



Progress Report 2004 and Application for Continuation in 2005

For research projects financed by grants from
The Directorate for Food, Fisheries and Agro Business
under the Danish Ministry of Food, Agriculture and Fisheries

1. Research program

Research in organic farming 2000-2005 (DARCOF II)

2. Project title and number

Production of organic milk of high quality considering the future demands for use of organically produced feed and natural vitamins (II-2)

3. Head of project

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6. Project period (month, year)

Start of project:0102

End of project:1206

7. Midterm description of the project, its results and progress, and application for continuation in 2005

Project summary

Future regulations for organic production of milk stipulate that all feed must be organically produced, and that the cow can only be fed natural vitamins and antioxidants. This leads to feeding changes of such importance that it must be presumed to influence the antioxidative capacity and thus the shelf-life of the milk. The project aims at elucidating the nature of the consequences that these new regulations for organic feeding and use of natural vitamins and antioxidants for cows in organic milk production will have on the antioxidative capacity of the milk. New studies show that uric acid in milk is an important antioxidant of significance for the oxidative stability of milk. During the project, it is the aim to increase the supply of selenium and the development of endogeneous antioxidants (uric acid and glutathione peroxidase) in order to produce oxidatively stable milk and dairy products.

Table A.1: Work package list (from application)

No.	Work package title	Participants*	Budget (1.000 DKK)	Start	End	Deliverable no(s):
1	Antioxidative capacity of raw milk	<u>Jacob Holm</u> <u>Nielsen</u>	1668	0102	1205	D1-D11
2	Cheese production and oxidative stability of cheeses with high levels of uric acid and selenium	<u>Leif H. Skibsted</u>	330	0104	1205	D5, D8 &D12

* Responsible participants are underlined

Objectives and expected achievements

To elucidate the effect of introducing new rules for organic feeding and use of natural vitamins and antioxidants on the antioxidative capacity of the milk from cows producing milk under organic conditions. Furthermore to increase the selenium supply and the formation of endogenous antioxidants (uric acid and glutathione peroxidase) of the cow in order to produce oxidatively stable milk and dairy products.

The aim consists of the below intermediate aims:

- To study the variation in content of pro- and antioxidants in organic milk from a number of herds in relation to regional variation, feeding and season
- To study whether the natural isomer of vitamin E is a more effective antioxidant than the synthetic isomer of vitamin E
- To study whether the natural carotenoid content in milk has an antioxidative effect
- To study whether cheese exposed to light shows improved antioxidative characteristics if it is based on milk with a high uric acid content
- To make guidelines on how to improve the oxidative stability of the organic milk through

feeding

Achievements

The project aims at investigating the possibility to create processed dairy products (cheese) with an oxidative stability up to the same standards as conventionally produced dairy products. The project will introduce the possibility of using endogenously formed antioxidants (uric acid and glutathione peroxidase) and natural vitamin E to improve shelf-life of organic dairy products. The project is holistic and comprises the chain from stable to table. It involves inter-institutional cooperation and is based on expertise at both KVL and at DIAS.

C. Midterm results and progress

C.1 Description (summary) of main results and conclusions

Survey of composition of conventional and organic milk

In the conventional milk production in Denmark, the use of corn silage has increased during recent years at the expense of the use of grass silage. This change in feeding strategy has affected the composition of the milk in a direction, where milk fat is more saturated while the content of important antioxidants as tocopherols and carotenoids has declined. However, in the organic milk production the use of grass silage is still very important, and on this background we found it interesting to make a survey of the general composition of untreated conventional and organic raw milk samples collected from silo tanks on organic and conventional dairy plants. The milk has been examined for the content of the following important anti- and prooxidants:

Fatty acid composition

Tocopherols

Carotenoids

Uric acid

Copper

The results clearly shows that organic milk has a lower content of the synthetic isomers of α -tocopherol. That organic milk contains significantly higher concentrations of *RRR*- α -tocopherol, *b*-carotene, lutein and zeaxanthine. There no significant difference in the content of CLA in the two milk types.

A study of the effect of synthetic α -tocopherol on the oxidative stability of milk with a high content of unsaturated fatty acids has been conducted.. After 16 days of supplementation with *all-rac*- α -tocopheryl acetate the ratio of the synthetic α -tocopherol stereo-isomers increased to 11.2% of the total α -tocopherol content in the milk. The cows were receiving in average 907 mg natural α -tocopherol from the feed per cow per day and 2282 mg *all-rac*- α -tocopherol from the vitamin supplement, which gave a total of 3189 mg *all-rac*- α -tocopherol per cow per day. If the contribution of α -tocopheryl stereo-isomers with no discrimination between the stereo-isomers had to be 37.4%, 26.8% and 35.8% of *RRR*- α -tocopherol, the *2S*-stereo-isomers, and the syn-

thetic 2*R*-stereo-isomers, respectively. However, the actual distribution of the isomers in the milk was 88.8%, 11.2% and 0.0% of *RRR*- α -tocopherol, the synthetic 2*R*-stereo-isomers, and the 2*S*-stereo-isomers, respectively. This result clearly show that only a minor part of the synthetic isomers are transferred to the milk. The amount of α -tocopherol in the milk increased with 29 % however the milk with the highest concentration of α -tocopherol was found to be more unstable to induced (light or copper) than the milk with the low α -tocopherol content. Kamal-Eldin *et al.* (1996) has described that α -tocopherol can act as a co-prooxidant when present at high concentrations together with known prooxidants like transition metal ions, lipid peroxides or other prooxidants. In the present study where the milk have a high content of polyunsaturated fatty acids and oxidation is accelerated by storage in light or by addition of copper(II) it can be hypothesed that the combination of oxidative stress and strong prooxidant as linoleic and linolenic acid, α -tocopherol will act as a co-prooxidant.

The results will be published in:

The effect of α -tocopherol supplementation to the feed of dairy cows on the oxidative stability of milk

Tina Lund-Nielsen^a, Ivan Nielsen^a, Hanne Damgaard Poulsen^b, Leif H. Skibsted^c, Jacob Holm Nielsen^{a*}.

Collecting and analysis of milk from herds using different feeding strategies have started and the samples have been analysed for

Fatty acid composition

Tocopherols

Carotenoids

Results will first be evaluated when Tina Lund-Nielsen returns from maternity leave.

WP2 Cheese production and oxidative stability of cheeses with high levels of uric acid and selenium

The first months have been used for simple production of cheese and improvement of analytical methods

C.2 Fulfilment of deliverables and milestones

(To be completed for each work package)

WP1 Antioxidative capacity of raw milk, DIAS	Time schedule	Deviations, if any*
Deliverables		
1 Report for the Danish Dairy Board	10/2002	Finished
2 Report for the Danish Dairy Board	04/2003	Finished
3 Report for the Danish Dairy Board	10/2003	Finished
4 Report for the Danish Dairy Board	04/2004	Finished
5 Report for the Danish Dairy Board	10/2004	Finished
6 Final report for FØJO and the Danish Dairy Board	12/2005	
7 Paper for Mælkeritidende	2003	Published
8 Paper for Mælkeritidende	10/2005	

9 The shelf-life of organic milk in relation to feeding (paper)	8/2003	See publication list
10 The antioxidative capacity of natural vitamin E in milk (paper)	6/2005	
11 Mechanistic study of vitamin E i MFGM (paper)	8/2005	
13 Report for the Danish Dairy Board	4/2005	
14 Report for the Danish Dairy Board	10/2005	
15 Report for the Danish Dairy Board	4/2006	
16 Report for the Danish Dairy Board	10/2006	
17 Defence of Ph.D. thesis	12/2006	
Milestones		
1 Characterization of milk from organic herds finished	5/2005	
2 Variation in the oxidative stability of milk	5/2005	
3 Feeding experiment with natural vitamin E finished	3/2005	
4 Model experiments with vitamin E incorporated in the fat globule membrane of milk	3/2005	
5 Light-induced processes in cheese are described in relation to the level of natural antioxidants	12/2004	
10 The antioxidative capacity of natural vitamin E in milk (paper)	6/2005	
11 Mechanistic study of vitamin E i MFGM (paper)	8/2005	
13 Report for the Danish Dairy Board	4/2005	
14 Report for the Danish Dairy Board	10/2005	
15. Report for the Danish Dairy Board	4/2006	
16 Report for the Danish Dairy Board	10/2006	
17 Defence of Ph.D. thesis	12/2006	
Milestones		
1 Characterization of milk from organic herds finished	5/2005	
2 Variation in the oxidative stability of milk	5/2005	
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4 Model experiments with vitamin E incorporated in the fat globule membrane of milk	3/2005	
5 Light-induced processes in cheese are described in relation to the level of natural antioxidants	12/2004	
WP2 Cheese production and oxidative stability of cheeses with high level of uric acid and selenium	Time schedule	Deviations, if any*
Deliverables		
5 Report for the Danish Dairy Board	10/2004	Finished
8 Paper for Mælkeritidende	10/2005	
12 Influence of urate and selenium on light-induced oxidative changes in cheese	12/2004	
Milestones		

5 Light-induced processes in cheese are described in relation to the level of natural antioxidants	12/2004	
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** Deviations are to be further discussed in D*

[Deliverables er forskellige former for offentligt tilgængelige produkter (artikler, rapporter, informationsmøder etc.) og det skal angives, hvilken form for produkt der er tale om. Milestones er væsentlige trin i forskningsprocessen. Der skal angives et tidspunkt for både deliverables og milestones (milestones ligger typisk tidligere end deliverables). Ændringer i deliverables og milestones forsynes med en notits om, at de er ændret i forhold til ansøgningen, og hvorfor de er ændret. Alle deliverables og milestones skal stå i rapporten, og de, der er passeret, mærkes af som udførte, eller der angives en ny dato. Større afvigelser kommenteres i D.]

