

Occurrence of *Ascaris suum* eggs in the danish swine breeding

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FOREWORD

Before I started as a veterinary student, I worked for two sessions on farms - broiler/pigs, pigs/dairy cows respectively. During these periods I learned that it is very important for the animal's welfare to physically clean and disinfect housing carefully.

When I now have the chance to investigate whether disinfection makes a difference, and because I like the universally applicable properties of slaked lime, [disinfects, pasteurisation, reduce odours, improves manure and stable conditions] and I have a good experience of Stalosan F, to use these two products as agents in this paper. Both products will, possibly, be accepted also in organic farming systems which I find is important.

I want to know something concrete, how roundworm problems can be tackled in practice without major use of medicines or chemicals. I hope to broaden my knowledge enough to advise the farmer in effective parasitic control without polluting the environment.

"What goes in, must come out" - this must also apply to the large quantity of anthelmintics that are used in Denmark each year. Despite anthelmintic use on Danish pig farms, I have observed in post-mortems, during my study of meat control, that a large proportion of slaughtered pigs have 'milk spots' in the liver [interstitial hepatitis] probably due to *Ascaris suum* larval migration.

I hope to find *Ascaris suum* eggs on a suitable farm then establish likely location of these eggs on other farms. Should there be time, I hope to be able to treat these farm buildings so that later investigations would find an absence of eggs.

I wish to acknowledge everybody who has made this work possible and for the help given by members of C.E.P. My supervisor, Allan Roepstorff, has been able to point me in the right direction when I was unable to move forward. Laboratory Assistants, Marlene Høg and Claus Dahl, together with all at C.E.P., have given fantastic help in solving a lot of practical problems. Head of Laboratory, Margrethe Pearman and lecturer Jesper Monrad, for help with microphotography. Chemical Consultant Hanne Sørensen, for help in protective clothing in handling slaked lime. Professor Arne Holm and lecturer Svend Clausen informed me of the nature of slaked lime. Faxe Kalkbrud, especially Marianne Ørbaek, Lotte Kjaergård, John Hansen and, not least, Jeanne Schubell, are acknowledged for samples, good ideas and relevant literature.

Stormøllen in Køge, Carl-Gustaf Holst and agronomist Kristian Smedegård for information on Stalosan F. Lecturer Anders Permin for giving me Stalosan F for the trial. Fie Graugård, Organics House, Århus, technician Niels Midtgård and secretary Anne Bisp. Torben W. Schou and Jaap Boes for help with statistics. Library Supervisor Anna Eslau Larsen for help in documentation. Clever and patient supervision in EDB [Computers] amongst others Jens Erik Jensen, Veterinarian Troels Christensen, Steff Houlberg, Ringsted for eleventh hour assistance in post-mortem results.

The Roskilde Slaughterhouse School for contacts with trial farm 2 and, not least, the farmers Willy Hansen, Tommy Mathiesen and families.

Amager, February 1999



Anna E. W. Ibsen

PREFACE

PURPOSE

I want to shed light on the occurrence of *Ascaris Suum* on a traditional Danish pig farm. I will focus on the morphology of *Ascaris Suum* eggs to establish the state of development and where in the housing they develop to the infective stage.

OCCURRENCE

Ascaris Suum is among the most common intestinal parasites in the Danish pig industry. According to Roepstorff and Jorsal [1989] 85 % of pig herds in Denmark are infected with *Ascaris Suum*. Prevalence throughout Denmark [Ropestorff et al. 1998] found to be 5.1 % piglets, 5.8 % weaners, 13.8 % growers, 20.8 % for finishers, 24.7 % gilts, 13.4 % dry sows, 10.4 % for lactating sows and 11.0 % for boars.

Ropestorff and Nansen [1994] report that it is almost impossible to find an uninfected swine farm in Denmark, not even among the most well managed. Furthermore, Eriksen et al. [1981] remarks that it is impossible to avoid a build up of *Ascaris Suum* in experimental work with this parasite, in spite of isolation of pigs, disinfection of all work clothing, high pressure cleaning and regular anthelmintic treatment of sows and pigs not directly involved in experimentation.

LIFECYCLE

A.suum has a direct lifecycle. After oral intake, the *Ascaris* eggs 'hatch' and L2 larvae penetrate the gut, from whence they migrate to the liver through the portal veins. In the liver they develop to L3 and, after 2-4 days, they leave the liver scar formed [milk spots][interstitial hepatitis]. The L3 go with the blood to the lungs where they enter the alveolus. The larva migrate through the trachea to the pharynx from where they are swallowed and enters the intestinal system approximately 10 days after the eggs were ingested. Two skin changes occur in the gut while larva increase in size. 6-8 weeks after ingestion, the infection is patent and the new eggs are shed in the faeces. The newly shed eggs are ovoid 50-80 μ m and have an amber brown outer appearance and small-knobly, thick surface. This surface is very resistant to many chemical treatments and temperatures up to 50 °C [description taken from Monrad and Nansen (1994)].

REACTION TO HEAT

According to Juris et al [1991], mechanical, chemical and biological waste treatment plant can extract all *A.suum* eggs. Tharaldsen and Helle [1989] found that all *A.suum* eggs are dead after 31 days in a slurry tank, agitated and kept at a constant 37 °C. Aerobic fermentation occurs. Tharaldsen and Helle [1989] referred to the article by Lündsmann [1972] that all *A.suum* eggs after 25 days at 40 °C are dead. Burden [1978] found that all the non embryonic eggs are dead after 30 minutes, and 15 minutes for embryonic eggs at 55 °C. The same result [Juris et al. 1992] reached. [Barnard et al. 1987] showed, that at 50°C, all of 1.000 eggs sampled were dead after 4 hours 45 minutes. Polprasert and Valencia [1981] refer to Feachem et al. [1978] for establishing total destruction of *A.suum* eggs: 1 minute at 68°C, 1 hour at 50 °C, 7 days at 46 °C, 30 days at 43 °C.

REACTION TO FERMENTATION

After 20 days of methanogenesis at 35-37 °C Juris et al. [1996] found that only 17-18 % of the A.suum eggs were damaged. Furthermore, Plachy et al. [1996] found 168 hours of waste water treatment with hydrated chalk [10 g/l] at 21-25 °C only reduces living eggs by 3.6 %. Juris et al. [1997] tried anaerobic fermentation and, after 42 days only 15.2 ± 4.02 % of A.suum non-embryo eggs were damaged. Matching this, Carrington et al. [1991] found anaerobic mesophilic fermentation [34 °C] did not have any effect on A.suum but a thermophilic aerobic fermentation can reduce the total eggamount to zero after 4 hours at 55 °C.

REACTION TO CHEMICALS

Polprasert and Valencia [1981] cites Feachem et al. [1978] that human urine is the most effective ovicide, compared to urine of other animal species. At concentrations of 10 %, ova will be killed in 16 hours [Hamdy 1970] [cf. Polprasert and Valencia 1981]. Nilsson [1982] found that swine urine has an ability to restrict development of A.suum. Hurley and Summerville [1981] found that iodine only inhibits the hatching of the eggs for a time and that traces of iodine can be found in most eggs, whether they have been stained by iodine or not and whether the time of exposure is 15 or 180 minutes. Eisenman et al. [1973] described that poly-iodine complexes can penetrate a lipid membrane. However Barret [1976] found that iodine cannot penetrate day-0 eggs even after 24 hour exposure.

Polprasert and Valencia [1981] describe lime as one of the cheapest disinfectants and that it also works as a deodorant when it is added to the faeces. They found that A.suum eggs were only reduced by 26.5 % in the most effective results and recommended that lime treatment of slurry should not take more than 3 hours because there would be only a small improvement over longer periods of time. Unfortunately, they did not use the exothermic energy in the slaking process and lost, therefore, the heating effect.

Strauch [1984] described lime as suitable for all types of waste; to settle toxic metals, take away organic material; destroy pathogens, reduce biochemical/biological oxygen; fix solids and take away odours. He found in his investigations non-slaked lime [CaO] was effective against A.suum eggs. Unfortunately, he did not record how effective.

EGG PROTECTION

To find the resistance of the *A. suum* egg, according to Wharton [1980] we have to examine the construction of the outer shell. It consists of four layers; the outer layer is amber brown, acidic and consists of mucopolysaccharide/protein. Next layer is vitellin, followed by a chitinous shield, inner layer is lipid [three inner layers are produced by the fertile oocyst].

The outer layer produces the lumpy surface of the egg. The chitin layer gives mechanical strength, improved by ribbed and connected conical infrastructure. This makes an impermeable egg, resisting water soluble substance intrusion although, under extreme conditions e.g. hyperosmotic solutions, water may pass. Also temperature is important for the rate of water loss [exponential]. This is caused by increased damp/water pressure and the temperature rises and a graduated 'melting' or transformation' of complex components that form the lipid layer of the egg. The inner lipid layer is giving the egg protection against chemicals.

Varying times of exposure and given temperatures [and if the egg is allowed to cool down before it is stained] have been shown to have influence on the structure of the eggshell [Wharton 1979] but over 63-65 °C, water loss is dramatic and the eggs are permanently damaged.

