

Project title

Name: **Tina Slots**
University: Copenhagen University, LIFE
Department: Dept. of Dairy and Food Science
Supervisor: Head of Research Unit Jacob Holm Nielsen, DIAS/Professor Leif H. Skibsted
Timescale: February 2003 – April 2008
E-mail/phone: tina.slots[a]agrsci.dk/89991248
Master's degree: Dairy Science

Background

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 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 endogenous antioxidants (uric acid and glutathione peroxidase) in order to produce oxidatively stable milk and dairy products.

Objective

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.

Progress – 2007

Tina Slots has returned from maternity leave on the 26th February 2007.

Survey of composition of conventional and organic milk (continued)

Data have been published in the International Dairy Journal.

Survey of milk composition in relation to shelf life in Danish milk (continued)

Data have been treated and is going to be published in the near future.

Survey of composition of conventional and organic milk (continued)

Data are about to be published.

Model experiment with α -tocopherol incorporated into the fat globule membrane of milk

There is a need to understand the mechanism of the antioxidative effect of α -tocopherol in the milk, and therefore a heterogenous model of the milk fat globule with α -tocopherol incorporated will be created. As a start, a model system consisting of α -tocopherol and riboflavin in a homogeneous system (with *tert*-butanol) has been created. The model system has been exposed to light or copper for a given period in order to start the oxidation, and the oxidation of tocopherol was followed by LC and LC-MS. A method to quantify the oxidation products of α -tocopherol was prepared as described by Nagata *et al.* (2004). The detection of tocopherol and the oxidation product tocopherylquinone was done by UV-detection at 268 and 290 nm with stepwise elution (0-7 min.

methanol, 7.1-20 min. methanol/ethyl-acetate (4:1) – 1 mL/min.). The detection of analytes on the MS-system was attained by negative-ion with an APCI interface using the selected-ion monitoring (SIM) technique. The Gas and vaporizer temperature was set at 350 and 450°C, respectively. The Capillary and the fragmenter voltage was set to 3000 and 160 V, respectively. The Corona Current was set to 15µA, the nebuliser pressure to 48 psig and the drying gas to 7.0 L/min. Selected-ion chromatograms at m/z 429 and 446 was used to calculate the peak area of α -tocopherol and tocopherylquinone, respectively. The data need to be evaluated further in order to quantify the oxidation product tocopherolquinone on the LC-MS system.

Plans - 2008

Survey of milk composition in relation shelf life in Danish milk (continued)

Data will be published in the near future.

Metal catalysed oxidation and photooxidation in milk (continued)

There is need for more research on this area, and this will be done as a part of the model experiment described below.

Model experiment with vitamin E incorporated into the fat globule membrane of milk

The heterogeneous model system will be exposed to different stress factors such as light or copper, and the degradation of α -tocopherol will be followed by LC-MS or other relevant analyses such as ESR.

Publications

Peer-reviewed and accepted

Slots, T.; Skibsted, L.H. and Nielsen, J.H. (2007) [The difference in transfer of all-rac- \$\alpha\$ -tocopherol stereo-isomers to milk from cows and the effect on its oxidative stability](#). *International Dairy Journal* 17(7):pp. 737-745.*

Stagsted, J. (2006) [Absence of both glutathione peroxidase activity and glutathione in bovine milk](#). *International Dairy Journal* 16(6):pp. 662-668.*

Not peer-reviewed

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

Slots, Tina; Leifert, Carlo; Butler, Gillian; Kristensen, Troels and Nielsen, Jacob Holm (2006) [Effect of dairy management on quality characteristics of milk](#). Paper presented at Joint Organic Congress, Odense, Denmark, May 30-31, 2006..

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

Lund-Nielsen, Tina; Mejer, Helena; Gunnarsson, Carina; Hansen, Preben Klarskov and Grigalaviciene, Ilona (2003) [How is ideologies related to actions?](#) Report, Unit for Learning and Interdisciplinary Methods, Royal Veterinary and Agricultural University.

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