D. Description of deviations and subsequent adjustments of plans

Due to maternity leave of the ph.d-student Tina Lund-Nielsen will the project be prolonged with 12 months to the end of 2006

Project publications and other products

1. Products from Organic Eprints archive

Submitted for peer-review but not yet accepted

Stagsted, Jan and Nielsen, Jacob H. (2004) [Purification of glutathione-binding proteins from bovine milk and identification of glutathione S-transferase](#). [preprint]*

Not peer-reviewed

Lund-Nielsen, Tina; Nielsen, Jacob H.; Skibsted, Leif and Nielsen, Ivan (2004) [The effect of a-tocopherol supplementation to the feed of dairy cows on the oxidative stability of milk](#). Working Paper.*

Nielsen, Jacob Holm; Lund-Nielsen, Tina and Skibsted, Leif [Higher antioxidant content in organic milk than in conventional milk due to feeding strategy](#). Online at <http://www.darcof.dk/enews/sep04/milk.html>>. Newsletter *

Østdal, Henrik; Weisbjerg, Martin; Skibsted, Leif and Nielsen, Jacob H. (2004) [Antioxidative capacity of milk with a high urate content](#). Working Paper.*

2. Other products (oral presentations, public meetings, field days, etc.)

* 25-75% financed by DARCOF

** 5-25% financed by DARCOF

F. Scientific education

Ph.d-student Tina Lund-Nielsen is employed at the project.

G. National and international cooperation

Since March 2004 participating in the EU programme “Improving quality and safety and reduction of cost in the European organic and “low input” food supply chains” coordinated by professor Carlo Leifert, University of Newcastle upon Tyne . Jacob Holm Nielsen will be responsible for a survey of milk composition in relation shelf life in Danish, Swedish, English and Italian milk.

Critical reflection on the project

The dairy industry is very interested in the results from the survey of organic and conventional milk and there will in the future be applied for money to continue developing feeding strategies in order to produce organic milk with another composition than the conventional

milk.

8. Budget

A. Account for any change in budgets

B. Budget for the whole project (1.000 DKK)

Total consumption of funds from DARCOF and expected consumption this year and coming years

Year:	Consumption before 2003	Consumption 2003	Expected consumption 2004	Budget 2005	2006	Total
Man-months						
Scientific personnel	2.9	5.5	9.8	7.4	3	28.6
Technical personnel	0.9	6.7	6	3.8	0	17.4

Year:	Consumption before 2003	Consumption 2003	Expected consumption 2004	Budget 2005	2006	Total
Salaries						
Scientific personnel	100	188	349	272	101	1010
Technical personnel	20	143	140	87	0	390
Other operational costs	20	78	120	27	20	265
Equipment						
Others (please specify)						
Direct costs	140	409	609	386	121	1665
Indirect costs (20% of direct costs)	28	91	122	77	15	333
Total	168	500	731	463	136	1998

Comments:

9. Signatures and stamps

Name	Institute	Date	Signature
Head of project			
Jacob H. Nielsen	Dept. of Food Science, DIAS		

Appendix I. Detailed budget

A. Budget for each participating institute (1.000 DKr)

Name of Institute and department: DIAS, Dept. of Food Science

Year:	Consumption before 2003	Consumption 2003	Expected Consumption 2004	2005	2006	Total
Man-months						
Scientific personnel	2.9	5.5	3	7.4	3	21.8
Technical personnel	0.9	6.7	6	3.8	0	17.4

Year:	Consumption before 2003	Consumption 2003	Expected consumption 2004	Budget 2005	2006	Total
Salaries						
Scientific personnel	100	188	124	272	101	785
Technical personnel	20	143	140	87	0	390
Other operational costs	20	78	70	27	20	215
Equipment						
Others (please specify)						
Direct costs	140	409	334	386	121	1390
Indirect costs (20% of direct costs)	28	91	67	77	15	278
Total	168	500	401	463	136	1668

Comments:

B. Budget for each participating department (1.000 DKK)

Name of Institute and department:

Year:	Consumption before 2003	Consumption 2003	Expected consumption 2004	Budget 2005	2006	Total
Man-months						
Scientific personnel			6.8			6.8
Technical personnel						

Year:	Consumption before 2003	Consumption 2003	Expected consumption 2004	Budget 2005	2006	Total
Salaries						
Scientific personnel			225			225
Technical personnel						
Other operational costs			50			50
Equipment						
Others (please specify)						
Direct costs			275			275
Indirect costs (20% of direct costs)			55			55
Total			330			330

Comments: