

**PhytoMilk**

**Final report**

for

**Project no. 2006-1921**

**PhytoMilk**

**Potential improvement of the salutary effects of organic milk by forage species and by supplementation**

**Period covered: (Final: 01.07.2007 – 30.06.2011)**

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## **Project Summary, including objectives and expected outputs**

Dairy milk and milk products have been challenged due to their relatively high concentration of saturated fatty acids (SFA) and, therefore, are considered to be associated with cardio-vascular disease (Pedersen et al., 2004) and cancer. However, recent research has revealed a number of ingredients in milk that may be salutary. Among these bioactive components are phytoestrogens (e.g. lignans and isoflavonoids), fatty acids (FA, e.g.  $c9t11$  C18:2 (CLA)), vitamins (e.g. vitamin A and E), and endogenous hormones and growth factors (e.g. estradiol, IGF-I and TGF- $\beta$ ).

Due to a higher proportion of forage in the ration, with high proportion of legumes and other herbs, organic milk quality is more and differently affected by the forage than conventionally produced milk that often is based on grass silage. There is limited knowledge regarding chemical and sensory characteristics of organic dairy milk internationally that has focused on organic grassland management and the milk salutary properties.

The objectives of the proposed project were to:

- investigate how different forage species affect the fatty acid composition of organic dairy milk, and the milk content of bioactive components such as tocopherols, carotenoids, selenium (Se) and phytoestrogens
- investigate the biological activity of the collected dairy milk samples from the Nordic countries on normal and cancer cells

# 1. Main results, conclusions and fulfilment of objectives

## 1.1 Summary of main results and conclusions

### Summary of main results

There are several ways to influence forage feed value and to affect dairy milk composition by feeding. The present project studied different forage related factors affecting milk concentration of substances with bioactivity in organic dairy production. The whole chain from field, through grazing, silage preservation, silage feeding, rumen metabolism, milk production and effects on milk products and human health (cancer cells) was investigated.

Starting at the field with the plant material, there are a number of possibilities to affect herbage fatty acid and phytoestrogen concentration. Results from the field study in Sweden (Umeå and Skara) and in Denmark (Foulum) showed that the fatty acid concentration in forage species varies with plant species, harvest date and site. Four forage species were harvested three times in primary growth (one week before heading of timothy, at heading, and one week after heading of timothy) and then after six and eight weeks of regrowth after each harvest time in the primary growth. Statistical analysis is not yet completed but according to preliminary results there were higher fatty acid concentrations in red clover and birdsfoot trefoil than in timothy and meadow fescue in the primary growth. There was a sharp decrease with delayed harvest in the primary growth in the total fatty acid concentration in all species. In the regrowth, there were smaller differences in fatty acid concentrations between species but, in general, lower concentration in the site furthest to the north, Umeå.

Herbage phytoestrogen concentration was also investigated in the field study. Phytoestrogen concentration was particularly high in red clover but the other forage species also contained phytoestrogens. In red clover, mainly the isoflavones formononetin and biochanin A were detected but also some daidzein, genistein and prunetin were observed. No phytoestrogens from the lignan group were found in red clover. Isoflavone concentration was higher in the primary growth than in the regrowth. In the primary growth, the concentration of formononetin decreased with maturity. In the regrowth there was higher formononetin concentration after six weeks than after eight weeks. In Denmark the highest isoflavone concentration was found in the third regrowth (in October).

In the silage preservation study the main factor affecting the fatty acid concentration were wilting; especially in the red clover-grass mixture the total fatty acid and the  $\alpha$ -linolenic concentration decreased. Silage additive affected mainly the fermentation characteristics of the silages; the acid additive resulted in higher concentrations of water soluble carbohydrates, inoculation increased the lactic acid concentration and decreased water soluble carbohydrates. In the silage without silage additive, the concentration of water soluble carbohydrates and the organic matter digestibility were lower and the pH was higher. There was also higher proportion of ammonia nitrogen and butyric acid indicating bad silage fermentation. There was higher  $\alpha$ -tocopherol concentration in birdsfoot herbage but in the silage there was higher in the red clover-grass mixture. However, there were no big effects of storage time on fatty acid,  $\alpha$ -tocopherol or  $\beta$ -carotene concentration.

Pasture type, red clover-grass or botanical diverse pastures, had no effect on milk yield, and there were no differences in content of fat, protein or lactose in the milk. However, milk from cows grazing the red clover pasture had a much higher content of phytoestrogens from the isoflavone group (1400 vs. 100  $\mu$ g/kg milk) than milk produced on the botanical diverse pasture. Differences in milk fatty acid composition and concentration of  $\alpha$ -tocopherol and carotene were small. In general, milk from both red clover and botanical diverse pastures had a high content of unsaturated fatty acids, conjugated linoleic acid and vitamins.

In the two silage-feeding experiments, the effects of different legume-grass silages on milk fatty acid composition, vitamin concentration and phytoestrogen concentration were studied. In Norway, the effects of short-rotational, red clover based ley silage (S3) compared to silage from long-term, botanical diverse grassland (L3) were studied. The effects of supplementation with natural  $\alpha$ -tocopherol on milk  $\alpha$ -

tocopherol concentration and oxidative stability were also tested. In Sweden, the effects of red clover-grass silage in a three-cut system (R3), in a two-cut system (R2) or birdsfoot trefoil-grass silage in a two-cut system (B2) were studied. Results showed that milk produced on diet S3 contained less fat, less free fatty acids and more urea than milk produced on silage L3. Milk on B2 contained more protein and less urea than on R2 and R3. The content of  $\alpha$ -tocopherol was lower in milk on S3 than on L3. As expected  $\alpha$ -tocopherol supplementation increased milk  $\alpha$ -tocopherol content but had no effect on milk's oxidative stability.

Feeding red clover containing diets (S3, R2 and R3) produced milk with less proportion of saturated fatty acids, higher proportion of  $\alpha$ -linolenic acid and sum of PUFA and lower ratio between n-6 and n-3 fatty acids than L3 and B2. The botanical composition of the silage affects the fatty acid composition of milk more than the botanical composition of pasture in these experiments. The reason for this may be difference in the activation of the enzyme PPO in red clover. For the enzyme to be activated it must come in contact with oxygen. Under grazing, the enzyme is exposed to oxygen for a very short time, but by pre-wilting the forage for silage making, the enzyme is exposed to plenty of oxygen and the exposure time for the enzyme to be activated is long.

The phytoestrogen concentrations in milk were also affected by silage treatment; red clover containing diets gave high milk isoflavones concentration. Equol accounted for most of the phytoestrogens in milk. Highest mean equol concentration was found on the two-cut red clover-grass silage in Sweden (1,500  $\mu\text{g}/\text{kg}$ ) but all diets with red clover as the dominating legume resulted in high milk equol concentration. However, despite similar intake of equol precursor formononetin and daidzein between the experiment in Norway and Sweden (S3 and R3), the concentration of equol on R3 was twice as high as on S3. A large interindividual variability in equol concentration between individual cows was seen. The content of lignans was lower in milk on S3 than on L3 and lower on R2 than on B2.

To understand more about the metabolism of fatty acids (biohydrogenation) and phytoestrogens in the rumen, rumen cannulated cows were fed four different silages; red clover grass (RK-S), botanic diverse (BA-S), timothy (TI-S) and perennial ryegrass (RG-S). More trans-vaccenic acid (C18:1t11) was formed in the rumen in cows fed RK-S and BA-S than RG-S and TI-S, and about half of the C18:1t11 were transferred to milk. Passage of CLA (C18:2c9t11) to milk followed the same pattern as C18:1t11. Effect of diet on CLA and trans-vaccenic acid may be due to change in rumen microbial composition. Silage from RK-S had a lower content of  $\alpha$ -linolenic acid, and feed intake of  $\alpha$ -linolenic acid on RK-S was lower than on BA-S and RG-S. Still, RK-S produced milk with higher proportion of  $\alpha$ -linolenic acid than BA-S and RG-S. The results showed that hydrogenation increases with increasing number of double bonds in unsaturated C18-fatty acids. Hydrogenation of unsaturated C18-fatty acids was lower for RK-S than RG-S, and there was a tendency for greater passage of unsaturated C18 fatty acids to milk for RK-S and BA-S compared with the grass-based silage type with RG-S and TI-S. The isoflavone formononetin and daidzein were to a great extent metabolised to equol and on to unknown compounds in the rumen. Recovery of isoflavonoid was greatest in manure and least in milk. Intake of lignans was higher on the TI-S than on the other diets, but there was no effect of diets on excretion into milk. Recovery of mammalian lignans was higher than the intake of lignans from the feed on all diets. The reason is probably that there are other (not analysed) lignans in the total diet (from the concentrate) that also are transferred into the known and analysed mammalian lignans in the milk.

Results from the Finnish farm study showed that farms that both had organic field management and produced organic milk had higher proportion of EPA (C20:5 n-3) and lower proportion of total omega-6 fatty acids than farms practicing only organic field farming. Consequently, on farms with organic field and milk production there was lower omega-6/omega-3-ratio than on only organic field farms. In the study in Finland, both farm groups had red clover as the predomination legume in the silages and this resulted in relative high phytoestrogen (especially equol) concentrations, which increased with increased silage clover content. For the vitamins, farms with organic field farming had three times higher vitamin A supplementation in their diets which gave higher retinol concentration and lower retinol/betacarotene-quota in milk than in farms with both organic fields and organic milk. Farm group had no effect on milk  $\alpha$ -tocopherol concentration, but the average level was moderate.

The selenium feeding experiment showed that the form of supplemented selenium had high effect on milk selenium concentration. Selenised yeast doubled milk selenium concentration compared to sodium selenite. Mixture (50:50) of selenised yeast and sodium selenite resulted in almost similar levels as the selenised yeast treatment. The results show that commercial feed supplemented with selenium-yeast is a good way for organic farms to fulfil cow selenium requirements and to reach the target level in milk.

To evaluate the effect of bioactive compounds in milk, the effect of different phytoestrogens were examined in different cell-based models. Milk samples from the different experiments were collected and tested on human breast cancer cells, prostate cancer cells and on colon cancer cells. Results from the grazing experiment in Norway, the silage feeding experiment in Sweden, and the field study in Finland all showed no or only small differences in proliferative activity in bioassays between different dietary treatments. However, an anti-estrogenic effect of whey was seen in milk samples from both the Swedish and the Finnish study.

## **Conclusions**

Harvesting at an earlier stage of maturity increased the fatty acid and the phytoestrogen concentration in herbage, especially in the primary growth. This was probably partly due to higher leaf to stem ratio but also the season might have effect. Wilting for silage making decreased the fatty acid concentration in the herbage but the changes during ensiling and storage were small. Vitamin concentration in silage was affected by the botanical composition of the mixture. Silage additives highly improved the hygienic quality of the silages.

It is possible to alter the fatty acid composition in milk by feeding different silages but the numerical changes in the milk were small. With red clover in the diets, the proportion of polyunsaturated fatty acids, mainly  $\alpha$ -linolenic acid, increased and the biohydrogenation of unsaturated fatty acids in the rumen were reduced. The beneficial effects of red clover on milk fatty acid composition are more distinct when feeding silage than when grazing; probably due to activation of the enzyme polyphenol oxidase in red clover. Supplementation with  $\alpha$ -tocopherol increased milk concentration but the value of this may be questioned as there was no difference in milk oxidative stability. However, higher milk  $\alpha$ -tocopherol concentration may be beneficial for the consumer of the product.

The Finnish farm study confirmed the necessity of fat soluble vitamin (A, E) supplementation during winter feeding at Finnish organic dairy farms and showed that supplementation with selenium yeast was an efficient method to increase organic milk selenium concentration.

For phytoestrogens, the content, intake and excretion of isoflavones were in all studies greatest on diets including red clover. Several experiments also showed very high equol concentrations in milk. Higher lignan concentrations in milk from cows on white clover-grass or birdsfoot trefoil-grass diets than on red clover-grass diets might be explained by higher grass proportion in the mixture, the legume species or differences in rumen passage rate. High phytoestrogen concentrations in milk did not affect cell proliferation but whey had a clear anti-estrogenic effect on some dietary treatments.

## **1.2 Fulfillment of objectives**

To what extent did the project achieve its objectives?

The project fulfilled its objectives. The statistical analyses and publication of parts of this project are delayed because the PhD students that are partly financed by the project have more than one year left until their dissertation which will occur in late spring semester in 2012 (Steffen Adler) and autumn semester in 2012 (Annika Höjer).

## 2. Milestones and Deliverables status, *actual* compared to planned

### Milestones:

Milestone no:	WP	Description	Planned time	Actual time
M1	WP 1-6	Workshop 1, Planning animal production experiments and survey, Kick-off	02 2007	02 2007
M2	WP 1-2	Establish grassland	02 - 03 2007	02-03 2007
M3	WP 4	Plant analysis	02 2009	
M4	WP 5	Silage experiment and chemical analysis	02 2007 – 02 2008 03 - 04 2008	02 2007 – 02 2008
M5	WP 3	Survey including milk sampling and feed and questionnaire data collection	01 2008 – 01 2009	01 2008 – 01 2009
M6	WP 1	Grazing experiment	02 – 03 2008	02 – 03 2008
M7	WP 1	Silage feeding experiment, including sampling at harvesting and conservation	03 2008 – 02 2009	03 2008 – 02 2009
M8	WP 2	Rumen biohydrogenation study	01 – 02 2009	03 2009
M9		Bioassay experiment on in vitro cell cultures	03 2008 – 03 2009	02 2011
M10	WP 4	Workshop 4 for planning scientific publication	02 2009	03 2009
M11	WP 1-6	Scientific publication	WP 1-2: 02 2009- 04 2009, WP 3: 02 2009, WP: 04 2007, WP: 04 2008, WP: 04 2009	WP 1-6: Publication is in progress
M12	WP 1-6	Dissemination on websites, meeting, papers etc.	04 2007 - 04 2008	On going
M13	WP 1-6	Workshop 2 for planning common experiments	Not planned	03 2007
M14	WP 1-6	Workshop 3 for planning common experiments	Not planned	02 2008
M15	WP 1-6	Final open seminar and final workshop	Not planned	06 2011

### Deliverables, status on deliverables planned in the application:

Deliverable no/ reference number in Organic E-prints:	WP	Description	Planned time	Actual time
1	WP 1	Adler et al. Botanical composition of pasture and quality of organically produced milk – Effect on fatty acid composition and vitamin and phytoestrogen content and oxidative stability	03 2009	01 2012
2	WP 1	Höjer, Adler et al. Effect of different legume-grass silages on fatty acid and vitamin composition in milk	04 2009	04 2011

3	WP 1	Höjer, Adler et al. Effect of different legume-grass silages on phytoestrogen concentration in milk	04 2009	04 2011
4	WP 1	Adler, et al. Effect of different legume-grass silages and vitamin E supplementation on milk oxidative stability	02 2010	Will be included in delivery 2
5	WP 2	Adler et al. Milk fatty acid composition and associated fatty acid hydrogenation when feeding organically produced silage with different botanical composition	01 2011	01 2012
6	WP 3	Okker et al. Increasing selenium concentration of organic milk by replacing sodium selenite supplementation totally or partly with selenised yeast	04 2009	02 2012
7	WP 4	Höjer et al. Effects of weather and latitude on the dynamics of fatty acid composition and phytoestrogens in organically managed legumes and grasses	02 2009	02 2012
8	WP 5	Höjer et al. Effects of storage time and preservation method on fatty acid composition and vitamins in organically managed silage	01 2011	02 2012
9	WP 6	Purup et al. Biological activity in organic milk with suggested health beneficial effects stability	03 2010	04 2011
10	WP 1-2	Höjer et al. Contribution at European Grassland Federation Congress in Kiel , Germany, 2010 <a href="http://orgprints.org/18295/">http://orgprints.org/18295/</a>	03 2010	03 2010
11	WP 1-2	Adler et al. Contribution at European Grassland Federation Congress in Kiel , Germany, 2010 <a href="http://orgprints.org/17493/">http://orgprints.org/17493/</a>	03 2010	03 2010
12	WP 1, 2, 3	NJF: Fostering healthy food systems through organic agriculture – focus on Nordic-Baltic region”, 25-27 August 2009, Tartu, Estonia <a href="http://orgprints.org/19081/">http://orgprints.org/19081/</a>	03 2009	03 2009
13		Contribution on NJF seminar “Healthier milk and functional dairy products” , in March 2010, Dröbak, Norway NJF workshop	01 2010	This seminar has been postponed and has not been held yet
14	WP 6	Contribution on European Federation of Animal Science (EAAP) in Crete, Greece, 2010 Is put into organic e-print but we have not received the address yet	03 2010	03 2010
15	WP 1	Contribution on NJF seminar “The potential of forage legumes to sustain a high agricultural productivity – a Nordic perspective” June 2010, Hvanneyri, Iceland <a href="http://orgprints.org/18829/">http://orgprints.org/18829/</a>	02 2010	02 2010
16	WP 1	Contribution on Regional Agricultural Conference, 16-17 March, Umeå, Sweden <a href="http://orgprints.org/18830/">http://orgprints.org/18830/</a>	01 2011	01 2011

**Additional comments (in case of major changes or deviation from the original list)**

### 3. Work package description and results:

<b>WP 1</b>	<i>Pasture and silage botanical composition - effect on milk quality</i>
<b>Responsible partner: "partner no 1, SLU, Anne-Maj Gustavsson and partner no 2, Bioforsk, Håvard Steinshamn"</b>	
<b>Description of work:</b> Two feeding experiments were carried out at the Animal Production Centre, University of Life Science, Norway (APC-UMB) and at Field research centre, Röbbäcksdalen, SLU, Umeå.  <u>Grazing experiment APC-UMB:</u> A continuous grazing trial was carried out during the grazing season 2008, with the first 2 weeks as pre-experimental period where the cows grazed the same pasture. Two groups of 8 cows (Norwegian Red Cattle) each, in mid lactation, was formed on the basis of their performance before the grazing season, calving date and lactation number. After the 2-week adaptation to grazing, three 4-week experimental periods followed; early (May), mid (June/July) and late (August) summer, during which each group grazed one of the two organic pasture management treatments; botanical diverse long term pasture (BA-B) and clover-rich short term pasture (RK-B). The long term pasture had a relatively high botanical diversity and high proportion of other herbs than red clover, whilst the short term managed pasture had a high proportion of red clover (on average 28 % of total DM yield). The cows received small and equal amounts of concentrate (3 kg per cow and day), a mineral additive but no vitamin supplements. The groups received similar herbage allowance, by adjusting the area, so that the cows had the same energy intake.  <u>Silage feeding experiment (APC-UMB and SLU-Umeå):</u> Two silage feeding experiments were carried out during 11 weeks of the housing period 2008/ 2009, with the first 2 weeks was a transition period where all cows received the same forage. The number of cows, stage of lactation and allocation to treatments was similar to and carried out in the same way as in the grazing experiment. The experiment was conducted using a change-over design with three 3-week periods. Two weeks for adaptation to the experimental feed and one week for measurement and sampling. The silage was fed <i>ad libitum</i> with a small and equal amount of concentrate supplementation (6 kg per cow and day). In the experiment conducted at APC-UMB, the treatments was round bale silage prepared from two contrasting organically managed grasslands; L3: long term ley with non red clover herbs like <i>Taraxacum officinale</i> and <i>white clover</i> and S3: short term rotational ley with high proportion of sown species rich in red clover. In the experiment at SLU-Umeå the treatments was two cut system silage with red clover-grass (R2), three cut system silage with red clover-grass (R3) and two cut system silage with birdsfoot trefoil-grass (B2).	
<b>Final report on work carried out and results compared to the original plan/WP aims:</b>	
<b>A- work carried out and results obtained:</b> <u>The grazing experiment:</u> The grazing experiment at APC-UMB was carried out the summer 2008 as planned.  Dairy cows that grazed on RK-B or BA-B had the same milk yield, and it was not found differences in content of fat, protein and lactose in milk. Milk from RK-B had a much higher content of phytoestrogens from the isoflavone group (1355 vs. 102 µg/kg milk) than milk produced on the BA-B. Differences in milk FA composition and concentration of α-tocopherol and carotene were small. In general, milk from both RK-B and BA-B had a high content of unsaturated fatty acids, conjugated linoleic acid and vitamins.  <u>The silage feeding experiment:</u> Silages were prepared during summer 2008 and in the housing period 2008/2009 the two feeding experiments was carried out. At APC-UMB, 16 multiparous dairy cows were fed four silage diets. Treatments were short-term grassland silage without α-tocopherol supplementation (S3E0), short-term grassland silage with α-tocopherol supplementation (S3E+), long-term grassland silage without α-tocopherol supplementation (L3E0), and long-term grassland	

silage with  $\alpha$ -tocopherol supplementation (L3E+). At SLU-Umeå, 24 dairy cows were fed red clover-grass silage in a three-cut system (R3), in a two-cut system (R2), or birdsfoot trefoil-grass silage in a two-cut system (B2).

$\alpha$ -tocopherol supplementation increased milk  $\alpha$ -tocopherol concentration but it did not affect milk fatty acid composition or phytoestrogen concentration. There was higher  $\beta$ -carotene concentration on red clover diets R2 and R3 compared to B2 and higher  $\alpha$ -tocopherol concentration on L3 than on S3. There was higher proportion of polyunsaturated fatty acid in milk from cows fed red clover. Proportion of CLA (C18:2c9t11) was higher in milk from R3 than from B2 and proportion of CLA (C18:2t10c12) was higher in milk from L3 compared to S3. Diet with red clover gave milk with lower (more beneficial) omega-6/omega-3 milk fatty acid ratio.

There was higher phytoestrogen concentration in milk from silage mixtures with red clover (S3, R3 and R2). Especially equol concentration was exceptional high (1,494  $\mu\text{g}/\text{kg}$  milk on R2). Lignan concentration (mainly enterolactone) in milk was higher in L3 and B2, possibly due to higher grass proportion in those silages or because of differences in the rumen metabolism or passage rate.

