



Midterm Status Report 2002 and Application for Continuation in 2003

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

1. Research program

Research in organic farming 2000-2005 (DARCOF II)

2. Project title and number

Bacterial infection risk associated with outdoor organic pig production

- with special reference to *Salmonella* and *Campylobacter* infection
 - SaCaFree FØJO II project no. II.10
-

3. Head of project

Dorte Lau Baggesen, Senior research officer, Ph.D.

Danish Veterinary Institute

27 Bülowvej

DK-1790 Copenhagen V

Denmark

Phone +45 35 30 02 07

Fax +45 35 30 01 20

E-mail dlb@vetinst.dk

4. Participating institutes

Danish Veterinary Institute

27 Bülowvej

DK-1790 Copenhagen V

Denmark

Phone +45 35 30 01 00
Fax +45 35 30 01 20
E-mail vetinst@vetinst.dk

5. Other project staff

Eva Møller Nielsen, Senior research officer, Ph.D.
Danish Veterinary Institute
27 Bülowvej
DK-1790 Copenhagen V
Denmark
Phone +45 35 30 02 03
Fax +45 35 30 01 20
E-mail emn@vetinst.dk

Annette Nygård Jensen, Cand. Scient.
Danish Veterinary Institute
27 Bülowvej
DK-1790 Copenhagen V
Denmark
Phone +45 35 30 03 28
Fax +45 35 30 01 20
E-mail anj@vetinst.dk

6. Project period (month, year)

Start of project:	1 st . July 2002
End of project:	30 th . June 2004

7. Midterm description of the project, its results and progress, and application for continuation in 2003

A. Project summary

The modern consumers are becoming increasingly critical of the quality of meat and other types of products of animal origin. Focus is put on the animal husbandry and the way of production concerning animal welfare. This leads to increasing interest for organic, free-range or other kinds of animal-friendly production systems where the animals benefit from a low animal density and good possibilities for expressing normal behaviour. In general, consumers also expect products from these kinds of systems to be of a higher microbiological quality compared to products from conventional production systems. However, today there is no documentation for a lower level of the most common zoonotic bacterial infections (e.g. *Salmonella*, *Campylobacter* and *Yersinia*) in organic or other alternative production systems.

The objective of this project is to improve the knowledge on the risk of outdoor pig production in relation to spread and persistence of *Campylobacter* and *Salmonella* infections. For *Salmonella* the specific objectives are to evaluate the survival of *Salmonella* Typhimurium in soil and grass of contaminated pastures used for outdoor pig production, measurement of the infectivity of naturally *S. Typhimurium* contaminated pastures in relation to time, and in the case of high infectivity, evaluation of the pathogen reducing effect of soil treatment. For thermophilic *Campylobacter*, the objectives are to describe the infection dynamics of natural *Campylobacter* infections over time in outdoor pigs, including time of colonisation, level of excretion in faeces, species distribution in the group and in the individuals, interaction with the environment, and to describe the possible changes in prevalence and species distribution in relation to time and environmental contamination.

Experimental pastures for production of outdoor organic piglets will be set up in 2003. Experimentally infected piglets will be grouped together with uninfected tracer piglets in order to monitor the transmission of infection. In addition, soil samples will be analysed to determine the level of contamination. After a period of two months, the pastures will be vacated, infected piglets slaughtered and tested. A new set of uninfected piglets will be introduced in each pasture to monitor if these animals become infected due to their habitation in the *Salmonella* contaminated pastures. The pastures and animals will be sampled and monitored (bacteriological (qualitatively and semi-quantitatively), serological and by necropsy examinations). Based on the results obtained in the second period, a third period of two months will be initiated for further monitoring of the infectivity of the *Salmonella* contaminated pasture environment, including the preventive effect of ploughing the pastures before new tracer animals are introduced.