MATERIALS AND METHODS

STRATEGY

Farm 1 farrowing pens are investigation [ultimo June 1998] for occurrence of *A. suum* eggs on the floor and the lactating sows for excreted eggs [rectal samples] made. This is done to follow the possible transmission of disease from piglets to the finishing pens to which they are moved at 12 weeks and to judge the extent of infection in the farrowing pen and to find where in the pens the *Ascaris* eggs will develop

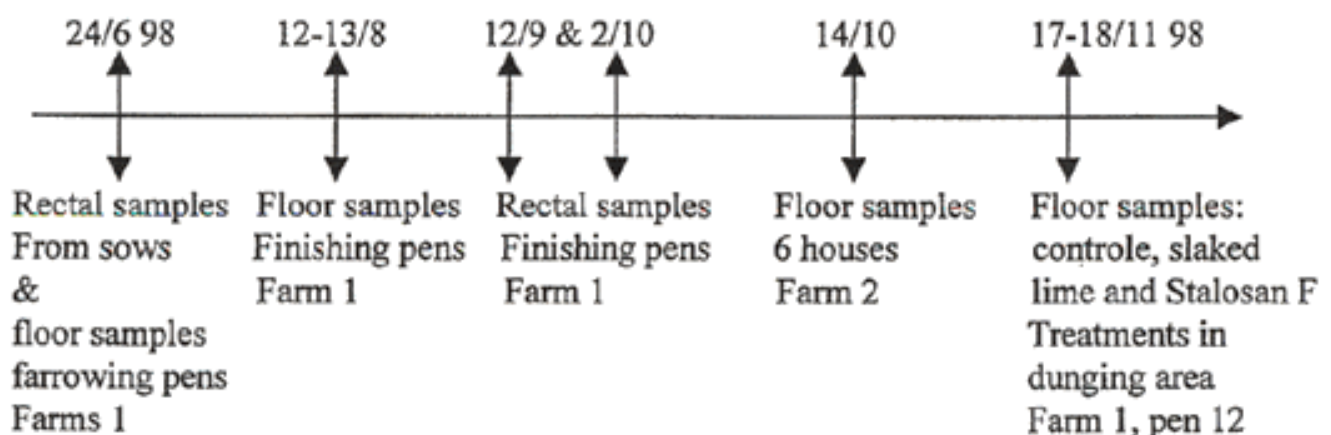
Shortly after growing pigs are placed in finishing pens [early August 1998] floor samples from the pens are taken to judge the extent of infection and where eggs are developing

About 5 weeks after entering the pens [mid September 1998] rectal samples from all pigs in the pens are investigated to evaluate carryover of infection from farrowing pens. Approximately 8 weeks after entering the finishing pens [early October 1998] a further series of rectal samples from all pigs were examined. At this time it should be possible to find eggs originating from both farrowing and finishing pens

An investigation of the efficacy of two disinfectants [slaked lime and Stalosan F] was completed [mid November 1998] in one pen in Farm 1.

In Farm 2 floor samples from 6 different stables at different cleaning stages were taken. In this way differences in buildup of *A. suum* at differing stages of the cleaning cycle can be established

Calendar of events



DESCRIPTION OF FARMS

CHOICE OF FARM

Farm 1 was chosen based on a recommendation of C.E.P. [Center of Experimental Parasitology], K.V.L.[Royal Veterinary school], Denmark, who have long experience that this farm "is a good place to find A.suum".

Furthermore, the farmer would like to help 'science'. The majority of samples were taken on this farm.

Farm 2 was found by Roskilde Slaughterhouse school following my request for a contact for a well managed farm which often delivered finishes with more than 10 milk spots in each liver. In this farm floor samples were taken from 6 different houses at varying stages of the cleaning cycle.

SIZE OF FARM

Farm 1 has 12 sows and 1 boar. They produce about 225 finishes per annum. Sows are homebred Landrace/ Yorkshire x Duroc or Hampshire though sometimes gilts are purchased to renew blood lines. Boar is currently Duroc. Often AI is used.

Farm 2 is modified SPF rated. 250 finishes produced weekly. All weaners bought in at 25 kg. Breed type are normally LY x Duroc or Hampshire.

HOUSING

Farm 1: sows in dry sow house placed in boxes [0.7 x 1.9 m] with iron frame on concrete floor. Cleaning by hand daily. Straw given daily. Approximately 2 weeks before farrowing moved to farrowing house consisting of 12 pens on concrete, wooden dividers between individual pens and sow/piglet areas. Dividers between sow and piglet partition is raised to 20 cm. Heating lamps are used for first few days after farrowing to maintain high temperature and to encourage piglets to lay in their own area. Four to five days after farrowing, the lamp is swapped for a bale of straw suspended to keep the area warm. Sow is free in the pen moving between resting and dunging area. This applies to piglets as well. Cleaning is manual performed twice weekly. Straw applied daily. Weaning is at 12 weeks, one litter at a time directly to finishing pens [12 pens in house] similarly with concrete resting area and slats in dunging area. Partitions are of wood, steel gates separating the dunging passage. This allows contact between litters. Chopped straw is provided daily. 8 x 15 kg barley straw per housed is used [equivalent to one forkful per litter per day]. Restricted water and feed.

Farm 2: 6 stables - 5 of which are new, slats in the dunging area. 6th stable in traditional style - concrete throughout. Partitions are made of wood, no contact between pens. Small quantities of chopped straw applied daily. Old fashioned stable used for sickly piglets or litter runts or separated pigs. Food and water ad libitum.

