione		Methionol	
Esters	Ethyl butanoate		Carboxylic acids (organic acids)	Acetic acid
	Ethyl hexanoate			Butanoic acid
Furans	Furaneol	3-Methylbutanoic acid		
	Homofuraneol	Propanoic acid		
Lactones	$\gamma$ -decalactone	Inorganic	Ammonia	
Pyrazine	Tetramethylpyrazine		Hydrogen sulfide	

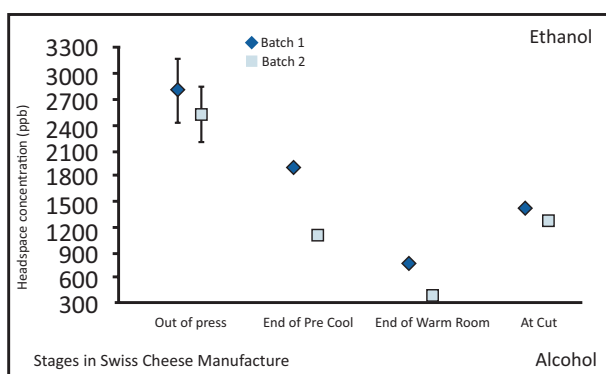
**Table 4.** Volatile organic compounds (VOC) isolated as flavors in cheese

Adapted from Marilley and Casey (2004) and Curioni and Bosset (2002)

### 3.3. Alcohol

The formation of ethanol, as a common fermentation product and main alcohol detected in cheese (Mc Guban and others 1975), is mainly from the activity of heterofermentative lactobacilli and/or yeasts (Thierry and others 2006). The availability of ethanol (and methanol) is important for ethyl (and methyl) ester formation. Thierry and others (2006) have shown that ethanol is the limiting factor of ethyl ester synthesis in

Swiss cheese. The headspace concentration of ethanol (Figure 2A) was highest at the out-of-press samples for both batches of Swiss cheese. Its concentration decreases until it reaches a minimum at the end-of-warm room samples and increases at the at-cut/packaging stage but with lower concentrations, on average, than the end of pre-cool.

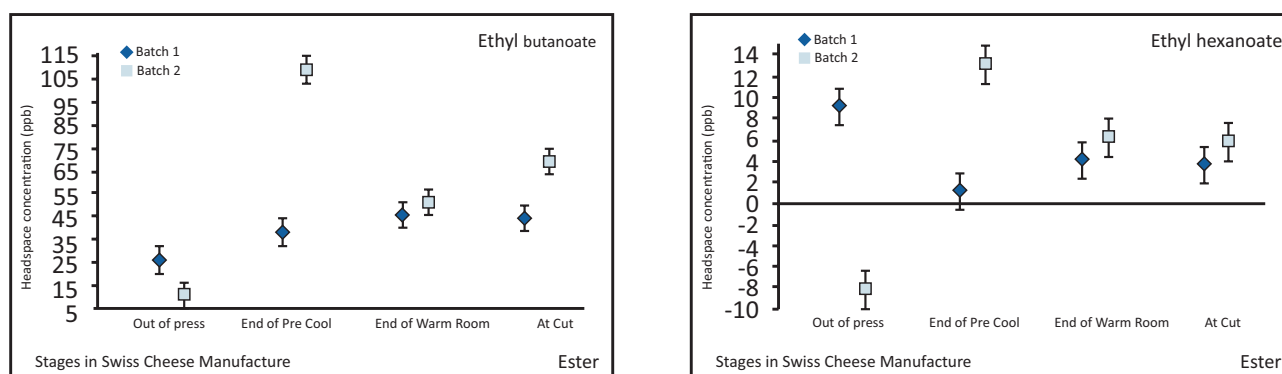


**Figure 2A.** Headspace concentration profile of ethanol, from different stages of Swiss cheese manufacture

### 3.4. Esters

Earlier studies have assumed a direct relationship between ethanol and ethyl esters in different varieties of cheese, including Swiss cheese (Thierry and others 2006; Mc Gugan and others 1975; Langler and others 1967). Ester formation is believed to arise from several biochemical pathways including esterification between acid and alcohol, reaction of another ester with either alcohol or acid, or by the reaction of two esters (transesterification) (McSweeney 2004; Holland

and others 2005). In Figure 2B, ethyl butanoate's headspace concentrations are relatively higher than ethyl hexanoate at the four stages of manufacture. Ethyl hexanoate's concentration is quite constant, while that of ethyl butanoate has a general trend of increasing, although insignificantly. For both ester compounds, the highest concentration was registered from cheese samples at the end of pre-cool, especially for the second batch of samples.



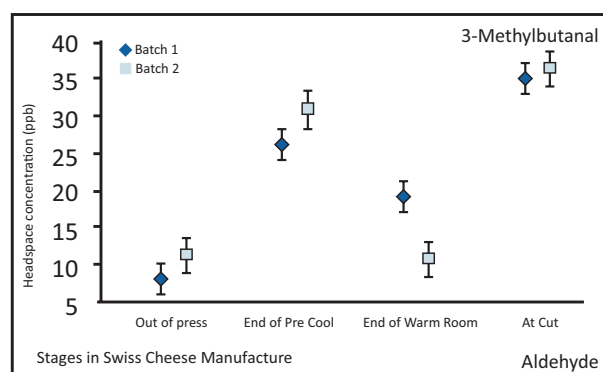
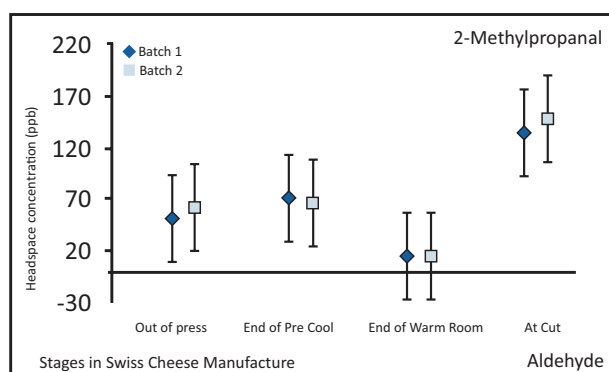
**Figure 2B.** Headspace concentration profile of ester groups, from different stages of Swiss cheese manufacture

### 3.5. Aldehydes and aromatic aldehydes

Aldehydes in cheese are produced, in general, via amino acid catabolism (Fox and others 1995; McSweeney and Sousa 2000). They are formed by the subsequent degradation through carboxylation or Strecker reaction of an imide – an intermediate formed by an enzyme-catalyzed transamination of free amino acid (Polo and others 1985; Fox and others 1995; Urbach 1995; Molimard and Spinnler 1996; McSweeney and Sousa 2000). Cheeses do not accumulate high concentrations of aldehydes because of their succeeding reduction to alcohol or oxidation to corresponding acids (Dunn and Lindsay 1985; Lemieux and Simard 1992). Moreover, with lower pH at the beginning of cheese ripening, amino acids are decarboxylated to amines; however, these amines are consequently oxidized to aldehydes with an increase in pH at a later stage of ripening, through the Strecker degradation reaction. The compounds, 2-methylpropanal and

3-methylbutanal, which are characterized by a malty flavor, are derived from their respective precursor amino acids valine (2-methylpropanal) and leucine (3-methylbutanal).

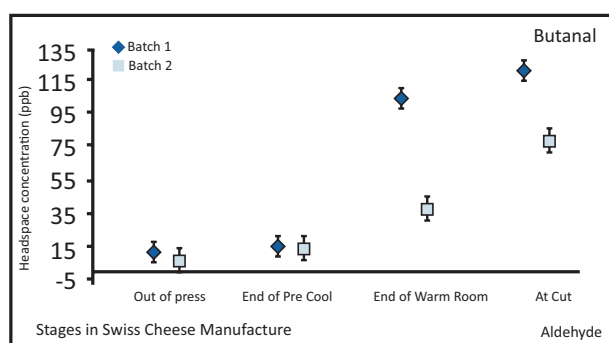
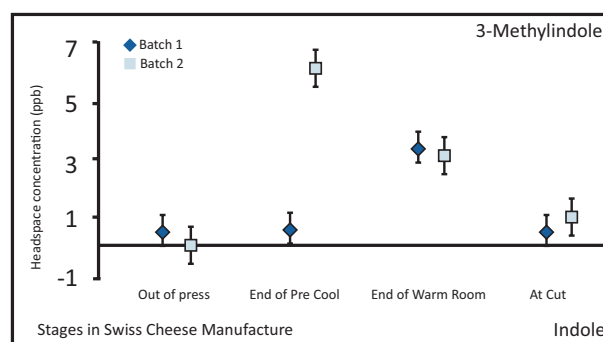
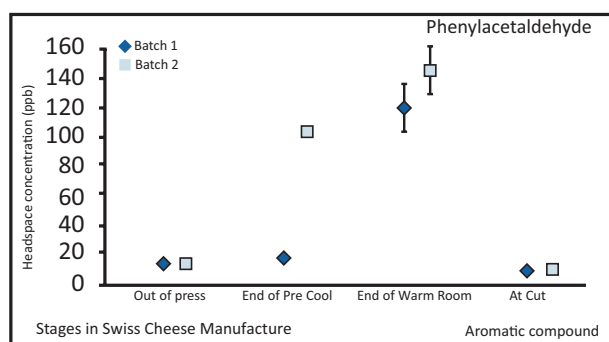
Figure 2C shows the profile of these two aldehydes at different stages of the manufacturing process. Both of these compounds have highest concentrations from the cheese samples at the time of cutting and packaging. The concentration of 2-methylpropanal at this stage has reached about 4x that of 3-methylbutanal's concentration. In all previous stages, the concentration of 2-methylpropanal is constant. The headspace concentration of 3-methylbutanal, however, is elevated at the end of pre-cool then decreases at end of warm room and finally increases at the time of cutting and packaging.



**Figure 2C.** Headspace concentration profile of branched-chain aldehydes, from different stages of Swiss cheese manufacture

The aromatic aldehydes derived from the catabolism of aromatic amino acids have great impact on cheese aroma. The mechanism produces aromatic compounds from tryptophan, tyrosine and phenylalanine. Several researchers have described, in detail, the mechanism of production of aromatic compounds in cheese including the microorganisms involved in their production (Gao and others 1997; Yvon and others 1997; McSweeney and Sousa 2000; Marilley and Casey 2004). Phenylacetaldehyde is characterized by a honey-like, floral, rosy and violet-like aroma, while indole compounds produce a fecal putrid and musty odor – an aroma often associated with the formation of unclean flavors in dairy products (Thierry and Maillard 2002; Curioni

and Bosset 2002; Marilley and Casey 2004). The headspace concentration levels (Figure 2D) of phenylacetaldehyde had increased significantly at the end of warm room stage for both batches of Swiss cheese samples. The samples from the other stages of manufacture have relatively the same concentration of phenylacetaldehyde, which was markedly lower than for the end of warm room samples. On the other hand, 3-methylindole exists at a very low, but detectable, level in the cheese samples from all stages of manufacture (Figure 2D). Although there are significant differences in the concentration among the stages of manufacture, the levels are very low compared to the other aldehyde compounds.



**Figure 2D.** Headspace concentration profile of phenylacetaldehyde, 3-methylindole and butanal.

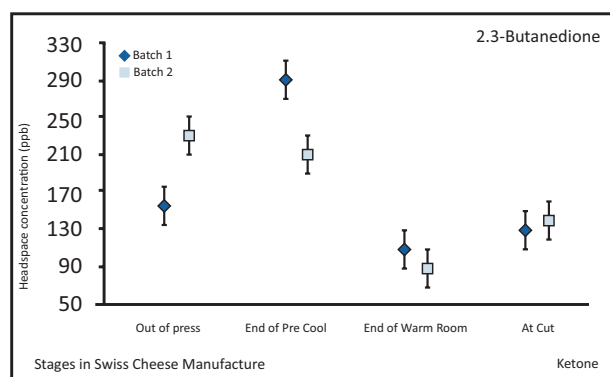
Butanal (butyraldehyde) and some straight-chain aldehydes like heptanal and nonanal, may be formed through the  $\beta$ -oxidation of unsaturated fatty acids. High levels of these straight chain aldehydes produce green grass-like aromas (Collins and others 2004). The concentration of butanal, as shown in Figure 2D, increases as the cheese progresses from the out-of-press towards the

cutting and packaging stage of manufacture. There was a 5- to 10-fold increase in concentration from the cheese samples at cut relative to the samples from the out of press stage. This result indicates a possible dynamic lipolytic activity in the cheese samples obtained, especially at the latter stages of manufacture.

### 3.6. Ketone

Diacetyl (2, 3-butanedione) was detected at a relatively higher concentration in the cheese samples from the two earlier stages of manufacture (out-of press and end of pre-cool) (Figure 2E). Its concentration was markedly decreased during the two latter stages, where the level reached a minimum from the end of warm room stage. The concentration level of diacetyl is higher during the initial stages of manufacture, coinciding with the metabolism of citrate, the

biochemical pathway that produces diacetyl (McSweeney 2004). An appreciable amount of diacetyl has a lethal effect on propionibacteria (Fröhlich-Wyder and Bachmann 2004). The minimal amount of diacetyl detected in cheese samples from the warm room stage is consistent and supportive of the fact that the rapid growth of propionibacteria occurs during this stage in Swiss cheese manufacture (Reinbold 1972).

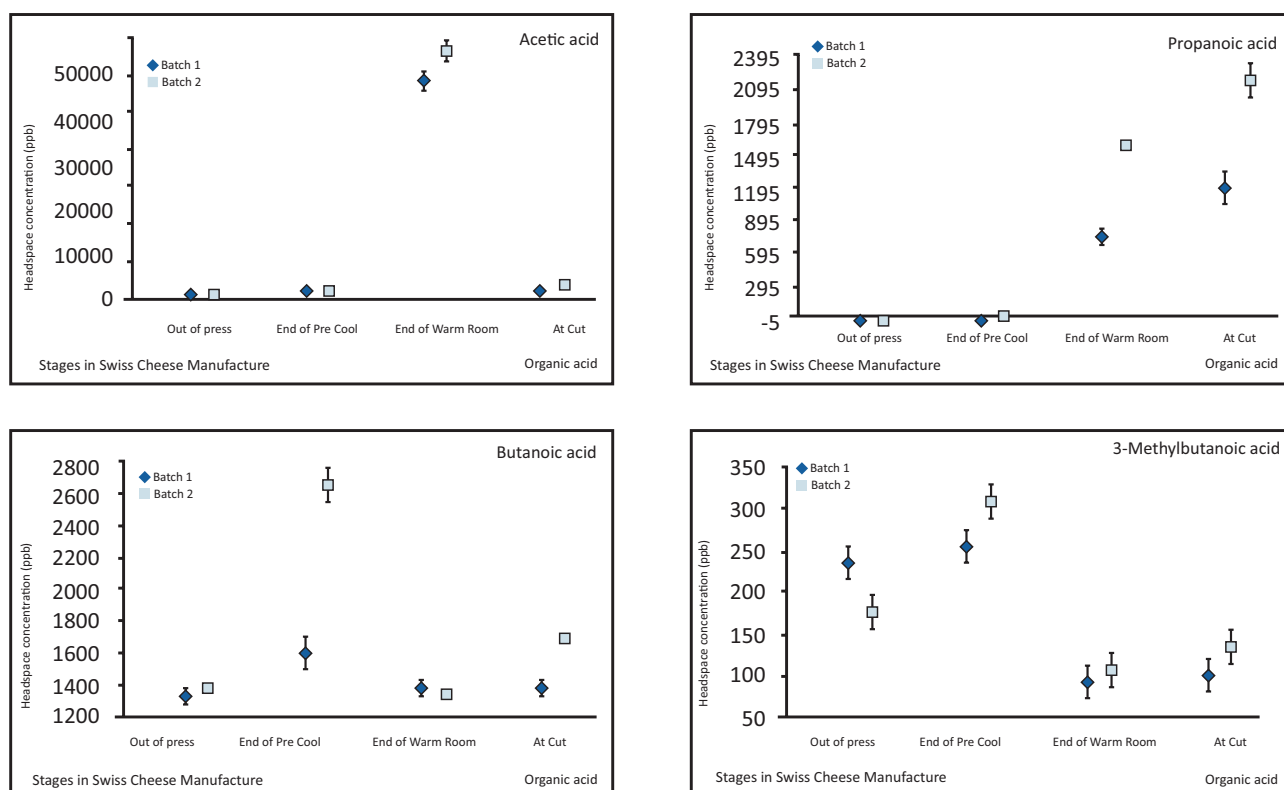


**Figure 2E.** Headspace concentration profile of 2,3-butanedione, from different stages of Swiss cheese manufacture

### 3.7. Organic acids

The organic acid group of compounds (Figure 2F) is produced from varied biochemical pathways in cheese. The propionic acid fermentation pathway, which is believed to mostly contribute to the important characteristic eye and the nutty flavor of Swiss cheese, generates acetic and propionic acids along with  $\text{CO}_2$  and adenosine triphosphate (ATP) (Fröhlich-Wyder and Bachmann 2004). Propanoic acid has an increasing concentration profile, where the cheese samples from the time of cutting and packaging have been detected to have the highest concentration relative to

the cheese samples from the previous stages of manufacture. Acetic acid concentration, on the other hand, was highly detected in cheeses from the end of warm room stage and decreased significantly during the final stage of processing.



**Figure 2F.** Headspace concentration profile of organic acid group, from different stages of Swiss cheese manufacture

An appreciable amount of butanoic acid, which could be produced from lipolysis and/or butyric acid fermentation (Steffen and others 1993; Chamba and Perreard 2002), was detected in all the cheese samples at all stages. There was a significant increase in butyric acid concentration (Figure 2F) from the second batch of cheese sample at the end of the pre-cool stage, but its concentration was comparatively stable in other stages.

The formation of isovaleric acid (3-methylbutanoic

acid) from the oxidation of the 3-methylbutanal, derived from the Strecker degradation of leucine (McSweeney and Sousa 2000), gives the cheesy, sweaty, old socks, rancid, fecal and rotten fruit aroma note in cheeses (Singh and others 2003). In Figure 2F, the concentration profile of 3-methylbutanoic acid is observed to have a similar profile to that of diacetyl, such that its concentration is higher during the earlier stages and was detected at a lower concentration from the cheese samples during the end of warm room stage and at the time of cutting and packaging.

### 3.8. Sulfur compounds

The low molecular weight sulfur compounds (Figure 2G), that must have originated primarily from the catabolism of methionine (Harper and others 2011; McSweeney and Sousa 2000), are considered essential contributors to cheese flavor, mainly because of their very low threshold values in the parts-per-billion (ppb) to ppt range (Harper and others 2011; Landaud and others 2008; Burbank and Qian 2005). These sulfur-containing VOCs generate the cabbage, sulfurous, onion and garlic aroma notes in cheeses (Singh and others 2003). Most of these compounds have

similar behavior profile. There was, however, a significant variation in the concentration of the sulfur compounds between batches, especially at the pre-cool stage. Methional, which has emerged as one of the key discriminating compounds in this study, was detected at a high concentration (~800–1000 ppb) from cheeses at the end of warm of room stage. One possibility for its high concentration is the reversible conversion of methionol to methional, especially if the environment is oxidizing (Escudero and others 2000).

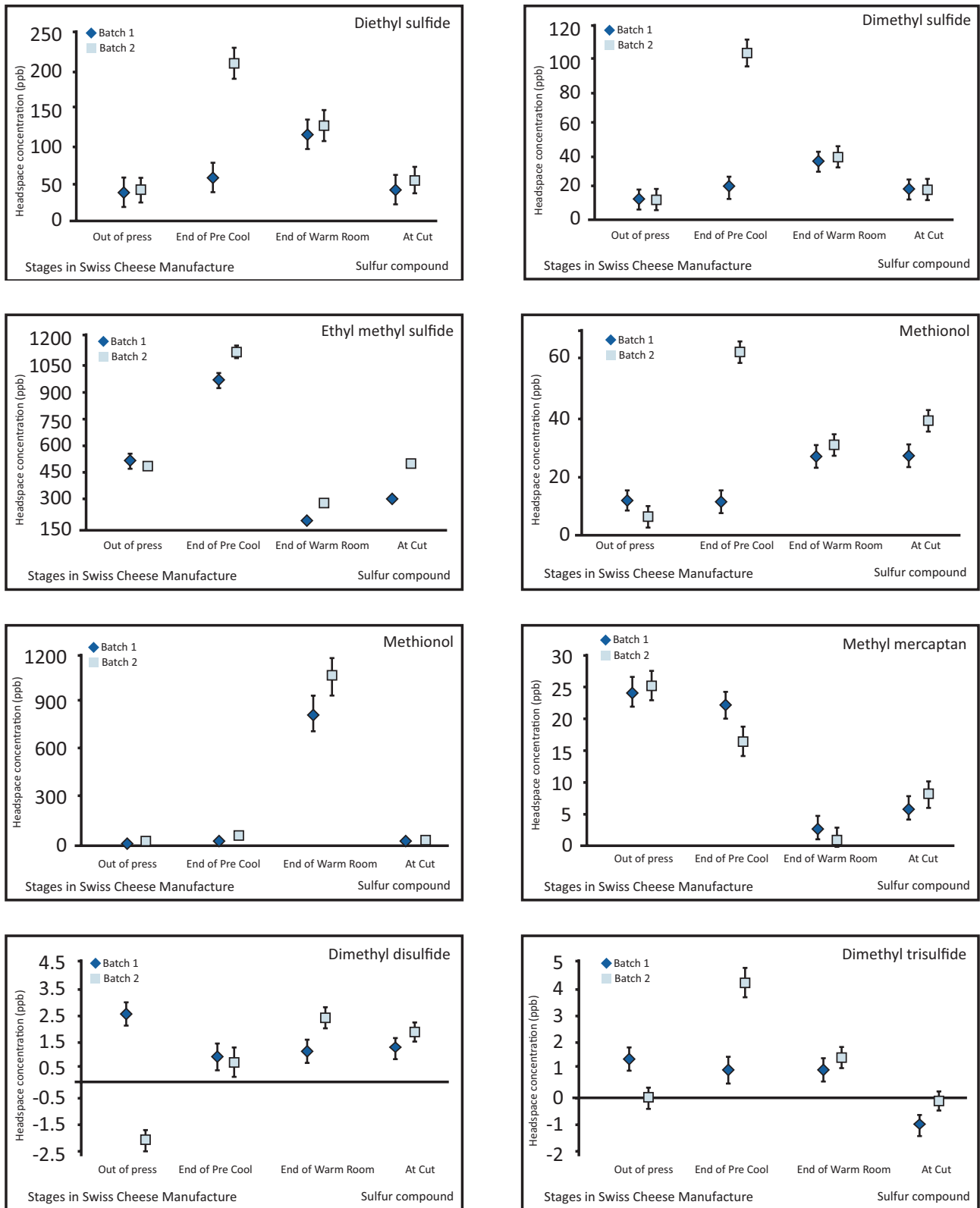
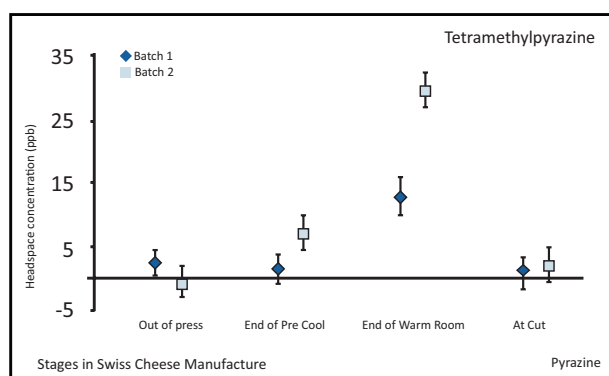


Figure 2G. Headspace concentration profile of sulfur compounds, from different stages of Swiss cheese manufacture

### 3.9. Pyrazines and other compounds

Tetramethylpyrazine, an alkylpyrazine, is considered to be a favorable contributor to the natural cheese flavor (Sloot and Hofman 1975). Some compounds of pyrazine, including tetramethylpyrazine, have been analyzed in the outer layer of Emmental and Gruyere cheeses, where they were presumably formed by microorganisms in the smear (McSweeney

and Sousa 2000). The headspace concentration of tetramethylpyrazine (Figure 2H) was significantly higher from the end of warm room cheese samples. All other stages did not show significant differences in the concentrations of the compound, which were very low (<5 ppb) compared to the concentration level from the end of warm room stage (~30 ppb).



**Figure 2H.** Headspace concentration profile of tetramethylpyrazine, from different stages of Swiss cheese manufacture

As for the other compounds, there were no significant differences detected in the concentration levels of furans, lactones and inorganic compounds among the four stages of manufacture. Some of the compounds in these categories were below the detectable range capability of the SIFT-MS instrument.

Cheese is considered to be the most diverse group of dairy products, with very challenging yet interesting biophysical and chemical characteristics. The dynamic biological and biochemical activities in cheese as well as the varying physical and environmental factors (temperature, pH, humidity, salt and water content) make its system integrally unstable and thus create inhomogeneity in the texture, flavor

and aroma, and other different physical and chemical aspects between the resulting cheese products (Fox and McSweeney 2004). In terms of the VOCs, which impart the flavor and aroma in cheese, their production and behavior can be understood much more clearly by tracing back their biochemical pathways of production through specific precursor compounds and back to their macromolecule of origin (i.e., citrate, lactate, triglyceride, casein) (McSweeney PLH 2004). These chemical pathways, and therefore VOC production, are also highly correlated with the microbiological activities and fermentation changes to the milk and curd, so it is also important to note and observe the differences and changes in the microbial profile/flora (i.e., population, strain, activities, etc.) in relation to the different stages of production in cheese manufacturing.

### 3.10. Vat-level VOC profile variability

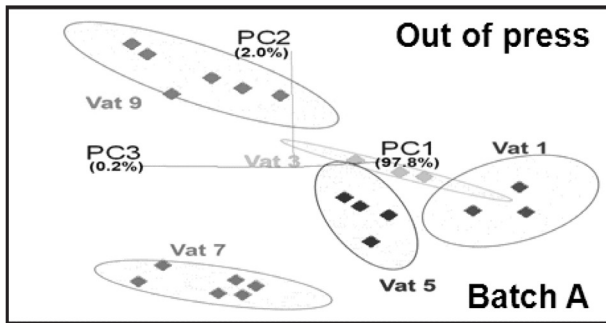
Perhaps one of the most interesting results in this study is the behavioral profile differences in VOCs between vats of cheese made on the same make-date and following through their profile at each stage of manufacture.

Figures 3A and 3B show the results of SIMCA class projections of the VOC profile along with the corresponding interclass distances between vats for the two batches of cheese samples. As suggested by the results, there is significant

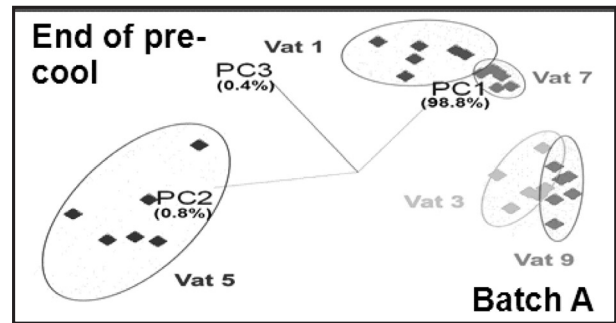


discrimination of VOC profile between the different vats at all the stages of manufacture, as indicated by the ICD values, which are all greater than 3 at a 95% confidence interval. Upon further analysis, the interclass distances of the VOC profile

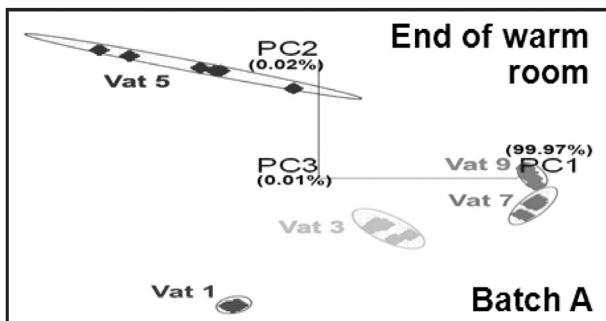
between vats could be observed to increase during successive processing stages. The VOC profile of Swiss cheese manufactured in the same batch but different vats became more differentiated as the cheeses reached the final stage of manufacture.



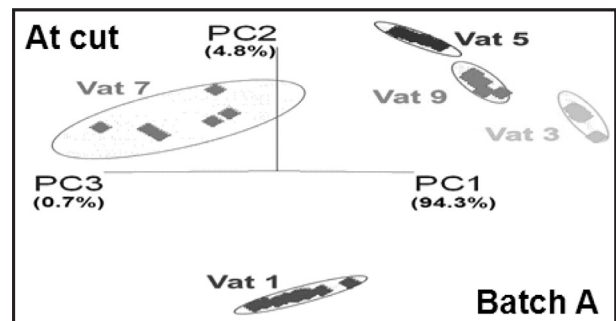
ICD	1	3	5	7	9
1	0				
3	3.0	0			
5	4.2	3.5	0		
7	6.2	9.4	13.3	0	
9	8.8	8.4	13.2	19.4	0



ICD	1	3	5	7	9
1	0				
3	9.8	0			
5	12.8	19.9	0		
7	4.6	7.6	16.3	0	
9	8.8	3.7	16.4	7.8	0

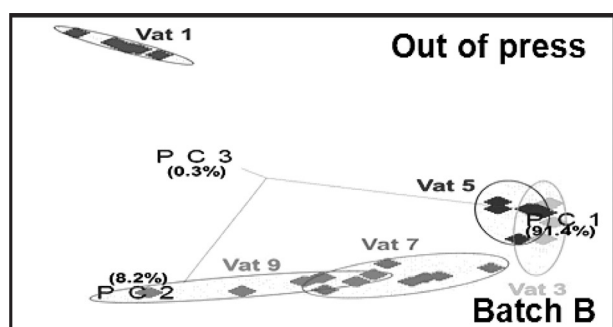


ICD	1	3	5	7	9
1	0				
3	27.4	0			
5	39.4	24.8	0		
7	25.7	17.7	15.4	0	
9	32.6	23.5	33.3	16.3	0

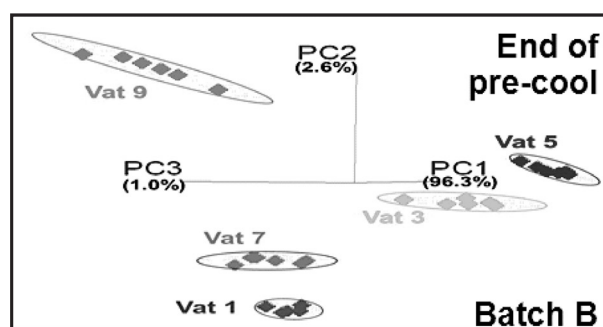


ICD	1	3	5	7	9
1	0				
3	53.9	0			
5	98.8	60.0	0		
7	34.8	11.8	15.6	0	
9	50.9	34.4	11.6	4.8	0

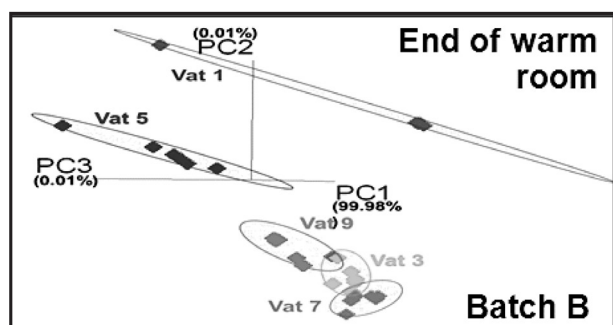
Figure 3A. SIMCA class projections of VOC profile from Swiss cheese made in different vats but on the same make-date at different stages of manufacture (Batch A)



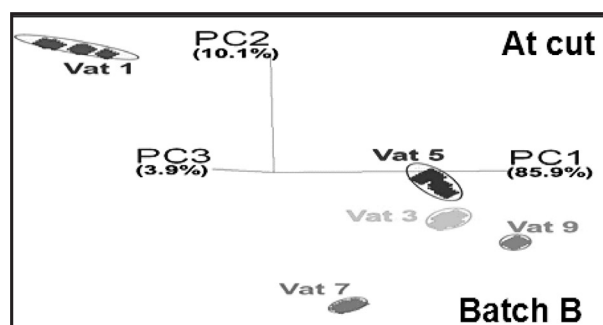
ICD	1	3	5	7	9
1	0				
3	34.9	0			
5	20.5	5.0	0		
7	33.4	12.8	4.3	0	
9	39.9	17.3	6.9	2.8	0



ICD	1	3	5	7	9
1	0				
3	24.3	0			
5	29.7	15.3	0		
7	8.2	12.1	21.3	0	
9	32.7	22.5	18.3	26.5	0



ICD	1	3	5	7	9
1	0				
3	73.3	0			
5	65.7	12.3	0		
7	95.4	11.3	17.7	0	
9	87.4	8.6	14.3	12.8	0



ICD	1	3	5	7	9
1	0				
3	112.8	0			
5	114.3	20.3	0		
7	122.1	108.4	86.1	0	
9	106.3	25.8	28.7	50.6	0

Figure 3B. SIMCA class projections of VOC profile from Swiss cheese made in different vats but on the same make-date at different stages of manufacture (Batch B)

This variability would mean increased inhomogeneity, and hence inconsistency, in the flavor and aroma among the Swiss cheeses manufactured within the same make-date in the same factory.

Consistency in the VOC profile of manufactured cheeses is important because not only does it dictate the flavor and aroma of the final products but it also gives an indication of consistency in the processes and techniques involving the manufacture of cheese. Such inconsistencies could lead to the production of sub-standard quality and lower grade of manufactured cheeses, varying flavor and aroma profile, and could cause potential detrimental effects such as the production of splits and crack defects that eventually downgrade the

cheese product and its market value. The latter effect could be due to the overproduction of gas (CO<sub>2</sub>) from sources other than the propionic acid fermentation. Our previous study showed key differentiating VOCs in the split segment, relative to the eye and blind areas (Castada and others 2013). Each of these compounds has accompanying CO<sub>2</sub> by-product that is associated with their production. Such possibility of excessive gas production from secondary fermentation sources other than the desired propionic fermentation have been previously discussed in an important review article by Daly, McSweeney and Sheehan (2010) on split defect formation.

## 4. CONCLUSIONS

Analysis and profiling of the behavior of VOC from different stages of Swiss cheese manufacture using SIFT-MS was proven to be effective and efficient. Statistical evaluation of the levels of VOCs from different stages has identified key discriminating compounds and shown that the levels of VOCs at the warm room stage are highly discriminatory. These results are indicative of the varied biochemical activity in relation to the microbiology and biochemistry in the different stages of manufacture. Significant VOC profile differences have also been identified in the same batch of Swiss cheeses made in different vats. Vat-level VOC profile variability could result in the inconsistency of flavor and aroma of the final Swiss cheese products manufactured. Such differences could be caused by irregularities in the different production variables, processing techniques and manufacturing processes. With the proper measures and coherent manufacturing procedures, regular analysis and evaluations, an efficient and effective control in cheese production could be achieved, leading to a better control of quality and a reduction in the formation of splits and cracks in Swiss cheese.

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# 5 Why children love milk? Milk companies and organizations together for school milk success, Polish experience

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The Polish Chamber of Milk (PCM) is the biggest dairy association in Poland. Today, Poland is a leader among the EU countries that benefit from subsidies for school milk. Many people have worked hard for this success. Obviously, the success is a result of numerous promotional and educational activities. One of the most important tasks of PCM is to promote milk consumption among younger generations. We educate and inform children, teenagers and adults about this by publishing books and films, organizing events and so on. "Milk with Class" is a joint project of Polish milk producers, the Council for the Promotion of Healthy Nutrition and Tetra Pak. The cooperation is very important for children's education and the success of "Milk with Class" and other programmes promoting milk. The newest project important and loved by children is "Milky Kids" (targeting 3–6 year olds). The next programme "I am crazy about milk" was started in 2012. This is a joint effort of PCM and the Polish Federation of Cattle Breeders and Dairy Farmers. The project is subsidized by the EC, the Polish national budget and the Milk Promotion Fund. Why do children in Poland like drinking milk? The answer is simple: it is a result of joint efforts by the dairy association, farmers, producers and companies like Tetra Pak.

The Polish Chamber of Milk is the biggest dairy association in Poland. This is an association or platform for discussion and exchanging opinions as well as for making strategic decisions for the dairy sector. We are a strategic partner for the dairy industry the EU, national institutions and government agencies. The Polish Chamber of

Milk is an association especially for milk dairy cooperatives and for processing and packaging companies. We are cooperating with farmers, especially with the Polish Federation of Cattle Breeders and Dairy Farmers. Together we work for promoting milk products.

The School Milk Scheme was implemented in Poland in 2004 when my country joined the European Union. The School Milk Scheme is administered by the Agricultural Market Agency. The aim of the scheme is to promote healthy eating among school children by encouraging consumption of milk and milk products, which are offered to school children free of charge or at reduced prices, because it is very important to shape a habit of milk consumption.

The programme is financed from three sources:

- The European Union budget – to support the consumption of milk and milk products by children attending kindergartens, primary schools and junior secondary schools (since 2004)
- The Milk Promotion Fund – to support consumption of milk and milk products by children in kindergartens and junior secondary schools (since 2005)
- The Polish budget – to support the consumption of milk and milk products by children in primary schools

In 2007, the Polish Government introduced National Aid for a glass of milk, which had a huge impact on milk consumption in schools. Thanks to the Polish Government's decision to finance the School Milk System, Poland has become a market leader in school milk consumption in Europe. It has been estimated that since 2004 Polish children have drunk 1 billion, 515 million glasses of milk.

In Poland, about 14 500 educational establishments are participating in the School Milk Scheme, including 12 000 primary schools. Milk and milk products are consumed by 2.5 million school children (40% of all pupils in Poland), of which 2.1 million are primary school children (96% of primary school children in my country).

In Poland there are a few milk companies, which produce milk for School Milk Scheme. The biggest milk producer is MLEKPOL dairy cooperative.

Today, Poland is a leader among the EU countries that benefit from subsidies for school milk. Many people worked hard for this success. Obviously, the success is a result of numerous promotional and educational activities. The School Milk Scheme is very popular in Poland and has become a regular part of school's everyday life. One of the most important tasks of PCM is to promote milk consumption among the younger generation. We educate and inform children, teenagers and adults about this by: publishing books, films, organizing events and so on.

The Polish Chamber of Milk supports and co-organizes the best milk campaigns in my country. For example "Milk with Class" was the first project for primary schools. This project is highly evaluated by schools and the Polish Government. That is why it is so important for us to work closely with the programme "Milk with Class"; we all complement each other to increase milk consumption in Poland.

"Milk with Class" has existed since 2006. It is a joint project of Polish milk producers, the Council for the Promotion of Healthy Nutrition and the Tetra Pak company. Since the launch of the programme, over 9000 schools and 1 million kids have cooperated with the programme. We estimate that this is the most successful enterprise supporting the School Milk Scheme in the country. The Polish Chamber of Milk and the "Milk with

Class" programme work together to organize events, contests and promotions for children in many Polish cities each year. The programme includes special lessons for children not only about the nutritional value of milk but also about, for example, waste segregation. Today lessons about milk and waste segregation are very popular among teachers and students in Poland.

Children also like the milk packaging of school milk. Milk for schools is delivered in small packages such as cartons of 0.2 or 0.25 l. This packaging is exclusively for the School Milk Scheme.

The cooperation between milk companies and dairy associations is very important for children's education and the success of "Milk with Class" as well as other programmes promoting milk and milk products.