A method will be developed and evaluated for determining the species composition (*C. jejuni* and *C. coli*) in faecal samples and subsequent isolation of the species in minority. The groups of pigs serving as controls in the experimental *Salmonella* infection study will also be used for studying the dynamics of natural *Campylobacter* infection in outdoor pigs. The piglets will be monitored for natural colonisation of thermophilic *Campylobacter* species, and the ratio between *C. jejuni* and *C. coli*. Likewise, environmental samples are analysed throughout the experimental periods. Isolates from animals and environment will be identified to the species level and serotyped to monitor the dynamics of the infection: number of different strains in each pig and persistence/exchange of strains over time.

The achievements obtained will be formulated in practical guidelines directly applicable for the

organic pig producers in order to minimise the risk of zoonotic infection in organic pig herds. In addition, results will be available for the scientific community through publication in reviewed journals. The large amount of quantitative data obtained in this project will deliver the necessary information for use in quantitative risk assessment of zoonotic infection in organic pig production.

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

No.	Work package title	Participants*	Budget** (1.000 DKK)	Start	End	Deliverable no(s):
WP1	Investigation of introduction and spread of <i>Salmonella</i> in outdoor pigs	<u>Dorte Lau Baggesen</u> ; Eva Møller Nielsen; Annette Nygård Jensen	1,325	2002	2004	1 – 6
WP2	Investigation of the dynamics of natural <i>Campylobacter</i> infection in outdoor pigs	<u>Eva Møller Nielsen</u> ; Dorte Lau Baggesen; Annette Nygård Jensen	675	2002	2004	7 – 11

* Responsible participants are underlined

** All expenses concerning the animal experiments (756,000 Dkr.) are covered by WP1. These experiments form the basis for both WP's.

B. Objectives and expected achievements

The overall objective of the project is to improve the knowledge on the dynamics behind environmental spread of two important zoonotic bacteria – *Salmonella* and *Campylobacter* – within outdoor pig production systems.

Good management procedures – good farming practice – based on scientific evaluation of the risks of introduction and persistence of zoonotic infections is essential for the redevelopment of the organic production. The present proposal focuses on studies to describe the dynamics of zoonotic infections in outdoor pigs and evaluate the risk of outdoor production in relation to spread and persistence of *Campylobacter* and *Salmonella* infections in pigs. This include

- evaluation of the survival of *Salmonella* Typhimurium in soil and grass of contaminated weaning and fattening pastures for outdoor pig production systems
- measurement of the infectivity of naturally *S. Typhimurium* contaminated pastures in relation to time, and
- in the case of high infectivity, evaluation of the pathogen reducing effect of soil treatment
- *description of the prevalence of natural campylobacter infection over time in outdoor pigs including description of the species distribution in the group and in the individuals*
- evaluation of the possible changes in prevalence and *Campylobacter* species distribution in relation to time and environmental contamination

Through the described project new important knowledge regarding salmonella and campylobacter infection in outdoor pig production systems will be obtained. This includes information on the infectivity of *Salmonella* under natural conditions, e.g. on the risk for healthy piglets to be infected from infected animals or contaminated environment. The investigations will provide quan-

titative information on the infection and contamination levels, which is presently very limited. In the case of a very high contamination level in the pastures the study will provide additional information on the pathogen reducing effect of the ploughing pastures. In addition, new information on the infection dynamics of thermophilic campylobacters in outdoor pigs is expected especially in relation to the level of colonisation, distribution of different species and types, the possible co-infection of *C. coli* and *C. jejuni*, and the transmission between animals and the environment.

The achievements obtained will be formulated in practical guidelines directly applicable for the organic pig producers in order to minimize the risk of zoonotic infection in organic pig herds. In addition, results will be available for the scientific community through publication in reviewed journals. The large amount of quantitative data obtained in this project will deliver the necessary information for use in quantitative risk assessment of zoonotic infection in organic pig production. This achievement will be strength by the interaction with other projects carried out by the present group (Wildlife as a source of salmonella infection in food-animal production, FØSI00-SVS-6, which has been extended by *Campylobacter* investigations; ”Grundlag for rådgivning vedr. foderets betydning for salmonellainfektion i økologiske svinebesætninger” [The significance of feeding in relation to salmonella infections in organic pig herds] , J.nr. 93S-2462-Å01-00981).