CLEANING

Farm 1:

As the pens are emptied of pigs [one cycle 2 months] they are cleared of manure manually. Ideally, when brush cleansing is completed, high pressure hose with hot [80°C] water followed by slaked lime painted on walls and floors. Generally no further cleaning is completed after manure is shovelled out and hose cold water on slats. Always a disinfectant powder chosen on the basis the 'the cheapest is as good as the most expensive' is used. The powder presently used is dissolved in water and spread with a garden syringe sprayer containing 100 gms dilute in 10 litres [1:100]. Effective after 5-30 minutes. Slurry channel in finishing pens is cleared when waste is 10-15 cm deep. Urine from beef herd is pumped through the channel to remove solids to slurry tank.

Farm 2:

In the stable with concrete floor, cleaning is daily, the remaining 5 cleaned between batches. These are thoroughly cleaned; manual removal of solids is followed by water sprayed over 24 hours. Further cleaning manually follows with high pressure hose [hot water at 80 °C]. 2-3 days later the walls, ceilings and floors are painted with lime. 2-3 days later a new batch of pigs come in.

On the day samples were taken, one pen was emptied and, in the process, soaked with cold water. The second pen had been cleaned one week previously and contained new pigs. The third pen had been cleaned six weeks previously, the fourth 8-9 weeks before, the fifth more than 16 weeks earlier and the sixth cleaned daily.

PARASITE CONTROL

Farm 1:

Farm 1:

Treatment Panacur® [Fenbendazole] since 1988 administered when piglets are 12 weeks old [weaning] and the sows one month before farrowing. Not always done! Doses are measured according to weight of pig [estimated] and mixed with dry food in individual pen feeder troughs. Troughs are communal in finishing and farrowing pens but individual in dry sow house.

Farm 2:

Treatment for parasites is completed shortly after the pigs are bought in. Choice of drug is on a rotating basis changed after each batch of pigs. Furthermore, emphasis is given to a very high level of hygiene.

FARM MANAGEMENT

Farm 1 - traditional father/son

Farm 2 - modern rational management by married couple both full time on the farm

SAMPLES

TIME PERIOD

All samples are collected in the period 24/6/98 to 18/11/98

FLOOR SAMPLING TECHNIQUE

Samples were taken from cracks in the floor where edges meet e.g. floor meets partition walls using screwdriver and putty knife, useful tools for the exercise. Each sample's characteristics [moisture, colour and consistency] together with location were noted. Samples were placed in numbered sealed plastic boxes and refrigerated. See Fig1 and Fig2 for location of samples.

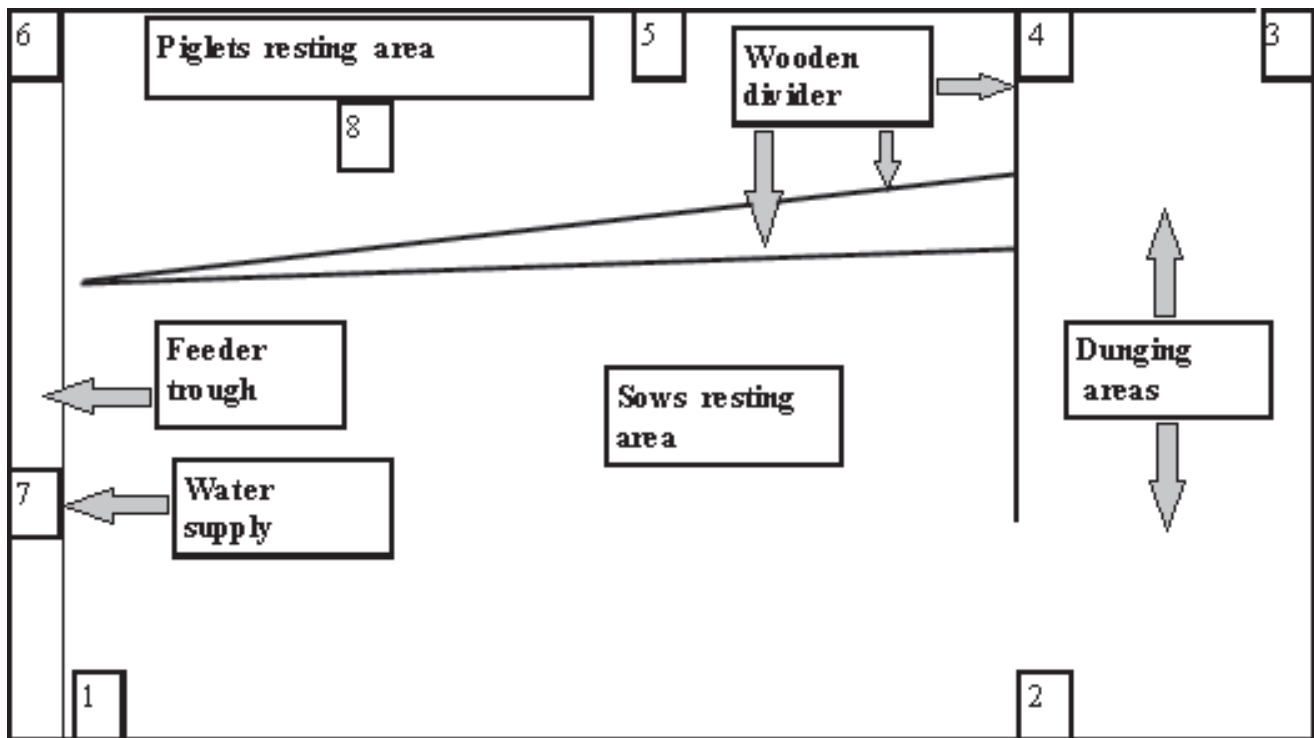


Fig.1 Farrowing pen, farm 1, exact location of samples taken.

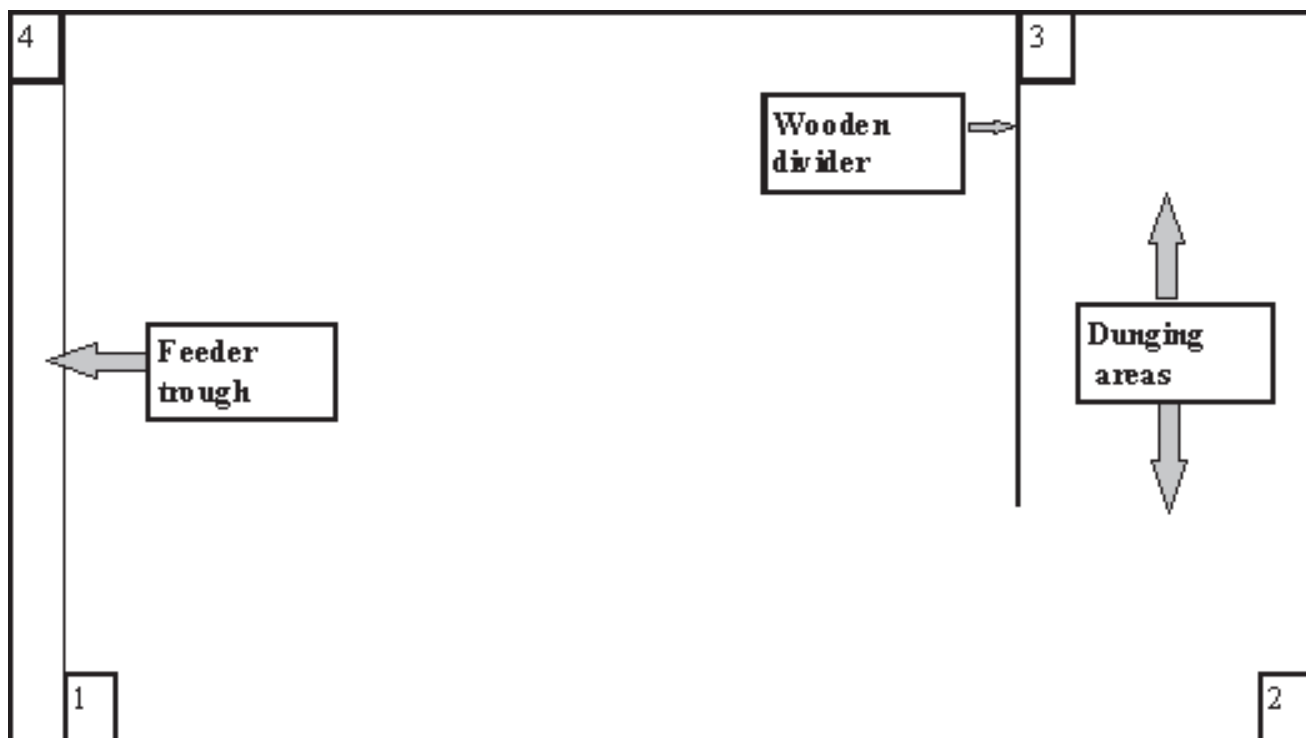


Fig. 2 Pen in finishing house, farm 1. Exact location of samples taken.

ANALYSIS OF FLOOR SAMPLES

Weighing $5 \pm 0,1$ gm of samples into 50 ml centrifugal glass containers. 0.5M NaOH up to 50 ml. After 16-18 hours refrigerated, samples were rinsed through 63 μ m and 20 mm sieves with hand hot water [38 - 40 °C]. By this method all material between 20 mm and 63 mm is retained and transferred to a 10 ml graduated centrifugal glass tube. After cleansing all samples the glasses were balanced by topping up with tap water. Centrifuge for 7 minutes at 1200 rev/min. The supernatant was removed and 0.5 ml of the residue was transferred to a clean 10 ml graduated tube. A sugar/salt suspension [density 1.28] was added as a flotation media. 1.3 ml of this well mixed suspension was transferred to a McMaster counting chamber. The counting of *Ascaris* eggs was done at 100 x magnification. Stage of development was noted for each egg though estimates were made when numbers exceeded 200.

PIG SAMPLING [RECTAL] TECHNIQUE

Rectal samples taken with gloved hand dipped in water immediately before sampling. In a few instances no faeces were found in the rectum, this pig being colour marked, sampling retried after 15 minutes. Samples stored in used glove numbered and refrigerated.

ANALYSIS OF PIG SAMPLES

4.0 ± 0.1 gms of faeces were placed in plastic drinking cups, 56.0 ml of tap water added and sample mixed carefully with a wooden spatula. After 30 minutes at room temperature the samples were individually stirred and passed through a single layer of rough gauze into a numbered plastic container. From here, 10 ml of sample transferred into a 10 ml centrifugal tube and placed in a centrifuge for 7 minutes at 1200 rev/min. The supernatant was removed from all samples and the flotation media [density 1.28] was topped up to 4.0ml, two at a time. After further mixing 1.3 ml was placed in a McMaster counting chamber [magnification 100x]. Within the chamber, both quadrants were counted and recorded [all eggs within and on borderlines]. In samples over 200, results were estimated.

STATISTICS

All statistics were executed in Graph Pad Prism.

MATERIALS USED

- Testo 635 Thermohygrometer
- Ruler
- Plastic cups and tops
- Plastic gloves
- Soup spoon
- Screwdriver
- Spatula
- Personal protective clothing [slaked lime]
- Slaked lime
- Stalosan F

SLAKED LIME

Slaked lime with added water is traditionally used to whiten houses, farms and churches and as a basis for cement. Chalk is mined as CaCO_3 and heated in an oven to $1100\text{ }^\circ\text{C}$ to produce CaO [slaked lime] and CO_2 . Adding water to slaked lime produces Ca(OH)_2 [hydrated lime] pH 12.4 and heat. After a short period of time, hydrated lime reacts with the CO_2 in the atmosphere and produces CaCO_3 [calcium carbonate] pH 9.1, water and heat. Slaked lime appears in two forms 'light slaked' that reacts with water within 2 minutes and 'hard slaked' that completes reaction with water in 20 minutes. This information is from Faxekridt [1998] and from Thøgersen [1982]. In my investigations, I used 'light slaked' because it gave the most useable results in laboratory [C.E.P.] environment. Because slaked lime is an aggressive irritant and can cause lung oedema after inhalation, I used a lot of personal protection in this experiment:

- 4H(PE/EVA/PE laminate) gloves
- 3M 8835 (FFP3SL EN-1490) dust mask
- Protective goggles
- Rubber boots
- Bucket - clean cold water
- Eye bath - sterile 0.9 % NaCl
- White coat

STALOSAN F

Stalosan F is a pink powder with a faint smell of chlorine. It was patented 5/10/66 [5142] and, according to the producer, it is built on synergy principles - ingredients improve each other's effects. The product was, until 1990, monitored by the Danish Veterinary Service and no negative effects were notified. Stalosan F is declared as non-toxic and displays no negative effects. After the expiry of the patent, a new product was developed [Stalosan F] more expensive than the original and more effective. The following information is taken from Stormollen's production manual [1998].

Stalosan F according to Mehtling et al. [1996] contains:

| | |
|---------|--|
| 85 % | phosphate complex |
| 10,10 % | Hydro aluminiumsilikat (inactive carrier) |
| 2,50 % | CuSO_4 |
| 2,10 % | FeSO_4 , Fe_2CO_3 |
| 0,25 % | active chlorine |
| 0,05 % | perica oil |

In a 32 % solution pH 2.51 [Mehltling et al. 1996] and in a 0.5 % solution pH 3.45. After making a very detailed protocol and trial report, the final conclusion describes Stalosan F as a 'most valuable hygiene product'.

HANDLING THE DISINFECTANT MATERIALS

Samples taken before and after treatment with lime and Stalosan F.
Location: Farm 1 - dunging area, pen 12, finishing house. See Fig 3.

Method: Underneath the slats in this slatted floor, in the dunging area, at the edges at each end, slats rest upon. In this area deposits build up with dung and A.suum eggs. These cavities measured 10 cm x 2 cm x 4 cm. Before treatment the upper half of each of 20 cavities was removed and kept in sealed and numbered plastic containers. Stalosan F was placed into cavity no. 7-13 [no. 1-6 control] for 24 hours as recommended by the producer. Slaked lime in no. 14-20 the next day. 150 ml of cold tap water [13.5 °C] was added and stirred into each cavity - the temperature being recorded. After 30 minutes all 7 cavities were completed. Then the 20 post samples were taken.