The newest project, important and loved by children is "Milky Kids" – in Polish *Dzieciaki Mleczaki*. This is project that targets 3–6 year old children. The Polish Chamber of Milk, with the support of members and subsidies from the Milk Promotion Fund, are the organizers of milk promotion for pre-school children. "Milky Kids" is a very simple project for parents, teachers but most of all for little kids. The project provides kids with books on milk and educational booklets. Moreover, "Milky Kids" offers meetings in kindergartens and a lot of information about milk and milk products for teachers and parents. The programme "Milky Kids" offers the website [www.dzieciakimleczaki.pl](http://www.dzieciakimleczaki.pl), where children have a lot of interesting, interactive and easy-to-use information about milk and milk products. This website is very popular among pre-school children for many reasons. For example, children like it because they can create and download stories and pictures.

The next programme "I am crazy about milk" (in Polish *Mam kota na punkcie mleka*) was started in 2012. This is a joint effort of the Polish Chamber of Milk and the Polish Federation of Cattle Breeders and Dairy Farmers. The project is subsidized by the European Commission, the Polish national budget and the Milk Promotion Fund. Again, the campaign advertises milk and milk products and educates about them on a professionally designed website, [www.mamkotanapunkciemleka.pl](http://www.mamkotanapunkciemleka.pl),



which is popular among children and teenagers. Children like using this website because they can find a lot of interesting games, quizzes, stories and information about milk and milk products. A crazy Pink Cat is the hero of this project and children know him and love him. All the material, games, information and films used in all the projects promoting milk are created in cooperation with experience teachers and school psychologists. Our professional approach eliminates all the possible dangers and misinformation.

## RESUME

Why do children in Poland like drinking milk? The answer is simple: It is a result of the joint efforts of the dairy association, farmers, producers and companies like Tetra Pak.

Together we organize interesting, very well planned and colourful campaigns to win children`s hearts for milk. And Polish children love milk and drink it with a happy smile.



# 6 The history of milk in Japanese school lunches

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## ABSTRACT

The history of milk in school lunches, the physical development of pupils, and changes in food culture will be reviewed. Basic literature and statistics about school lunches and milk were examined. Questionnaires about school lunches and milk were conducted with pupils, their parents and students.

The use of large quantities of skim milk for school lunches after World War II was due to acknowledgement that milk was highly effective in improving the nutrition of children, which was seen in increases in body weight and height. Domestic milk began to be used from about 1957, and school lunches played a large role in the consumption and expansion of drinking milk. In 1976, when school lunches began to include rice and Japanese dishes, there were complaints that milk did not fit with the meals. However, the nutritional value of milk caused a change in dietary culture to accept milk with rice meals.

## 1. INTRODUCTION

The introduction of milk in school lunches in Japan began in 1946 with skim milk [1, 2]. Skim milk and wheat flour bread became a basic part of school lunch menus. By the late 1960s, skim milk was replaced with the use of domestic milk. In 1976, school lunches began to include rice along with milk. Since 1951, school lunches have primarily included the combination of bread, milk and Western-style dishes. Present day Japanese school lunches include rice, bread and noodles as a staple food, and a variety of main dishes, side dishes, and milk. The menus are planned by the

school nutritionist. According to a national survey performed in 2010 [3], more than 70% of Japanese pupils answered that they liked school lunches because school lunches were delicious, because they could eat with friends, and because school lunches were well-balanced nutritious meals. In this study, research on the history of Japanese school lunches and milk was conducted. The purpose of this study is to review and clarify the history of Japanese school lunches and milk, the physical development of pupils, and also changes in food culture.

## 2. METHODS

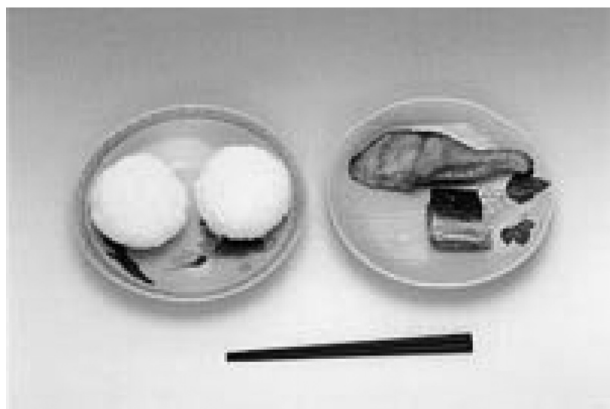
Basic literature and statistics about school lunches and milk were collected and analyzed. Questionnaires about school lunches and milk were conducted with 40 elementary school children and their parents (95.2% response rate) and 81 junior college students (100% response rate) in Nagano Prefecture.

## 3. RESULTS

### 3.1. The history of school lunches

Beginning in 1889 at a private elementary school in Tsuruoka-City, Yamagata, school lunches were provided to encourage attendance and nourish poor children. Rice balls, a grilled fish and pickles were offered on the school lunch menu (Figure 1). Until the 1940s, rice (in some regions mixed with barley or other grains) was the dominant ingredient in the Japanese diet, supplying as much as 80% of the total caloric intake (385 g of rice a day in 1935) [4]. Before World War II, traditional rice-centered school lunches such as boiled rice

containing vegetables and miso soup, or boiled rice, a grilled fish and cooked spinach, to aid poor children appeared in various regions.

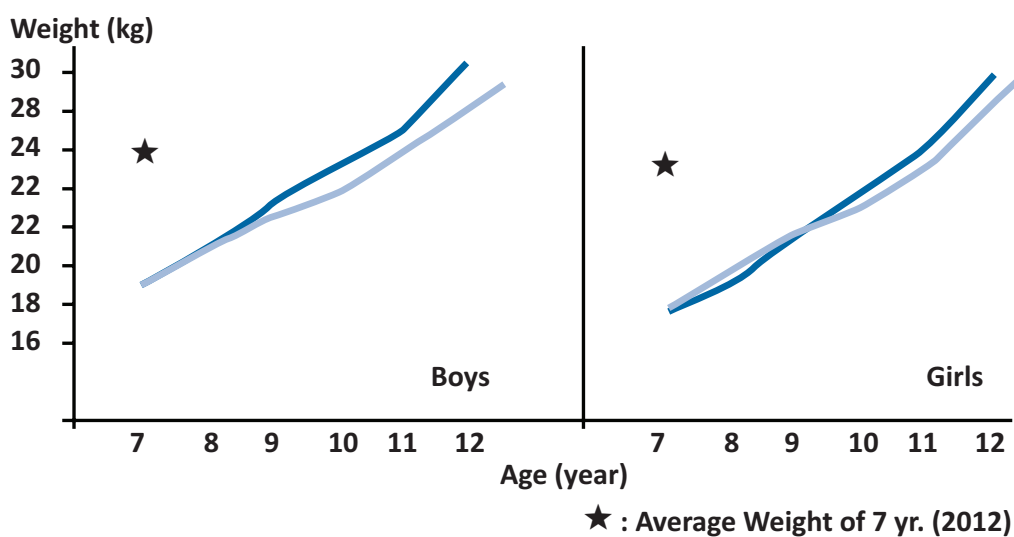


**Figure 1:** Replication of the first school lunch offered in Japan  
 Note: Rice balls, a grilled fish and pickles were offered at a private elementary school in Tsuruoka-City, Yamagata, in 1889

Source: Japan Sport Council

During World War II, almost all foodstuffs were subject to rationing. Rice rationing was initiated in 1941 and allotted 330 g a day per for a standard civilian adult. Further rice rationing decreased to 300 g a day, or less, in July 1945. In addition, the conventional school lunches were not continued because of lack of food. Rice was substituted with other foods made of wheat flour, sweet potatoes, corn, winter squash and various grains. A substitute food made of wheat flour, such as Suito, was offered in school lunches.

Figure 2 shows growth curves for the average weight of elementary school children before and during World War II [5]. The left side of the graph shows the growth curves for boys and the right side shows the growth curves for girls. The blue lines shows growth curves for sixth grade pupils in 1936, and the red lines are for 1943; the stars mark the body weight in 2012. These plots of average weight of children before and during World War II show serious malnutrition of children. After World War II, children suffered even more serious malnutrition.



**Figure 2:** Growth curves of average weight of sixth grade pupils in 1936 (blue) and 1943 (red) at Tokyo Sanya Elementary School

Source: "A report on an investigation into the present situation of national nutrition during the war, 1943" (1998)

In view of the serious situation, former US President Hoover moved Crawford Sams, GHQ Chief of Public Health and Welfare, to organize a school lunch program [4, 6]. The ministries of education, welfare and agriculture directed school lunches for all elementary school children for

physical development and nutrition education. By this official notice from three ministries in 1946, school lunch programs spread throughout the whole country.

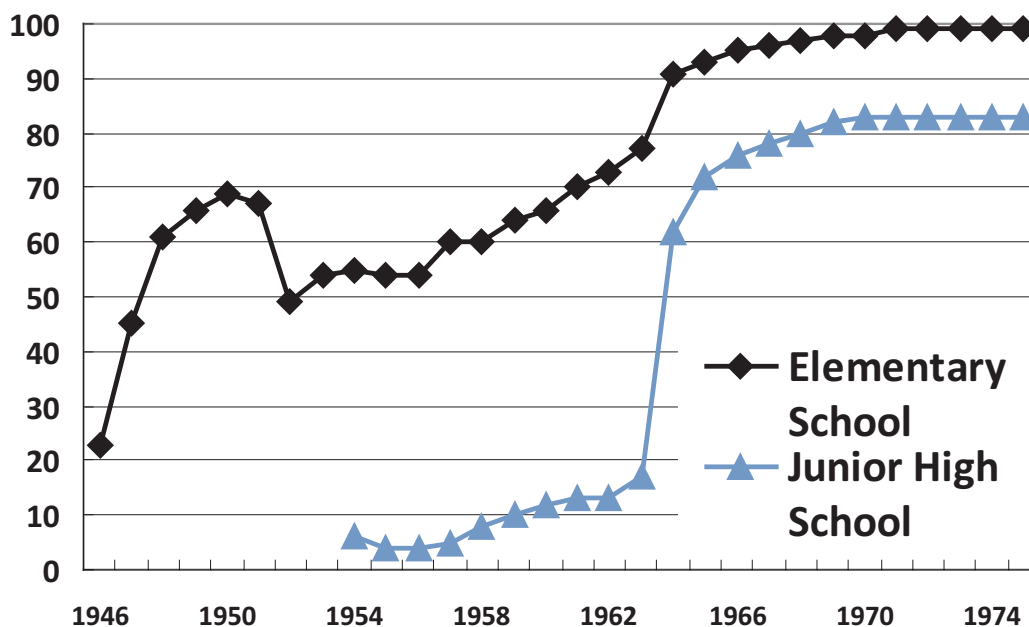
The prescription for the nutrition standards of school lunches was established in 1946 [1, 2]. The program prescribed that each school lunch should provide approximately 600 kcal, and 25 g of protein. At that time, Japan was still plagued by food shortages. Stockpiled canned foods from the Japan Imperial Army and relief supplies from LARA (Licensed Agencies for Relief in Asia) and UNICEF (United Nations Children’s Fund), such as powdered skim milk were used. School lunches were able to continue due to LARA and UNICEF relief supplies. Miso soup or tomato stew, powdered skim milk and wheat products such as sliced bread, doughnuts, or a long roll instead of rice were offered.

In 1954, the law of school lunch programs was established and it was legal grounds for the school lunch program as a part of school education. The aim of the law was nutritional improvement, increase in health, right understanding and desirable habits about meals, and a rich school life of sharing meals [1, 2]. After the end of relief supplies in 1951 and until the late 1960s, powdered milk was still one of the most cost-effective ways of improving childhood nutrition. It was purchased at a discount from the USA and by state subsidy [1, 2].

In the school lunch program law, the staple food was fixed as wheat flour bread. Wheat flour was provided at a discount by the government, which was obliged to rely on shipments of surplus wheat flour from the USA [1, 2].

Almost all school lunches from the 1950s through the early 1960s, consisted of bread, powdered skim milk, and main and side dishes. These school lunches were distinctly different from the typical Japanese rice-centered meals served in children’s homes. The staple food was fixed as wheat flour bread but main and side dishes were not only Western-style dishes, but also Japanese and Chinese-style dishes offered with skim milk. The popular and common school lunch menus were curry stew (pork, potatoes, carrots, and onions in a curry-flavored stew), whale tatsuta-age (whale meat marinated in soy sauce, coated in starch and deep-fried) and age-pan (deep-fried long rolls, coated in sugar or a combination of sugar and soybean flour).

**Rate of implementation (%)**



**Figure 3:** Changes in the rate (in numbers of children) of implementation of school lunches  
 Note: The rate of implementation in elementary schools in 2010 was 99.2% and the rate for junior high schools was 85.4%

Source: “Development of school lunches” (1976)

Figure 3 shows the changes in the rate (in numbers of children) of implementation of school lunches [2]. The black line shows elementary school children and the red line shows junior high school students. More than 90% of elementary school children since 1964 ate school lunches, so school lunch programs had a large influence on domestic meals and the food preferences of children.

Due to an increase in the domestic supply of milk and the general dislike of the smell and taste of

powdered skim milk, domestic milk started to be used in some school lunches in 1957. By the late 1960s, milk had replaced skim milk. The volume of milk per child in school lunches increased from 180 ml to 200 ml in 1970, in order to increase consumption of milk. Figure 4 shows the changes in school lunch supplies of milk, wheat flour and powdered skim milk [1, 2]. From 1964 to 1972 consumption of milk in school lunches clearly increased.

### School lunch supply (tons)

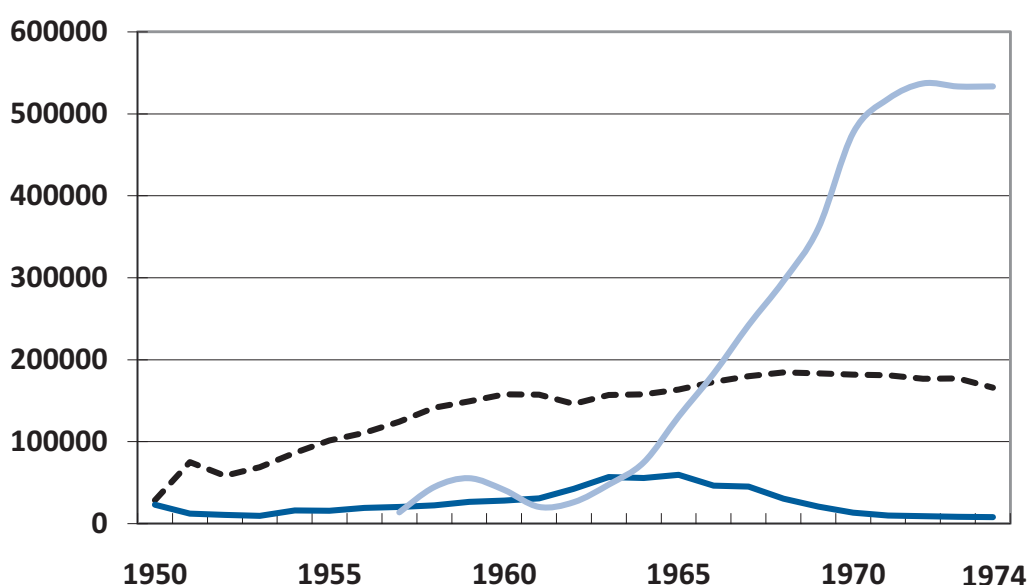


Figure 4: Changes in school lunch supplies of milk, wheat flour and powdered skim milk

Source: "Development of school lunches" (1976)

In 1976, the use of rice to dispose of surplus rice started. Thanks to the induction of rice, various main and side dishes began to be served. A smorgasbord was also conducted in school lunches for pupils to learn about quantity and nutritional

balance. Pupils learn about food and nutrition from everyday school lunches. Various dishes including Japanese dishes and milk are always served together.

### 3.2. The growth of Japanese children and the introduction of milk

The graphs in Figure 5 and Figure 6 show the growth of Japanese children from school health statistics. These graphs show changes in standing height (Figure 5) and weight (Figure 6) of boys and girls of 6 years old and 14 years old from 1900 until 2010. A comparison of children's height and weight before World War II and in 2010 shows that height increased by about 10 cm and weight

increased from 3 to nearly 10 kg. The increase in height and weight is seen clearly once children started drinking skim milk and milk. Improvement in general nutrition could contribute to these increases in height and weight. The increase in milk intake might also have a great influence on children's growth.

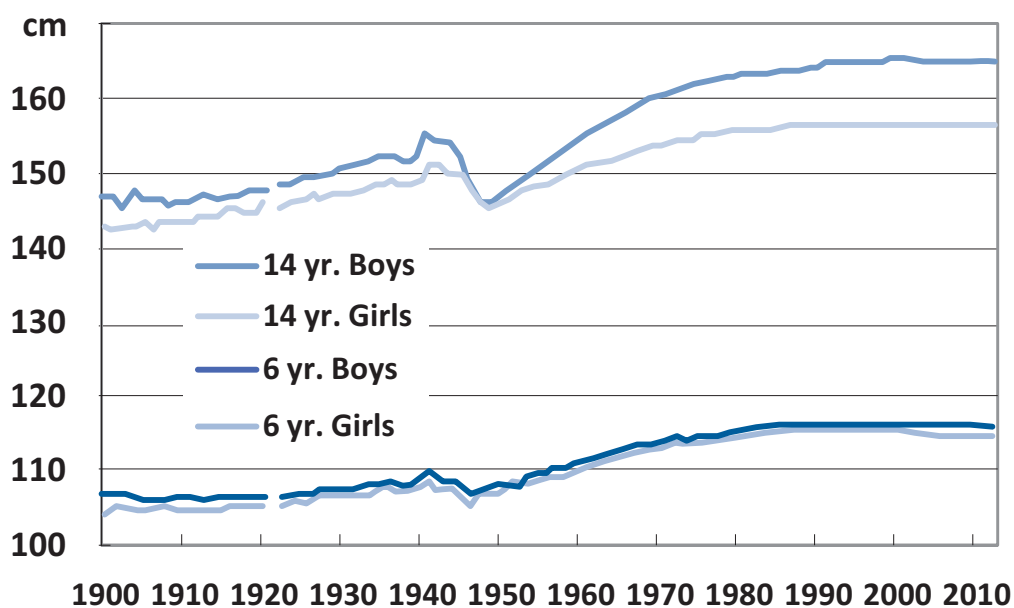


Figure 5: Changes in standing height of Japanese children

Source: “Development of school lunches” (1976) up to 1975 and “School health statistics” (2013) from 1976 onward

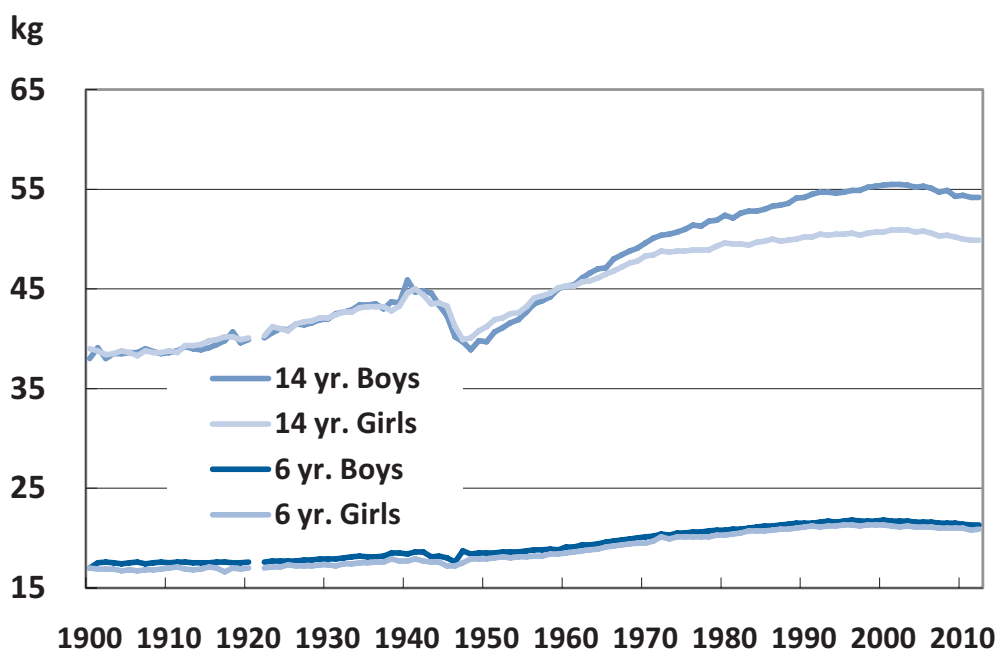


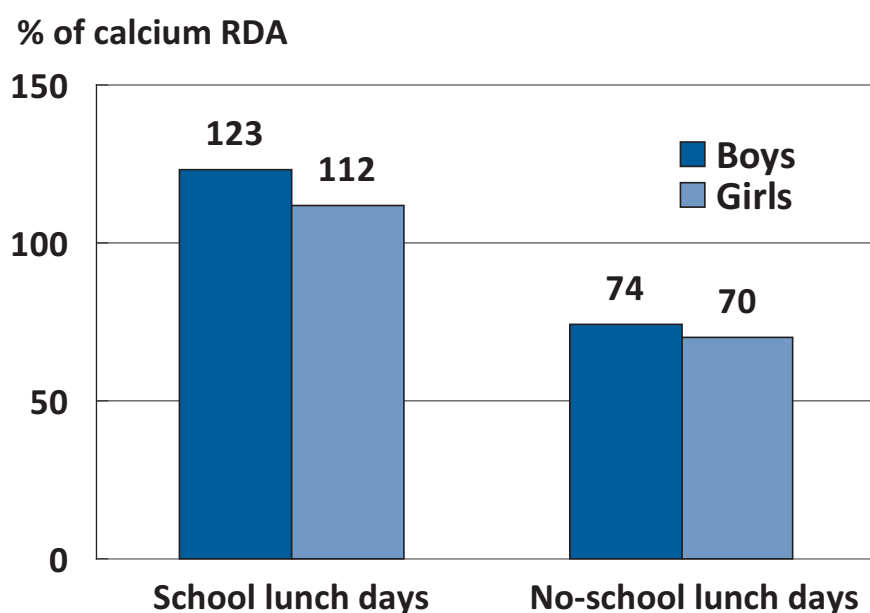
Figure 6: Changes in weight of Japanese children

Source: “Development of school lunches” (1976) up to 1975 and “School health statistics” (2013) from 1976 onward

### 3.3. Intake of calcium and milk in school lunches

Because calcium is a micronutrient that is apt to be lacking in a Japanese meal [7], 50% of the daily recommended amount of calcium [8] is to be provided in school lunches. Based on the latest results of research [3], the recommended

amounts of calcium for 6- to 11-year-old children in school lunches changed from 600 mg (1954) to 300–400 mg (2013) [1, 2, 8].



**Figure 7:** Percentage of calcium RDA of 10-year-old children

Note: *RDA* Recommended dietary allowance; calcium RDA for 10-year-old children is 700 mg

Source: "The report on the eating habits survey of children, 2010" (2012)

The graph in Figure 7 shows the present percentage of recommended calcium intake of 10-year-old children on the school lunch days (from Monday to Friday) and on no-school lunch days (Saturday and Sunday). The blue bar shows boys and the red bar shows girls. The recommended calcium intake (RDA, recommended dietary allowance) for 10-year-old Japanese children is 700 mg per day [8]. According to the bar graph, we compared the calcium intake on days with school lunch and without school lunch. There was little calcium intake of children on a day

without school lunch, and it clearly did not satisfy the recommended intake. Thus, we can conclude that many Japanese children could drink milk in school lunches and that milk in school lunches is an important source of calcium for school children. Therefore, the use of milk in school lunch programs is recommended because it can provide calcium effectively. A volume of 200 ml a day of milk is served in school lunches. A subsidy is issued for milk for school lunches. Milk for school lunches accounts for 10% of milk production nationwide.

### 3.4. Results of questionnaire about milk and rice in school lunch programs

The graph in Figure 8 shows answers to the questions, "What is an elementary school child's experience of drinking milk?" and "What is a college student's experience of drinking milk?"

Almost all children and college students drank milk without difficulty with rice and Japanese dishes in school lunches.



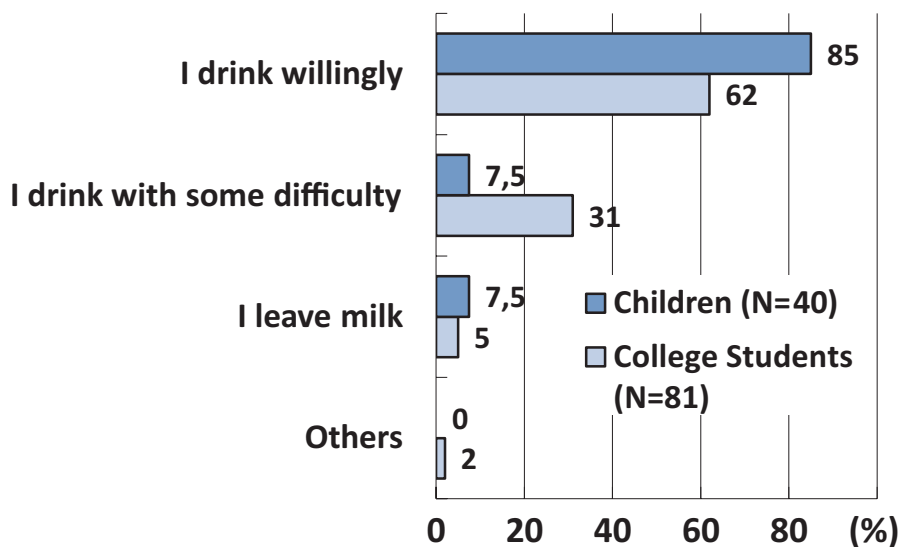


Figure 8: Experience of drinking milk with rice and Japanese dishes in school lunches

When college students were asked, “Do children need milk in school lunches?” 84% answered that school children needed milk at every meal in school lunches, 12% answered that school children needed milk every few days, and only 4% answered that school children did not need milk.

When college students were asked, “Why do children need milk in school lunches?” 46% of college students answered that children needed milk for intake of calcium and 15% said they needed it because rich nutrition was not available at home.

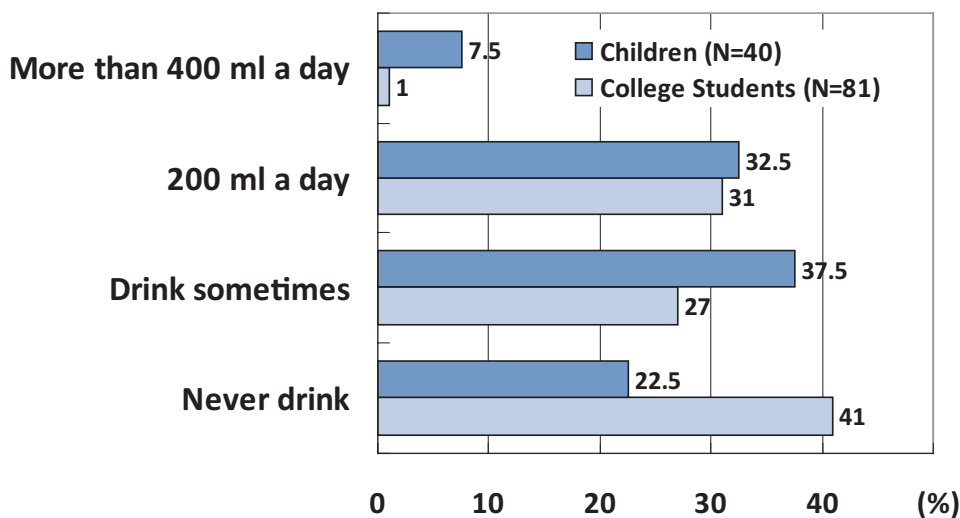


Figure 9: Amount of milk drunk at home

The graph in Figure 9 shows the answers to the question, “How much milk does an elementary school child or college student drink at home?” 22.5% of children and 41% of students never drank milk at home, and 37.5% of children and 27% of students sometimes drank milk at home. Only about 30% of children and students drank 200 ml of milk a day at home. Thus, this graph shows that there is not a habit to drink milk at home.

A minority opinion among college students and parents is that milk is not necessary in school lunches. The reasons were: a desire to receive calcium in other ways, drinking milk made the stomach too full, and that milk did not fit with Japanese meals.

### 3.5. Changes in Japanese food culture

The food preference was examined by asking college students about their favorite dishes in school lunches. Their responses show that the top seven dishes were not Japanese dishes: (1) fried chicken (23.5%), (2) gratin (22.5%), (3) curry (18.5%), (4) hamburger (13.6%), (5) noodle salad (9.9%), (6) meat sauce pasta (9.9%), and (7) fried bread (6.2%). Food preference in school lunches of 10-year-old children, according to a national survey [3] found: (1) curry and rice, (2) bread, (3) noodles, (4) dessert, (5) deep-fried food, (6) soup, (7) boiled rice mixed with vegetables etc., (8) salad, (9) hamburger, and (10) stewed vegetables. The top 10 dishes were almost all Western dishes. On the other hand, the worst 10 dishes in school lunches for 10-year-old children, according to the national survey [3] were: (1) vegetables, (2) salad, (3) seafood, (4) sautéed dishes, (5) soup, (6) dessert, (7) stewed vegetables, (8) bread, (9) vegetables with Japanese dressing, and (10) beans. Many of the dishes children disliked in school lunches were typical Japanese dishes.

According to the national survey [3], the favorite top 10 dishes for 10-year-old children were: (1) sushi, (2) curry and rice, and hashed rice, (3) omelet containing fried rice, (4) ramen, Chinese noodles in soup, (5) dessert, (6) pizza, (7) hamburger, (8) steak, (9) sashimi, fresh slices of raw fish, and (10) gratin. The top 10 dishes were almost all Western dishes, except sushi and sashimi. On the other hand, the worst 10 dishes for 10-year-old children were almost all typical Japanese dishes [3]. The worst 10 dishes for 10-year-old children were: (1) liver, (2) grilled eel, (3) salad, (4) pickles, (5) grilled fish, (6) boiled fish, (7) sweet-and-sour pork, (8) vegetables with Japanese dressing, (9) sashimi, and (10) stir-fried vegetables. Considering the questionnaire results, Japanese children's food preferences have changed and many Japanese children like Western dishes such as a meat dishes, while disliking typical Japanese dishes, such as seafood and vegetable dishes with boiled rice.

#### Daily intake (g)

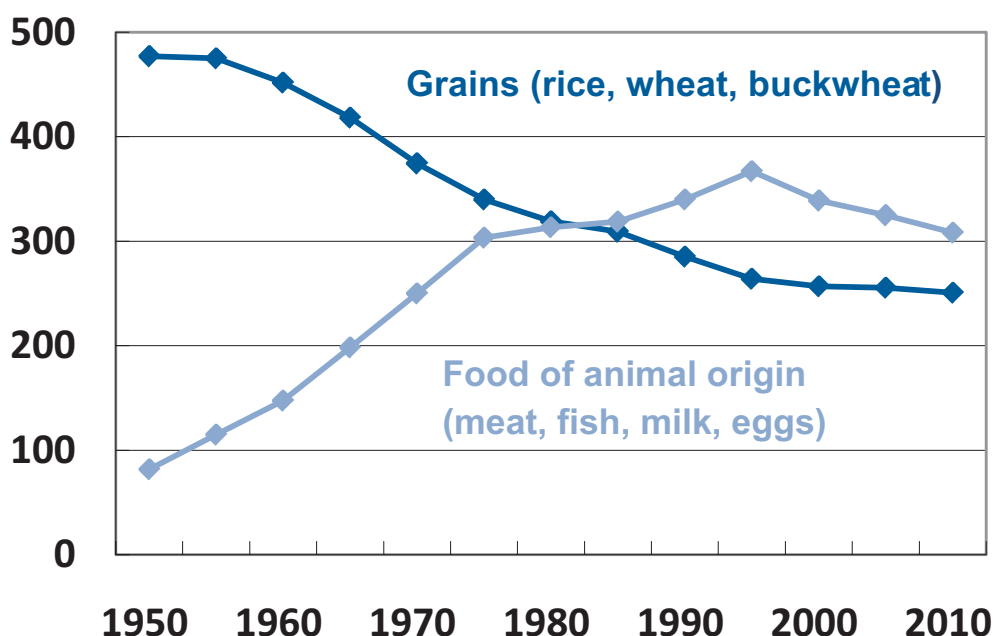


Figure 10: Changes in daily intake of grains and food of animal origin

Source: "National health and nutrition survey Japan, 2010" (2011)

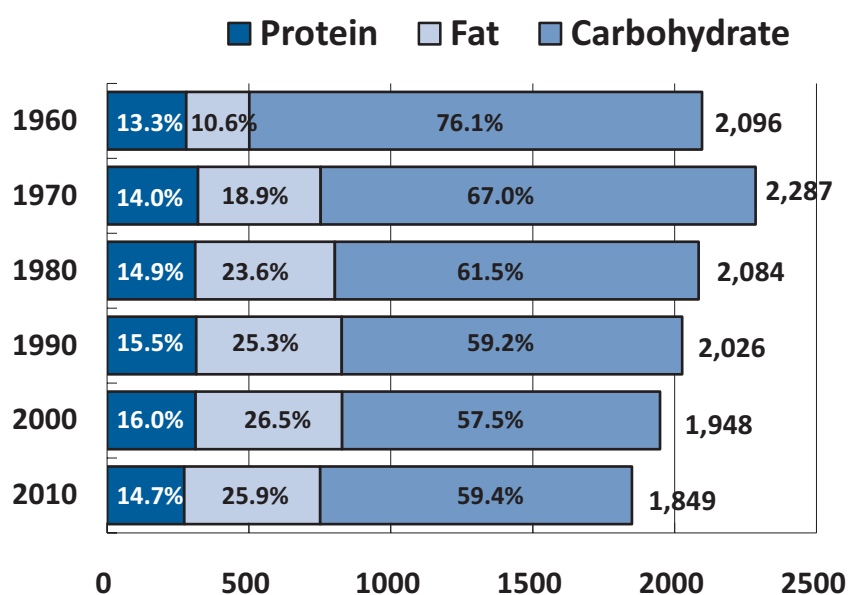


Figure 11: Changes in caloric intake of three major nutrients

Source: “National health and nutrition survey Japan, 2010” (2011)

Therefore, Japanese daily intake of grains and food of animal origin has changed. The graph in Figure 10 shows that, generally, the daily intake of grains decreased and the daily intake of food of animal origin increased from 1950 to 2010. In addition, caloric intake has changed. The graph in Figure 11

shows changes in Japanese caloric intake of three major nutrients from 1960 to 2010. It shows that, generally, the caloric intake rate of carbohydrate decreased and the rate of fat increased. The rice-centered Japanese meals have changed to Western-style meals.

### 3.6. Current issues in school lunch programs

The 2009 revision in school lunch programs attached great importance to the promotion of food education. It emphasized that school lunches are a practical teaching material, and that we should teach children right knowledge about dietary habits. In addition, understanding local traditional food culture or production, distribution and consumption of food are included.

Because of changes of eating habits and lack of exercise, life-style related diseases are increasing rapidly in Japan. School lunches influence the future eating habits and health of children. Current special issues in school lunches are the response to children with food allergies, safe security of food (food poisoning, contamination) and the use of local crops that activate local industry.

Concerning the future of milk in school lunches, I recommend the increased use of milk as a cooking ingredient. Milk is being suggested as

an ingredient that can reduce salty seasonings in Japanese dishes. Milk can also be used for new flavors in cooking.

## 4. CONCLUSION

The need to improve children’s nutrition after the war led to the introduction of milk in school lunches. School lunches changed children’s food preferences and eating habits. Even now, the nutrition from milk in school lunches has a broad influence.

## 5. ACKNOWLEDGMENTS

This study was performed with financial support by the Research Network of Dairy Social Culture and Japan Dairy Association (2012). The author truly appreciates Dr. Ayako Ehara (Emeritus Professor, Tokyo Kasei Gakuin University) for her insightful suggestions.

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# 7 The school milk scheme of the European Union

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The EU School Milk Scheme (SMS) is an overarching framework that is differently implemented in each EU Member State participating to it. The main objective of the SMS is to encourage consumption of healthy dairy products by children. This is accomplished by providing quality products for children and by contributing to a healthy way of living and to nutritional education with better knowledge on products.

In the school year 2011–2012, 26 EU Member States participated in the SMS. A total of 20 357 745 pupils received milk and dairy products with the SMS and 312 706 tons of milk products were delivered. The EU spent € 68 858 000 on subsidizing milk and dairy products for schools, and EU Member States spent € 41 440 000 to contribute to paying the difference between the EU subsidy and the actual prize. In fact, the EU subsidy does not cover the total costs and additional funding has to be found when milk and dairy are provided to school kids for free.

The SMS was established in 1977. Back then, its main aim was to ensure revenue for farmers in times of instable market, to avoid public storage of too many dairy products and to subsidize provision of milk in schools to “alleviate” the market.

Over time, the objectives of the SMS have changed and several revisions have been carried out. With the revision in 2008, several changes have been introduced to modernize the application and implementation of the SMS and to take into account the changes in eating habits and life style behaviour. For example, new and more innovative milk and dairy products have been added to the list of products that could receive

funding from the SMS budget. On the other hand, stricter nutritional criteria were introduced, e.g. the allowance for added sugars for milk and milk drinks was reduced from 10% to 7%.

The current rules for the SMS are laid down in Commission Regulation (EC) No. 657/2008. The main elements are that educational establishments from nursery schools/other pre-school establishments to primary and secondary schools can benefit from the SMS; that the maximum daily quantity per pupil on which aid can be granted is equivalent to 250 ml milk; that limitations are set on the amount of added sugars allowed in subsidized products; and that the list of eligible products takes into account different consumption habits of milk products in the EU.

There are a series of milk and dairy products that are eligible for the subsidy provided by the SMS. They are listed in the annex of the Regulation. There are five different categories of products that can be subsidized: from plain milk, to fermented yoghurt with fruit, to fresh, processed and hard cheese. There are conditions set for each category, e.g. referring to the content of added sugars in flavoured milks and fermented milks as well as the content of non-lactic ingredients. Different aid rates apply for each category.

EU Member States participating in the SMS provide information on the usage of the SMS per school year, such as the total quantity of milk and milk products provided via the SMS, the quantity of dairy products that could have been subsidized overall in that country, the number of EU pupils participating in the SMS, the amount of money spent by the EU and the additional national support.

During the last five years, there has been a 10-fold difference between the total amount of milk and milk products subsidized and what could actually be subsidized. This means that the SMS is underperforming and that not all pupils eligible to receive school milk and dairy actually do. There are several reasons for that.

The SMS provides a framework for the provision of milk and dairy products in schools with great flexibility for implementation on national level. Government bodies of the EU Member States apply for funding by the EU Commission, which provides the subsidy depending on the number of pupils receiving the milk and dairy products and which also controls the correct application of the SMS. The government bodies then work with dairies or schools directly to provide the products, the EU subsidy and in some cases additional national funding. In some EU countries, however, the parents have to pay the difference between the EU subsidy and the actual price. The products are delivered by the dairies or by retailers who also provide and maintain the facilities necessary for storage and cooling.

EU Member States can decide which milk and dairy products provided for by the SMS they would like to provide in schools. Some EU Member States only support plain milk, some plain and flavoured milk and fresh cheeses, again others support all five categories from the list of eligible products. In addition, some EU Member States apply stricter nutritional criteria than those foreseen by the SMS.

In 2011, the European Court of Auditors has carried out an evaluation of the two school food schemes subsidized by the European Union: the SMS from 1977 and the school fruit scheme established in 2009. The results showed that both schemes are not running effectively and efficiently due to various reasons. There are some issues coming from the current set up and organization of the schemes. There are also external influences such as changes in the society and the global environment that have an impact. Therefore, the European Court of Auditors asked to improve the efficiency and effectiveness of the two schemes, for greater coordination and synergy between the two schemes, for higher relevance of educational aspects and for a harmonized approach to nutrition.

In response, the EU Commission is carrying out a revision of the school food schemes this year. They have set several objectives, which can be summarized into three different areas: Firstly, to contribute to support demand and sustainable consumption of milk and dairy in the long term, to contribute to stabilize and further develop the market to support farmers, to boost local economies and support short supply chains and to promote EU agriculture. The second area covers the aim to educate children about good nutrition and the importance of healthy eating as well as where food comes from and how to value food and agriculture. Another main objective is obviously to reduce administrative burden and improve effectiveness for the application of the schemes.

In spring this year, the EU Commission carried out a public consultation, asking all interested to provide their view on the current problems of the SMS and the EU school fruit scheme, the reasons for those problems (both endogenous and exogenous) and to comment on different options suggested by the EU Commission on how to restructure the schemes and their organization in order to improve their performance. Three specific options have been proposed: separate improvements for both schemes; a greater synergy between schemes but separate financing systems; or a new framework that would support a wider variety of agricultural products, distribution of those foods free of charge, focus on local/seasonal products and short marketing chains. In addition, greater focus would be put on supporting measures for education and awareness-raising on healthy eating and nutrition, avoiding food waste and respecting the environment.

In response to the public consultation, which closed in the spring of this year, 347 replies were received. As regards to the three options proposed, none of them was supported most by all the respondents. In fact, each option was equally supported by around one third of the respondents. Dependent on the interest and focus of the respondents, added value was seen with one or other of the options. However, all respondents mentioned the importance of supporting educational measures for both schemes.

EDA also replied to this public consultation and provided concrete suggestions on how to improve the current organisation and administration of the SMS. EDA mentioned the importance of eating dairy in positively influencing children's eating habits and of introducing support for educational measures. In EDA's view, the proposed merger of the two schemes does not support the objectives set out by the EU Commission.

Some of the respondents supporting the interests of fruit and vegetables producers questioned whether milk and dairy should be provided in schools. They consider that the nutritional benefits of fruits and vegetables are undisputed while the benefits of milk and dairy products are not universally confirmed. They consider that the health message of milk is weaker than for fruit and vegetables.

The task for the EU Commission is now to analyse all the replies and to identify the advantages and disadvantages of the three options. An impact assessment of the different options is currently being carried out. In parallel with the impact assessment, the EU Commission is also analysing the results of an external evaluation of the SMS that was carried out by a consultancy company over the past few months. The results should be available within the coming months.

For EDA and the European dairy industry, it is important to continue to point out the nutrition and health benefits of milk and dairy for children, to advocate for free milk and dairy in schools, to increase the number of pupils benefiting from the SMS, to ensure maximum usage possibilities for dairy products in the SMS and to reduce the administrative burden and financial loss for dairies.

These activities have to be carried out within a rather challenging environment. In 2014, a new EU Parliament will be elected and the college of EU Commissioners will be renewed. This means that the focus and objectives of the schemes could be adjusted and emphasis put on different aspects. In addition, a revision of the EU Common Agricultural Policy is in progress, which might have an impact on the school food schemes. There is, for example, the ending of the milk quotas in 2015 and a proposed limitation of intervention measures. Also, there is discussion ongoing about the next

EU budget running from 2014–2020. There might not be sufficient financial means available to provide milk and dairy products free at schools *and* to support a broad range of educational measures to support the SMS.

There is also discussion ongoing about the nutritional implications of providing certain milk and dairy products in schools. More specifically, there are concerns about the provision of flavoured milks in schools due to the content of added sugars. In fact, some EU Member States already limit the provision of flavoured milks in schools. There are also considerations to use the SMS to re-teach children the good taste of plain milk. Many children are used to and prefer flavoured milks, which contribute to the intake of added sugars. However, when flavoured milk can no longer be distributed to schools, will pupils switch to plain milk or maybe not drink any milk at all in schools? And will those children maybe miss out on the many important nutrients contained in milk and dairy?

Our aim at EDA is to ensure an optimal regulatory environment that allows school children in the European Union to have access to nutritious milk and dairy products in order to encourage sustained consumption of milk and dairy products now and later in life.





# 8 Development of the School Milk Programme in China

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## ABSTRACT

The School Milk Programme (SMP) in China was launched in 2000, led by the central government. With the aim of ensuring high quality milk for children, the SMP puts every part of the dairy industry value chain under scrutiny, from dairy farms and manufacturing to delivery. The programme helped drive the healthy development of China's dairy industry. Moreover, the SMP continues to carry out a wide variety of student's nutrition and health education activities in schools. Systematic tests have been running to monitor the result of the programme. It has been proven that drinking milk at schools greatly improves children's physical and health conditions, especially in terms of stature and bone mineral content, etc. Milk has been widely adopted in China for its high nutrition and proven benefits to children's health. In 2012, SMP was extended to 60 000 schools in 28 provinces, and 18 million students received milk every day in school.

400 mg in 1992, 388 mg in 2002 and 500 mg in 2010 [1]. The survey data show that the dietary calcium intake of Chinese adults remained at a relatively low level over the past 30 years as compared with the Chinese dietary reference intake value for adults (800 mg per day) [2]. As well as the low dietary calcium intake among Chinese adults, it is also low among Chinese children. From 1992 to 2002, the average daily calcium intake of Chinese primary school students reached just 30–40% of the recommended adequate intake (AI) values for this age group (see Figure 1) [1]. Milk and milk products are a good food source of dietary calcium because of its high content and bioavailability [3]. However, milk and milk products play a less important role in traditional Chinese diet than do other food groups. The study indicated that milk and milk products only contribute 4.3% of the total dietary calcium intake in Chinese diet. Meanwhile, vegetable contributes 35%, rice contributes 20% and beans contribute 14% of the total dietary calcium intake [4].

## 1. INTRODUCTION

### 1.1. Low dietary calcium intake and milk consumption in Chinese population

The dietary calcium intake is at a low level in the Chinese population. By reviewing the data of Chinese national nutrition surveys from 1982 to 2010, the average daily intakes of dietary calcium of adults was calculated to be 600 mg in 1982,

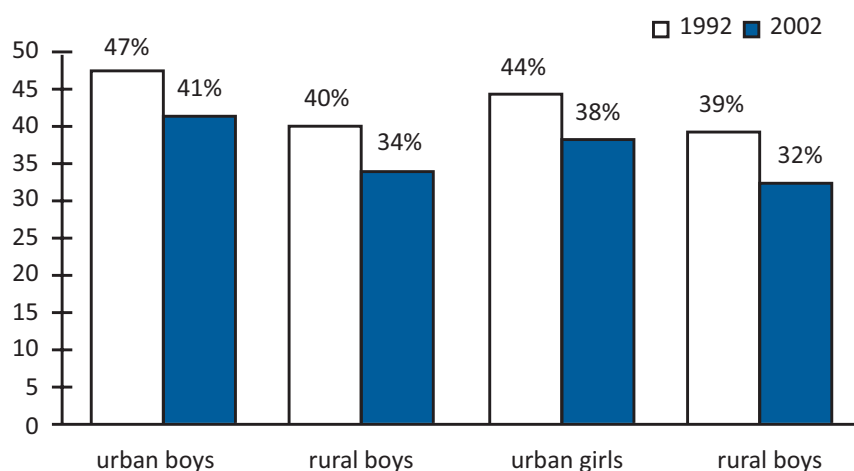


Figure 1: Daily calcium intake (percentage of adequate intake) of Chinese primary school students (1992–2002)

## 1.2. Low vitamin D level and poor bone health in Chinese children

An adequate vitamin D status is necessary for vitamin D activity within bone in order to establish a healthy skeleton. However, vitamin D deficiency is common among Chinese children. A study showed that the serum vitamin D concentration of 718 Tibetan and Han children in Sichuan Province was 15.5 ng/ml for boys and 12.5 ng/ml for girls (aged  $12.8 \pm 3.0$  years), and significant gender difference was observed ( $p < 0.05$ ) [5]. The local synthesis of 1,25-dihydroxyvitamin D within bone is necessary to modulate bone resorption and promote bone formation. Current evidence suggests that serum 25-hydroxyvitamin D levels of between 20 and 80 nmol/l (8–32 ng/ml) are associated with decreased bone mineral content (BMC) [6]. In China, the poor vitamin D status combined with low calcium intake leads to low BMC in children. Thus, the poor bone health of Chinese children needs to be paid more attention.

## 1.3. Evidence of proven benefits of milk to children's health

Recent studies provided more strong evidences of the benefits of milk to children's health. An interesting Chinese study proved the beneficial effects of calcium and vitamin D fortified milk supplementation on bone mass accretion in Chinese adolescents. In this study, 757 girls aged 10 years were randomly divided into three groups. Two intervention groups were given 330 ml calcium-fortified milk or 330 ml calcium

and vitamin D fortified milk every day. The control group was not given any supplementation. A significant total body bone mineral content (BMC) gain, height gain and sitting height gain were observed during the 2-year milk supplementation period ( $p < 0.05$ ) [7]. However, no significant total body BMC gain was observed 3 years later when the milk supplementation was stopped for all groups.

Results of a systematic review proved the positive effect of milk intake on bone mineral density (BMD) and growth in children. Compared with the control group, increasing milk intake did significantly promote growth and development of children compared to a calcium supplemented group [8].

## 2. DEVELOPMENT OF THE SCHOOL MILK PROGRAM

### 2.1. The organizers and working mechanism

Based on the previous survey and study results, the Chinese government noticed the significance of providing milk supplementation to school children, and the School Milk Program (SMP) was officially launched by the central government in November 2000 [9]. The National School Milk Office (NSMO) was established in 2000 and consists of seven ministries including the Ministry of Agriculture, Ministry of Education, Ministry of Health, Ministry of Finance, the state administration of Quality, National Development and Reform Commission, and China Dairy Industry

Association.

The SMP was organized by the central government and monitored by the regional school milk administrations. A collaborative working mechanism was established in the SMP system. The NSMO was the policy maker, the provincial SMP offices were programme coordinators, the municipal SMP offices were programme supervisors and regulators, and the licensed dairy producers were programme promoters and distributors.

## 2.2. National legislations and policy to ensure high quality of school milk

Safety, nutrition, convenience, and affordability were four key elements and characteristics of the SMP. With the aim of ensuring high quality milk for children, the SMP puts every part of the dairy industry value chain under scrutiny, from dairy farms to manufacturing to delivery. It helped drive the healthy development of China's dairy industry. Series of national standards and regulations were legislated to regulate and manage every stage of the school milk production and distribution process. For example, to ensure the safety and convenience of school milk, the regulations require that students should be provided with UHT milk; to protect the farmer's interest, the regulations require fresh raw milk must be used for production; to ensure the affordability and lower the price, the regulations require no bar-code on packages of school milk and school milk cannot be used for commercial purpose; to keep the stability of school milk market, only licensees were permitted to supply the market. In addition, the standardized process was not only required during the production process but also extended to in-school management [10].

## 2.3. Relevant activities of nutrition and health education in schools

To improve the nutrition and health knowledge and to ensure compliance with the SMP in students, a series of nutrition and health activities were promoted in schools nationwide. On one hand, the governments promoted many national nutrition and health activities to build a good school environment, such as the State Kids Drawing Competition 'Milk and Health' organized by the Ministry of Education. On the other hand,

joint publicity with some non-governmental organizations and companies played an active part in the activities in schools and achieved great progress. A successful example was the Physical Health Enhancement Programme funded by Tetra Pak China Education. It won the Best School Milk Initiative, International Dairy Federation (IDF) Dairy Innovation Awards 2010 for its remarkable achievement.

Meanwhile, the academic institutions provided strong technical support for the SMP. For example, nutrition professionals from the National Institute for Nutrition and Food Safety, Chinese Center for Disease Control and Prevention (NINFS, CCDC) edited and published many teaching materials for nutrition education in schools, for instance the Healthy Campus textbooks, Student's Nutrition Guidebooks (for different areas), a set of school posters focusing on the theme of 'Drinking milk is good for your health' etc.. The experts from NINFS also provided food safety and nutrition lectures and training programmes for students, school teachers, parents and the public.

## 2.4. Government attention and positive outcomes of regional trials inspired the promotion of SMP

In 2006, the Prime Minister Wen Jiabao suggested that Chinese children should have 500 g of milk every day. It brought the attention of local governments to children's health. Regional trials implemented in Chongqing and Xinjiang had positive outcomes and provided other local governments with confidence and models to copy. The government's increasing attention and successful experiences in trial areas inspired the promotion of the SMP, and the financial subsidy by central or/and local governments increased. The most recent national nutritional intervention for school children in China was the Nutrition Improvement Programme for Rural Compulsory Education Students (NIPRCES). In November 2011, the General Office of the State Council launched the NIPRCES. It is a school feeding programme targeting compulsory education students, normally aged from 6 to 15 years old, from the rural poverty areas. This programme annually subsidizes a total of 16 billion RMB to provide free food for students to improve their nutrition and health status by increasing their dietary intake. A total of 23 million rural students were covered

in 2012. In the NIPRCES, milk has been widely adopted for its high nutrition value and proven benefits to children's health [11].

## 2.5. Achievements and the future of SMP

With the substantial inputs and efforts made together by the government, non-governmental organizations, schools, and the public, the SMP has achieved remarkable progress in the past 12 years. In 2012, a total of 20 million students received milk every day in school. The SMP has been extended to 28 provinces, 660 cities and 60 000 schools in China [11]. In the future, the SMP will be integrated with more nutrition improvement programmes for school children in China, such as the NIPRCES. The SMP will reach and cover more schools, and more children will benefit from this national programme.

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# 9 Preventive effects against infections by intake of yogurt fermented with *Lactobacillus delbrueckii* ssp. *bulgaricus* OLL1073R-1

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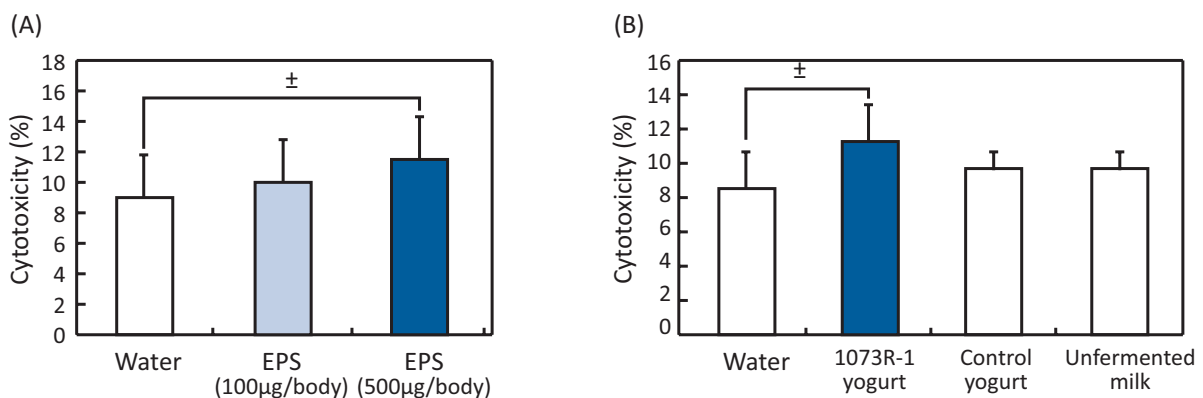
## ABSTRACT

*Lactobacillus delbrueckii* ssp. *bulgaricus* OLL1073R-1 (1073R-1) is an exopolysaccharide (EPS)-producing lactic acid bacterial strain, the EPS of which has been reported to stimulate immune cells *in vitro*. We confirmed that the oral administration of either EPS or yogurt fermented with this strain (1073R-1 yogurt) enhanced the activity of natural killer (NK) cells in mice. We also investigated whether the intake of 1073R-1 yogurt had an inhibitory effect on respiratory tract infections. In the animal study, survival periods were longer, the activity of NK cells was higher, and anti-influenza virus antibody titers were larger in the group administered 1073R-1 yogurt and EPS orally than in the water-treated groups. In the human study, the risk of catching common colds and the flu was significantly lower in the group given 1073R-1 yogurt than in the group given milk.

NK cell activity also increased in subjects who had low NK cell activity prior to consuming 1073R-1 yogurt.

## 1. PROMOTION OF NK CELL ACTIVITY IN MICE

To investigate whether the oral administration of exopolysaccharide (EPS) stimulated the immune system *in vivo*, we measured the activity of natural killer (NK) cells in mouse spleen cells. BALB/c mice were administered EPS orally at a dose of 100 µg/mouse/day or 500 µg/mouse/day. A dose-dependent increase in NK cell activity was detected in mice administered EPS. NK cell activity was significantly higher in mice administered 500 µg/body/day than in control mice administered distilled water ( $P < 0.05$ ) (Figure 1A).



**Figure 1.** Effects of orally administered EPS produced by *L. bulgaricus* OLL1073R-1 (A) and 1073R-1 yogurt (B) on NK cell activity in mouse splenocytes



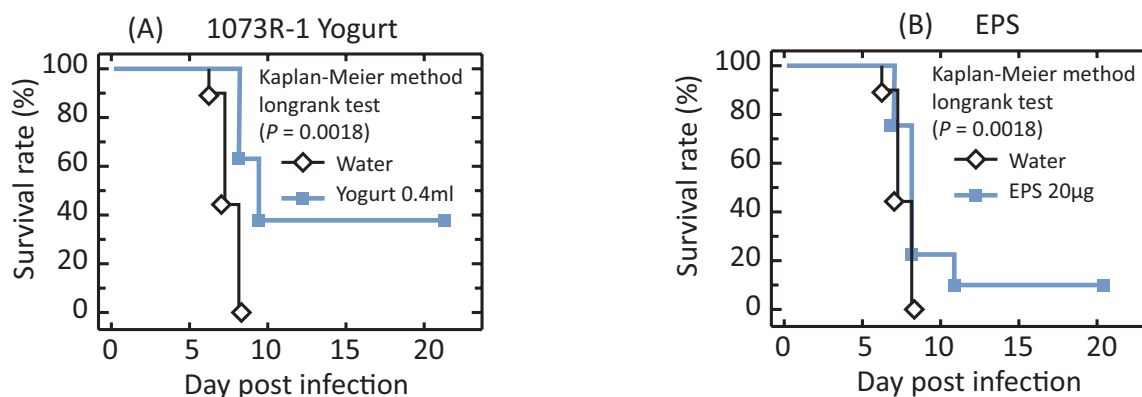
We also examined whether yogurt fermented with *L. bulgaricus* OLL1073R-1 had a similar *in vivo* immunostimulatory effect to that of EPS. Yogurt fermented with *L. bulgaricus* OLL1073R-1 and *Streptococcus thermophilus* OLS3059 (1073R-1 yogurt) were lyophilized and administered at 200 mg/day for 4 weeks. NK cell activity was

significantly higher in the spleen cells of mice administered this yogurt than in those of mice administered distilled water ( $P < 0.05$ ); however, this effect was not observed for another yogurt fermented with *L. bulgaricus* OLL1256, *S. thermophilus* OLS3295, and unfermented milk (Figure 1B) [1].

## 2. ANTI-INFLUENZA VIRUS EFFECTS IN MICE

NK cells are considered to play an important role in early defense against viral infections, and individuals with low NK cell activity were shown to be more susceptible to catching the common cold, including the influenza virus [2]. Therefore, we evaluated the anti-influenza virus activities of EPS. BALB/c mice were administered EPS orally at a dose of 20  $\mu\text{g}/\text{mouse}/\text{day}$  from 21 days before to 6 days after influenza virus infection.

Mice were infected with the A/PR8 virus by intranasally dropping 20  $\mu\text{l}$  of a viral suspension containing twice the LD50 dose, and survival rates were measured for 14 days. The survival rates of mice administered distilled water and EPS were 12.5%, and 38.9%, respectively. These results demonstrated that the oral administration of EPS significantly protected mice from influenza virus infection ( $P < 0.05$ ) [3].



**Figure 2.** Effects of 1073R-1 yogurt and EPS prepared from the culture supernatant of *L. bulgaricus* OLL1073R-1 on the survival rate of influenza virus-infected mice. BALB/c mice were orally administered water, yogurt (A), or EPS (B)

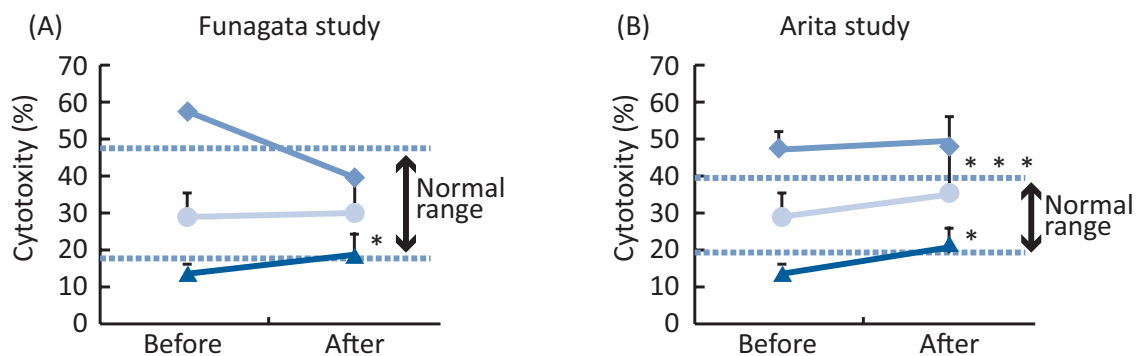
The oral administration of 1073R-1 yogurt (0.4 ml/mouse/day) or EPS at the same amount as the yogurt were also shown to prolong survival periods following influenza virus infection in mice (Figure 2) [3]. Furthermore, all mice were still alive 4 days post infection. The infectious viral titer of the bronchoalveolar lavage fluid (BALF), NK cell activity in the spleen cells, and anti-influenza virus antibody titer in the BALF were evaluated. The viral titers of yogurt- and EPS-administered mice were significantly lower than that of the water-administered group. NK cell activity in the spleen cells and anti-influenza virus IgA and IgG<sub>1</sub> antibody titers in the BALF of both the yogurt-treated and EPS-treated groups were higher than those of the water-treated group [3].

These results demonstrated that the oral administration of 1073R-1 yogurt protected mice against influenza virus infection and that these effects were partly dependent on immunostimulatory EPS. Whether NK cell activity or anti-influenza virus antibody titers largely contribute to these anti-influenza virus effects has yet to be determined.

### 3. REDUCTION IN THE RISK OF CATCHING COMMON COLDS AND THE FLU IN HEALTHY ELDERLY INDIVIDUALS

In a human study, the consumption of some probiotics was shown to enhance cell-mediated immune responses in the elderly [4, 5] and also to promote the NK cell activity of peripheral blood mononuclear cells in healthy people with low NK cell activity [6]. The number of studies investigating the effects of probiotics on respiratory infections has recently increased [7,

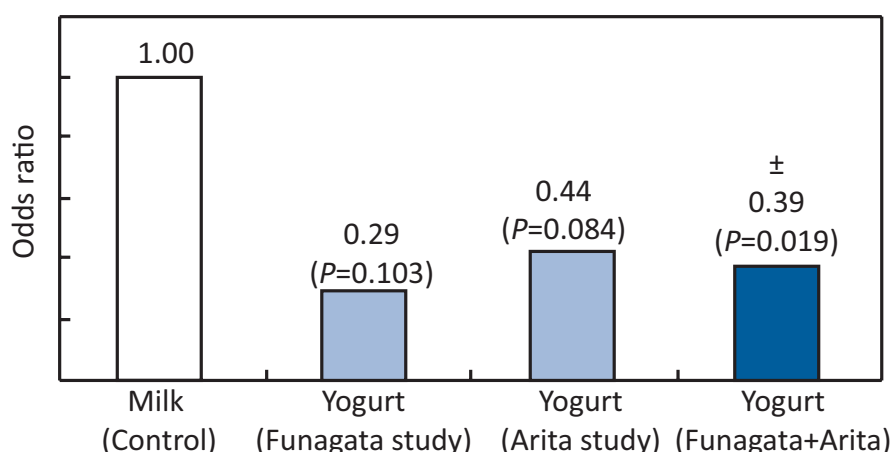
8, 9]. However, whether yogurt that contains probiotics exerts these effects has not yet been investigated. Therefore, we conducted two independent studies to evaluate whether the intake of 1073R-1 yogurt containing immunostimulatory EPS promoted NK cell activity and also had an inhibitory effect on respiratory tract infections.



**Figure 3.** NK cell activity in the 1073R-1 yogurt groups in the Funagata (A) and Arita (B) study. Subjects were categorized into low activity (▲), normal activity (●), and high activity groups (◆) according to their NK cell activity prior to consuming yogurt

A total of 57 (median age 74.5 years, range 69–80) and 85 healthy elderly people (median age 67.7 years, range 59–85) participated in these studies. Subjects were divided into two groups based on age and gender and were instructed to eat 90 g of 1073R-1 yogurt or drink 100 ml of milk once a day over an 8- or 12-week period. In the first study (Funagata study), no significant change was observed in NK cell activity before and after the intake of both milk and 1073R-1 yogurt. However, when subjects in the 1073R-1 yogurt group were classified into normal, low, and high-activity subject groups according to their NK cell activity at the beginning of the intake period, NK cell activity in the low-activity subject group significantly increased following the intake of 1073R-1 yogurt until normal ranges were reached (Figure 3A). Such an increase in NK cell activity in the low-activity subject group was also observed in the second study (Figure 3B) (Arita study), which indicated that the intake of 1073R-1 yogurt containing immunostimulatory EPS can increase NK cell activity in low-activity subjects [10]. The common cold or influenza virus infection was observed in 3 subjects in the 1073R-1 yogurt group and 8 subjects in the milk group in the

first study, whereas no influenza virus infection was noted in either group in the second study. The common cold was observed in 17 subjects in the 1073R-1yogurt group and 25 subjects in the milk group. The risk of catching the common cold or influenza virus in the first and the second study were about 3.4 times (OR = 0.29,  $P = 0.103$ ) and 2.3 times (OR = 0.44,  $P = 0.084$ ) lower in the 1073R-1 yogurt group than in the milk group, respectively (Figure 4). We performed meta-analysis by integrating the results of these two studies, the results of which revealed that the risk of catching the common cold or influenza virus was approximately 2.6 times (OR = 0.39,  $P = 0.019$ ) lower in the combined 1073R-1 yogurt group than in the milk group (Figure 4) [10].



**Figure 4.** The risk of catching the common cold in the milk and 1073R-1 yogurt groups in the two independent studies. Meta-analysis was performed by integrating the results of these two independent studies using the Mantel–Haenszel method

## 4. CONCLUSIONS

The oral administration of immunostimulatory EPS produced from *L. bulgaricus* OLL1073R-1 prolonged survival period in mice, and this may have occurred due to increases in NK cell activity and influenza virus-specific antibodies in the BALF. These results suggest that EPS plays an important role as a functional component in yogurt. Yogurt containing EPS fermented with *L. bulgaricus* OLL1073R-1 exerted anti-influenza virus effects similar to EPS in mice. Furthermore, the consumption of this yogurt promoted NK cell activity and reduced the risk of catching the common cold and flu in elderly individuals.

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# 10 Milk consumption promotion in Chile: the PROMOLAC experience

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## ABSTRACT

Promolac is the Chilean institution in charge of promoting milk consumption in the domestic market. This institution is jointly owned by milk processing companies and the National Dairy Farmer association. Following a consumer's study in 2001, a long-term milk consumption campaign was developed in two stages: The "Image Campaign", dating from 2002 to 2007, and The "Motivation and Education Campaign", which ran from 2008 to date. In 2010, an evaluation of the campaign showed that the fluid milk consumption per person increased by 19.8% for the 2000–2010 period compared with a 3.6% rise in total dairy product consumption in the same period. In 2011 after considering these evaluations, it was decided to broaden the campaign from milk to dairy products, incorporating cheese and yogurt in this first stage.

## 1. PROMOLAC BACKGROUND INFORMATION

Promolac is a Chilean institution created with the objective of promoting milk and dairy product consumption in the long run in the domestic market. Promolac was born in 1986, when it made its first campaign to promote the consumption of milk. In 2000, the dairy farmers association showed the potential for growth in milk consumption in the domestic market, in the context of a consumption of less than 130 kg per person and an increasing milk production, at an approximate ratio of 5% average between 1990 and 2000. The involvement of the dairy farmer's association and the milk processing

industry in the ownership of Promolac, and the financial support in the first stages by the Chilean Ministry of Agriculture, was an innovation for Chile. The ownership of the institution is: Dairy Processors (50%), National Dairy Farmers Association Fedeleche (30%), Association of Dairy Cooperatives (16%), and others farmer's associations (4%). The administration is conducted by a board whose members are from the processor companies and the National Dairy Farmers Association (Fedeleche). At the executive level, there is a Marketing Committee with marketing people from the processors and the dairy farmers and a general manager from Fedeleche. The executive level is in charge of the relationship with the publicity agencies. Currently, the institution is financed through a voluntary levy from the dairy farmers, an amount of money that is matched by the processors. The Chilean Ministry of Agriculture partially financially supported the institution until 2006.

## 2. THE BEGINNING

In 2001, after a complete consumer study conducted by Promolac, a long term campaign was designed. This campaign is still active to date, although many adjustments have been made following the changes detected in consumer trends through the years. The study targeted liquid white milk-like products and the main aims were to perceive the advantages and disadvantages of its consumption by consumers and the perception of the image of the product. The advantages detected by consumers were that the product was perceived as a healthy and natural product, nutritive, rich in mineral and vitamins, with a

strong relationship between milk and bone health and growth. The disadvantages were bad taste or flavorless, not sweet, very greasy, and not refreshing. Regarding product image, the study differentiated between fluid milk, fluid flavored milk and yogurt, where the fluid milk appeared with the most “boring” and “serious” image followed by flavored milk and then yogurt.

Some of the conclusion of this study showed that the main limitation in fluid milk consumption was related to the characteristics of the product in terms of flavor and consistency, and its perceptual image among young people. These limitations have restricted consumption occasions to breakfast and mid-afternoon, being replaced by other dairy products that taste better and appear more entertaining (and therefore easier to give to children). Specifically in terms of image, fluid milk was related to maternal care, maternal authority over the child, inability to take care of oneself, and elder people. It was seen as a traditional food, with few associations of interest to teens and young adults. In addition, it was also found that white fluid milk had less popularity than other dairy products, the main substitute being yogurt, and that there was no presence of milk outside the supermarket or place of consumption outside the home.

The conclusion of the study suggested that the next steps were to create an image that will reverse the current perception among children and young people aged 11–18 years with regard to milk, using communication elements that give attraction to the product and displace the attention of children from the characteristics of the product such as the taste and consistency to related attributes focused on external attributes as entertainment. The following step was to create a long-term campaign with the commercial goal being to increase milk consumption and with a strategy that included a first stage called the “Image Campaign” with the objective to change the perception of milk from a product related to mothers, old and boring to make it an attractive food, fresh and modern. The second stage was called the “Motivation and Education Campaign” with the objective being to expand and strengthen consumption occasions, forms, and reasons. The campaign was designed to be implemented annually with a budget of around 1.2 million dollars (USD). It included the production of around

ten TV advertisements played 522 times on open TV (mainly in prime time), 260 advertisements on cable TV, 48 000 contacts on supermarket activities, and 35 000 contacts on school activities, celebration of the World School Milk Day and World Milk Day, sponsorship of sport activities (mainly basketball and skiing), presence in seminars, dairy farmers markets, and gifts.

### 3. FIRST STAGE: THE IMAGE CAMPAIGN

The first stage of the campaign started in 2002 and finished in 2006 and included a study to evaluate consumers’ perceptions of the products.

The 2002 campaign was called “Confessions of a Celebrity” and had the short term advertising objectives of strongly increasing awareness of the product and positioning milk as a priority and relevant food for Chileans. The strategy was based in giving visibility and salience to milk and to impact the public, in particular young people through a clear, direct and surprising message and also overthrow negative perceptions associated with milk, making it a young, modern and intelligent drink. To implement the campaign, two famous actors, two famous actresses, one TV presenter, two sport players and two Chilean rock band singers were hired to participate in the media productions. For the TV commercial, the singer of the most famous Chilean rock band at the time (Beto Cuevas) shot a two-part commercial (teaser and resolution). In the teaser, he declared “I confess that I drink” leaving this statement open to interpretations. In the resolution part of the commercial played at the end of a prime time program, he appeared saying “I confess that I drink milk” adding all the good reasons why he did. This commercial had a big impact on TV audiences at the time and in the media, in general because it was the first time that a generic campaign for milk appeared on TV at prime time with very famous people involved and also for the strategy used.

The 2003 campaign was called “The Truth: Everyone Drinks Milk” and had the short term advertising objectives of continuing to increase awareness and keep the importance of milk as a relevant food priority for Chileans. The strategy included maintaining visibility and salience of

the first year of campaign, followed by knocking down the filters that stop milk consumption, enlarging the identification of young people with their opinion leaders, and installing the concept “The truth is that everyone drinks milk”. Following the same strategy of the year before, one famous actor, two famous actresses, one TV presenter, two football players and four musicians were hired to participate in the media productions. This year, the idea was to increase the variety of the celebrities in order to strengthen the concept. Additionally, during this year the production of merchandising and gifts to farmers were implemented to get them involve with the campaign. This strategy showed to be a strong way to make farmers proud of the campaign and also to participate in field activities.

The 2004 campaign was called “Nude Benefits” and had the short term advertising objectives of renewing impact and beginning to highlight the benefits of drinking milk. The strategy was based in on bold and provocative campaign that included the word “irreverent” as the key success of the campaign, especially among young people. For this campaign, the logo was improved, becoming a visually powerful and recognizable symbol to clearly identify its concept and intention “Yo Tomo” (“I Drink”). This campaign got a lot of attention from the consumers and from the media due to the fact that in the TV commercial the celebrities appeared showing part of their bodies to reinforce the concept of opinion leaders demonstrating the benefits of consuming milk reflected in their own bodies. This approach had a poor evaluation, even though that the campaign was much commented on and attracted a lot of attention, because the real message behind it was not well communicated.

The 2005–2006 campaign was called “Cool Milk; Young People and the Subcultures” and had the short term advertising objectives of identifying milk with youth and to start socializing milk consumption (from home to meeting places). This time, the strategy kept building the image of milk as youthful, but now oriented towards socialization and increasing consuming instances, reinforcing the concept from the individual to the collective, from indoors to outdoors, from boring to fun. Finally, the aim was to maintain the good achieved with previous campaigns: brand, claim, and new image. This time, new celebrities

representing different youth interests were hired and, for the first time, an internationally renowned signer, Shakira, was included. This campaign included several activities in summer holiday locations for young people.

In 2006, an evaluation study was conducted by Promolac to detect consumer perceptions of the product. This study showed that some of the most important obstacles toward consumption identified in the 2002 study, such as milk being a product related to boring people and sick people and its consumption relegated to within the home, were gradually becoming less important. Furthermore, the predominant associations of children, women, and the elderly, were also gone. Therefore, those results suggested that milk products were being perceived in new scenarios and in youth social spaces, spaces that until now had belonged exclusively to soft drinks, juices, coffee, and alcohol.

#### 4. SECOND STAGE: THE MOTIVATION AND EDUCATION CAMPAIGN

The second stage of the campaign started in 2007 and is still ongoing, although changes were made since 2011 following the outcomes of the consumer studies. The strategy of the campaign was to transcend from being “different/cool” to being “smart”, to conceptually understand the relevant aspects of young people’s lives so that they can be positively related to the consumption of milk, and to start making clear the “other” product’s strengths in order to bring new consumer motivations.

The 2007 campaign “I Drink, My World”, had the short term advertising objective of promoting new reasons and occasions for milk consumption and to keep milk identification with youth, with the new and modern. The strategy was to maintain the presence of milk around youth and the ideas of “Milk is always with me” and “Milk is part of my lifestyle”. This campaign maintained the media approach, although this time it did not use celebrities to support the campaign. This time, the TV commercials were made with digital images related to rock music and skateboard culture.

That year, a deeper consumer study was conducted by Promolac and showed that: those who do not consume milk do it for organoleptic reasons; the basic reason for temporary abandonment of consumption is due to lack of money (in low income families); all consumers know and value the virtues of milk as food; one of the main reasons for consumption of milk are health reasons; the supermarket is the place to buy milk; consumption had increased in all segments; low income segments were more vulnerable to price; biggest changes in consumption occurred in the section aged 26–35 years, and consumption dropped in the segment aged 50 year and over. Following these results, another study was conducted to explore the meanings around the finding related to low incomes families and their relation to milk consumption. This study showed that, according to preliminary information, consumption fell since 2008 due the loss of real income in families, added to the fact that the prices of both milk and other basic products had increased significantly during that time. However, other interesting findings were that, for these families, milk was essential and linked to such concepts as “Milk makes you beautiful”, “It is a tool towards being somebody” and “It is an investment for the future”.

The 2008–2010 campaign was called “Thanks Mom, the Thanks of the Celebrity” and had the short term advertising objective of mitigating the loss of milk consumption in low income segments generated by declining incomes. The strategy was to give more value to milk consumption and increase its priority over other products through empowering milk as a staple food, irreplaceable and of permanent consumption. For this campaign, the target group were women aged 20–55 years, who mostly do the shopping and decide what to buy, and also pre-teens and teens (8–15 years) both from low income segments. For this campaign, a very famous football player was hired who appeared in the TV advertisement with his mother. The idea of the ad was for the celebrity to thank his mother for giving him milk when he was young because that allowed him to succeed in sports.

In 2010, an evaluation of the impact of the long-term campaign on fluid milk and dairy consumption was conducted. This study showed that the campaign had been a success and had

made a real contribution to the increase in fluid milk consumption per person (19.8%) for the 2000–2010 period, compared with a 3.6% rise in total dairy product consumption in the same period.

## 5. 2011–2013: FROM MILK TO DAIRY PRODUCTS

In 2011, after considering the last evaluations of the campaign, it was decided to extend the communication from milk to milk products, incorporating cheese and yogurt in this first stage, which is still on going. The idea was transfer the concept of “Good milk” to “Good milk products” evolving from the original logo of “Yo Tomo” (“I Drink”) to “Yo Tomo, Yo Como” (“I Drink, I Eat”). The strategic approach for the campaign was that there was more choice of products, more consumption occasions, and multiple benefits associated with the different milk products. Moreover, the target group was expanded from young people to adults. Also, the “I Drink, I Eat” campaign maintained a style that constructed an identity. The objective was to communicate to consumers the diversification offered by dairy products (variety for everyone), suggest more consuming occasions, communicate that dairy products offer the same benefits as milk and the idea that dairy consumption is essential for a “good life”.

## 6. THE PROMOTION CAMPAIGN HISTORY

The campaign throughout time was focused on improving the “emotional bond” with milk, making it something “Indispensable” by a two-stage campaign, which is showed in the time line of Figure 1.

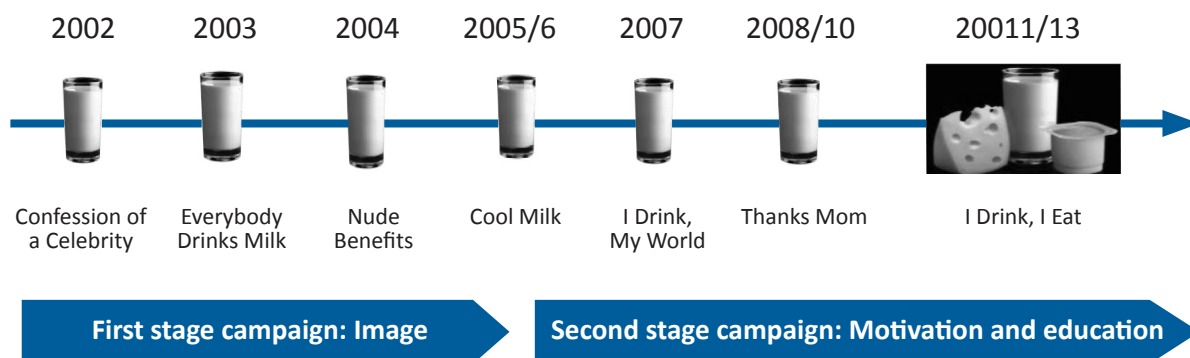


Figure 1: Time line of the milk promotion campaign in Chile

## 7. CONCLUSIONS

Promolac has achieved its goal of increasing milk consumption of Chilean consumers through an innovative campaign spanning more than 10 years. This success was possible thanks to the work of an institution with a clear and single goal, with steady funding, and with the participation of farmers and milk processing companies in its management. During the long-term campaign, the continuous research conducted through the years on the consumer's perception of milk and milk products was crucial in modifying the campaign strategies in time to meet the changing views and needs of consumers.





# 11 Biodiversity in Japanese dairy systems: Floral diversification mechanisms in grazed grasslands

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## ABSTRACT

In Japan, semi-natural grasslands were traditionally used as extensive communal grazing systems in most agricultural regions. However, following agricultural intensification or abandonment, areas of semi-natural grasslands have decreased, transformed to pasture or converted to conifer plantations. This loss of semi-natural grasslands has resulted in the listing of familiar plant species in the Red Data Book. Using experimental field trials in Hokkaido and Kyushu, this study investigated the effect of grazing and burning on maintaining plant diversity. At each site, the plant species' composition and soil properties were compared between the grazed and ungrazed treatments. This study suggests that grazing is an effective method for increasing plant species diversity in semi-natural grasslands. The study showed that modification and heterogenification of soil properties derived from livestock grazing influences environmental niche differentiation and plant diversity. The use of semi-natural grasslands and indigenous plants for dairy production would be an effective strategy for biodiversity conservation.

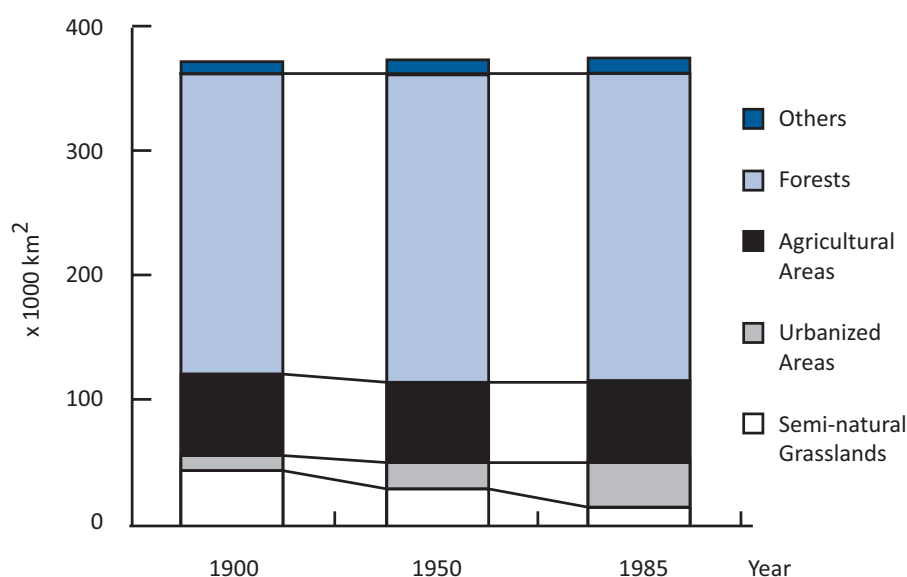
## 1. INTRODUCTION

Globally, grasslands are one of the most common terrestrial ecosystems. However, the unambiguous decline of grasslands has occurred, particularly in temperate zones. In Japan, three types of grassland occur: (1) cultivated grasslands, composed of sown grass or legume species introduced mainly from Europe and planted after the removal of native plant communities using herbicides; (2) semi-natural grasslands, composed of native herbaceous

plant species with anthropogenic disturbances such as grazing, burning or cutting, which stop the successional process to forests; (3) natural grasslands, composed of native herbaceous plant species without human disturbances because of climatic or edaphic constraints. This study investigated semi-natural grassland biodiversity and grassland use for agricultural activities including dairy production.

In Japan, semi-natural grasslands were traditionally used for extensive communal grazing systems in most agricultural regions. However, as a result of agricultural intensification or abandonment during the last 50 years, semi-natural grasslands have declined. Many semi-natural grassland areas have been transformed into artificially sown grasslands composed of introduced grass species or conifer plantations [1]. In Japan, the decline of semi-natural grasslands has resulted in many familiar plant species in the rural regions being listed in the Red Data Book. In addition, there is the cultural loss because these grasslands have been part of Japanese rural culture for decades. In 2010, as part of the National Biodiversity Strategy, the Ministry of the Environment announced that the loss of species in semi-natural grasslands occurred from changes in habitat. The issue was described as part one of the "second crisis" for Japanese Biodiversity [2].

In this paper, the author will explain how grazing maintains plant diversity in the semi-natural grasslands of Japan, presenting two case studies conducted in Hokkaido, the northernmost main island of Japan [3] and in Kyushu of Japan's south-west [4]. The author will discuss which type of dairy systems could contribute to improved rural biodiversity in Japan.



**Figure 1:** The decline of semi-natural grasslands in Japan during the last century. Modified from Himiyama et al. [1]

## 2. METHODS

To investigate the effects of grazing on plant communities, grazed and ungrazed sites (treatments) were compared. Field surveys were conducted in the grazed semi-natural grasslands in Hokkaido and Kyushu. At each field, the plant species composition and soil properties were compared between the grazed and ungrazed treatments.

### 2.1. Study site

This study sites and treatments were as previously described [3]. The study consisted of three sites: day-grazed (24 h), night-grazed (17:00–09:00) and ungrazed (control). The sites were situated on the horse-grazed boreal semi-natural grassland on the summit plain of a coastal terrace, approximately 100 m above sea level at Akkeshi, eastern Hokkaido (42°59'1"N, 144°55'37"E). The closest meteorological station to the site showed that the mean annual precipitation was 1065 mm and the mean annual temperature was 5.5°C. The soil types at the site are cumulic fine Andosols derived from fine layers of volcanic ash deposited from the Akan and the Mashu volcanoes.

The second study [4] was conducted in the Aso area, central Kyushu (33°N, 131°E). From 1995 to 1999, the mean annual precipitation and mean annual air temperature were 3 120 mm and 13.1°C, respectively, at the nearest weather

station (497 m above sea level [a.s.l.]). Mount Aso, (1592 m a.s.l.) is an active volcano. The Aso somma, forming one of the world's largest caldera basins, is 18 km wide (east to west) and 25 km long (north to south). The climax natural vegetation in most of this area consists of broad-leaf evergreen forest, except at the hilltops and the top of the somma. For this experiment, four sites (treatments) were used at the northern foot (approximately 700 m a.s.l.) of the central crater hill of Mt. Aso: grazed (by breeding beef cows) and burned; grazed and unburned; ungrazed and burned; ungrazed and unburned.

### 2.2. Vegetation survey and soil sampling

At the Hokkaido site, ten 1×1 m plots were established every 10 m along a 100-m transect for each site. At the Kyushu site, two 100-m transects were established and ten 1×1 m plots were established every 10 m. In each plot, the coverage and maximum height of each vascular plant species, total plant coverage and community height (mean height of canopy top) were measured and recorded. The accumulated dominance value (coverage × maximum height) for each species or species group was calculated. Soil surface hardness was measured using a soil hardness tester (Push-Cone, DIK-5 553; Daiki, Tokyo, Japan) at five different points at the centre of each plot and the median value calculated.

In each plot, the relative photon flux density at ground level was measured and compared with full photon flux density above the sward. At the end of the field survey, a 100-cm<sup>3</sup> soil core of undisturbed surface soil was taken from the centre of each plot.

Laboratory experiments were conducted to test the physical properties of the soil samples. Volumes of the liquid and solid phases of the soil samples were determined using a volumenometer (Three-Phase Meter, DIK-1 130; Daiki), with the air phase calculated from the remainder of the sample. Samples were oven dried at 105°C for ≥24 h, with the moisture (volumetric water content) of the samples determined. Soil particle density and bulk density were calculated. The saturated hydraulic conductivity, an indicator of water permeability, was tested using the falling-head method. Using a pressure plate apparatus (Multi-Fold pF Meter, DIK-3 440; Daiki), the water retention characteristics of the samples were determined at desorption (from -3 to -157 kPa). The air phase was determined after gravitative drainage (non-capillary pore space), an indicator of aeration. The available water-holding capacity of the soil (capillary pore space), an indicator of water availability, was also calculated.

### 2.3. Statistical analyses

The effects of grazing on species richness and the measured or calculated environmental parameters were analysed using ANOVA. The significance of differences between site means was examined using Tukey's honestly significant difference (HSD) test. The analyses on the exponentially distributed variables (that is, relative photon flux density and saturated hydraulic conductivity) were performed after the data were log-transformed. For a broader overview of multiple species, environments and plant community relationships, canonical correspondence analyses (CCA) were used.

## 3. RESULTS

### 3.1. Vegetation and species richness of plants

At the Hokkaido site, 60 plant species were identified, with 46, 36 and 37 species recorded in the day-, night-grazed and ungrazed sites, respectively. The day-grazed site showed significantly higher species richness than the other sites (Table 1). The accumulated dominance value for *Iris setosa*, a hygrophilous species targeted for conservation at Akkeshi, was significantly higher in the day-grazed site compared with the other two sites. In contrast, the dominance value for forbs at the day-grazed site was significantly lower compared with the other two sites (Figure 2).

Site	Species richness
Day-grazed	22.8 ± 0.6 <sup>A</sup>
Night-grazed	16.4 ± 1.3 <sup>B</sup>
Ungrazed	15.4 ± 1.3 <sup>B</sup>

**Table 1:** The mean species richness (m<sup>-2</sup>) for each site [3]

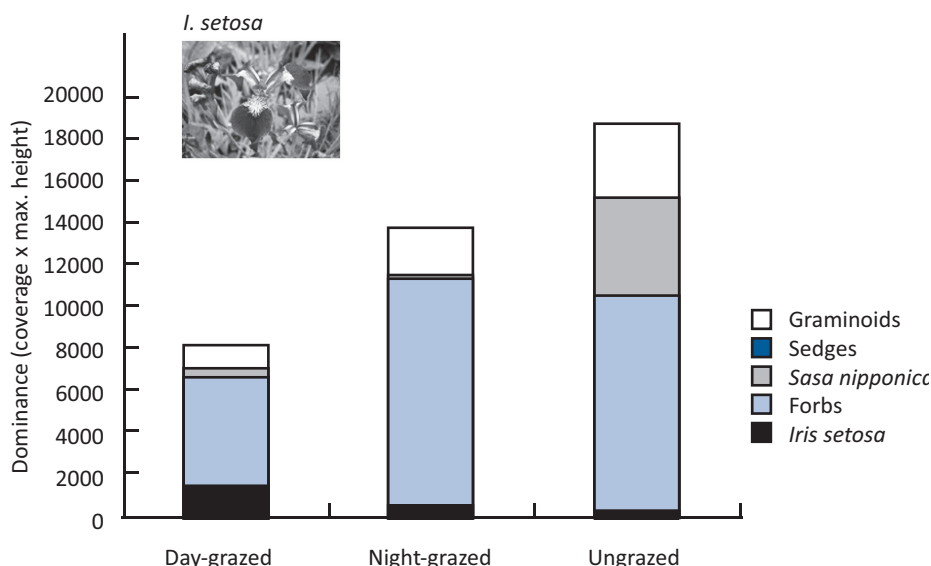


Figure 2: Accumulated dominance value for each species group [3]

At the Kyushu site, 77 species were identified in plots with higher species richness ( $m^{-2}$ ) in the grazed compared with the ungrazed site (Figure 3). Both grazing and burning significantly increased species richness, but a significant negative

interaction occurred between the two treatments. *Miscanthus sinensis* (tall-type C4 grass) was dominant in 38 plots at all the sites and *Zoysia japonica* (short-type C4 grass) was dominant in 12 plots at the grazed and burned site (Figure 4).

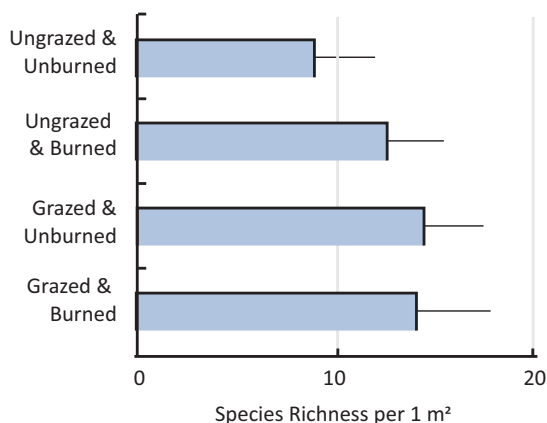


Figure 3: The mean species richness ( $m^{-2}$ ) for each site [4]

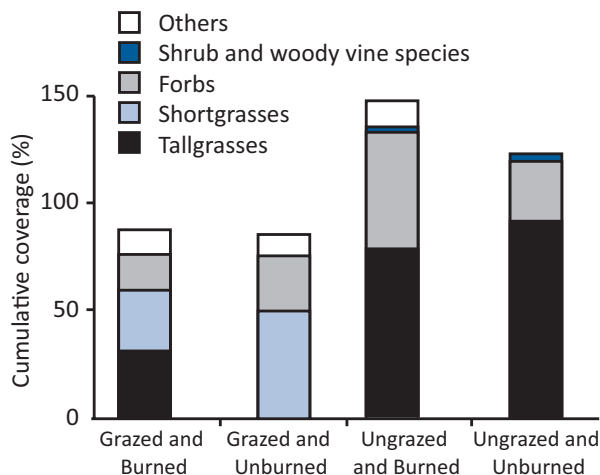


Figure 4: The mean coverage of each species group among sites [4]

### 3.2. Light conditions and soil properties

At the Hokkaido site, light intensity at ground level was slightly higher in the day-grazed site compared with the other two sites. This difference was not significant (Table 2). Aeration (air phase after gravitative drainage) and water permeability (saturated hydraulic conductivity) of the surface

soil were significantly lower in the day-grazed site compared with the other two sites. The results suggest that wetter soil conditions were caused by grazing on the day-grazed site. There was no significant difference between sites for the other soil properties.

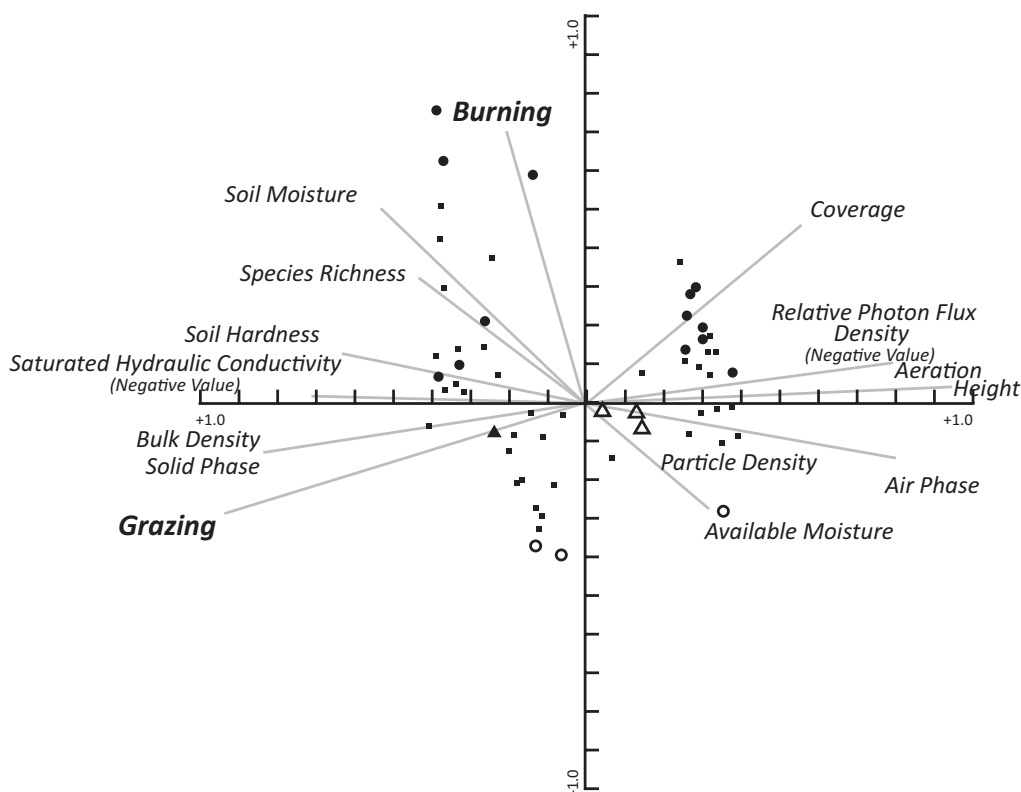
Site	Relative photon flux density	Air phase after gravitative drainage	Saturated hydraulic conductivity (log-transformed)
Day-grazed	0.042±0.014	7.3±0.6 <sup>A</sup>	2.92±0.09 <sup>A</sup>
Night-grazed	0.028±0.013	14.7±2.1 <sup>B</sup>	2.36±0.13 <sup>B</sup>
Ungrazed	0.031±0.011	15.6±2.5 <sup>B</sup>	2.33±0.21 <sup>B</sup>

**Table 2:** Light conditions at the ground level and soil physical properties for each site [3]

At the Kyushu site, the relative photon flux density at ground level significantly increased with grazing and burning, probably because the C4 grasses suppressed growth. There was no significant interaction between grazing and burning. The non-capillary pore space, which contributes to soil aeration, declined in the grazing and burning treatments, with a significant interaction between grazing and burning. Grazing significantly decreased the hydraulic conductivity, an indicator of water permeability. In contrast, burning significantly increased the hydraulic conductivity. A significant interaction between grazing and burning decreased the hydraulic conductivity.

### 3.3. Results of the canonical correspondence analyses

At both study sites the canonical correspondence analyses (CCA) suggested that each plot for the different sites showed a distinctive distribution. The grazing (and burning at the Kyushu site) caused different species composition and environmental conditions. For the Kyushu site, the results indicating grazing and burning effects cross nearly perpendicular to each other, with the bar indicating species richness on the interior angle (Figure 4). This suggests that grazing and burning enhance species richness but in different ways.



**Figure 4:** Canonical correspondence analysis showing species–environment relationships. Each solid circle, open circle, solid triangle, open triangle and dot in the diagram indicates colourful-flowering, shrub, short-type grass, tall-type grass and other species score, respectively [4]

## 4. CONCLUSION

This study suggests that grazing is an effective method of increasing plant species diversity in semi-natural grasslands in Japan. Furthermore, grazing is not an alternative method to burning because of the different effects between grazing and burning on plant species' composition. At present, fewer dairy cow herds are grazed in semi-natural grasslands and animal production depends on intensified systems. The wise use of semi-natural grasslands and indigenous plants for dairy production would be an effective method for biodiversity conservation.

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# 12 Revision of Codex Microbiological Criteria guideline

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## ABSTRACT

The Codex Alimentarius “Principles for the establishment and application of microbiological criteria (MC) for foods” (CAC/GL21-1997) was revised in 2013 to update the principles in line with the latest knowledge and practices, to introduce the new risk management metrics and other quantitative microbiological limits and to provide guidance on the relationship between MC and the new microbiological risk management (MRM) metrics. This article outlines the changes made during revision of the document.

## 1. INTRODUCTION

Microbiological Criteria is a risk management tool. The Codex Alimentarius “Principles for the establishment and application of microbiological criteria (MC) for foods” (CAC/GL21-1997) was adopted in 1997. After the adoption of emerging microbiological risk management metrics in Annex II of the “Principles and guidelines for the conduct of microbiological risk management (MRM)” (CAC/GL 63-2007), the Codex Committee on Food Hygiene (CCFH) decided to undertake a new work on the revision of the Codex MC document to update the principles in line with the latest knowledge and practices, to introduce the new risk management metrics (food safety objective, performance objective and performance criteria) and other quantitative microbiological limits (e.g. process control criteria, testing for hazard analysis and critical control point, HACCP, verification), and to provide guidance on the relationship between MC and the new MRM metrics. The CCFH began revision of the MC document in 2010, and the new main document was adopted at the 36th

session of the Codex Alimentarius Commission meeting in July 2013. During the course of the elaboration, seven practical examples on establishing and applying MC were developed by seven working groups. Each working group consisted of a lead country and three to five mentee countries. This initiative is a better use of the Codex Trust fund because it enhanced the involvements of developing countries from the drafting stage, strengthened capacity building in each participating developing country, and facilitated understanding of the contents of the main document.

## 2. TITLE

The title was modified from “Principles for the establishment and application of microbiological criteria (MC) for foods” to “Principles and guidelines for the establishment and application of microbiological criteria related to foods”. This is due to the fact that the document includes both principles and guidelines. The original MC document focused on the lot acceptability, but the new document also covers the evaluation of food safety control system or process control, which is a part of the food safety control system. Therefore “MC for foods” was modified to “MC related to foods”.

## 3. STRUCTURE

The new document is restructured and has (a) a new Section 3 on general principles; (b) combines Sections 4 and 5 into a Section entitled “Establishment and application of MC”; and (c) creates in Section 4 a new sub-section “Purpose” to highlight the multiple purposes of MC.



Sections 4.9 “Moving window” and 4.10 “Trend analysis” were created, based on the experiences gained during the preparation of the practical examples. These two subsections are kept

separate because trend analysis is not part of an MC and should preferably be described in a stand-alone section in order to avoid confusion.

Table of contents in 1997	Table of contents in 2013
Introduction 1. Definition of microbiological criterion 2. Components of microbiological criteria for foods 3. Purposes and application of microbiological criteria for foods 3.1.1 <u>Application by regulatory authorities</u> 3.1.2 <u>Application by a food business operator</u> 4. General considerations concerning principles for establishing and applying microbiological criteria 5. Microbiological aspects of criteria 5.1 Microorganisms, parasites and their toxins/metabolites of importance in a particular food 5.2 Microbiological methods 5.3 Microbiological limits 6. Sampling plans methods and handling 7. Reporting	1. Introduction 2. Scope and definitions 2.1 Scope 2.2 Definitions 3. <u>General principles</u> 4. Establishment and application of microbiological criteria 4.1 General considerations 4.2 Purpose 4.3 <u>Relationship between microbiological criteria other microbiological risk management metrics and ALOP</u> 4.4 Components and other considerations 4.5 Sampling plan 4.6 Microbiological and/or other limits 4.7 Analytical methods 4.8 <u>Statistical performance</u> 4.9 <u>Moving window</u> 4.10 <u>Trend analysis</u> 4.11 <u>Actions to be taken when the microbiological criterion is not met</u> 4.12 Documentation and record keeping 5. <u>Review of microbiological criteria for foods</u>

**Table 1:** Comparison of the tables of contents of CAC/GL21-1997

## 4. DEFINITION

In order to reflect broad scope and current use of MC, the definition of MC was modified as follows:

- Old: A microbiological criterion for food defines the acceptability of a product or a food lot, based on the absence or presence, or number of microorganisms including parasites, and/or quantity of their toxins/metabolites, per unit(s) of mass, volume, area or lot.
- New: A microbiological criterion is a risk management metric which indicates the acceptability of a food, or the performance of either a process or a food safety control system following the outcome of sampling and testing for microorganisms, their toxins/metabolites or markers associated with pathogenicity or other traits at a specified point of the food chain.

The important change is that previously the MC focused on the acceptability of a specific lot, but now the MC determines not only acceptability of a lot, but also the performance of either a process or a food safety control system.

## 5. GENERAL PRINCIPLES

The revised document has eight general principles, which highlight key principles for the establishment and application of MC:

- A MC should be appropriate to protect the health of the consumer and where appropriate, also ensure fair practices in food trade.
- A MC should be practical and feasible and established only when necessary.
- The purpose of establishing and applying a MC should be clearly articulated.



- The establishment of MC should be based on scientific information and analysis and follow a structured and transparent approach.
- MC should be established based on knowledge of the microorganisms and their occurrence and behaviour along the food chain.
- The intended as well as the actual use of the final product by consumers needs to be considered when setting a MC.
- The required stringency of a MC used should be appropriate to its intended purpose.
- Periodic reviews of MC should be conducted, as appropriate, in order to ensure that MC continue to be relevant to the stated purpose under current conditions and practices.

## 6. TOWARDS A RISK-BASED APPROACH

Risk analysis is being used increasingly in the CCFH. There must be a link between the level of hazard in a food, and the risk for the consumer.

A MC can be linked directly to the ALOP, without explicit articulation of a food safety objective (FSO) or a performance objective (PO). One approach involves testing the acceptability of individual lots and evaluating the relative risk to public health of the lot as compared to the appropriate level of protection (ALOP). Another approach is to link a MC directly to an ALOP, using a risk assessment model to estimate the reduction in public health risk as a result of applying corrective actions to lots or processes that do not conform to the microbiological criterion.

Ongoing efforts to reduce the complexity of risk assessment may facilitate the development of “true” risk-based MC.

Statistical models can be used to translate a PO or FSO to a MC. The link between the PO or the FSO and the ALOP should also be demonstrated. To establish such a MC for a food, an assumption needs to be made regarding the distribution of the target microorganism in the food. A log-normal distribution is often assumed and a default value for the standard deviation applied.

Furthermore, the maximum frequency and/or concentration of the hazard need to be defined in the FSO or PO. If a concentration is used as

a limit, also the proportion (e.g. 95%, 99%) of the distribution of possible concentrations that satisfies this limit should be defined.

## 7. RELATIONSHIP BETWEEN MC, ALOP AND OTHER MRM METRICS

MC may be used by competent authorities and FBOs, to operationalize the ALOP either directly or through other microbiological risk management metrics (e.g. PO, FSO). This requires the use of quantitative risk assessment. The risk estimation (itself based on source attribution studies) should include a combination of factors such as the prevalence and concentration distribution of target microorganisms, as well as any changes in these after the step for which the MC has been set.

The risk assessment should include a characterization of the variability inherent to the food production system and express the uncertainty in the risk estimate.

## 8. APPROACH

When considering the establishment of microbiological criteria, a variety of approaches can be used depending on the risk management objectives and the available level of knowledge and data.

These approaches can range from developing microbiological criteria based on empirical knowledge related to good hygiene practice (GHP), to using scientific knowledge of food safety control systems such as through HACCP, or by conducting a risk assessment.

The choice of the approach should be aligned with the risk management objectives and decisions relating to food safety and suitability

## 9. PURPOSE

There may be multiple reasons for establishing and applying MC. The purposes of MC include, but are not limited to, the following:

- Evaluating a specific lot of food to determine its acceptance or rejection, in particular if its history is unknown
- Verifying the performance of a food safety control system or its elements along the food chain, e.g. prerequisite programs and/or HACCP systems
- Verifying the microbiological status of foods in relation to acceptance criteria specified between food business operators
- Verifying that the selected control measures are meeting POs and/or FSOs
- Providing information to food business operators on microbiological levels, which should be achieved when applying best practices.

## 10. MOVING WINDOW

In a moving window approach, a sufficient number of sample units ( $n$ ) are collected for a defined period of time (the “window”). The results of the latest  $n$  sample units are compared with the microbiological limits ( $m$ ,  $M$ ) using the acceptance number  $c$ .

As a new result from the sampling period is available, it is added to the window while the oldest result is removed, creating the “moving window”.

This approach can also be applied to a set of results, e.g. results obtained during a week. The window, always consisting of  $n$  results, moves one result or set of results forward in time.

The moving window approach is a practical and cost-beneficial way of checking continuous microbiological performance of a process or a food safety control system. As in the traditional point-in-time approach commonly used in connection with MC, the moving window determines the acceptability of the performance so that interventions can be made in case of unacceptable shifts in control.

The length of the moving window should be appropriate to enable corrective action to be taken in a timely manner. If more than  $c$  out of  $n$  results is above the limit  $m$ , or the limit  $M$  is exceeded, then corrective action is required.

The moving window approach is a specific application of trend analysis used in process control that employs the addition of a decision rule to determine when the process can be considered to be out of control.

## 11. TREND ANALYSIS

Trend analysis is a procedure to detect a change in the patterns of observations over a period of time (usually over a relatively long period of time, often not predefined). It can be applied to many types of information including results of microbiological testing against a microbiological criterion. Trend analysis can detect a gradual loss of control that might not be detected by a moving window approach, as well as a more sudden loss of control.

Trend analysis may show changes or patterns in the data that are a result of unwanted changes in the manufacturing process, enabling the food business operator to take corrective actions before the food safety control system is out of control. The trends (or patterns) can be visualized, e.g. by displaying the test results graphically.

## 12. CONCLUSIONS

MC could be utilized as a risk management tool by both competent authorities and food business operators, and should be used in combination with GHP and HACCP. A risk-based MC could be established and achieve risk reduction, which is estimated by microbiological risk assessment. Further guidance on the mathematical and statistical aspects of MC will be developed to provide more detailed information of the development and implementation of MC in foods.

# 13 Feasibility and issues in the application of HACCP at the farm level in Japan

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<sup>3</sup>Japan Food Hygiene Association

<sup>4</sup>Inoue Dairy Farm Company

## ABSTRACT

HACCP criteria have been verified over the past three years on pilot farms in terms of producing safe fresh milk and improvements in animal health and milk production. Based on certification criteria, we conducted a hazard analysis of routine activities on the model farms according to the Codex General Principles of Food Hygiene, identified hazards, and specified control measures. Then, we established a HACCP plan for CCPs, and assessed the effectiveness of implementation of the plans with viable bacteria counts, somatic cell counts of fresh milk, contamination of antibiotics, or from the cows' condition and milk production. With comparison of pre- and post-implementation of the HACCP system for the pilot farms, the safety and productivity of raw milk have been improved.

Moreover, productivity improvement includes increased milk production, decreased ratio of cows with abnormal milk production, and increased survival ratio of cows at 12 months old.

## 1. APPLICATION OF FARM HACCP TO DAIRY FARMS

### 1.1. Establishment of hygiene control system

The certification criteria of farm HACCP (hazard analysis and critical control point) consist of (1) establishing an organizational structure that enables management systems to work properly, and (2) establishing pre-requisite programmes and HACCP plans (see Table 1).

[I] Establishment of organization structure that allows its management system

- Hygiene control and goal setting
- Conduct of instruction and training
- Analysis, evaluation, improvement, update of information
- Internal verification, management review

[II] Preparation for hazard analysis

- Establishment of prerequisite programs
- Hazard analysis
- Establishment of HACCP plans
- Document control and recordkeeping

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**Table 1:** Requirements for farm HACCP certification

Model farm owners assembled HACCP teams and established hygiene management systems according to their hygiene management policies and goals. The Japan Food Safety Verification Organization (JVO) supported the teams as an advisor.

Prior to hazard analysis, we documented the traffic patterns of employees, equipment and cows in flow diagrams, and the routine (regular and irregular) activities in order to exactly assess the existing condition of hygiene control. On that basis, we conducted a hazard analysis in accordance with the eight requirements of the general principles of animal hygiene, which are based on the Codex general principles of food hygiene.

Considering the aspects of hygiene management for feeding, milking, and raw milk, we completely

identified and listed hazards. We identified the hazards and developed written procedures like SOPs (standard operating procedures) and SSOPs (sanitation standard operating procedures) that define how and what to perform in order to prevent hazards. Following comprehension of all the hazards corresponding to the eight requirements of the Codex general principles, we determined critical control points (CCPs) and established HACCP systems in line with the HACCP system and the guidelines for its application.

Many of “the countries with advanced HACCP system” adopt good manufacturing practice (GMP) or good agricultural practice (GAP) as the prerequisite program (PRP); however, there are no such standards for dairy farms in Japan. Therefore, we adopted preventive control measures (procedures) for hazards, except for the above-mentioned CCPs as PRP (see Table 2).

Code of practices	Procedures	Recordkeeping
Facility maintenance / hygiene control	Tank cleaning	Inspection
	Excreta disposal/cleaning	
Pest control		Deratization and confirmation of efficacy
Waste management	Disposal of dead animals/bugs	
Hygiene control for employees/visitors		Disinfection of visitors (incl. vehicles)
Breeding management, etc.	SSOP, Artificial insemination, etc.	Work log Vaccination Observation checklist, etc.
Total: 13	Total: SSOP:1 Procedures: 23	Total: 16

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**Table 2:** Prerequisite program

## 1.2. Determination of CCPs in establishing HACCP systems

The CCPs used were (1) identification of cows to be milked to prevent contamination by drug residues and (2) raw milk temperature in bulk tanks.

### **Identify cows to be milked**

Critical limit: Milk is antibiotic/antimicrobial residue-free or of below detection limit.

Monitoring includes identifying cows treated with drugs for mastitis or other troubles by means of red duct tape on the tail and hind legs. These are checked against ear-tag numbers written on a white board, and number agreement is confirmed between red mats and red-taped cows. When discrepancy occurs, the cow is milked into a separate bucket and the milk is dumped (see Figure 1).

**Critical limits • Monitoring**

**Critical limits**

drug residues, e.g. antibiotics, antimicrobials zero or below measurable limits

**Monitoring**

- Match between the no. on a whiteboard and the ear tag no. of cows w/red tapes
- Match between the no. of cows w./no milk shipped and the no. of red mats

**Corrective action**

**Corrective action**

- The milk should be discarded after bucket milking



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Figure 1: HACCP plan 1. CCP: choose cows to be milked

**Bulk tank temperature**

Critical limit: Milk temperature is less than or equal to 8°C one hour after milking.

Monitoring includes visual checking and recording temperature of the tank by milkers at the completion of milking and checking that there is no deviation from critical limits by using a self-recording thermometer. In case of a deviation, the milk is dumped.

Even though a program may be good in content, inadequate training brings no change in the workplace. We drew up thorough programs that were even and moderate in volume, and offered training. Employee’s comprehension was determined by multiple choice tests or skill tests; and if found incomplete, we repeated the training.

**2. APPLICATION OF HYGIENE MANAGEMENT SYSTEM AND THE RESULTS**

**2.2. Effectiveness of farm HACCP**

**2.1. Training of employees**

When applying farm hygiene management system, we provided training for employees using our own training programs. The program for less-experienced employees covered codes but not SSOP procedures; the program for mid-career and experienced employees covered every procedure including SSOPs contained in PRP, codes, and the HACCP plan.

To evaluate the effectiveness of farm HACCP, we compared the results for 2012 when the training was completed and the application started, with those for 2010. The year 2011 was a training period and was therefore excluded from the comparison.

Viable bacterial counts, somatic cells counts, and drug residues including antibiotics were measured as an index of safety. Average milk production per cow, cow numbers with abnormal milk production, and survival rate of 12-year-old cows were measured as an index of productivity (see Table 3).

## 1) Protect safety of raw milk

1. Prevent increases in viable bacterial counts, e.g. *Staphylococcus aureus*
2. Prevent drug residues, e.g. antibiotics, antimicrobials
3. Prevent increases in somatic cells counts

## 2) Improvement in productivity (1)

1. Increase in milk production (average per cow)
2. Decreasing ratio of cows w/abnormal milk production
3. Stability in survival rate of 12-month-old cows

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**Table 3:** Effectiveness of implementing HACCP at pilot farms

Viable bacterial counts, somatic cells counts, and drug residue levels showed slight improvement after implementation of HACCP, but no significant differences. This is because these parameters were already low in 2010. No drug residues were detected either before or after the implementation.

Average milk production, cow numbers with abnormal milk production, and survival rate of 12-year-old cows after application significantly improved compared to pre-application (see Table 4)

	Before instruction/training (2010)	After instruction/training (2012)	Statistical test
Average milk production/cow (kg)	31.4	34.0 (**)	0.0028
Ratio of cows w/abnormal milk production (%)	3.22	1.90 (**)	0.0045
Survival rate of 12-year-old cows (%)	94.0	96.6	—

$$t_o = \frac{|\bar{X} - \bar{Y}|}{\sqrt{U_e \left( \frac{1}{m} + \frac{1}{n} \right)}} \quad P < 0.01 **$$

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**Table 4:** Improvement in productivity

### 2.3. Qualitative assessment by owners

After application of farm HACCP, employees' attitudes to work changed: from the feeling of being forced to do work, to working under their own motivation. Employees understand the significance of hygiene management and what each task means, and proactively make suggestions for improvement.

## 3. DISCUSSION

On 4 January 2011, the Food Safety Modernization Act (FSMA) was signed into law in the USA and the FDA published a draft proposal on HACCP. In the near future the operation will start. Consequently, only those food products (including agricultural

and livestock products) produced or manufactured under the management of a HACCP system can be marketed in the USA. This will inevitably be applied to foreign-produced products, and Japan must export products produced under the hygiene management system in compliance with the FSMA.

HACCP will probably be required as a government mandate in "countries with advanced HACCP system" in the near future. Along with such worldwide trends, Japan needs to accelerate the nationwide implementation of HACCP.

Experts from around the world highly appreciate what we showed during the presentation, such as the usefulness of farm HACCP, adequate analysis,



bulk temperature and identification of cows to be milked as CCPs, development of unique means such as distinguishing cows by red tape and mats. They recognize that a world-class HACCP system has been developed and implemented appropriately.

#### 4. ACKNOWLEDGMENTS

We have established a promotion committee so that the farm owners were able to conduct model business adequately and effectively. We would like to express appreciation to committee members for the worthwhile support and guidance from a comprehensive viewpoint. We would also like to thank co-researchers who actively participated in the model project and promoted it, and the people in Inoue Dairy Farm, Gumma, who understood the idea of the project and willingly worked with us. In addition, we thank officials of Animal Health, Ministry of Agriculture, Forestry, and Fisheries of Japan for offering us the opportunity to work on the project.

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COMPILATION OF SOME PRESENTATIONS FROM THE IDF WORLD DAIRY SUMMIT 2013  
YOKOHAMA, JAPAN

## ABSTRACT

This Bulletin contains thirteen papers presented at conferences during the IDF World Dairy Summit in Yokohama, Japan from 28 October to 1 November 2013. The conferences topics were Children and milk, Animal health and welfare, Dairy farming, Nutrition and health, Marketing, Environment and Food safety.

*Keywords: Biodiversity, Cheese, Chile, China, Food safety, HACCP, Japan, Marketing, Metabolomics, Milk, Poland, School milk, Thailand, Turkey, Volatile organic compounds, Yoghurt*

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Example: 1 Singh, H. & Creamer, L.K. Aggregation & dissociation of milk protein complexes in heated reconstituted skim milks. *J. Food Sci.* 56:238-246 (1991).

Example: 2 Walstra, P. The role of proteins in the stabilization of emulsions. In: G.O. Phillips, D.J. Wedlock & P.A. William (Editors), *Gums & Stabilizers in the Food Industry* - 4. IRL Press, Oxford (1988).

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"	Usually double quotes and not single quotes
? !	Half-space before and after question marks, and exclamation marks
±	Half-space before and after
microorganisms	Without a hyphen
Infra-red	With a hyphen
et al.	Not underlined nor italic
e.g., i.e.,...	Spelled out in English - for example, that is
litre	Not liter unless the author is American
ml, mg,...	Space between number and ml, mg,...
skimmilk	One word if adjective, two words if substantive
sulfuric, sulfite, sulfate	Not sulphuric, sulphite, sulphate (as agreed by IUPAC)
AOAC <u>INTERNATIONAL</u>	Not AOAC!
programme	Not program unless a) author is American or b) computer program
milk and milk product	rather than "milk and dairy product" - Normally some latitude can be allowed in non scientific texts
-ize, -ization	Not -ise, -isation with a few exceptions
Decimal comma	in Standards (only) in both languages (as agreed by ISO)

No space between figure and % - i.e. 6%, etc.

Milkfat	One word
USA, UK, GB	No stops
Figure	To be written out in full
1000-9000	No comma
10 000, etc.	No comma, but space
hours	∅ h
second	∅ s
litre	∅ l

the Netherlands

Where two or more authors are involved with a text, both names are given on one line, followed by their affiliations, as footnotes

for example A.A. Uthar<sup>1</sup> & B. Prof<sup>2</sup>  
<sup>1</sup> University of .....  
<sup>2</sup> Danish Dairy Board .....

IDF does not spell out international organizations

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