#### **B- comments on deviations from the original plan:**

The grazing experiment: Because of annual weeds a fourth measuring period was added, and period 1 – 4 were measured. The chemical analyses of herbage and milk samples were done on samples collected at period 2 – 4. Milk samples from the experiment were used to examine the treatments effect on lipid oxidation, both chemically and with fluorescence spectroscopy, after exposure to light.

Both silage feeding experiments was carried out in three-week periods instead of four-weeks, due to less herbage yields than expected. Supplementation with natural  $\alpha$ -tocopherol was added as a treatment at APC-UMB. This was done without any negative effects on the original study. At SLU-Umeå the experiment was enlarged by introducing a treatment with three cuts for the red clover mixture to be able to compare the effect of different regrowth intervals.

Delivery 1, 2, and 3 will be published in late autumn semester in 2011 and in early spring semester in 2012. Delivery 4 will be included in delivery 2.

<b>WP 2</b>	<i>Silage botanical composition – effect on rumen FA hydrogenation and milk FA composition (Department of Animal and Aquacultural Sciences – IHA/UMB)</i>
<b>Responsible partner: " partner no 2, Bioforsk, Håvard Steinshamn and partner no 3, UMB, Erling Thuen"</b>	
<p><b>Description of work:</b></p> <p>The experiment was carried out during the housing period 2008/2009 at IHA/UMB. Four types of silages ensiled in round bales were prepared from the primary growth in 2008: Perennial ryegrass (RG-S); timothy and meadow fescue mixture (TI-S); Red clover-grass mixture (RK-S); non red clover herb-grass mixture (BA-S). The RG-S and TI-S were included as control treatments. The RK-S and BA-S silages was the same as used in WP 2, and the varieties used in the TI-S and RK-S treatments was the same as the one used in the field trials in WP 4 and 5. The silages were offered ad libitum with small and equal amount of concentrate supplementation. The experimental diets were fed to 4 cannulated dairy cows in a Latin square design with 4 periods of 24 days (14 days adaptation to the diet and 10-day measurement period for milk sampling and collection of faeces, rumen and duodenal fluid). Markers were used in order to estimate digesta flow at the duodenum. Faecal output was measured on days 15 to 20, and duodenal digesta were collected every 4 h on days 21 and 22. Samples of rumen fluid were taken every 2 h over a 24-h period on day 23. Rumen fluid was analysed for pH, ammonia and volatile fatty acids. Digesta flow and apparent digestibility of fatty acids in the rumen was calculated after mathematical reconstitution of true digesta, and the biohydrogenation of C:18 PUFAs was estimated by simple difference between feed and duodenal flow.</p>	
<p><b>Final report on work carried out and results compared to the original plan/WP aims:</b></p> <p><b>A- work carried out and results obtained:</b></p> <p>It was formed more trans-vaccenic acid (C18:1t11) in the rumen of RK-S and BA-S than on the RG-S and TI-S, and about half were transferred to milk. Passage of CLA (C18:2c9t11) to milk followed the same pattern as C18:1t11. Effect of diet on CLA and trans-vaccenic acid may be due to change in rumen microbial composition. Silage from RK-S had a lower content of <math>\alpha</math>-linolenic acid and feed intake of <math>\alpha</math>-linolenic acid was lower than in silage from the BA-S and RG-S. Still, RK-S gave higher proportion of <math>\alpha</math>-linolenic acid in milk than BA-S and RG-S. There was no effect of silage type on the apparent hydrogenation of oleic and linoleic acid. Recovery from feed to milk of <math>\alpha</math>-linolenic acid was about 70 % higher on RK-S than on the other diets.</p> <p>The reason for reduced hydrogenation of RK-S may be the activity of the enzyme PPO in red clover. The results showed that hydrogenation increases with increasing number of double bonds in unsaturated C18-fatty acids. Hydrogenation of unsaturated C18-fatty acids was lower for RK-S than RG-S, and there was a tendency for greater passage of unsaturated C18 fatty acids to milk for RK-S and BA-S compared with the grass-based silage type with RG-S and TI-S.</p> <p>The content, intake and excretion of isoflavones were as expected greatest on red clover based silage. The isoflavone formononetin and daidzein were to a great extent metabolised to equol and on to unknown compounds in the rumen. Intake of lignans was higher on the TI-S than the other diets, but there was no effect of diets on excretion to milk. Recovery of mammalian lignans was higher than the intake of lignans from the feed on all diets. The reason is probably that there are other lignans in the diet that we do not know and which is also transferred into the mammalian lignans that were analysed.</p> <p><b>B- comments on deviations from the original plan:</b></p> <p>The DS sward had much less proportion of unsown herbs than anticipated (see report from WP1). We therefore decided to postpone the experiment from winter 2008 to autumn 2009. Postponing the experiment had no negative effect.</p>	

**WP 3** *Effects of Se supplementation on milk quality***Responsible partner: "partner no 5, University of Joensuu, Eeva Kuusela"****Description of work:**

Current situation: A survey was conducted in Eastern Finland in cooperation with the local ProAgria advisory centre and local organic farms. In the beginning of year 2008 tank milk samples was collected from 50 organic farms (25 practicing certified organic animal production and 25 farms practising only organic field farming). Representative samples of silage and concentrate mixture were taken from each farm. Number of cows and average daily milk yield was recorded. Farmers were interviewed for their current feeding and Se and vitamin supplementation.

Improved Se situation: All farmers were introduced to Se yeast. Twenty voluntary farms switched to Se yeast supplementation (3 mg Se/cow) for 4 weeks. Tank milk samples were collected in the end of experimental period.

**Final report on work carried out and results compared to the original plan/WP aims:****A- work carried out and results obtained:**

The main objective was to investigate specific milk quality (fatty acids, vitamins, phytoestrogens, selenium) during winter feeding at organic farms. The main aim was to find a tool to improve milk selenium content at organic farms. The farm investigation (22 certified organic milk farms and 23 farms practicing only organic field farming) was carried out in the end of January 2008. The selenium feeding experiment was performed between 6.2. – 29.4.2008. Total 12 weeks consisted from three four-week periods (Latin square model). Treatments were organic selenium (yeast), mineral (sodium selenite) + organic selenium and mineral selenium. Additional aims were to provide information of salutary effect of organic milk, provide a selenium feeding recommendation for organic dairy farms and on current data basis to discuss the necessity of vitamin supplementation.

Our results showed that organic feeding had favourable effects on milk fatty acid content. Both farm groups resulted to similar relative high, proportion of monounsaturated fatty acids and total omega-3 fatty acids reflecting positive effects of red clover. Farms producing organic milk resulted to higher proportion of EPA (C20:5 n-3) and to lower proportion of total omega-6 fatty acids than farms practicing only organic field farming. Consequently all organic farms resulted to lower omega-6/omega-3-relation compared to field organic farms (2.7 vs. 3.3). Field organic farms had three times higher vitamin A supplementation in their feeding and resulted to higher retinol concentration and lower retinol/betacarotene-quota in milk than all organic farms. Farm group had no effect on milk vitamin alphotocopherol concentration, but the average level was moderate. Current farm and literature studies confirmed the necessity of fat soluble vitamin (A, D, E) supplementation during winter feeding at Finnish organic dairy farms. Both farm groups resulted similarly to relative high phytoestrogen especially equol concentrations, which increased with increased silage clover content. The selenium feeding experiment showed that the form of selenium supplementation was crucial for milk selenium concentration. Sodium selenite resulted to the lowest selenium concentration. Selenised yeast more than doubled milk selenium concentration near to similar level with Finnish conventional milk. Mixture (50:50) of selenised yeast and sodium selenite resulted to almost similar level than selenised yeast. Selenised yeast is powerful tool for organic farms in increasing milk selenium concentration and it is needed if the target is to reach the level of Finnish conventional milk. Selenium yeast containing commercial feeds are convenient way to ensure cow's selenium.

**B- comments on deviations from the original plan:**

Effect of selenium supplementation on milk oxidative stability measurements were rejected since the lack of an expert laboratory for this measurement.

<b>WP 4</b>	<i>Effects of latitude and harvest time on FA composition and phytoestrogen concentrations on four organically managed forage species</i>
<b>Responsible partner: "partner no 1, SLU, Anne-Maj Gustavsson and Elisabet Nadeau and partner no 2, DIAS, Sören Krogh-Jensen"</b>	
<p><b>Description of work:</b></p> <p>In an ongoing collaboration, the seasonal dynamics of herbage tocopherols, carotenoids, lignin, and ash content and herbage carbohydrate and crude protein fractions and digestibility in timothy, meadow fescue, red clover and birdsfoot trefoil are investigated together with plant development and plant morphology investigations on three sites (SLU-Umeå, Sweden), (SLU, Skara, Sweden) and (DIAS-Foulum, Denmark). From this experiment, already collected samples were used to investigate site and harvest time effects on FA composition and concentration of phytoestrogens. From each site, freeze dried plant material from two years harvested at nine different harvest times (three in primary growth and 2*3 in regrowth) separated in pure species were analyzed.</p>	
<p><b>Final report on work carried out and results compared to the original plan/WP aims:</b></p> <p><b>A- work carried out and results obtained:</b></p> <p>Among the collected samples, samples for fatty acid and phytoestrogen analysis were selected and analysed. Four forage species were harvested three times in primary growth (1 week before heading of timothy, at heading, and 1 week after heading of timothy). Regrowth was harvested six and eight weeks after each cut in the primary growth.</p> <p>Statistical analysis is not yet completed but preliminary results show higher fatty acid concentration in the legumes (red clover and birdsfoot trefoil) than in grasses (timothy and meadow fescue) in the primary growth. There was a sharp decrease in total fatty acid concentration as the plant matured in the primary growth. Meadow fescue had a higher fatty acid concentration in regrowth than in the primary growth, especially with shorter regrowth interval. For the other species we will present more details for secondary growth when the statistical analysis is completed. There seemed to be lower fatty acid concentration in general in plant material from the site furthest to the north, Umeå.</p> <p>Phytoestrogen concentration was only measured in red clover. Phytoestrogens detected was mainly the isoflavones formononetin and biochanin A and lower concentrations of daidzein, genistein and prunetin. No lignans were found. Isoflavone concentration was higher in primary growth than in the regrowth. In the primary growth, the concentration of formononetin decreased with maturity. In the regrowth there was higher formononetin concentration after six weeks than after eight weeks. In Denmark there was highest isoflavone concentration in the third regrowth (in October).</p> <p><b>B- comments on deviations from the original plan:</b></p> <p>Samples from DIAS-Foulum were harvested in four harvests during each season and the species were red clover, white clover, birdsfoot trefoil, timothy and perennial ryegrass.</p> <p>The results from this WP are only preliminary because the data from these experiments are not yet analysed with a full statistical model. The full analysis will be done in early spring semester in 2012 and delivery 7 will be published in late spring semester in 2012.</p>	

<b>WP 5</b>	<i>Effects of storage time and silage preservation methods on FA composition and concentrations of tocopherols and carotenoids on four organically managed forage species</i>
<b>Responsible partner: "partner no 1, SLU, Elisabet Nadeau"</b>	
<b>Description of work:</b>	
<p>In the same experiment as in WP 4, a silage experiment was conducted at one site. Primary growth of a birdsfoot trefoil/timothy mixture and a red clover/timothy mixture was harvested at the heading stage of timothy in 2007. Each mixture was replicated three times in the field. Wilting to 30% dry matter before ensiling was a target. Forage mixtures were ensiled, without an additive or with an addition of a bacterial inoculant or an acid, in laboratory silos. The silos were opened after 1.5, 3, 6 and 9 months. Forages were analysed for FA composition, <math>\alpha</math>-tocopherol and carotenoids before and after wilting and after each storage period. Chemical composition of wilted forage before ensiling and of silage at three months of storage (incl. fermentation characteristics) was assessed according to the NorFor system.</p>	
<b>Final report on work carried out, results, deviations from the original plan/WP aims:</b>	
<b>A- work carried out and results obtained:</b>	
<p>The experiment was carried out as planned.</p>	
<p>Results on fermentation characteristics indicated good silage quality in the two treatments with additives, and relatively poor quality of the control silage (no additive) (higher pH, especially in red clover-timothy mixture, and relatively high proportion of ammonia N in both control silages).</p>	
<p>Fatty acid concentration and composition was mainly affected by wilting; it decreased fatty acid concentration in red clover-timothy mixtures and it was mainly the unsaturated fatty acids that were affected. During storage, the fatty acid concentration did not change. <math>\alpha</math>-tocopherol concentration was higher in birdsfoot trefoil-timothy herbage but there was no difference in the silage. <math>\beta</math>-carotene concentration was not affected by wilting or by silage additive.</p>	
<b>B- comments on deviations from the original plan:</b>	
<p>The forages were wilted to 22 % dry matter before ensiling instead of 30 % because of cloudy and rainy conditions during wilting.</p>	
<p>The results from this WP are only preliminary because the data from this experiment are not yet analysed with a full statistical model. The full analysis will be done in early spring semester in 2012 and delivery 8 will be published in late spring semester in 2012.</p>	

**WP 6** | *Bioactive components in organic milk with suggested health beneficial effects*

**Responsible partner: "partner no 4, DIAS, Stig Purup"**

**Description of work:**

The milk samples were collected from the Nordic countries in this project represented a repository of milk samples collected from cows at different breed and lactation- and pregnancy stages, and from cows fed rations high in clover, grass, birdsfoot trefoil, corn etc. *In vitro* cell-based models were used for assessing the biological activity in specific human tissues. Cell-based models include normal- and cancer cell lines of gastro-intestinal, mammary or prostate origin. Different cellular endpoints were evaluated. Milk samples having the ability to inhibit proliferation of human breast, prostate and intestinal cancer cells received highest priority. The milk samples tested were selected mainly based on the content of bioactive components.