C. Midterm results and progress

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

The project has just started and no results have been obtained

C.2 Fulfilment of deliverables and milestones

(To be completed for each work package)

WP1: Investigation of introduction and spread of <i>Salmonella</i> in outdoor pigs	Time schedule according to application	Deviations, if any*
Deliverables		
1. Determination of detection limit for bacteriological methods	Jan. 03	
2. Information on the risk for <i>Salmonella</i> transmission to uninfected piglets constituted by infected animals and contaminated pasture environment representing different infection pressures	Sept. 03	
3. Information on whether survival of <i>Salmonella</i> in the environment under naturally conditions is high enough to cause/transmit infection in uninfected animals	Sept. 03	
4. Information on the pathogen reducing effect of ploughing pastures will be available if the infection level established is sufficiently high for this evaluation	Nov. 03	
5. Scientific and international publication of results	June 04	
6. Practical guidelines for minimising the risk of salmonella infection in outdoor pig production	June 04	
Milestones		
Determination of detection limit for the bacteriological method applied	After 1. quarter of 03	

First experimental period – spread of infection between experimentally infected animals and uninfected tracer animals	After 2. quarter of 03	
Second experimental period – spread of infection from pasture environment to uninfected tracer animals	After 3. quarter of 03	
Third experimental period – <i>either</i> spread of infection from pasture environment to uninfected tracer animals and effect of soil treatment <i>or</i> spread of infection between experimental highly infected animals and uninfected tracer animals	After 3. quarter of 03	
Scientific and international publication of results and formulation of practical guidelines for minimizing the risk of salmonella infection in outdoors pig production	After 2. quarter of 04	

* *Deviations are to be further discussed in D*

WP2: Investigation of the dynamics of natural <i>Campylobacter</i> infection in outdoor pigs	Time schedule according to application	Deviations, if any*
Deliverables		
7. Development and evaluation of a new approach for determining the species composition (<i>C. jejuni</i> and <i>C. coli</i>) in faecal samples and subsequent isolation of the species in minority	Jan. 03	
8. Publication of method in the form of a short paper (note/letter)	Apr. 03	
9. Elucidation of the infection dynamics of thermophilic campylobacters in out-door organic pigs, including time of colonisation, excretion level in faeces, number of different strains co-colonising one piglet during time, persistence of strains, and the ratio between <i>C. jejuni</i> and <i>C. coli</i> in individual animals	Oct. 03	
10. Information on the interaction between individual pigs and between the pigs and environment with respect to <i>Campylobacter</i> infection	Nov. 03	
11. Scientific and international publication of results	June 04	
Milestones		
Implementation of a PCR-based method for determination of the composition of <i>Campylobacter</i> species in faecal samples and the subsequent isolation of the species in minority. Determination of detection limit of the method.	After 2. quarter of 03	
Results are obtained on the infection dynamics of thermophilic campylobacters in outdoor piglets repeated during three seasons	After 1. quarter of 04	
Results are obtained on the interaction between outdoor piglets and the environment with respect to <i>Campylobacter</i> infections	After 1. quarter of 04	
Publication of results in international journal	After 2. quarter of 04	

D. Description of deviations and subsequent adjustments of plans

The project has just been started and no deviations are made. A Ph.D. studium has been established based on the project. This has caused adjustment of the budget (see 8. A & B). In addition, related activities will be performed just after this project period.

E. Project publications and other products

1. Articles in international, scientific journals with review procedures

None

2. Papers presented at congresses, symposiums, etc.

None

3. Reports, articles in agricultural journals, etc.

None

4. Oral presentations, public meetings, field days, etc.

None

F. Scientific education

A Ph.D. studym has been established based on the project. This has been possible due to an additional grant given by Research School for Organic Agriculture and Food Systems, The Royal Veterinary and Agricultural University and Danish Research Centre for Organic Farming.

The Ph.D. study lasts three years from 1st of October 2002 to 31st of September 2005. In addition to the activities described in this project the Ph.D. study will include model investigations on the surveillance of pathogens in the extra intestinal environment. The model investigations have the aim to validate hypothesis described through the activities carried out in this project.

Cand. Scient. Annette Nygård Jensen has been employed as a ph.d.-student. Associate professor Anders Dalsgaard, DVM, Ph.D., Institute of veterinary microbiology is main supervisor whereas Dorte Lau Baggesen, Senior research officer, Ph.D. and Eva Møller Nielsen, Senior research officer, Ph.D. are co-supervisors.

G. National and international cooperation

The outdoor experiments will be carried out at the research animal facilities of The Danish Veterinary and Agricultural University.

H. Critical reflection on the project

The amount and quality of expected achievements will increase because of the establishment of a Ph.D. study with additional activities. The contact to other researchers in the area of organic farming will be improved through the participation in the Research School for Organic Agriculture and Food Systems.

8. Budget

A. Account for any change in budgets

Within the time schedule and the budget of this project some adjustments have been necessary to fulfil the requirements of the Ph.D. study. The investigations carried out in this study will cover the main part of a ph.d. study (see pkt. F).

B. Budget for the whole project (1.000 DKK) – NB ! AJUSTED BUDGET

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

Year:	Consumption before 2002	Expected consumption 2002	2003	2004	2005	Total
Man-months						
Scientific personnel		3	12	9		24
Technical personnel			19			19

Year:	Consumption before 2002	Expected consumption 2002	2003	2004	2005	Total
Salaries						
Scientific personnel		80	320	244		644
Technical personnel			430			430
Other operational costs		10	287	50		347
Equipment		25	130			155
Others (please specify) Ph.d. fee (RVAU)			45	45		90
Direct costs		115	1.212	339		1.666
Indirect costs (20% of direct costs)		23	242	68		333
Total		138	1.454	407		1.999

Comments:

9. Signatures and stamps

Name	Institute	Date	Signature
Head of project Dorte Lau Baggesen	Danish Veterinary Institute	25. september 2002	

Appendix I. Detailed budget

A. Budget for each participating institute (1.000 DKr)

Name of Institute:

Year:	Consumption before 2002	Expected consumption 2002	2003	2004	2005	Total
Man-months						
Scientific personnel						
Technical personnel						

Year:	Consumption before 2002	Expected consumption 2002	2003	2004	2005	Total
Salaries						
Scientific personnel						
Technical personnel						
Other operational costs						
Equipment						
Others (please specify)						
Direct costs						
Indirect costs (20% of direct costs)						
Total						

Comments:

B. Budget for each participating department (1.000 DKK)

Name of Institute and department:

Year:	Consumption before 2002	Expected consumption 2002	2003	2004	2005	Total
Man-months						
Scientific personnel						
Technical personnel						

Year:	Consumption before 2002	Expected consumption 2002	2003	2004	2005	Total
Salaries						
Scientific personnel						
Technical personnel						
Other operational costs						
Equipment						
Others (please specify)						
Direct costs						
Indirect costs (20% of direct costs)						
Total						

Comments:

C. Budget for co-financing from each participating institute (1.000 DKK)

Name of Institute:

Year:	Consumption before 2002	Expected consumption 2002	2003	2004	2005	Total
Man-months						
Scientific personnel						
Technical personnel						

Year:	Consumption before 2002	Expected consumption 2002	2003	2004	2005	Total
Salaries						
Scientific personnel						
Technical personnel						
Other operational costs						
Equipment						
Others (please specify)						
Direct costs						
Indirect costs (20% of direct costs)						
Total						

Comments: