Technical Annex

Scientific description of the project

Title: Organic cropping Systems for Vegetable production, - product Quality, natural Regulation and Environmental effects

Acronym: VEG-QURE

Duration: **From:** November 2005 **to:** October 2010

Participating scientists:
(Titles, names and brief institutional affiliations of scientists in the project. Head of project is written in bold, participants responsible for work packages are underlined).

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English summary (1 page, suitable for publication):
The objective of this project is to contribute to our understanding of the effects of organic cropping systems on vegetable quality and on the environment. Producing food of a better quality and in a more environmentally friendly way are some of the main goals of organic farming. Sales and public support for organic farming are based on the expectation that these things are achieved. In order to increase the support for organic farming, there is a clear need to show that the goals are actually achieved.

This is difficult to document, as the results of many years of research have shown. This is difficult for two main reasons: 1) effects on quality and on the environment are highly complex and thus difficult to study, and 2) organic farming is not just one well defined cropping system; within the rules for organic farming, many very different systems can be made. Organic farming is developing and changing, and this can make consumers and politicians question whether it is moving away from its original ideas.

To answer some of these questions, we will compare four different cropping systems for vegetable production, one conventional and three organic systems. All of the organic systems will comply with the regulations of organic farming, but in very different ways. One system will be much like the conventional, though using organic fertilizers and no pesticides. In the second organic system, green manure or catch crops will be grown in the autumn as often as possible. In the last system, stripes of the green manure will be left at soil tillage, to grow as intercrops between the rows of the vegetable crops, in order to improve the conditions for organisms involved in natural regulation.

Together, these systems represent large variations in growth conditions for the crops. There are systems with or without use of chemical inputs, but also large variations in the biological surroundings which the crops will meet in terms of insect pests, fungi, beneficial organisms and other plants. There will also be large differences in nutrient balances and the availability of plant nutrients, creating effects on the environment and on crop quality.

Vegetable quality will be studied in a number of ways, to cover a range of this highly complex question. We will measure contents of secondary metabolites in cabbage, lettuce and carrot, focussing on metabolites which are known to affect human health. We will measure quality parameters important for product marketing, but also taste and volatile aroma compounds. Aspects of product storability will be measured. Carrots from the different cropping systems will be stored, and the interactions between their physiology, metabolic activity, contents of secondary metabolites and the development of fungal diseases will be studied. With the disease liquorice rot, detailed studies of the interactions during the storage time will be made.

To get an even broader picture of the effect of cropping systems on the crop, we will examine them with the methods of proteomic analysis. These methods show which proteins are expressed in the plants, and for which proteins the level of expression is affected by the cropping methods. Subsequently, the function of the responding proteins can be elucidated. The results of the proteomic analysis will supplement the more specific measures of quality obtained by the other methods.

Natural regulation mechanisms in the field will be studied with a focus on insect pathogenic fungi in the soil, and prevalence and diversity of insects. We will examine how the insect pathogenic fungi interact with pest insects. By studying this in the four systems, we can see how the use of pesticides and inorganic fertilizers, growing of autumn green manures or intercropping affects natural regulation.

In addition the effects of the cropping systems on nutrient balances, root growth and effect on nitrogen leaching losses will be studied. The conventional and two of the organic systems are already ”well developed” systems, but the system with intercropping is not. This system holds many promising possibilities, but also problems. Therefore, we will work on developing this system further. We will study the yield and cost of the different systems, and compare this to their effects on quality, biodiversity, natural regulation and the environment. In this way we will illustrate the cost of the systems, and the tradeoffs between quality, environment and production economy which are likely to be found. While many of the quality measures are specific, we get many different measures. We will use this to study whether more general effects on crop composition and quality can be found.

A.0 Introduction, state of the art and objectives of the project (max. 3 pages):

The main objective of this proposal is to study effects of different types of organic cropping systems. We want to study effects on product quality and composition of vegetables, on natural regulation of pests and on environmental effects.

When consumers buy organic products, or when politicians give support for organic farming, they expect that organic products are of better quality and that organic production systems are more environmentally friendly than conventional systems. While some aspects of this are obvious (e.g. effects on pesticide residues in food) others are not. There are two main problems making such studies and conclusions difficult, 1) the effects on quality and environment are highly complex, and therefore difficult to measure, and 2) organic farming is not a well defined system, it can be done in very different ways, leading to very different effects on quality and the environment. Though it is difficult to study, it is highly important for organic farming to be able to document benefits; only in this way will it be feasible to maintain and even increase the confidence and interest from the public. Some studies have indicated that organic farming products are better for human health than conventional products. If this can be confirmed in further studies, it can give an important boost to the market for organic vegetables and other products.

Organic farming is continuously developing and changing. Organic farms become larger, more specialised and often more commercially oriented. Much of this development may be necessary to increase the organic food production, and to keep the price premium as low as possible, thereby favouring the sales of organic products. However, the development may reduce some of the benefits from organic farming, or at least make the public suspect that modern organic farming becomes “less organic”. This is an important question regarding the integrity of organic farming. It is important to reach the goals set and communicate to the public and it is equally as important to show that organic farming continually works towards fulfilment of its goals. All in all, it is becoming important to compare not only conventional vs. organic production methods, but also to compare the effects of different approaches to organic farming.

Comparisons of different organic cropping systems are therefore important for future development of organic farming. If the absence of pesticides and inorganic fertilizers are the main reasons for positive effects on quality and environment, then organic farming can develop rather freely within the current set of regulations. But if there are significant differences among organic production systems, this will be important for the directions which further development of organic farming must take.

The effects of avoiding chemicals are the direct result of compliance with the regulations, and do not depend on the way in which organic farming is practiced. However, many other effects of organic farming do not follow automatically from compliance with the regulations. Regulations limit the access to nitrogen, and this is assumed to lead to a lower nitrogen surplus and a more careful use of the limited resources, thereby reducing excess and loss. However, excessive fertilization can also be made in organic farming, leading to nitrogen leaching losses and effects on crop quality. As vegetables are high value crops, farmers tend to prioritize them. For this reason, excessive fertilization seems to happen frequently in some types of organic vegetable production.

High input of organic matter to the soil is not guaranteed by the rules of organic farming either. Although the absence of pesticides will favour the biological activity in the fields, other aspects of the cropping systems, which favour field biology, are not guaranteed. Reaching many of the goals of organic farming therefore requires more than just compliance with the regulations; active strategies to farm in a better way are also needed.

We will address these issues by comparing one conventional vegetable production system (C1) to three different organic cropping systems (O1, O2 and O3) clearly varying in the aspects listed above. All the organic systems will comply with the rules for organic farming, but in one system (O1) little else will be changed compared to the conventional system. In the second system (O2) we will grow green manures as often as possible during the autumn and winter season, thereby reducing nitrogen leaching losses, reducing the need for nutrient import and improving the organic matter input to the soil and the living conditions for the field biology. The third system (O3) will also include green manures as O2, but we will allow stripes of the green manure from one autumn to remain as intercrops between the vegetable crop rows in the next year. Leaving the intercrop stripes will allow more soil animals to survive compared to full soil tillage (e.g. Axelsen and Thorup-Kristensen, 2000) and give much better conditions for population regrowth after soil tillage. The O3 system will have the same advantages as the O2 system, but allow even better conditions for natural regulation mechanisms. It may also lead to less leaching loss and better pre-crop effects of the vegetable crop, as the intercrops will function as green manure crops after harvest. However, the O2 and O3 systems are increasingly complex to handle, with increasing cost and risk of yield loss; this is especially true for the O3 system.

Work in previous and current projects (some mentioned under international cooperation), and especially the current DARCOF project VegCatch has shown how substantial possibilities for improving organic cropping systems for vegetable production. By optimal use of crop rotation, catch crops and green manures, systems which are less dependent on external inputs, loose less nutrients and offer better living conditions for the microorganisms and fauna of the soil and fields can be developed (Thorup-Kristensen *et al.* 2003).

There are many reasons why organic crops may posses a different and better quality than conventional crops, and why differences in effects on field biodiversity and other environmental effects may be expected. Herbicides are plant toxins, and are known often to harm also the crops they are meant to protect, though harming them at sub-lethal levels. Fungicides and insecticides strongly affect the "biological environment" of the crops as they are meant to do, and reduce the need for the plant to defend itself. Systemic fungicides spread within the plant vascular system, and they are likely to affect mycorrhizas and other microorganisms living in the rhizosphere of the crop roots. Inorganic fertilizers can give a strong and unbalanced nutrition of the crop. With a high nutrient availability, the crops are induced to grow at maximum rates, and thereby to use practically all of their photosynthesis products for growth. This leaves little surplus for other functions of the plants, such as the production of secondary metabolites (Stamp, 2003).

A diet rich in fruits and vegetables has a protective effect against cancer and cardiovascular diseases (van Poppel, 1996). A variety of secondary metabolites (bioactive compounds) other than those conventionally recognized as nutrients may contribute to this protection (Johnson et al, 1994; Picman, 1986). Many of the same compounds also have important effects on taste. It is therefore important to know how our crop production methods affect the content of these compounds in our crops.

Some of the most important groups of secondary metabolites in vegetables are polyacetylenes (PAs), sesquiterpene lactones (SQLs) and glucosinolates (GSLs). SQLs found e.g. in lettuce (Hansen and Boll, 1986). These compounds have anti-tumour effects (Sessa et al, 2000) and are also known for anti-fungal and antibiotic properties and their contribution to bitter taste.

Aliphatic C_{17} PAs of the falcarinol-type are found in carrots and related vegetables where they seem to have a defensive role, against invading fungi (Brandt et al., 2004). They are highly toxic to mammalian cells (Brandt et al., 2004; Bernart et al., 1996; Hansen et al., 2003), and falcarinol isolated from carrots have been shown to reduce the development of cancer in rats (Kobæk-Larsen et al., 2005). Further, PAs have recently been recognised as the most important bitter principles in carrots.

GSLs occur in brassicas. GSLs and their breakdown products (isothiocyantes) have long been known for their fungicidal, bacterial, and nematocidal effects (Hecht, 1999). In particular the isothiocyanates have been shown to protect against carcinogenesis, and today the GSLs are among the best-proven cancer-preventing principles in vegetables. GSLs have also been shown to have a great impact on the flavour of cabbage.

The cropping systems we propose to study, will affect the plant nutrition and the biological environment of the vegetable crops in very different ways, and through this, they are likely to affect the production of these important secondary metabolites within the plants, and many other aspects of plant quality.

The studies of secondary metabolites in vegetables from the four cropping systems will show us examples of the effects of cropping systems. However, it will be examples of effects on single compounds. The results are likely to show a scattered picture of increasing as well as decreasing content due to the organic farming methods (Nørbæk *et al.* 2003), and they may tell little about the more basic reactions of the plants to the cropping systems. The methods of proteomic analysis (Fey and Larsen, 2001; Dierick *et al.* 2002) can help us towards a more basic understanding of this. It can be used to show which proteins in the plant are affected by the treatments, and to identify them and their function in the plant.

Building on previous experience in working with for example crop fertility (sugar beet roots) and growth stress due to drought and elevated temperature (poplar leaves, Dierick *et al.* 2002), new methods will be developed specifically for the vegetables in this study, optimised to obtain quantitative extraction of proteins while removing cellulose, phenolic and other compounds which would otherwise prevent high resolution proteome analysis. The methods to be used allow up to 20,000 proteins to be separated and quantified, and further analysis of the proteins, which are then found to respond to differences in cropping system.

Studying storability of products is another broad method of looking at cropping system effects on quality. The storability of carrots depend on a number of factors, such as lesions due to pest attack in the field, the specific fungi carried from the field to storage, content of secondary metabolites and physiological status of the plants. Physiological status is related to the content of energy reserves such as sugars, and to the metabolic activity of the products. Calorimetric measures of the heat production rate can be used to quantify even small changes in metabolic activity (Criddle et al., 1991).

The relative storability and disease development carrots will be compared in relation to the different organic cropping systems. It is a traditional idea of organic farming that the “energy balance” of the products is better, which will allow e.g. improved storability. This could be another aspect of the effects of inorganic fertilizers boosting crop growth and reducing the surplus of assimilates for other processes (Stamp, 2003).

Some post harvest diseases are caused by “cold” pathogens, which are transmitted from the field via soil adhering to the carrots. Recent studies from Danish soils liquorices rot to be the most important of these diseases. It develops after 4-5 month of storage at temperatures even as low as 0-2°C (Knudsen et al, in prep). Resistance to late infection depend on many different plant factors eg. antifungal substances (Davies et al. 1981). Among the secondary metabolites studied in this project, especially falcarinol have been shown to be important for disease development during storage (Davies & Lewis, 1980).

According to the IFOAM principles natural regulation of pests is among the most desired wishes for organic production. Natural regulation of pest insects is dependent on the action of two major groups of organisms: a) insect pathogenic microorganisms and b) predators and parasitoids. More sophisticated knowledge on the action of natural regulation and the actions needed to support the involved organisms is therefore essential for successful organic cropping. The principles behind natural regulation are similar to the concept of ‘conservation biological control’ (Eilenberg *et al.,* 2001) so the protection and enhancement of natural enemies is a key element (Landis *et al*., 2000).

Among the microorganisms, insect pathogenic fungi are significant in natural regulation of insects (Eilenberg and Meadow, 2002). These fungi occur more frequently in organically grown fields than in conventional fields (Klingen *et al.,* 2002), and they seem to be affected by crop rotations (Meyling *et al*., 2004) Obviously, a cropping system with rotation and intercropping will strongly influence natural regulation, but there is a lack of studies over several years in plots with precise information about environmental factors.

In previous projects it has been shown that efficient use of catch crops and green manure crops can supply much of the nitrogen needed by organically grown crops (Thorup-Kristensen *et al.* 2003). At the same time, these fertility building crops can efficiently reduce nitrogen leaching losses and improve the living conditions for the soil biology. The results make it obvious to develop organic rotations relying increasingly on green manure crops. However, there are also costs involved, the cropping systems become more complex to handle, in some yield losses will also occur. To increase the use of green manure and catch crops, and make use of all the advantages they offer, developments towards better methods of employing green manure crops are needed. Some cost can be accepted if the benefits are great enough, and this also needs further studies. Advantages and disadvantages in agronomic terms have been shown, but benefits in terms environmental effects and especially of product quality need further study. If these advantages are large enough, some costs in agronomic terms are acceptable. This is a crucial aspect of the ideas of organic farming, where considerations of environment and quality do to a certain extent have higher priority than economy.

In this project it is our objective to compare the quality and environmental consequences of different cropping systems. We will for the first time measure many factors including effects on many specific secondary compounds in the vegetables combined with broader analysis of taste, metabolic activity, storability and protein expression of products from different organic cropping systems. The specific compounds of study will be chosen among compounds with known effects on human health and vegetable taste. We will study insect biodiversity and natural regulation mechanisms in the field, with the inclusion of molecular and ecological tools as well as cropping system effects on nutrient balances and nitrogen leaching. For the first time, these results will also be synthesized to achieve a full understanding about the most significant effects (and novel interactions) of the cropping systems.

A.1 Technical content of the research activity

(The work plan should be divided into work packages (WP1 to WPn), each with different objectives, milestones and deliverables.

The organizational core activity of the VEG-QURE project is the cropping system in the field. From the cropping system, plant material, field plots and measurement data must be made available for the other work packages. Most of the other activities and the main objectives of the project depend on the success of this. This requires not only that the cropping system is grown as planned, but also a considerable amount of organization and management, to make sure that the deliverables from the cropping systems are made available for the other work packages at the right time, amount, form etc. Therefore, the task of running the cropping systems is combined with the task of managing the VEG-QURE project in WP1.

The fact that the project is built around the cropping system has other effects on organization and planning as well. Some of the cropping system effects are "present" already in the first year of vegetable cropping, but others are rotational effects which are not. To reduce this problem, we have initiated the treatments on our own funding already from August 2005. But even so, the vegetables which will be grown in 2006 are not really representative of the cropping systems. Therefore the activities of 2006 will be focused on method development and initial studies such as the mapping of the "start occurrence of insect pathogenic fungi" in the soil.

Then, during the years 2007, 2008 and 2009, vegetables will be produced which should be representative for the cropping systems. The studies in the field as well as in the lab will have to work with vegetables from all three years, to cover the year to year variation always encountered in field studies. This means that even where relatively fast laboratory methods are used, the work will still have to be spread out over the whole period. This also leaves the publication activities for the last period of the project. For this reason we have scheduled the project to continue for a full 5-year period until October 2010, to have a final year with little experimental activity leaving time for the publication work.

There are many interactions between partners in the work packages. However, there are still one partner who are the main responsible for each work package, and responsible for the direct coordination of this. Some of the objectives and deliverables of the project are obtained mainly from single work packages, though off course based on the cropping system in WP1. Other objectives and deliverables are synthesis across the work packages. Some involve two work packages, e.g. the involvement of the proteomic analysis in the other work packages. A main objective of the project is synthesis across the whole project, and the deliverables related to this are collected in WP6.

Workshops including the whole group will be held once a year, to exchange results and ideas and promote further cooperation which is not already planned. At these workshops it will also be important to make a follow up on the project parts and make adjustments if needed. The meetings will also be used to plan and coordinate project communication towards users and others. The discussions in the group as a whole will mainly focus on scientific aspects as well as practical matters and publication plans.

A "Management group" consisting of the responsible scientist from each of the five partners will be formed to formal aspects of the work, and to follow the overall progress of the project and discuss what to do where things may not be proceeding as planned. This will save time for discussions of scientific aspects when the whole group meets, and concentrate discussions of management to a smaller and more directly responsible group. This group will meet in connection with the yearly project workshops. Apart from these meetings, this group will discuss via electronic communication or meet ad hoc if needed.

In the project we have planned to have one PhD project. The PhD study is planned to form part of the activities of WP4, and funding for this is part of the WP4 budget. The plan is to apply for additional funding form the research school for organic farming (SOAR) and from other sources. We hope that this will be possible, but if we cannot get the additional funding as planned, we will use the same funds to try to finance a PhD study in one of the other WPs.

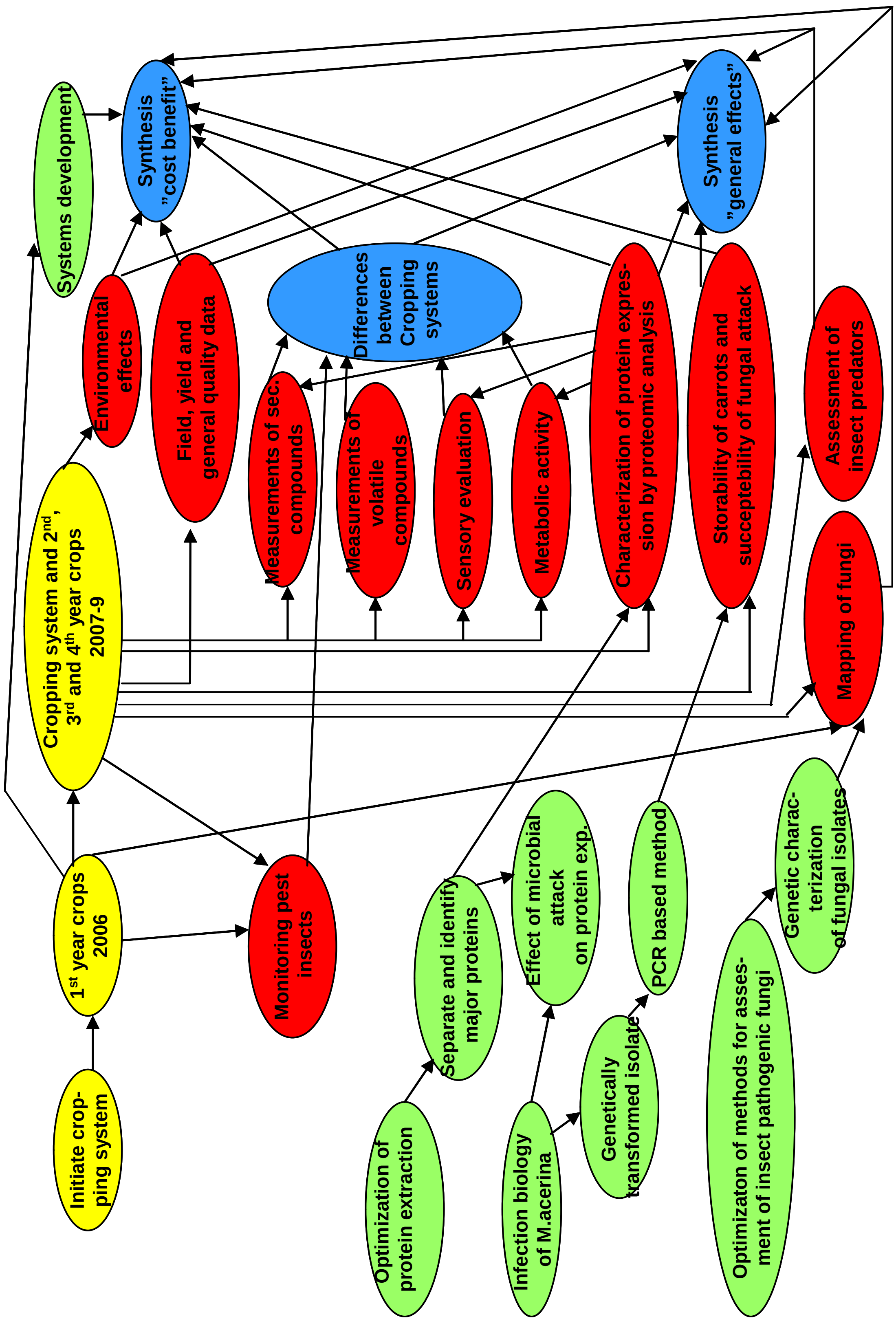
Gantt diagram showing the timing of tasks in the VEG-QURE project

	TASKS	2005 (months 8 10 12)	2006 (months 2 4 6 8 10 12)	2007 (months 2 4 6 8 10 12)	2008 (months 2 4 6 8 10 12)	2009 (months 2 4 6 8 10 12)	2010[3] (months 2 4 6 8 10)
	WP 1: Cropping systems and project management						
1	Coordination of the Veg-Qure project	X X	X X X X X X X X X X X X	X X X X X X X X X X X X	X X X X X X X X X X X X	X X X X X X X X X X X X	X X X X X X X X X X
2	Arrange international workshop				X X X X	X X X	
3	Designing and growing the cropping systems[1]	X X X X X	X X X X X X X X X X X X	X X X X X X X X X X X X	X X X X X X X X X X X X	X X X X X X X X X	
4	Documentation of the cropping systems and deliveries for other WPs		X X X X X X X X X	X X X X X X X X X X X X	X X X X X X X X X X X X	X X X X X X X X X X X X	
5	Single factor experiments[2]			? ? ? ? ? ? ?	? ? ? ? ? ? ?	? ? ? ? ? ? ?	
	WP 2: Effect of cultivation systems and storage on bioactive compounds, sensory quality and metabolic activity						
1	Isolation, identification and quantification of bioactive compounds from vegetables		X X X X X X X X X X	X X X X X	X X X X X	X X X X X	X X
2	Sensory evaluation of vegetables			X X X	X X X	X X X	
3	Isolation, identification and quantification of volatile compounds			X X X	X X X	X X X	
4	Metabolic activity during storage		X X	X X X X X X X	X X X X X X X X X	X X X X X X	
5	Relationship between bioactive compounds, volatiles, sensory attributes and cropping systems, metabolic activity and resistance to pathogens					X X X X X X X X	X X X
	WP3: Characterisation of Protein Expression in Vegetable Crops grown under different conditions						
1	Optimise extraction procedures		X X X X				
2	Separate proteins by two dimensional gel electrophoresis		X X X X X X	X X	X X	X X	
3	Identify some of the major proteins		X X X X X	X X	X X	X X	
4	Compare the vegetables grown under different conditions and harvested at different years.			X X X X X	X X X X X	X X X X X	
5	Identify effects of microbial attack on stored carrots			X X X	X X X	X X X	
6	Carry out bioinformatic analysis		X X	X X	X X	X X	
	WP 4: Storage product quality						
1	Characterization of the infection biology of M. acerina		X X X X X X X X X X	X X X X X X X X X X X X	X X X X X X X X X X X X		
2	Development of a genetically transformed isolate of M. acerina		X X X X X	X X X X			
3	Development of PCR based method			X X X X X X X X X X X X			
4	Investigation of the role of agricultural system input			X X X X	X X X X X X X X X X	X X X X X X X X X X X X	X X X
	WP5: Natural regulation of insects with particular focus on soil living stages and the influence of cropping system						
1	Optimisation of methods for assessment of insect pathogenic fungi		X X X X X X X X X X X X	X X X X X			
2	Genetic characterization of isolates		X X X X X	X X X X X X X	X X X X X X X X X X X X		
3	Mapping of occurrence of fungi		X X X	X X X X X X X	X X X X X X X X X		
4	Assessment of insect predators			X X X X X	X X X X X	X X X X X	
5	Elucidating the effects of selected proteins			X	X X X X	X X	X X
6	Monitoring pest insect species			X X X X X X	X X X X X X	X X X X X X	
	WP 6: Cropping systems, environment, crop composition and system development						
1	Cropping system effects on nutrient leaching losses		X X X X X X X	X X X X X X X X X X X X	X X X X X X X X X X X X	X X X X X X X X X	
2	Development of cropping systems		X X X	X X X X X X X X X	X X X X X X X X X X X		
3	Analyse general effects of cropping systems on product quality and natural regulation					X X X X X X X X X	X X X X X X X X X X
4	Quantification and presentation of tradeoffs					X X X X X X X X X	X X X X X X X X X X

1: Task 1.3 will be started on own funding in August 2005, as in this way we can clearly improve the initiation of the cropping systems for 2006

2: Specific plans will be based on results from other WPs

3: Due to the nature of the field studies, much of the publication work must necessarily be postponed until the resutls of 2009 are finished, therefore 2010 is reserved mainly for publication work


Systems development
Synthesis
"cost benefit"
Synthesis
"general effects"
Environmental
effects
Field, yield and
general quality data
Differences
between
Cropping
systems
Measurements of sec.
compounds
Measurements of
volatile
compounds
Sensory evaluation
Metabolic activity
Characterization of protein expres-
sion by proteomic analysis
Storability of carrots and
succeptebility of fungal attack
Assessment of
insect predators
Mapping of fungi
Cropping system and 2nd,
3rd and 4th year crops
2007-9
1st year crops
2006
Monitoring pest
insects
Initiate crop-
ping system
Optimization of
protein extraction
Separate and identify
major proteins
Effect of microbial
attack
on protein exp.
Infection biology
of M.acerina
Genetically
transformed isolate
PCR based method
Genetic charac-
terization
of fungal isolates
Optimizaton of methods for asses-
ment of insect pathogenic fungi

Work Package list

WP No.	WP title	Responsible scientist	Budget DKK	Start	End	Deliverable No.
1	Cropping systems and project management	Senior Scientist Kristian Thorup-Kristensen	2.460.000	Nov. 2005	Oct. 2010	D1.1-D1.8
2	Effect of cultivation and storage on bioactive compounds and sensory quality	Senior Scientist Lars Porskjær Christensen	2.840.000	Jan. 2006	Oct. 2010	D2.1-D2.4
3	Characterisation of Protein Expression in Vegetable Crops grown under different conditions	Associate Professor Peter Mose Larsen	1.000.000	Jan. 2006	Oct. 2010	D3.1-D3.4
4	Storage product quality	Associate Professor Dan Funck Jensen	2.010.000	Jan. 2006	Oct. 2010	D4.1-D4.5
5	Natural regulation of insects with particular focus on soil living stages and the influence of cropping system	Professor Jørgen Eilenberg	2.340.000	Jan. 2006	Oct. 2010	D.5.1- D5.5
6	Cropping systems, environment, crop composition and system development	Senior Scientist Kristian Thorup-Kristensen	2.850.000	Jul. 2006	Oct. 2010	D.6.1-D6.4
Total			13.500.000			

(Please give month and year for start and end)

Deliverables list

Deliverable No	Deliverable title	Lead scientist	Delivery date	Allocated scientific person mothts	Type of deliverable
D1.1	Yearly status reports.	KTK	Continuous	3	R
D1.2	Popular papers on the cropping systems and the results obtained	KTK	Continuous	1 (2)	P
D1.3	Project homepage, basic project information and current topics, updated at least four times a year.	KTK	Feb. 06 + continuous	2 (3)	O
D1.4	International workshop on the overall themes of the project	KTK	Apr. 09	2 (3)	C
D1.5	Presentation of the project at meetings, open field days etc.	KTK	Continuous	2 (3)	C
D2.1	Oral or poster presentations of the results at national and international workshops/congresses.	LPC	Before Oct. 09	1.5	C
D2.2	Manuscript 'Effect of organic cultivation systems on secondary metabolites in selected vegetables' will be published in an international reviewed journal.	LPC	Jun. 10	7	S
D2.3	Manuscript 'Effect of organic cultivation systems on flavour compounds and sensory properties in selected vegetables' will be published in an international reviewed journal.	LPC	Jun. 10	7	S
D2.4	One or two manuscripts on correlation between secondary metabolites, volatiles, sensory attributes and the metabolic activity, resistance towards pathogenic attack during storage	LPC	Oct. 10	8	S
D3.1	Methods description for the preparation of protein samples from the vegetables.	SF	May 06	1 (3)	R
D3.3	List of proteins whose expression is affected by the growth conditions at the end of each growing season. Aimed at possible patent application(s)	SF	Aug. 07, Aug. 08, Aug. 09	2 (6)	R
D3.4	Two or three manuscript describing the effect of growth conditions on the protein composition in lettuce, carrot and cabbage.	SF	Aug. 10	2 (6)	S
D3.5	Presentation of the data at scientific meetings and congresses	SF	Continous	1	C
D.4.1	Paper on a method to inoculate carrots and a defined plant assay	IK	Apr. 07	6	R
D 4.2	Paper on a connection between physiologically status and etiology of *M.acerina*	BJ	Dec. 08	12.5 (15)	S
D 4.3	Paper on the role of agricultural system on storage quality	DFJ	Mar. 10	10	S

D5.1	Laboratory protocol for assessing insect pathogenic fungi in soil in small plots	NVM	Aug. 06	4 (6)	R
D5.2	Report on genetic characterisation of insect pathogenic fungi	NVM	Oct. 07, Oct. 08	6	R
D5.3	Contribution to FØJO publication on insect pathogenic fungi in organic agro-ecosystems	NVM	Oct. 06, Oct. 07	6	O
D5.4	Scientific paper on analysis of three years prevalence of insect pathogenic fungi in soil	NVM	Feb. 09	10	S
D5.5	Presentation of preliminary project results at international conference (SIP/IOBC)	NVM	Apr. 07, Oct. 08	4	C
D5.6	Scientific paper on biodiversity assessment (in cooperation with WP6)	NVM	Feb. 09	6 (8)	S
D5.7	Scientific paper on analysis of insect population regulation with emphasis on soil living stages	JE	Dec. 09	5 (6)	S
D6.1	Scientific paper on the effect of cropping systems on nutrient balances and N leaching loss	KTK	Dec. 09	11	S
D6.2	Scientific paper on methods for limiting competition and yield loss by intercropping in vegetable production	KTK	Feb. 09	11	S
D6.3	Scientific publications analysing general effects of cropping systems on crop composition and quality and on field biology. This will be at least one publication, but may be more if data allows this.	KTK	Aug. 10	15	S
D6.4	Report analysing and illustrating costs and benefits of the four cropping systems	KTK	Feb. 10	6	R

(The nature of the deliverables must be indicated by S = publication in scientific journal with peer review; P = publication in journals without peer review; R = reports; C = presentation at meetings and congresses or O = other types of deliverables, e.g., prototypes, models, websites, etc.).

Abbreviations: KTK: Kristian Thorup-Kristensen, LPC: Lars Porskjær Christensen, SF: Stephen Fey, NVM: Nicolai V. Meyling, DFJ: Dan Funck Jensen, BJ: Birgit Jensen, JE: Jørgen Eilenberg

Milestones list

Mile-stone No	Milestone title	Lead sci-entist	Delivery date
M1.1	Finally deciding on crop species, crop management practices and green manure crops for the four cropping systems	KTK	Mar. 06
M1.2	Delivery of vegetable products for other WP2, mainly WP2, WP3 and WP4	KTK	2006-09
M1.3	Making field plots in the cropping systems available for other WPs, mainly WP5 and WP6	KTK	2006-09
M1.4	Delivery of soil, crop and management data for all other WPs	KTK	2006-09
M1.5	Delivery of yearly report with detailed presentation of field data from the cropping systems	KTK	Dec. 06, Dec. 07. Dec. 08, Dec. 09
M1.6	Vegetables or field plots from simple single-factor experiments made available for other WPs, to be planned during the project	KTK	2007-09
M1.7	Field experiments finalized and final data delivered to other WPs	KTK	Jan. 10
M1.8	Organizing contributions from the project partners to DARCOF electronic communication, popular presentations and publications	KTK	2005-2010
M2.1	Analytical methods for isolation and quantification of bioactive and volatile secondary metabolites have been established and optimized	LPC	Aug. 06
M2.2	A sensory profile of important attributes for carrot, lettuce and white cabbage has been developed	LPC	Sep. 06
M2.3	Preliminary results of bioactive and volatile secondary metabolites and sensory analysis of crops from the cultivation years 2007, 2008 and 2009 are completed	LPC	Dec. 09
M2.4	Testing of the potential of calorimetry for determination of metabolic activity in carrot during storage has been completed	LPC	Apr. 07
M2.5	Preliminary results of bioactive and volatile secondary metabolites and metabolic activity from post harvest storage of carrot for two cultivation years	LPC	Jun. 09
M2.6	Data for resistance of pathogens, cultivation, metabolic activity, protein expression, sensory evaluation and bioactive and volatile secondary metabolites has been correlated	LPC	Apr. 10
M3.1	Selection of the optimal method for sample preparation	SF	Apr. 06
M3.2	Initiation of the image databases for each of the vegetables	SF	Jun. 06
M3.3	Identification of the 25th protein for the database	SF	Oct. 06
M3.4	Image database including some proteins identified.	SF	Dec. 07
M3.5	Completion of the yearly analysis of lettuce	SF	Sept. 06, Sept. 07, Sept. 08, Sept. 09
M3.6	Completion of the yearly analysis of carrots	SF	Sept. 06, Sept. 07, Sept. 08, Sept. 09

M3.7	Completion of the yearly analysis of cabbage	SF	Sept. 06, Sept. 07, Sept. 08, Sept. 09
M4.1	A plant bioassay has been developed	IK	Aug. 06
M4.2	A fluorescent transformant of *M. acerina* has been developed and tested stabile	BJ	Apr. 07
M4.3	A method for further studies of liquorices rot development has been developed	IK	Apr. 07
M4.4	A method to investigate natural populations of *M. acerina* in situ during infection has been developed	BJ	Dec. 07
M4.5	Interaction between physiologically status and etiology of *M. acerina* has been evaluated	IK	Dec. 08
M4.6	The role of agricultural system on storage quality has been described	DFJ	May 10
M5.1	Development of method for quantification of populations of insect pathogenic fungi in soil	NVM	Jul. 06
M5.2	Selection and testing of molecular markers for identification of insect pathogenic fungi at isolate level	NVM	Aug. 07
M5.3	Evaluation of data on insect pests and their natural insect enemies related to the cropping system (for WP6)	NVM	Oct. 08
M5.4	Evaluation of cropping factors on natural regulation of insect populations by insect pathogenic fungi in organic agriculture	NVM	Apr. 09
M5.5	Evaluation of effects of selected compounds (from WP3) on selected insect pathogenic fungi *in vitro*	JE	Dec. 09
M6.1	Define "hot spots" with high N leaching risk to be studied in the cropping systems	KTK	Aug. 06
M6.2	Define intercropping systems to be tested in exps. during 2007/08. First intercrops must be established in the spring of 2006.	KTK	Mar. 06
M6.3	Asses effects of different intercropping systems on crop yield and on N competition between crops and intercrops	KTK	Dec. 08
M6.4	Agree with project partners on which data can and should be included in paper(s) on general system performance (D6.3)	KTK	Oct. 09

Abbreviations: KTK: Kristian Thorup-Kristensen, LPC: Lars Porskjær Christensen, SF: Stephen Fey, NVM: Nicolai V. Meyling, DFJ: Dan Funck Jensen, BJ: Birgit Jensen, JE: Jørgen Eilenberg

Description of work packages (this page should be copied for each WP**)**

WP No.: 1 Cropping systems and project management

	Start date or starting event: (please give month and year) Nov. 2005								
Partner id.	DIAS-Hort								
Person-months per participant	10 VIP 33 TAP								
Total PM VIP **Total PM:**	**10** **43**								

Objectives:

- Manage the VEG-QURE project, coordinate activities, reporting, communication of results
- Design and run the vegetable cropping system experiment with four different cropping systems
- Document methods and results of the cropping systems as background for the other WPs
- Make crop material, field data and field plots from the cropping systems available for the other WPs
- Perform small single-factor experiments for specific studies in other WPs
- Make crop material available for the work in the DARCOF III project "BioTrace"

Description of work:

Task 1: Coordination of the VEG-QURE project: This includes coordination of activities the production of status reports. Further, it includes securing that general information about the project is communicated. This will be done through a project homepage describing the project and its development, presenting results through the DARCOF electronic communication channels, popular articles about the project in general, and arranging open field days and other communication activities directed at farmers, advisors, and others. A yearly workshop, where the participants of the project will meet and exchange results and ideas will be arranged. In 2009 we will arrange an international workshop on the overall theme of the project.

Task 2: Designing and growing of vegetable cropping systems: Four different cropping systems for vegetable production will be made, in a randomized experimental design with three replicates. Vegetable crops of lettuce, white cabbage, carrots and onion will be grown within each cropping system, in rotation with cereal crops. The systems will be designed as one conventional (C1) system and three organic systems (O1, O2 and O3). The systems will be made as uniform as possible (i.e. same sequence of crops) except that in:
C1: Pesticides and inorganic fertilizers will be applied according to commercial practice
O1: Crop nutrition will be based mainly on import of slurry, imports will be made according to the regulations of organic farming
O2: Crop nutrition will be based mainly on autumn green manures and catch crops, limited manure import will be directed only at the most demanding of the vegetable crops
O3: As O2, but rows of the green manure will be left to grow as intercrops between every second row of the vegetable crops to improve natural regulation mechanisms and environmental effects

Effects of the cropping systems on vegetable quality, field biology and nutrients will be present already in the first cropping year of 2006, but the pre-crop effects will not be there until the 2007. To minimize this problem for the work in the other WPs, we will use some of our own resources to initiate cover crop and green manure treatments already in the summer of 2005, before we know whether the project will be funded.

Task 3: Documentation of cropping systems and deliveries for other WPs: The results of the cropping systems will be documented with crop and soil sampling to analyze yield, nutrient uptake and nutrient balances. Crop material, data, and field plots for studies of natural regulation will be made available for the other WPs. This task includes delivery of material to the "BioTrace" if both projects receive funding.

Task 4: Single factor experiments: Comparison of crop quality or other effects of the cropping systems, involves variation in many factors at once, and this can make interpretation difficult. To allow studies of some of the important single factors, e.g. fertilizers and nutrient availability, small single-factor experiments will be made outside the four cropping systems according to plans in other WPs. The specific experiments will be planned during the project depending on the results achieved and questions that arise.

Deliverables:

D1.1	Yearly status reports .
D1.2	Popular papers on the cropping systems and the results obtained
D1.3	Project homepage, basic project information and current topics, updated at four times a year.
D1.4	International workshop on the overall themes of the project, planned for April 2009
D1.5	Presentation of the project at meetings, open field days etc

Milestones:

M1.1	Finally deciding on crop species, crop management practices and green manure crops for the four cropping systems (Mar. 06)
M1.2	Delivery of vegetable products for other WP2, mainly WP2, WP3 and W (2006-09)
M1.3	Making field plots in the cropping systems available for other WPs, mainly WP5 and WP6 (2006-09)
M1.4	Delivery of soil, crop and management data for all other WPs (2006-09)
M1.5	Delivery of yearly report with detailed presentation of field data from the cropping systems (Dec. 06, Dec. 07, Dec. 08, Dec. 09)
M1.6	Vegetables or field plots from simple single-factor experiments made available for other WPs, to be planned during the project (2007-09)
M1.7	Field experiments finalized and final data delivered to other WPs (Jan. 10)
M1.8	Organizing contributions from the project partners to DARCOF electronic communication, popular presentations and publications (2005-2010)

WP No.: 2 Effect of cultivation systems and post harvest storage on bioactive compounds, sensory quality and metabolic activity

	Start date or starting event: January 2006 – March 2010									
Partner id.	DIAS-Food	SDU-CPA	KVL-PlBio							
Person-months per participant	20.5 VIP 20 TAP	1 VIP 4 TAP	2 VIP							
Total PM VIP **Total PM:**	**23.5** **47.5**									

Objectives:

- To determine the effect of different organic cultivation systems on the content and composition of bioactive compounds in carrot, lettuce and white cabbage.
- To determine the effect of different organic cultivation systems on the sensory quality of carrot, lettuce and white cabbage.
- To correlate the content of flavour compounds (volatiles and bioactive compounds) to the sensory quality of carrot, lettuce and white cabbage.
- To determine the variation in content and composition of bioactive and volatile compounds and sensory quality in carrot during post harvest storage.
- To assess interactions between attack of pathogens and insects, and the content of bioactive compounds.
- To correlate the content of bioactive compounds to the protein expression in carrot, lettuce and white cabbage at harvest.
- To correlate the metabolic activity in carrots measured by calorimetry to changes in protein expression, bioactive and volatile compounds and pathogen attack during post harvest storage.

Description of work:

Samples of carrot, lettuce and white cabbage grown in different organic cultivation systems in WP1 during 2006 – 2009 will be collected for analysis.

Carrot, lettuce and white cabbage will be analysed at harvest for:
- Bioactive compounds (sesquiterpene lactones (SQLs) in lettuce, polyacetylenes (PAs) in carrot and glucosinolates (GSLs) in white cabbage) (**task 1**)
- Important sensory attributes (odour, flavour, texture and appearance) will be evaluated by a trained sensory panel (**task 2**)
- Volatile compounds (VOCs) (**task 3**)

Carrot stored under different conditions will be evaluated with respect to:
- Bioactive compounds (PAs in carrot) (**task 1**)
- Changes in important sensory attributes (odour, flavour, texture and appearance) during storage will be evaluated by a trained sensory panel (**task 2**)
- Changes in the content of VOCs during storage (**task 3**)
- Changes in the metabolic activity in relation to storage potential and resistance to pathogenic attack (**task 4**)

The relationship between bioactive compounds, volatiles, sensory quality and cultivation systems, storage potential, metabolic activity and resistance to pathogens will be determined by multivariate data analysis (**task 5**).

Task 1. Isolation, identification and quantification of bioactive compounds from vegetables
Bioactive SQLs and PAs of the falcarinol-type will be isolated from extracts from lettuce and carrots, respectively, by open-column chromatography and by preparative HPLC and identified by spectro-

scopic methods such as 1D and 2D-NMR techniques and mass spectrometry. The SQLs and PAs are quantified in plant extracts by analytical HPLC and gas chromatography (GC), respectively, using the purified compounds as external standards. GSLs are extracted and isolated from white cabbage according to Rodrigues & Rosa and identified by liquid-chromatography-mass spectrometry (LC-MS). The glucosinolates are quantified by analytical HPLC.

Task 2. Sensory evaluation of vegetables
The sensory quality of carrot, lettuce and white cabbage from different cultivation systems from WP 1 will be evaluated by a trained sensory panel of 8-10 assessors. A sensory profile of important attributes, which include attributes of odour, flavour, taste, texture and appearance, will be developed by the sensory panel. Using a pointed scale, the developed profile of attributes will by used in evaluating the raw vegetable samples after they have been trimmed and shredded. Thus, conducting a sensory analysis lead to measure differences in selected sensory attributes in an objective way and specific attributes related to the cultivation system can be revealed.

Task 3. Isolation, identification and quantification of volatile compounds from vegetables
VOCs are isolated from raw, shredded plant material by dynamic headspace technique as described by Kjeldsen *et al.*, and quantified by GC using internal standards. The individual volatile compounds will be identified by GC-MS.

Task 4. Development and testing of a calorimetric method for expressing changes in metabolic activity during storage of vegetables.
Carrots will be stored for 8-10 months at low temperatures and samples will be taken for analysis of metabolic activity and changes in bioactive compounds, volatiles, storage potential and resistance to pathogens. Samples with disease symptoms will be analysed for plant pathogens (Partner 4, PLBio, KVL) The metabolic activity of carrots will be studied at laboratory scale to determine the potential of calorimetric measures in relation to pathogen development

Task 5. The relationship between bioactive compounds, volatiles, sensory quality and cultivation systems, storage potential, metabolic activity and resistance to pathogens
Multivariate data analysis will be used on data obtained from all WP's with the aim to correlate recordings from the cultivation systems, analysis of bioactive and volatile compounds, sensory attributes, resistance to pathogens and insects, metabolic activity and protein expression. These analyses will contribute to knowledge about different cultivation systems will influence the final quality of raw and stored vegetables.

Deliverables:

D2.1 Oral or poster presentations of the result at national and international workshops/congresses.

D2.2 Manuscript 'Effect of organic cultivation systems on bioactive compounds in selected vegetables' will be published in an international reviewed journal.

D2.3 Manuscript 'Effect of organic cultivation systems on flavour compounds and sensory properties in selected vegetables' will be published in an international reviewed journal.

D2.4 One or two manuscripts on correlation between bioactive compounds, volatiles, sensory attributes and the metabolic activity, resistance towards pathogenic attack during storage.

Milestones:

M2.1 Analytical methods for isolation and quantification of bioactive and volatile secondary metabolites have been established and optimized (Aug. 06)

M2.2 A sensory profile of important attributes for carrot, lettuce and white cabbage has been developed (Sep. 06)

M2.3 Preliminary results of bioactive and volatile secondary metabolites and sensory analysis of crops from the cultivation years 2007, 2008 and 2009 are completed (Dec. 09)

M2.4	Testing of the potential of calorimetry for determination of metabolic activity in carrot during storage has been completed (Apr. 07)
M2.5	Preliminary results of bioactive and volatile secondary metabolites and metabolic activity from post harvest storage of carrot for two cultivation years (Jun. 09)
M2.6	Data for resistance of pathogens, cultivation, metabolic activity, protein expression, sensory evaluation and bioactive and volatile secondary metabolites has been correlated (Apr. 10)

WP No.: 3 Characterisation of Protein Expression in Vegetable Crops grown under different conditions.

	Start date or starting event: January 2006								
Partner id.	SDU-CPA								
Person-months per participant	6 VIP 20 TAP								
Total PM VIP **Total PM:**	**6** **26**								

Objectives:

- Optimise methods for the extraction of proteins from the vegetable crops under study
- Analyse the protein extracts by two dimensional gel electrophoresis and select differences within each crop which depend on the different cropping systems
- Identify the selected proteins and interpret the changes using bioinformatics

Description of work:

Task 1: Optimise extraction procedures for defined parts of each of the crops: lettuce (leaf), carrot (root and leaf) and cabbage (leaf). Methods that will be explored will be grinding in liquid nitrogen, fine shredding (ultraturrax), sonication, and differential solubilisation (to separate proteins from phenolic compounds).

Task 2: Separate proteins by two dimensional gel electrophoresis, visualise the proteins using quantitative fluorescent dyes to detect the proteins and initiate an image database for each vegetable.

Task 3: Identify some of the major proteins (looking especially for proteins involved in energy metabolism) by mass spectrometry (MS).

Task 4: Compare the vegetables grown under different conditions and harvested at different years. Select the statistically significant differences and identify these proteins by MS, (both matrix assisted laser desorption ionisation MS and nanoelectrospray tandem MS).

Task 5: Identify effects of microbial attack on the protein expression in stored carrots. To be able to do this, proteins from selected disease fungi from the studies in WP4 will be analysed.

Task 6: Carry out bioinformatic analysis of the data to interpret the changes seen in terms of the growth conditions and other factors measured (e.g. flavour, colour, nutritional value).

Deliverables:

D3.1 Methods description for the preparation of protein samples from the vegetables .
D3.2 List of proteins whose expression is affected by the growth conditions at the end of each growing season. Aimed at possible patent application(s)
D3.3 Two or three manuscript describing the effect of growth conditions on the protein composition in lettuce, carrot and cabbage.
D3.4 Presentation of the data at scientific meetings and congresses

Milestones:

M3.1 Selection of the optimal method for sample preparation (Apr. 06)
M3.2 Initiation of the image databases for each of the vegetables (Jun. 06)
M3.3 Identification of the 25th protein for the database (Oct. 06)
M3.4 Image database including some proteins identified (Dec. 07)
M3.5 Completion of the yearly analysis of lettuce (Sept. 06, Sept. 07, Sept. 08, Sept. 09)
M3.6 Completion of the yearly analysis of carrots (Sept. 06, Sept. 07, Sept. 08, Sept. 09)

M3.7	Completion of the yearly analysis of cabbage (Sept. 06, Sept. 07, Sept. 08, Sept. 09)

WP No.: 4 Storage product quality

	Start date or starting event: Jan 2006-March 2010								
Partner id.	KVL-PlBio	DIAS-Food	SDU-CPA						
Person-months per participant	26.5 VIP 6 TAP	1 VIP 1 TAP	1 VIP 4 TAP						
Total PM VIP **Total PM:**	**28.5** **39.5**								

Objectives:

- To study disease progress of liquorices rot in carrots during post harvest storage in relation to changes in parameters expected to be expressed by agricultural system: root physiology, bioactive compounds and metabolic activity
- To clarify if there are effects of the agricultural systems described in WP 1 on physiology and bioactive compounds and development of post harvest diseases in carrot and if these parameters are related. Special focus on liquorices rot.

Description of work:

Task 1: Characterization of the infection biology of *M. acerina* in relation to root physiology and bioactive compounds of carrots during cold storage

a. A well characterized plant assay (a pathosystem) will be developed for studies of infection biology of *M. acerina*. Carrots are artificially inoculated on wounds using a *M. acerina* strain (IK2013) isolated from liquorice rot in carrots. Apparently healthy roots, naturally wounded as well as artificially wounded carrots will be inoculated at defined sites with well defined wound sizes and depth. Disease development will be studied under controlled storage temperatures. The most appropriate pathosystem will be selected. (KVL-PlBio)

b. Carrots are harvested at defined time in the first experimental year (delivered from WP1), and apparently healthy/wounded carrots are artificial inoculated at different time of storage and followed in a time course study using the pathosystem developed in (a). Pathogen infection and mycelia development in latent and developing infections are studied by use of a genetically transformed isolate of *M. acerina* with a fluorescent gene (developed in task 2) and a PCR based method (task 3) (KVL-PlBio). Samples of carrots with latent and developing symptoms will be delivered to partners (SDU-CPA and DIAS-Food). CDU-CPA will measure differences in proteomics as described in WP3. Pure cultures of *M. acerina* and natural colonizers in carrots will also be analyzed as references. DIAS-Food will analyze plant metabolites with the methodology described in WP2.

Results from task 1 makes it possible to define the timing and numbers of assessments for the survey (task 4)

Task 2. Development of a genetically transformed isolate of *M. acerina.* The pathogen used for inoculation (task 1) will be transformed with the *DsRed* and *gfp* gene, respectively, to facilitate histopathological studies especially of latent infections. (KVL-PlBio). *DsRed (*DsRed Ti-plasmid mod. after Mikkelsen et al. 2003) and *gfp* (*gfp* Ti-plasmid construction by Solveig Christiansen, *unpubl.*) genes under constitutive promoters, together with the hygromycin resistance gene, are available for *Agrobacterium* mediated transformation of fungal spores (Protocol mod. after Bundock *et al.*, 1995). Transformants will be intensively screened for selection of those resembling the wild-type in growth rate, pathogenicity on carrot and metabolite profile on various substrates.

Task 3. Development of PCR based method For use in naturally infected carrots (task 5) a molecular PCR based method will be developed for detection and quantification of *M. acerina* in carrot roots from harvest and during storage. (KVL-PlBio) The method will be based on existing *M. acerina* specific primers (Patent application WO2004/040017 A2). KVL-PlBio has experience with PCR detection of pathogens in carrot root tissue.

Task 4. Investigation of the role of agricultural system input on the disease development and the resulting quality of the carrots after storage. Carrots from each agricultural system in WP 1 will be characterized at harvest and a survey of post harvest diseases will be done at different time points throughout the storage period. Special attention will be given to disease caused by *M. acerina.* Samples of healthy carrots are artificially inoculated with *M. acerina* and the susceptibility of carrots from the different agricultural systems compared.
A general characterization of carrot roots regarding physiological status, wounds from pest attacks and disease level based on symptoms will be given by DIAS-Hort (WP1). From these carrots representative samples are delivered for following studies:

- Classification of the pathogens (KVL-PlBio)
- Protein characterization using proteomics and the related bioinformatics: (SDU-CPA)
- Physiological characterization and profile of bioactive compounds: (DIAS-Food)

Deliverables:
D4.1 Paper on a method to inoculate carrots and a defined plant assay
D4.2 Paper on a connection between physiologically status and etiology of *M.acerina*
D4.3 Paper on the role of agricultural system on storage quality

Milestones:
M4.1 A plant bioassay has been developed (Aug. 06)
M4.2 A fluorescent transformant of M. acerina has been developed and tested stabile (Apr. 07)
M4.3 A method for further studies of liquorices rot development has been developed (Apr. 07)
M4.4 A method to investigate natural populations of M. acerina in situ during infection has been developed (Dec. 07)
M4.5 Interaction between physiologically status and etiology of M. acerina has been evaluated (Dec. 08)
M4.6 The role of agricultural system on storage quality has been described (May 10)

WP No.: 5 Natural regulation of insects with particular focus on soil living stages and the influence of cropping system

	Start date or starting event: January 2006								
Partner id.	KVL-Eco	SDU-CPA							
Person-months per participant	41 VIP	2 TAP							
Total PM VIP **Total PM:**	**41** **43**								

(Please give Institution or Department as partner id.)

Objectives:

- To optimise methods for assessing the presence of insect pathogenic fungi in the soil
- To quantify the presence and the function of these fungi in the cropping systems over time
- To study natural regulation by beneficial insects in the plots

Description of work:

Task 1: Optimisation of methods for *in vivo* and *in vitro* assessment of insect pathogenic fungi in small plots in soil. Soil samples 0-10 cm will be taken with a core sampler and semi-selective sterile media and bait insects (*Galleria* or *Tenebrio*) will be used for isolation. Insect pathogens present will be determined and quantified.

Task 2: Genetic characterization of fungal isolates. DNA extraction, then the PCR profile will be determined. Data compared with Gene-bank data.

Task 3: Mapping of the fungi. The occurrence of the fungi will be mapped (by the SADIE programme, Rothamsted Research) in the plots and in their surroundings. Data from task 1 and 2 will be used for the mapping.

Task 4: Assessment of insect predators, with particular reference to small plots. In each crop the relevant predators (Coleoptera, Hemiptera and possibly also Hymenoptera and Aracnida) will be sampled (pitfalls, netting) and quantified. The species selected will obviously differ between years and crop.

Task 5: Elucidating the effects of selected proteins from the crops (to be determined in WP 3) and the effect on spore germination of insect pathogenic fungi in soil.

Deliverables (reports):

D5.1 Laboratory protocol for assessing insect pathogenic fungi in soil in small plots
D5.2 Report on genetic characterisation of insect pathogenic fungi
D5.3 Contribution to FØJO publication on insect pathogenic fungi in organic agro-ecosystems
D5.4 Scientific paper on analysis of three years prevalence of insect pathogenic fungi in soil
D5.5 Presentation of preliminary project results at international conference (SIP/IOBC)
D5.6 Scientific paper on biodiversity assessment (in cooperation with WP6)
D.5.7 Scientific paper on analysis of insect population regulation with emphasis on soil living stages

Milestones:

M5.1 Development of method for quantification of populations of insect pathogenic fungi in soil (Jul. 06)
M5.2 Selection and testing of molecular markers for identification of insect pathogenic fungi at isolate level (Aug. 07)
M5.3 Evaluation of data on insect pests and their natural insect enemies related to the cropping system (for WP6) (Oct. 08)

M5.4	Evaluation of cropping factors on natural regulation of insect populations by insect pathogenic fungi in organic agriculture (Apr. 09)
M5.5	Evaluation of effects of selected compounds (from WP3) on selected insect pathogenic fungi in vitro (Dec. 09)

WP No.: 6 Cropping systems, environment, crop composition and system development

	Start date or starting event: (please give month and year)				July 2006				
Partner id.	DIAS-Hort	DIAS-Food	SDU-CPA	KVL-PlBio	KVL-Eco				
Person-months per participant	36 VIP 4.5 TAP	2 VIP	1 VIP 2 TAP	2 VIP	2 VIP				
Total PM VIP **Total PM:**	**43** **49.5**								

Objectives:

- Study effects of cropping systems on nutrient balances and nitrogen leaching loss
- To improve our basis for growing intercrops between main crops with limited yield loss. The intercrop systems must allow significant intercrop growth to gain the advantages of intercropping
- Study whether there are general effects of the different cropping systems on crop quality, content of secondary metabolites, expression of proteins and energy metabolism
- Study cropping system effects on field insect biology
- To quantify production tradeoffs to effects on quality and environment in the cropping systems

Description of work:

Task 1: Cropping system effects on nutrients and leaching losses: We will study root growth of main crops and fertility building crops (using the minirhizotron method, extending to 2.5 m soil depth where relevant), and the effects of cropping systems on soil nitrogen dynamics and nitrogen losses. Studying this in detail in all crops in all rotations will not be possible, but based on previous work we will identify hot spots where most of the losses will occur, and direct the studies at these hotspots. Simulations with the DAISY model will then be used to estimate total nitrogen leaching losses across the rotations.

Task 2: Development of intercropping systems: In current projects we are working with methods to reduce the yield loss typically encountered with intercropping systems. Based on this, we will study effects of root pruning of the intercrops and the spatial arrangement of crops and intercrops. Competition between vegetables and intercrops will be studied with the ^{15}N method and with direct observation of root growth with the minirhizotron method. In 2006 this will only include measurements in the cropping systems, during 2007 and 2008 separate experiments will be made with intercrops.

Task 3: General effects of cropping systems on crop quality and natural regulation
The study of specific quality factors or contents of specific compounds in the vegetables are likely to show a scattered picture. Specific cropping systems may increase the content of some compounds and reduce others. A similar result is likely to be obtained from the studies of insects and soil fungi. However, there may still be more general patterns of effects. Based on results from WP2 and WP3 and WP4, we will study whether more general effects of the cropping systems on crop quality can be found. Based on data from WP1, WP4 and WP5, we will study general effects on field biology. The question is: Does organic production lead to generally higher or more diverse content of factors such as secondary compounds in the vegetables? General changes in quality and storability, in protein expression or in energy metabolism? Higher or more diverse populations of soil fungi of insects in the field? Are there differences among vegetables from the three organic cropping systems in these respects?

Task 4: Quantification and presentation of tradeoffs
Based on the data on marketable yield, field management operations, and other costs from WP1, and on data on quality, biodiversity, nutrient balances and nitrogen leaching losses and general nutrient balances from all the other WPs, we will analyse and illustrate the cost of obtaining advantages in terms of quality and environment by the different cropping systems. As we cannot put economic value to the different advantages, and certainly not value e.g. leaching loss against higher contents of specific secon-

dary compounds, what we can do is to illustrate the costs and effects of the systems, to allow others to evaluate which systems seem to give most “value for money”.

Deliverables:

D6.1 Scientific paper on the effect of cropping systems on nutrient balances and N leaching loss

D6.2 Scientific paper on methods for limiting competition and yield loss by intercropping in vegetable production

D6.3 Scientific publications on general effects of cropping systems on crop composition and quality and on field biology. This will be at least one publication, but more if data allows this

D6.4 Report analysing and illustrating costs and benefits of the four cropping systems

Milestones:

M6.1 Define “hot spots” with high N leaching risk to be studied in the cropping systems (Aug. 06)

M6.2 Define intercropping systems to be tested in exps. during 2007/08. First intercrops must be established in the spring of 2006 (Mar. 06)

M6.3 Asses effects of different intercropping systems on crop yield and on N competition between crops and intercrops (Dec. 08)

M6.4 Agree with project partners on which data can and should be included in paper(s) on general system performance (D6.3) (Oct. 09)

A.3 Project resources and budget overview

(Short description, max. 1 page, of partners, key competences and management of the project. Overview of budget according to tables given below)

Kristian Thorup-Kristensen *et al.*, Research group Vegetables (Partner DIAS-Hort)
The group has more than 15 years experience in development of organic cropping systems for vegetable production. The main topics have been crop root growth, nitrogen utilization, catch crops, green manures and crop rotations. Main methods have been field studies with e.g. minirhizotron and ^{15}N studies of root growth and function, and use and development of plant-soil simulation models. During the last three years, studies of intercropping systems combining main crops and green manures have become an important activity.

Kristian Thorup-Kristensen has previously been project leader of interdisciplinary research projects involving field studies of vegetable production methods as the basis for studies by various disciplines involving product quality aspects, plant nutrients and field and soil biology.

Lars Porskjær Christensen *et al.*, Dept. Food Science, (Partner DIAS-Food)
The group has key competences within natural product chemistry, including analysis of bioactive compounds and aroma/flavour compounds, and it has great experiences within sensory analysis and post harvest storage of vegetable foods.

Peter Mose Larsen *et al.*, Center for Proteome analysis (Partner SDU-PCA)
The group has large experience in proteomic analysis, analysing and quantifying tissue protein expression and its responses to various factors. The methods allow separating up to 20,000 proteins, and to analyze them quantitatively using fluorescent dyes. They have developed software (ProteoMiner) to find which proteins' expressions are significantly changed. These proteins are then identified by mass spectrometry (MALDI). In cases where the protein cannot be identified (using the available genomic data) MALDI time-of-flight time of flight (MALDI tof-tof) or nano-electrospray ionisation tandem mass spectrometry (NESI-MS-MS) is used to allow search for homologous proteins in the databases. Most of the work has been directed at human health issues, such as human diseases or effects of toxins. However, the group has also in previous and current projects been involved in studies of plants, and been able to show clear effects of growth conditions on protein expression.

Dan Funck Jensen *et al.* Section for Plant Pathology (partner KVL-PlBio)
The group has key competences within basic studies of biological and molecular interactions between plants, pathogens and environmental factors. This work includes methodology in classical and molecular plant pathology using e.g. genetic transformation, exploitation of reporter gene technology with filamentous fungi, gene cloning, gene expression, PCR methods and bioinformatics. These methods are used routinely. Another field include histopathological studies of infection and pathogenesis by use of advanced microscopically techniques as confocal microscope and bioimaging.

Jørgen Eilenberg *et al.* the Zoology Group (partner KVL-Eco)
The group has more than 20 years of experience in natural regulation of insects. Focus on population interactions and conservation biological control.

Competences in: Insect quantitative sampling and determination; isolation *in vivo* and *in vitro* of insect pathogens from environment; diagnosis of insect pathogens; phenological and genetical characterization of insect pathogens from organic grown crops; analysis of horizontal and vertical distribution of organisms in the environment; biodiversity studies (functional, diversity indices); parameters influencing growth *in vitro* of microorganisms

Table for person month allocated on WP's and partners

Partner / WP	1 (DIAS-Hort)	2 (DIAS-Food)	3 (SDU-CPA)	4 (KVL-PlBio)	5 (KVL-Eco)	Total
WP1	10					10
WP2		20.5	1	2		23.5
WP3			6			6
WP4		1	1	26.5		28.5
WP5					41	41
WP6	36	2	1	2	2	43
Total	46	23.5	9	30.5	43	152

(Please give Institution or Department as partner id.)

DIAS-Hort: Danish Institute of Agricultural Sciences, Department of Horticulture
DIAS-Food: Danish Institute of Agricultural Sciences, Department of Food Science
SDU-CPA: University of Southern Denmark, Centre for Proteomic Analysis
KVL-PlBio: Royal Veterinary and Agricultural University, Department of Plant Biology
KVL-Eco: Royal Veterinary and Agricultural University, Department of Ecology

Table for breakdown of total budget on partners and different cost categories (DKK)*

Participating institution	Responsible scientist	Salaries		Equip-ment	Operational expenses	Total budget DKK
		Academic	Techn. adm.			
DIAS, Department of Horticulture	Kristian Thorup-Kristensen	2.541.000	1.162.300	0	305.035	4.810.000
DIAS, Department of Food Science	Lars Porskjær Christensen	1.238.589	690.980	0	398.023	2.840.000
SDU, Centre for Proteomic Analysis	Peter Mose Larsen	339.217	702.212	0	333.662	1.650.000
KVL, Department of Plant Biology	Dan Funck Jensen	1.209.000	195.000	15.000	131.000	1.860.000
KVL, Department of Ecology	Jørgen Eilenberg	1.666.000	0	0	284.000	2.340.000

*Breakdown of the part of the budget which we apply for

A.4. Dissemination of scientific results

Please enclose information on:

- Planned education of scientists including Ph.D. and post-docs
- Stays abroad, guest researchers etc.
- Communication of results. Publication strategy, contributions to national or international workshops etc.

Communication

Communication towards farmers and the public in general will be made in several ways. We will make a homepage to describe the project and its participants. Though it will of course be updated at intervals, it is not meant to hold the most current news about the project. DARCOF has developed several electronic means of communicating results, which are well suited for the continuous communication of news and results from the project. The project and its results will be presented at meetings, at open days and through papers in farmers' journals and other magazines.

The scientific dissemination is described in the application, through the scientific publications to be made and the workshop which will be arranged in the fourth year. Further, we will present results at other meetings and workshops, and through the research contacts we have (see international cooperation).

Education
A PhD study is also planned as part of WP4, and we have reserved funding for this in the WP4 budget. The plan is to apply for additional funding form the research school for organic farming (SOAR) and from other sources. We hope that this will be possible, but if we cannot get the additional funding for this PhD study as planned, we will use these funds to try to finance a PhD study in one of the other WPs.

In WP5 Nicolai V. Meyling will be employed in a post-doc position.

The project will contribute directly to education, as three of the partners are university partners from The Royal Veterinary and Agricultural University and from University of Southern Denmark. Results can be used directly in the education and form the basis for a number of bachelor and master thesis. Also the two non-university partners (DIAS, Department of Horticulture and Department of Food Science) will involve master students in the work as they have done in previous DARCOF studies.

Apart from all the dissemination work planned within the project, it should be remembered that all of the participants have other channels of communication which will also be used. Much will be communicated through meetings, homepages and publications organized by institutions, organizations or by DARCOF.

Partner 4 (PlBio, KVL) will also contribute to a new Ph.D.-course at KVL on post harvest biology teaching in post harvest diseases of fruit and vegetables.

Stays abroad and guest scientists
No specific plans for this have been made in the project. However, the scientists involved in the project have intensive contacts with scientists from other countries, also in direct project co-operations, some of which are mentioned below. This should lead to good interaction with researchers in other countries working with related topics.

A.5. Scientific collaborations

The project will be co-ordinated with the BioTrace project (also DARCOF III), delivering vegetable material and information to be used for feeding studies in the BioTrace project.

Kristian Thorup-Kristensen and others at the Research group for Vegetables are involved in the following relevant projects:

- EU project: **QualityLowInputFood**, We are working on environmental audits of cropping systems and modelling of nitrogen dynamics in organic vegetable production
- **Development of an environmentally friendly growing system for organic production of high quality wheat and rape by intercropping with autumn catch crops**, Project financed by EU trough Interreg II. Together with DIAS-Food and the University of Kiel (Ralph Loges), Germany
- **Ecology of the cropping system, Green manure crops as multifunctional tools in vegetable production.** Project financed by the Swedish Research Council, where we from DIAS work on control of competition in vegetable crops with intercrops. Head of project is Birgitta Rämert, SLU, Uppsala
- **Development of a model based decision support system to optimise nitrogen use in horticultural crop rotations across Europe**, Financed by EU. We work with modelling of nitrogen dynamics in vegetable production. Our primary responsibilities are modelling of root growth and modelling fertility building crops. Project is lead by Clive Rahn, HRI Warwick, UK

- **Cost action 631, Understanding and modelling plant-soil interactions in the rhizosphere environment", member of the management committee.** With partners from most European countries. Kristian Thorup-Kristensen is a member of the management committee of the network

Jørgen Eilenberg *et al.* cooperates in:

- **Danish Centre for Biological Control.** Jørgen Eilenberg is chair of this major national network, which include most teams in Denmark working with biological control. The centre has much international collaboration.
- **COST Action 862, Bacterial Toxins for insect control.** Jørgen Eilenberg is a member of management committee of this newly initiated network
- **Cornell University (Ann Hajek) and Rothamsted Research, UK (Judith Pell):** Co-operation on biological control and natural regulation

Peter Mose Larsen and Stephen Fey co-operates with:

- **Dr. S.P. Ferreira, Plant Biotech Lab, University of Lisbon, Portugal**. Use of proteomic analysis to study heat stress effects in poplar trees

Lars Porskjær Christensen and colleagues cooperates with:

- **Federal Centre for Breeding Research on Cultivated Plants,** Institute for Plant Analysis, Quedlinburg, Germany (Contact person: Prof. Dr. Hartwig Schulz)
- **Department of Food Engineering and Department of Building Materials**, Lund University (Contact persons: Dr. Federico Gómez, and Dr. Lars Wadsö).

Dan Funck Jensen and colleagues are involved in:

- **Environmentally friendly control of post-harvest diseases of carrots**. Program financed by DFFE (Innovation) Partners: Vilmorin, France, Danisco Seeds, Daehnfeldt a/s, Gulerodsgruppen a/s, Rosenfeldt Estate and DIAS-Flakkebjerg - Post-harvest program associated cooperation: Jürgen Kohl, Plant Research International, Business Unit Crop and Production Ecology, Wageningen; The Netherlands; Arne Hermannsen & Sonja Klemsdal, Planteforsk, Norway, Bodil Jönsson, SJV, Sweden.

A.6 Other issues

(E.g. special considerations concerning conducting of experiments in relation to principles of organic food and farming).

A.7 References

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Hansen SL, Purup S, Christensen LP (2003). Bioactivity of falcarinol and the influence of processing and storage on its content in carrots (*Daucus carota* L.). *J Sci. Food Agric.* **83**, 1010-1017.

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Meyling, N.V., Eilenberg, J. (2004). Distribution and occurrence of entomopathogenic fungi in the soil in a single agroecosystem in Denmark. 37th Annual Meeting, Society for Invertebrate Pathology, Helsinki, Finland, August 1-6.

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van Poppel G. (1996). Epidemiological evidence of b-carotene in prevention of cancer and cardiovascular disease. *Eur. J. Clin. Nutr.* 50, S57-S61.

A.8 Curriculum vitae

Curriculum Vitae for Kristian Thorup-Kristensen

Born: 25 September 1961

Education: 1987 M.Sc. in agriculture from The Royal Vet. and Agricultural Univ. (KVL)
1995 Ph.D. degree in Plant Nutrition and Soil Fertility, KVL

Employment: 1987 to 1988, Scientist at The Royal Vet. and Agricultural Univ.
1989 to 1997 Scientist at the Danish Inst. Agric. Sci.
Since 1997 Senior Scientist at the Danish Inst. Agric. Sci.
Since 1998 Head of Research group for Vegetable production, Danish Inst. Agric. Sci.

Other activities Key-supervisor at the research school for organic farming (SOAR) and at the research school for Horticulture.
Co-supervisor for four PhD students, three at KVL and one at The Swedish Agricultural University, Uppsala.
Referee at international scientific journals, Plant and Soil and others

Current research projects: 2000 to 2005: "Development of organic vegetable cultivation methods, and the use of catch crops to improve the production and protect the environment" under the second DARCOF initiative, as active scientist and project leader
2002-2005: Project financed by Interreg II, Development of an environmentally friendly growing system for organic production of high quality wheat and rape by intercropping with autumn catch crops
2002 to 2005: COST action 631: "Understanding and modelling plant-soil interactions in the rhizosphere environment", member of the management committee
2003 to 2006: "Development of a model based decision support system to optimise nitrogen use in horticultural crop rotations across Europe", EU project, as active scientist and leader of the Danish part of the activities.
2004 to 2009: "QualityLowInputFood", EU project, as active scientist and work package leader, working on environmental audits of cropping systems and modelling of nitrogen dynamics in organic vegetable production

Research interests: The main topics have been N utilization, root growth of vegetables and catch crops and effect of catch crops and green manure and organic crop rotations. The work has focused on growth, and the significance of differences in root growth for N utilization, on methods to study root growth also at larger soil depths, and on modelling of root growth. The results have been communicated through 31 papers in scientific journals, more than 30 papers in proceedings from workshops etc., and more than 50 papers in farmers journals etc.

Some recent publications

Kristensen, H.L. and Thorup-Kristensen, K. (2004) Root growth and nitrate uptake of three different catch crops in deep soil layers. Soil Sci Soc. Am. J. 68: 529-537

Thorup-Kristensen, K., Magid, J. and Jensen, L.S. 2003 Catch crops and green manures as biological tools in nitrogen management in temperate zones. Advances in Agronomy, 79: 227-301

Sørensen, J.N. and Thorup-Kristensen, K. (2003) Undersowing legume crops for green manuring of lettuce. Biol. Agric. Hortic. 21, 399-414.

Thorup-Kristensen, K., (2002) Utilising differences in rooting depth to design vegetable crop rotations with high nitrogen use efficiency (NUE). Acta Hort. 571, 249-254.

Thorup-Kristensen, K. (2001) Are differences in root growth of nitrogen catch crops important for their ability to reduce soil nitrate-N content, and how can this be measured? Plant and Soil, 230: 185-195

Curriculum Vitae for Lars Porskjær Christensen

Born: May 25, 1963 in Horsens, Denmark

Education:

1989 M.Sc. from Department of Organic Chemistry, University of Aarhus (UA). Majoring in analytical organic chemistry (natural product chemistry).

1992 Ph.D. from Department of Organic Chemistry, UA. Natural product chemistry and organic synthesis.

Employment:

1988–89 Part time teacher in chemistry at Horsens Statsskole (High school).
1990–92 Graduate scholarship from the Danish Natural Science Research Council (SNF).
1990–92 Teaching assistant, Department of Organic Chemistry, UA.
1993–95 Scientist, Research Group for Plant Breeding & Propagation, Department of Ornamentals, Danish Institute of Agricultural Sciences (DIAS).
1995–98 Senior scientist, Research Group for Plant Breeding & Propagation, Department of Ornamentals, DIAS.
1996–99 Associate professor (external), Department of Chemistry, University of Southern Denmark (USD).
1998–2002 Senior scientist, Department of Horticulture, Research Group for Food Quality and Natural Products Chemistry, DIAS.
2002–03 Head of research group, Department of Horticulture, Research Group for Food Quality & Health, DIAS.
2003– Head of research unit, Department of Food Science, Research Unit for Plant Food Science, DIAS.
2003– Associate professor (external), Department of Chemistry, USD.

Other activities

Reviewer activity/experience:
1993–present. Reviewer for the following international journals: **1.** *J. Nat. Prod.*, **2.** *Phytochemistry*, **3.** *J. Agric. Food Chem.*, **4.** *J. Food Sci.*, **5.** *J. Sci. Food Agric.*, **6.** *LWT Food Sci. Technol.,* **7.** *Industrial Engineering Chem. Res.,* **8.** *Contact Dermatitis*, **9.** *Proteins: Structure, Function, Genetics*, **10.** *Basic Clin. Pharmacol. Toxicol.*, **11.** *Environ. Experimental Bot.*, **12.** *European J. Hort. Sci.* and **13**. *Australian J. Agric. Res.*

Research projects and interests:

- Isolation, quantification and structure elucidation of primary and secondary metabolites, including isolation of bioactive natural products by bioassay and/or assay-guided chromatographic fractionation.
- Investigations of the biological activity of secondary metabolites and evaluation of their possible health-promoting effects.
- Investigations on the biosynthesis and taxonomic applications of secondary metabolites.
- Plant contact allergens, including direct release of contact allergens from plants and their possible role in airborne contact dermatitis.
- The physiological mechanisms behind volatile release.
- Identification of important aroma- and flavour compounds in fruits, vegetables and other food products using organoleptic evaluations such as aroma extract dilution analysis combined with GC-olfactometry.
- Natural colorants in fruit and vegetables in relation to cultivars, post-harvest, storage and processing.

Some recent publications

Publications: 62 Papers in peer reviewed international journals, **16** peer reviewed book chapters and proceedings, > **80** posters, abstracts and non-reviewed proceedings, and **38** popular papers and reports in Danish or English.

Selected relevant peer reviewed papers in international journals in the period 2000–2005:

Andreasen, M. F., **Christensen, L. P.**, Meyer, A. S., Hansen, Å. (2000). Ferulic acid dehydrodimers in rye (*Secale cereale* L.). *J. Cereal Sci.* **31**, 303–307.

Andreasen, M. F., Landbo, A.-K., **Christensen, L. P.**, Hansen, Å., Meyer, A. S. (2001). Antioxidant effects of phenolic rye (*Secale cereale* L.) extracts, monomeric hydroxycinnamates, and ferulic acid dehydrodimers on human low-density lipoproteins. *J. Agric. Food Chem.* **49**, 4090–4096.

Larsen, E., Kharazmi, A., **Christensen, L. P.** & Christensen, S. B. (2003). An antiinflammatory galactolipid from rose hip (*Rosa canina* L.) that inhibits chemotaxis of human peripheral blood neutrophils *in vitro*. *J. Nat. Prod.* **66**, 994–995.

Hansen, S. L., Purup, S., **Christensen, L. P.** (2003). Bioactivity of falcarinol and the influence of processing and storage on its content in carrots (*Daucus carota* L.). *J. Sci. Food Agric.* **83**, 1010–1017.

Kidmose, U., Hansen, S. L., **Christensen, L. P.**, Edelenbos, M., Larsen, E., Nørbæk, R (2004). Effects of genotype, root size, storage, and processing on bioactive compounds in organically grown carrots (*Daucus carota* L.). *J. Food Sci.* **69**, S388–S394.

Kobæk-Larsen, M., **Christensen, L. P.**, Vach, W., Ritskes-Hoitinga, J. & Brandt, K. (2005). Inhibitory effects of feeding with carrots or (–)-falcarinol on development of azoxymethane-induced preneoplastic lesions in the rat colon. *J. Agric. Food Chem.*, in press.

Curriculum Vitae for Ulla Kidmose

Born: December 1, 1961

Education:
1989 MsC in Food Science at the Royal Veterinary and Agricultural University
2004 Ph.D. in human nutrition/food technology from The Royal Veterinary and Agricultural University

Employment:

1990- 1996	Scientific associate, Danish Institute of Plant and Soil Science, Årslev
1996-	Scientist at Department of Food Science, Statens Planteavlsforsøg
2001-2004	Part time teacher at Dalum Collage of Food and Technology
February 2005-	Senior scientist, Danish Institute of Agricultural Sciences (DIAS), Department of Food Science

Maternity leaves
April 1995- January 1996, February 1998- October 1998.

Main research projects: I have participated in DARCOF III project "Organic food and health – a multigeneration animal experiment"with analysis of carotenoids in vegetables and have been project responsible for the project "Sensory evaluation of vegetables and potatoes cultivated in organic and conventional systems". I have participated in "Development of deep frozen organically grown vegetables food for children, where I was responsible for chemical analysis of bioactive compounds in spinach and carrots.

Research interests: My research field is sensory and nutritional quality of vegetables food in relation to raw material, storage and processing. In relation to nutritional quality, focus has been on quantification of compounds, e.g. vitamins, carotenoids and phenolics in fruits, vegetables, potatoes and cereals, which are related to aspects of human nutrition and health.

Some recent publications

Ph.D.Thesis: Kidmose U. (2005). Effects of storage, processing and household preparation on carotenoids in commonly consumed vegetables.

Kidmose, U. and Kaack, K. (1999). Changes in texture and nutritional quality of green asparagus spears (*Asparagus officialis* L.) during microwave blanching and cryogenic freezing. Acta Agriculturæ Scandinavica, 49,110-116.

Kidmose, U. and Martens, H.J. (1999). Changes in texture, microstructure and nutritional quality of carrot slices during blanching and freezing. Journal of Science of Food and Agriculture, 79, 1747-1753.

Kidmose, U., Knuthsen, P., Edelenbos, M., Justesen, U. and Hegelund, E. (2001). Carotenoids and flavonoids in organically grown spinach (*Spinacia oleracea* L.) genotypes after deep frozen storage. Journal of Science of Food and Agriculture, 81 (9) 918-923

Kidmose, U., Pedersen, L. and Nielsen, M. (2001). Ultrasonics in evaluating rheological properties of dough from different wheat varieties and during ageing. Journal of Texture Studies, 32, (5/6), 321-334

Thybo, A., Mølgaard, J.P. and Kidmose, U. (2001). Effect of different organic growing conditions on quality of cooked potatoes. Journal of Science of Food and Agriculture 82, 12-18

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Curriculum Vitae for Peter Mose Larsen

Born: 14.5.1957

Education:
Diploma at Holstebro High School
Major in Chemistry and Biology at Aarhus University
Cand. Scient. thesis in Biochemistry at Biostructural Chemistry Division, Chemistry Institute, Aarhus University

Employment:
1982-1984: Biostructural Chemistry Division, Chemistry Institute, Aarhus University
1984-1996: Institute of Medical Microbiology and Institute of Human Genetics, Aarhus University (Group Leader)
1996- : Founder of Centre for Proteome Analysis, International Science Park Odense, University of Southern Denmark, (Associate professorship)
2003- : Third Military Medical University and West China Hospital, Chongqing, People's Republic of China (Professor in Medical Proteomics)

Other activities
International reviewer for the Major National Research Facilities Program, Australia
Danish jury member for the EU competition for "Young Researchers and Inventors"
External evaluator for a research professorship for the NSW Cancer Council, Australia (1995-to date)
Jury member for the International Competition for Environmental Science and Biotechnology
International evaluator in biotechnology for Belgium; The Czech Republic and EU
Vice-president for the Danish organisation: "Young Researchers and Inventors"

Main research projects:
1998-2003: "Center for Experimental BioInformatics (CEBI)", The Danish National Research Foundation, (DKK 34,000,000)
1998-2002: "Improved Proteome Analysis in Medical Biotechnology" under the THOR (Techno-logy by Highly Oriented Research) program, the Danish Research Councils,(DKK 7,000,000)
2000-2008: "The Danish Biotechnology Instrument Centre: Proteome Analysis (DABIC)",(DKK 12,400,000)
2001-2005: "Genomics and Proteomics - New Methods in Pollution Research (GENIPOL)",EU contract under the 5th Frame Program, (DKK 17,000,000)

Research interests:
Application of cutting edge proteomics to major medical health and environmental problems. Improvement of the technology, education of young scientists

Some recent publications
S.J. Fey and P. Mose Larsen. ***"2D - or not 2D"***. Current Opinions in Chemical Biology **5**: 26-33; 2001.
K. Højlund, K. Wrzesinski, P. Mose Larsen, S.J. Fey, P. Roepstorff, Aa. Handberg, F. Dela, J. Vinten, J.G. McCormack, C. Reynet, K. Herick and H. Beck-Nielsen. ***"Proteome analysis reveals phosphorylation af ATP synthase beta –subunit in human skeletal muscle and proteins with potential roles in type 2 diabetes"***. J Biol Chem 278: 10436-42; 2003.
P. Mose Larsen, S.J. Fey, A.E. Karlsen and J. Nerup. ***"Human protein markers for Type I diabetes development"***. Patent Publication: Application No. PA 2001 01298
S. Ferreira, K. Hjernø, M. Larsen, G. Wingsle, P. Mose Larsen, S.J. Fey, P. Roepstorff and M.S. Pais. *"**Heat stress-induced proteome profiling of Populus euphratica through 2D-PAGE and mass spectrometry**"*. *Submitted;* 2005.

Curriculum Vitae for Stephen J. Fey

Born 23.10.1953

Education
B.Sc. Kings College, London University;
A.K.C. (Associate of King's College, London University
Ph.D. Department of Biophysics, Kings College, London University

Employment
Biostructural Chemistry Division, Chemistry Institute, Aarhus University 1980-1984
Institute of Medical Microbiology and Institute of Human Genetics, Aarhus University (Joint Group Leader) 1984-1996
Co-founder of Centre for Proteome Analysis, International Science Park Odense, University of Southern Denmark, (Associate Professorship) 1996-to date
Third Military Medical University and West China Hospital, Chongqing, People's Republic of China (Professor in Medical Proteomics) 2003-to date

Other activities
International reviewer for the Major National Research Facilities Program, Australia
Reviewer for several scientific journals inc. Electrophoresis and Proteomics.
Advisor to the EU on Future Policies, 2004-5
Co-organiser of 2nd International Conference on TCM, Chengdu, China 2005

Main research projects
"Center for Experimental BioInformatics (CEBI)", The Danish National Research Foundation, (DKK 34,000,000) 1998-2003
"Improved Proteome Analysis in Medical Biotechnology" under the THOR (Technology by Highly Oriented Research) program, the Danish Research Councils, (DKK 7,000,000) 1998-2002
"The Danish Biotechnology Instrument Centre: Proteome Analysis (DABIC)", (DKK 12,400,000) 2000-2008
"Genomics and Proteomics - New Methods in Pollution Research (GENIPOL)", EU contract under the 5th Frame Program, (DKK 17,000,000) 2001-2005

Research interests
Application of cutting edge proteomics to major medical health and environmental problems especially in the areas of diabetes and cancer. Development of new technologies for proteomics (including both pre- and post-proteomics)

Some recent publications
S.J. Fey and P. Mose Larsen. ***"2D - or not 2D"***. Current Opinions in Chemical Biology **5**: 26-33; 2001.
P. Mose Larsen and S.J. Fey. ***"Method for the manufacture of Separation Matrices and method for analysing biomolecules on ultra-long separation matrices"***. Patent publication No. WO200270111
P. Mose Larsen, S.J. Fey, A.E. Karlsen and J. Nerup. ***"Human protein markers for Type I diabetes development"***. Patent Publication: Application No. WO 2003078456
S. Ferreira, K. Hjernø, M. Larsen, G. Wingsle, P. Mose Larsen, S.J. Fey, P. Roepstorff and M.S. Pais. ***"Heat stress-induced proteome profiling of Populus euphratica through 2D-PAGE and mass spectrometry"***. *Submitted* 2005.
U. Bjerre Christensen, P. Mose Larsen, S.J. Fey, A.E. Karlsen, F. Pociot, J. Nerup and T. Sparre. ***"Different islet protein expression profiles during spontaneous diabetes development vs. allograft rejection in the BB-DP rats"***. *Submitted* 2005

Curriculum Vitae for Dan Funck Jensen

Born: 6. February 1955 in Århus, Denmark

Education:
KVL 27. February 1980 Candidatus Hortonomiae (cand. hort., HG).
KVL 12. August 1985 Licentiatus agronomiae (lic. agro.) (Ph.D.)

Employment:

1986	Post Doc at the Department for Plant pathology, KVL (Visiting scientist at Colorado State University for 7 months).
1987-88	Assoc. Professor (substitute) for 18 months at the Department for Plant Pathology, KVL.
1988-89	Post Doc. at the Department for Plant Pathology, KVL.
1998-99	Sabbatical at Horticulture Research International, Wellesbourne, UK.
1989-	Assoc. Professor at the Department of Plant Biology, Section for Plant Pathology, KVL

Research interests:
Biological control of plant diseases. Fungal ecology and studies of antagonist/pathogen interactions. Exploitation of reporter gene technology, molecular characterization and detection of fungi. Formulation and integrating BCAs with seed technology. Post harvest diseases in carrots and apple.

The list of publications include 3 book chapters, 35 papers in journals with peer review and 37 proceedings from international scientific meetings and chief editor of two proceedings from an OECD/EFPP and an IOBC/EFPP meeting that he organized respectively.

Some recent publications

Jensen, B., Knudsen, I.M.B, Jensen, D.F. (2002). Survival of conidia of *Clonostachys rosea* on stored barley seeds and their biocontrol efficacy against seed borne *Bipolaris sorokiniana*. Biocontrol Science and Technology 12:427-441.

Jensen, B., Knudsen, I.M.B, Madsen, M & Jensen, D.F. 2004. Biopriming of infected carrot seed with an antagonist selected for control of seedborne Alternaria spp. Phytopathology 94: 551-560

Jensen, D.F., & Schulz, A. (2003). Exploitation of GFP-technology with filamentous fungi. chapter 33.pp 441-451.*In*: Handbook of Fungal Biotechnology, 2edition, Dilip K. Arora, Poul D Bridge and Deepak Bhatnager (Eds.). Marcel Dekker Inc., New York.

Lübeck, M. and Jensen, D.F. (2002). Monitoring of biocontrol agents based on *Trichoderma* strains following their application to glasshouse crops by combining dilution plating with UP-PCR fingerprinting. Biocontrol Science and Technology 12: 371-380.

Lübeck, M., Knudsen, I.M.B., Jensen, B., Thrane, U., Janvier C. and Jensen, D.F. (2002). GUS and GFP transformation of the biocontrol strain *Clonostachys rosea* and the potential for using these marker genes in ecological studies. Mycological Research 106: 815-826.

Nielsen, K., Justesen, A.F., Jensen, D.F. and Yohalem, D.S. (2001). Universally primed polymerase chain reaction alleles and internal transcribed spacer restriction fragment length polymorphisms distinguish two subgroups in *Botrytis aclada* distinct from *B.byssoidea*. Phytopathology 91: 527-533.

Mikkelsen, L., Roulund, N., Lübeck, M. and Jensen, D.F. (2001). The perennial ryegrass endophyte *Neotyphodium lolii* genetically transformed with the green fluorescent protein (*gfp*) and visualization in the host plant. Mycological Research 105 (6): 644-650.

Mikkelsen, L., Sarrocco, S., Lübeck, M., and Jensen, D.F. (2003).
Expression of the red fluorescent protein DsRed-Express in filamentous ascomycete fungi. **FEMS Microbiology Letters 223 (1): 135-139.**

Yohalem, D.S., Nielsen, K., Green, H. & Jensen D.F. (2004) Biocontrol agents efficiently inhibit sporulation of Botrytis aclada on necrotic leaf tips but spread to adjacent living tissue is not prevented FEMS MICROBIOLOGY ECOLOGY 47 (3): 297-303 2004

Curriculum Vitae for Jørgen Eilenberg

Born: 14/7-1954

Education: Cand.Hort. 1981, PhD 1985, D.Sc. (Danish dr.agro.) 2002

Employment: Professor, Department of Ecology, The Royal Veterinary and Agricultural University, Thorvaldsensvej 40, 1871 Frb C, Denmark. jei@kvl.dk

Other activities

Chair of Danish Centre for Biological Control (www.centre-biological-control.dk)

Management Committee member and WG leader, COST Action 842 (insect pathogenic fungi), Management Committee member, COST Action 862 (bacterial toxins)

Chair of Fungus Division, Society of Invertebrate Pathology, 2004-2006

Editorial board 'Biological Control'

Steering committees, The Ministry of Environment, various projects on biocontrol

Main research projects:

- Natural occurrence of insect pathogenic fungi on the organic farm site 'Bakkegården'
- Horizontal and vertical transmission of entomopathogens and endosymbionts in aphid populations.
- Infection processes of insect pathogenic fungi in aphids
- Plant protection by using beneficial soil microorganisms

Co-operation with major Danish universisities and research institutes and abroad.

Research interests: Strategies for biological control; insect-pathogenic fungi; insect-pathogenic bacteria; *Bacillus*; Population interactions; host-pathogen interactions; field prevalence; infection processes; genetical characterization; field releases; effects on target; effects on non-target; field persistence; metabolites; strategies for biological control.

Some recent publications

Eilenberg, J.; Enkegaard, A.; Vestergaard, S.; Jensen, B. (2000): Biological control of pests on plant crops in Denmark: Present status and future potential. Biocontrol Science and Technology, **10**, 703-716.

.*Eilenberg, J.; Hajek, A.E.; Lomer, C. (2001)*: Suggestions for unifying the terminology in biological control. BioControl, **46**: 387-400.

Klingen, I., Eilenberg, J.; Meadow, R. (2002) : Impact of farming systems, field margins and bait insect on the findings of insect pathogenic fungi in soil. Agriculture, Ecosystem and Environment, **91**, 191-198.

Eilenberg, J.; Meadow, R.(2002): Fungi for Biocontrol of brassica root flies, *Delia radicum* and *Delia floralis*. In Upadhyay, R. (ed.): Advances in Microbial Control. Kluwer, 181-191.

Hughes, W.O.H.; Eilenberg, J.; Boomsma, J.J. (2002): Trade-offs in group living: transmission and disease resistance in leaf cutting ants. Proceedings of the Royal Society of London B, **269**, 1811-1819.

Nielsen, C.; Hajek, A.E., Humber, R.H.; Bresciani, J.; Eilenberg, J. (2003): Soil as an environment for winter survival of aphid-pathogenic Entomophthorales. Biological Control, 28: 92-100.

Jensen, .G.B.; Hansen, B.M.; Eilenberg, J.; Mahillon, J. (2003): The hidden lifestyles of *Bacillus cereus* and relatives. Environmental Microbiology, 5: 631-640.

Goettel, M.; Glare, T.; Eilenberg, J. (2004). Entomopathogenic Fungi and Their Role in Regulation of Insect Populations. In: Comprehensive Molecular Insect Science. Vol. 6 (L.I. Gilbert, K. Iatrou and S. Gill, Eds.) Elsevier. pp 361-406.

Eilenberg, J. & Hokkanen, H.T.M. (eds). An ecological and societal approach to biological control. Springer Verlag, to be published autumn 2005