**Final report on work carried out and results compared to the original plan/WP aims:**

**A- work carried out and results obtained:**

A repository of milk samples was collected from experiments performed in Denmark, Norway, Sweden and Finland. This repository of complex mixtures contains naturally occurring bioactive components with suggested health beneficial effects in human tissues. To elaborate these effects in relation to different feeding of cows in different Nordic countries, we first identified cell-based models for screening and testing representing relevant human tissues and cellular endpoints. A review entitled "Cell-based models to test the effects of milk-derived bioactives" discusses the choice of relevant cell-based models, sample preparation and endpoints (Purup & Nielsen, 2011, *Animal*, accepted). To evaluate the effect of milk samples in relation to the content of bioactive compounds, the effect of different phytoestrogens were examined in different cell-based models. These results showed significant differences in the potency of the different bioactive compounds, and furthermore that phytoestrogens found in cow's milk have the potential to contribute to a COX-inhibitory effect, suggesting that phytoestrogens have anti-inflammatory effects (Purup et al., 2010; Nielsen et al., 2010). The effect of different legume-grass silages on phytoestrogen concentrations and proliferative activity was studied in 45 milk samples from Sweden. A manuscript entitled "Proliferative effect of whey from cow's milk varying in phytoestrogen content in human breast and prostate cancer cells" has been submitted to *Journal of Dairy Research* (Nielsen, Höjer, Gustavsson, Hansen-Møller and Purup). These results showed no significant difference in the proliferative effect of whey from the different dietary treatments in neither breast- nor prostate cancer cells, and it was therefore not possible to discriminate between the effects of milk with high and low levels of phytoestrogens. However, interestingly, addition of milk clearly inhibited cell proliferation of breast cancer cells simultaneously treated with oestrogen. Although there obviously is a long step from *in vitro* measurements to *in vivo* effects, these results lend support to the idea that milk may inhibit the effect of high levels of endogenous oestrogen. This effect was independent of the dietary treatment of cows. In 64 milk samples collected from 16 cows in a grazing experiment performed in Norway, the bioactivity was also studied in breast- and prostate cancer cells and furthermore in human colon cancer cells. Milk samples represented cows fed either a "red clover pasture" or a "botanical diverse pasture". Despite significant differences in concentrations of phytoestrogens in milk samples, especially equol (1047 vs. 170 ng/ml), only small differences were seen in proliferative activity in bioassays between different dietary treatments. Milk samples collected from 45 farms in Finland, representing two groups: "organic field" and "organic field and milk", showed only small differences in phytoestrogen content between groups, and no dietary treatment effects were observed in proliferation of breast- and prostate cancer cells. As with milk samples from cow's fed legume-grass silages from Sweden, we observed an anti-estrogenic effect of whey independent of dietary treatment. These results will be published in the nearest future.

**B- comments on deviations from the original plan:**

No deviations from the original plan.

## 4. Publications and dissemination activities

### 4.1 List

Note: the report should contain all the publications and dissemination activities of the project. Publications should have been loaded in Organic Eprints, but some dissemination activities might not be in Organic Eprints and still have to be listed below. Part of the list required below can be extracted and pasted by doing a search in Organic Eprints, and others added manually. The resulting list should be clear and complete whether the tables below are used or a search in Organic Eprints.

#### *Project website(s)*

Address	Authors: (name + institution acronym)	When was it last updated	Language	Comments
PhytoMilk	<a href="http://www.coreorganic.org/research/projects/phytomilk/index.html">http://www.coreorganic.org/research/projects/phytomilk/index.html</a>	A-M Gustavsson SLU: NJV	English	Under construction

#### *Reviewed papers (with full reference)*

Planned / actual date	Title:	Authors: (name + institution acronym)	Name of Magazine, volume, pp. etc.	Language	Comments
03 2009 / 01 2012	Botanical composition of pasture and quality of organically produced milk – Effect on fatty acid composition and vitamin and phytoestrogen content and oxidative stability.	Adler et al.		English	In preparation
04 2009 / 04 2011	Effect of different legume-grass silages and $\alpha$ -tocopherol supplementation on fatty acid composition and fat-soluble vitamin concentration in organically produced bovine milk.	Höjer, A., Adler, S., Martinsson K., Krogh-Jensen, S., Steinshamn, H., Thuen, E. and Gustavsson, A.-M.		English	Manuscript
04 2009 / 04 2011	Milk phytoestrogen concentration when feeding different legume-grass forages to dairy cows	Höjer, A., Adler, S., Hansen-Møller, J., Purup, S., Martinsson K., Steinshamn, H. and Gustavsson, A.-M.		English	Manuscript
01 2011 / 01 2012	Milk fatty acid composition and associated fatty acid hydrogenation when feeding organically produced silage with different botanical composition.	Adler et al.		English	In preparation

<b>04 2009 / 02 2012</b>	Increasing selenium concentration of organic milk by replacing sodium selenite supplementation totally or partly with selenised yeast.	Kuusela et al.		English	In preparation
<b>02 2009 / 02 2012</b>	Effects of weather and latitude on the dynamics of fatty acid composition and phytoestrogens in organically managed legumes and grasses.	Höjer, A., Krogh-Jensen, S., Nadeau, E., Hansen-Møller, J. and Gustavsson, A.-M.		English	In preparation
<b>01 2011 / 02 2012</b>	Effects of storage time and preservation method on fatty acid composition and vitamins in organically managed silage.	Höjer, A., Nadeau, E., Krogh-Jensen, S., Lindqvist, H. and Gustavsson, A.-M.		English	In preparation
<b>03 2010 / 04 2011</b>	Cell-based models to test the effects of milk-derived bioactives	Purup, S. and Nielsen, T.S.	Animal	English	Accepted
<b>03 2011</b>	Proliferative effect of whey from cow's milk varying in phytoestrogens in human breast and prostate cancer cells.	Nielsen, T.S., Höjer A., Gustavsson, A.-M., Hansen-Møller, J. and Purup, S.	J. Dairy. Res.	English	Submitted
<b>2012 02</b>	Improving organic milk Se concentration by participating farm study in Finland.	Okker et al.		English	In preparation
<b>2011 02</b>	Characterisation of Finnish organic milk.	Kuusela et al.		English	In preparation
<b>2012 02</b>	Gastro-intestinal metabolism of phytoestrogens in lactating dairy cows fed silages with different botanical composition.	Njaastad, K.M., Adler, S., Hansen-Møller, J., Thuen, E. and Steinshamn, H.	Animal	English	In preparation
<b>2011 04</b>	Alpha-tocopherol and beta-carotene in legume-grass mixtures as influenced by wilting, ensiling and type of silage additive.	Lindqvist, H., Nadeau, E. and Jensen, S.K.	Grass and Forage Science doi: 10.1111/j.1365-2494.2011.00827.x	English	Published

**Presentations/papers at scientific conference** (oral presentations, papers, leaflets, posters, etc.)

<b>Planned / actual date</b>	<b>Type and Title of contribution:</b> (also mention if a partner was keynote speaker)	<b>Conference:</b>	<b>Partners involved:</b> (partner acronyms)	<b>Type of audience</b> (General public, higher education, researchers, industry, farm sector, advisors etc.)	<b>Size of audience</b>	<b>Countries addressed</b>
June 2008	Danielsson, H., Nadeau, E., Gustavsson, A-M., Jensen, S.K., Søgaard and Nilsdotter-Linde, N. 2008. Contents of $\alpha$ -tocopherol and $\beta$ -carotene in grasses and legumes harvested at different maturities. Grassland Science in Europe, Vol. 13, pp. 432-434. Oral presentation by Danielsson.	Grassland Science in Europe / European Grassland Federation (EGF), Uppsala, Sweden June 2008	SLU; HMM, NJV AU; DJF	Researchers, advisors	460	European
August 2009	Okker L., Kuusela, E. and Eurola M. 2009. Selenium concentration of Finnish organic milk. Oral presentation by Okker.	NJF: Fostering healthy food systems through organic agriculture – focus on Nordic-Baltic region”, 25-27 August 2009, Tartu, Estonia	UEF; MTT	Researchers, advisors	100	Nordic and Baltic
August 2009	Okker L., Kuusela, E. and Eurola M. 2009. Effect of selenite and selenised yeast supplementation on selenium concentration of Finnish organic milk – a farm survey. Poster.	NJF: Fostering healthy food systems through organic agriculture – focus on Nordic-Baltic region”, 25-27 August 2009, Tartu, Estonia	UEF; MTT	Researchers, advisors	100	Nordic and Baltic
August 2009	Kuusela, E., Okker L., Jensen S.K., Purup, S. and Hanssen-Moller, J. 2009. Fatty acid characterization of Finnish organic milk – a Farm survey. Poster.	NJF: Fostering healthy food systems through organic agriculture – focus on Nordic-Baltic region”, 25-27 August 2009, Tartu, Estonia	UEF; AU	Researchers, advisors	100	Nordic and Baltic
August 2009	Adler, S. Effect of pasture botanical composition on milk quality in organic production. Poster.	NJF: Fostering healthy food systems through organic agriculture – focus on Nordic-Baltic region”, 25-27 August 2009, Tartu, Estonia	Bioforsk, UMB, AU	Researchers, advisors	100	Nordic and Baltic

October 2009	H. Lindqvist, E. Nadeau and S. K. Jensen. 2009. Vitamin concentration of legume-grass silages treated with different additives. <i>Irish Journal of Agricultural and Food Research</i> Draft Issue: 135, pp. 135. Poster.	Proc. Int. Conf. "Forage legumes in temperate pasture-based systems"	SLU; HMM AU; DJF	Researchers, industry, farm sector, advisors	113	European
June 2010	Höjer, A., Martinsson, K., Krogh-Jensen, S. and Gustavsson, A.-M. PhytoMilk: Effects of botanical composition and harvest system of legume/grass silage on fatty acid, $\alpha$ -tocopherol and $\beta$ -carotene concentration in organic forage and milk. Oral presentation by Höjer.	NJF: The potential of forage legumes to sustain a high agricultural productivity - a Nordic perspective	SLU; NJV AU; DJF	Researchers	40	European
August 2010	Purup, S. and Nielsen, T.S. 2010. Cell-based models to test the effects of milk-derived bioactives. Oral presentation by Purup. Key note speaker.	EAAP/Biology of lactation in farm animals, August 23-27, Crete, Greece	AU	Researchers, industry, farm sector, advisors	500	European
August 2010	Purup, S., Nielsen, T.S. and Hansen-Møller, J. 2010. Phytoestrogens found in milk inhibit COX enzyme activity in murine macrophages. Poster.	EAAP/Biology of lactation in farm animals, August 23-27, Crete, Greece	AU	Researchers, industry, farm sector, advisors	500	European
August 2010	Nielsen, T.S. Hansen-Møller, J. and Purup, S. 2010. Milk phytoestrogens inhibit COX enzyme activity in murine macrophages. Poster.	EAAP/Biology of lactation in farm animals, August 23-27, Crete, Greece	AU	Researchers, industry, farm sector, advisors	500	European
September 2010	Höjer, A., Martinsson, K. and Gustavsson, A.-M. PhytoMilk: Effect of silage botanical composition and harvest system on organic milk composition. Poster.	European Grassland Federation (EGF), September 2010, Kiel, Germany	SLU; NJV	Researchers	500	European
September 2010	Adler, S., Dahl, A., Steinshamn, H., Thuen, E., Garmo, T. and Jensen, S.K. Effect of pasture botanical composition on milk composition in organic production. Oral presentation by Steinshamn.	European Grassland Federation (EGF), September 2010, Kiel, Germany	Bioforsk, UMB, AU	Researchers	500	European

**Presentations/papers at other conferences and meetings** (oral presentations, papers, leaflets, posters etc.)

<b>Planned / actual date</b>	<b>Type and Title of contribution:</b> (also mention if a partner was keynote speaker)	<b>Conference/title:</b>	<b>Partners involved:</b> (partner acronyms)	<b>Type of audience</b> (General public, higher education, researchers, industry, farm sector, advisors etc.)	<b>Size of audience</b>	<b>Countries addressed</b>
2009	Gustavsson, A-M. CORE organic pilot project PhytoMilk presentation. Oral presentation.	Joint meeting of the CORE Organic pilot projects coordinators and the CORE Funding Body Network, Rome Italy, 8 June 2009.	SLU	Decision makers, researchers	50	European
2009	Organic selenium for all bovine. Oral presentation in Finnish.	Finnish organic farming symposium 'Luomupäivät'	UEF	Researchers, Advisors, Farmers	100	Finland
2009	Höjer, A., Martinsson, K. and Gustavsson, A.M. Fettsyror och antioxidanter i vallfoder. Oral presentation by Höjer.	Svenska vallföreningens sommarmöte	SLU; NJV	General public, researchers, industry, farm sector, advisors	250	
2010	Jensen, S.K., Søegaard, K., Sehested, J., Lindqvist, H. och Nadeau, E. Indflydelse af høstmetode og konservering på vitamin- og fedtsyreindhold. Oral presentation by Jensen.	Temamöte FØJO	AU; DJF SLU; HMM	Researchers, industry, farm sector, advisors	50	Denmark
2010	Effect of all organic or field organic feeding on milk omega-3 and omega-6 milk fatty acid concentration. Oral presentation.	Finnish symposium of plant science	UEF	Researchers	100	Finland
2011	Adler, S., Dahl, A., Steinshamn, H., Thuen, E., Garmo, T. and Krogh-Jensen, S. Effekt av rødkløverbeite eller botanisk allsidig beite på kvalitetsegenskaper hos melk i økologisk drift. Oral presentation by Adler.	Husdyrforsøksmøtet 2009	Bioforsk, UMB, AU	Researchers, industry, farm sector, advisors	100	Norway
2011	Adler, S., Steinshamn, H, Dahl, A.V., Thuen, E., Jensen, S. K. and Hansen-Møller, J. Effekt av surfôr fra kortvarig eng eller langvarig eng på kvalitet i kumelk. Oral presentation by Adler.	Husdyrforsøksmøtet 2011	Bioforsk, UMB, AU	Researchers, industry, farm sector, advisors	100	Norway

2011	Höjer, A., Martinsson, K. and Gustavsson, A.-M. Fytoöstrogener i foder och mjölk – vad påverkar halterna? Oral presentation by Höjer.	14:e Regionala Jordbrukskonferensen för Norra Sverige	SLU; NJV	Researchers, industry, farm sector, advisors	100	Nordic
2011	Adler, St., Steinshamn, H., Thuen, E., Jensen, S.K. and Hansen-Møller, J. Hydrogenering av fettsyrer i vomma - effekt av botanisk sammenstening av surfôret Oral presentation by Adler.	Husdyrforsøksmøtet 2011	Bioforsk, UMB, AU	Researchers, industry, farm sector, advisors	100	Norway
2011	Steinshamn, H., Adler, S.; Njåstad, K.M., Thuen, E. and Hansen-Møller, J. Omsetjing av planteøstrogen hos mjølkeku - effekt av botanisk samansetjing av surfôret. Oral presentation by Njåstad.	Husdyrforsøksmøtet 2011	Bioforsk, UMB, AU	Researchers, industry, farm sector, advisors	100	Norway
2011	Höjer, A., Jensen, S.K., Nadeau, E., and Gustavsson, A-M. Latitude and harvest time affect fatty acid composition and phytoestrogen content of forages. Oral presentation by Höjer.	PhytoMilk Open seminar	SLU: NJV, HMM AU: DJF	Researchers, advisors, higher education	40	Nordic and Baltic
2011	Gustavsson, A-M. PhytoMilk – an overview. Oral presentation.	PhytoMilk Open seminar	SLU: NJV, HMM AU: DJF	Researchers, advisors, higher education	40	Nordic and Baltic
2011	Höjer, A., Lindqvist, H., Gustavsson, A-M., Jensen, S.K, And Nadeau, E. Storage time and preservation methods affect fatty acid composition and fat soluble vitamins in ensiled forages. Oral presentation by Höjer.	PhytoMilk Open seminar	SLU: NJV, HMM AU: DJF	Researchers, advisors, higher education	40	Nordic and Baltic
2011	Steinshamn, H., Adler, S., Jensen, S.K., Thuen, E., and Veberg Dahl, A. Effect of pasture botanical composition on milk composition. Oral presentation by Steinshamn.	PhytoMilk Open seminar	UMB AU: DJF Bioforsk	Researchers, advisors, higher education	40	Nordic and Baltic
2011	Höjer, A., Adler, S., Steinshamn, H., Jensen, S.K., Martinsson, K., and Gustavsson, A-M. Botanical composition of silage and quality of milk. Oral presentation by Höjer.	PhytoMilk Open seminar	SLU: NJV, HMM AU: DJF Bioforsk	Researchers, advisors, higher education	40	Nordic and Baltic
2011	Steinshamn, H., Adler, S., Jensen, S.K., and Thuen, E. Rumen fatty acid biohydrogenation. Oral presentation.	PhytoMilk Open seminar	UMB AU: DJF Bioforsk	Researchers, advisors, higher education	40	Nordic and Baltic

2011	Njaastad, K., Steinshamn, H., Adler, S., Hansen-Møller, J., and Thuen, E. Rumen transformation of phytoestrogens. Oral presentation by Njaastad.	PhytoMilk Open seminar	UMB AU: DJF Bioforsk	Researchers, advisors, higher education	40	Nordic and Baltic
2011	Kuusela, A. and Okker, L. Effect of Se supplementation of milk quality. Oral presentation by Kuusela.	PhytoMilk Open seminar	UJ	Researchers, advisors, higher education	40	Nordic and Baltic
2011	Bioactive components in milk and possible health effect. Purup, S. and Jensen, S.K. Oral presentation by Nielsen.	PhytoMilk Open seminar	AU: DJF	Researchers, advisors, higher education	40	Nordic and Baltic
2011	Invitation to Open seminar. Leaflet.	PhytoMilk Open seminar	SLU: NJV, HMH AU: DJF UMB Bioforsk UJ	Researchers, advisors, higher education	40	Nordic and Baltic

**Popular articles and other dissemination activities (press releases, interviews etc.)**

<b>Planned / actual date</b>	<b>Title of contribution:</b>	<b>Type of contribution</b> (presentation, leaflet, poster etc.)	<b>Partners involved:</b> (partner acronyms)	<b>Type of audience</b> (General public, industry, farm sector, advisors, policy makers, public authorities, higher education, researchers, etc.)	<b>Language</b>	<b>Countries addressed</b>
2008	Lindqvist, H., Nadeau, E., Gustavsson, A-M., Jensen, S.K., Nilsson-Linde, N., and Søgaard, K. 2008. Vitaminer i ekologiskt odlade vallbaljväxter och gräs (Vitamins in organically grown forage legumes and grasses).	Final report Stiftelsen Lantbruksforskning	SLU; HMM, NJV AU; DJF	Farm sector, advisors, policy makers, general public.	Swedish	Sweden
2008	Finnish organic milk Se concentration to similar level with conventional milk.	Press release	UEF	Farm sector	Finnish	Finland
2008	Finnish organic milk Se concentration to similar level with conventional milk.	Popular article	UEF	Farm sector	Finnish	Finland
2008	Organic milk selenium problem has been solved.	Popular article	UEF	Farm sector	Finnish	Finland
2010	Vitaminer og fedtsyrer i hø og ensilage – hvad sker der ved forvejrning og lagring? ICROFS nyt 2, 2010, p 6-7.	Popular article	DJF; SLU	Farm sector	Danish	Denmark
2010	Fatty acids of organic milk.	Popular article	UEF	Farm sector	Finnish	Finland

**Internal reports and proceedings, newsletters, web communication and other dissemination activities etc.**

<b>Planned / actual date</b>	<b>(No.) and title</b>	<b>Type:</b> Deliverable, proceedings, internal report, newsletter, web communication	<b>Partners involved:</b> (partner acronyms)	<b>Type of users addressed</b> (General public, higher education, researchers, industry, farm sector, advisors etc.)	<b>Language</b>	<b>Countries addressed</b>
2009	Fatty acids and vitamins in forages.	Internal report	SLU	Researchers	English	Sweden
2009	Vae, A.H. Effekt av botanisk sammensetning i beite på oksidativ stabilitet i kumelk.	Master Thesis	UMB, Bioforsk	Higher education	Norwegian	Norway
2010	Effekt av botanisk sammensetning av surfôr på metanproduksjon hos melkekyr.	Master Thesis	UMB, Bioforsk	Higher education	Norwegian	Norway
2010	Effect of organic feeding on milk fatty acid concentration.	Master Thesis	UEF	Higher education	Finnish	Finland
2010	Influence of production system to milk's retinol and beta carotene content.	Master Thesis	UEF	Higher education	Finnish	Finland
2010	Effect of organic farming system on milk phytoestrogen content.	Master Thesis	UEF	Higher education	Finnish	Finland
2010	Final report for MMM.	Final report	UEF	Researchers, advisors, education	English	Finland
2011	Njåstad, K.M. Metabolism of phytoestrogens in dairy cows - effect of botanical composition of silages.	Master Thesis	UMB, Bioforsk, AU	Higher education	English	Norway
2011	Vitamin E supply in organic dairy farming.	Master Thesis	UEF	Higher education	Finnish	Finland

#### **4.2 Further possible actions for dissemination**

- List publications/deliverables arising from your project that Funding Bodies should consider disseminating (e.g. to reach a broader audience)

A leaflet describing the main results from the project is planned together with the CORE organic administration.

- Indicate publications/deliverables that could usefully be translated (if this has not been done, and indicate target language)

Translations of the leaflet presenting the main results to the languages of the involved countries are planned together with the CORE organic administration.

### 4.3 Specific questions regarding dissemination and publications

- Is the project website up-to-date?

No, the new webpage is under construction. The old webpage is closed down by CORE organic.

- List the categories of end-users/main users of the research results and how they have been addressed/will be addressed by dissemination activities

Farmers and advisors:

- Publication of the results in farmers magazines
- Publication of the results on workshops
- Presentation of the results in a leaflet (planned to be done in 2012)
- Presentation of the main results on the project home-page (planned to be done in 2012)

Researchers:

- Presentation of the results in peer reviewed research journals
- Presentation of the results at research conferences
- Presentation of the main results on the project home-page (planned to be done in 2012)

General public:

- Presentation of the main results on the project home-page (planned to be done in 2012)
- Presentation of the results in a leaflet (planned to be done in 2012)

- Impact of the project in relation to main beneficiaries of the project results

Note: for the different categories of end-users/main users of the research results, explain how well the project has been able to reach these target groups, and any known impact

When the original home page was working it had many (hundreds) of visitors each month, we hope that the new home page will be as popular as the original.

Workshops and conferences have been well attended.

We hope that the peer-reviewed articles that will be a result of this project will be read and cited.