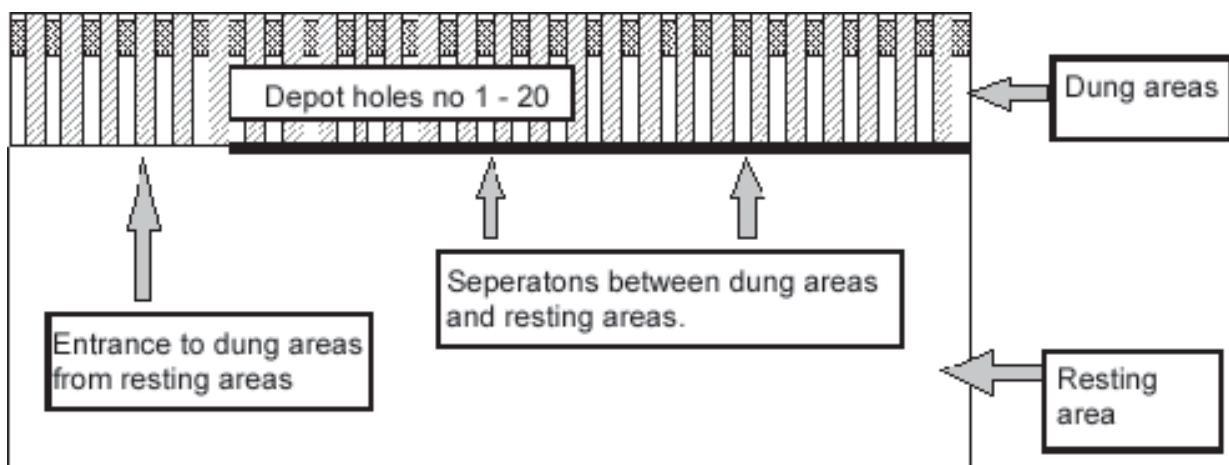
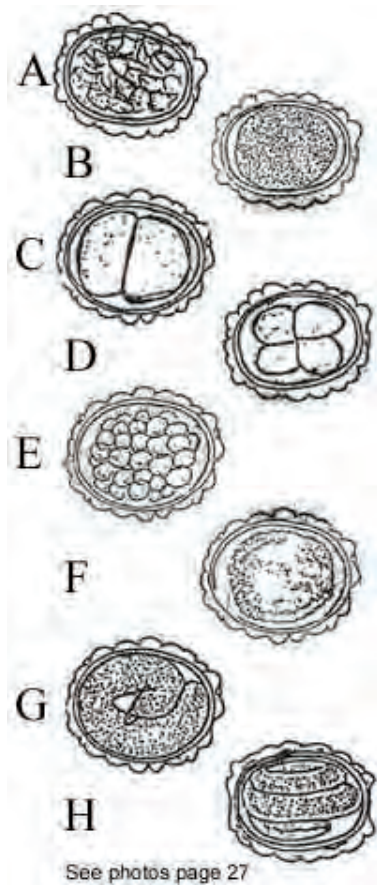


Fig.3 Dunging area in finishing pen. Farm 1.Exact location of samples taken (small rectangles at the top)

MORPHOLOGY OF Ascaris EGG

In all floor samples the stage of development and morphology of Ascaris eggs was evaluated. These following diagrams produced after Roepstorff [1996].

Diagrams of Ascaris eggs classification



- A The newly excreted or resting egg. Shell is totally filled with a heterogeneous mass.
- AB Egg contents have started to contract within the shell.
- B Egg centre is one homogeneous global cell, not completely filling the shell
- BC The last stage has begun to divide internally
- C Egg centre clearly divided into two cells joined together
- CD Start to divide again into 4
- D Clearly 4 cells
- DE Four cells have begun to divide further
- E 8 or more cells visible with distinct borders
- EF Border lines are no longer distinct
- F Centre of egg is an uneven mass without visible cell borders
- FG Cell mass is undergoing change perhaps visible larvae ends evolving

- G Short thick larva almost filling shell
- GH Little longer larva lying in a loose spiral
- H Infectious larva long, slim in tight spiral

- S Normal eggs with black spots clearly defined
- 0 Degenerated egg - sometimes cracked shell
- TOM Eggshell [empty]

Furthermore I decided to work with the following simplified groups:

- | | | | |
|-----------|----------------------------|-----------|---|
| A | not started development | G-H | almost/completely developed/ infective |
| AB- FG | developing | S- TOM | presumed dead |

RESULTS

FARM 1

On 24/6/98 rectal faeces samples for all 12 sows [lactating]. All sows had 0 EPG [eggs per gram]. There is reason to believe that all the sows on Farm 1 had been continually infected throughout their life.

Table 1 - location of A.suum eggs. Farm 1.
Floor samples from farrowing pens 24/6/98.
All samples 5 gm.

| Pen:sample | Exact location of sample (see also fig.1) physical appearance of sample. | E.P.G. |
|------------|---|--------|
| 1:1 | Corner (feeder trough/wall); composted (food, faeces, straw); slightly moist | 0 |
| 1:2 | Corner (resting area/dunging area/wall) slightly composted (food, faeces, straw), wet | 0 |
| 1:3 | Corner (dunging area/wall); (faeces, straw) dry | 0 |
| 1:4 | Corner (dunging area/resting area); faeces, straw), wet | 0,2 |
| 1:5 | Piglets resting area (beneath a hanging bale of straw); (dust, straw), dry | 0 |
| 1:6 | Corner (feeder trough(neighbour pen divider) piglets resting area; composted (straw, food, maggots); wet | 0 |
| 1:7 | Beneath watersupply dispenser in sow's area (straw, food); rotten | 0 |
| 1:8 | Center of piglets resting area; (straw, dust, faeces in "flakes"), dry | 0 |
| 2:1 | Corner (feeder trough/neighbour pen divider) composted (faeces, straw, food) moist (like fertile soil) | 0 |
| 2:2 | Corner (resting area/dunging area); (faeces, straw, crumbling wood, urine smelling); moist | 0 |
| 2:3 | Corner (dunging area/wall/neighbour pen divider) (faeces, straw); moist | 0,6 |
| 2:4 | Corner (dunging area(resting area/neighbour pen divider); (faeces, crumbling wood, maggots); wet | 0 |
| 2:5 | Piglets resting area (beneath a hanging bale of straw); (dust, straw) dry | 0 |
| 2:6 | Corner, piglets resting area (feed trough/neighbour pen divider); (straw, food, maggots); composted, slightly moist | 0 |
| 2:7 | Beneath watersupply (dispenser) in sow's area (straw, food); rotten | 0,6 |
| 2:8 | Center of piglets resting area (straw, dust, faeces in "flakes"); dry | 0 |
| 3:1 | Corner (feeder trough/neighbour pen divider); composted (faeces, straw, food) slightly moist | 0 |
| 3:2 | Corner (resting area/dunging area); (faeces, straw, dust); dry | 0 |
| 3:3 | Corner (dunging area/wall/neighbour pen divider) (faeces, straw, urine smelling), slightly moist | 0 |
| 3:4 | Corner (dunging area/resting area/neighbour pen divider); (faeces, crumbling wood); moist | 1,8 |
| 3:5 | Piglets resting area (beneath a hanging bale of straw); (dust, straw); dry | 0 |
| 3:6 | Corner, piglets resting area (feeder trough/neighbour pen divider); (faeces, straw, food, dust); slightly composted, slightly moist | 0,2 |
| 3:7 | Beneath watersupply dispenser in sow's area (straw, food); rotten, wet | 3,0 |
| 3:8 | Center of piglets resting area; (faeces, dust); dry | 0 |

From [table 1](#) one can see there was an excretion of Ascaris-eggs in the farrowing pens, but the amount of eggs is very low. This can be due to a low amount in the floor-material, but it can also be caused by taking the samples just beside the actual position of the eggs.

Samples taken from locations 1,2,5 and 8 [see [fig. 1](#)] in farrowing were negative. It was noted that no sows in the farrowing house excreted eggs on that day. Therefore, when sampling in the finishing house, the sampling technique focused on the most likely areas for infection. In [table 1](#) this rather simplistic table details are fully described: In all the following tables a uniformity will be used which raises the level of comprehension. When using the Aug 98 finishing pen results - the most descriptive results were selected [due to imbalance in numbers] . In all other tables all results are recorded.

Table 2

Egg stages expressed in percentage terms as defined in [fig. 4](#). Farrowing house, floor samples, farm 1 [see also [table 1 June 98](#)]

| | Total number found | % of total number |
|-----------------------|--------------------|-------------------|
| Eggstadium A | 24 | 80 |
| Eggstadium B | 3 | 10 |
| Eggstadium D | 1 | 3 |
| Eggstadium F | 1 | 3 |
| Eggstadium G | 1 | 3 |
| Total observed | 30 | 99 |

From [table 2](#) it can be seen that a large majority of eggs in Farm 1 had not developed

In the finishing house similar samples were taken. The pigs were 12-16 weeks old in Aug 98. The results from Aug 98 can be seen in [table 3](#). Standard deviations are very large due to the wide variation in the sample results. The results point out the status of the house i.e. Ascaris eggs are widely and consistently found. I therefore chose not to exclude the extreme results in order to reflect the true situation.

Table 3. Average and distribution in percentage in groups of egg stadium. Floor samples from finishing house farm 1 were taken 12&13/8/98. The exact position of sample location can be seen in [figure 2](#). Definition of egg stadia see [figure 4](#). Values express average EPG [eggs per gramme]. Percentages are estimated on the basis of the total of the individual samples. See as well [tables 4; 7 & 8](#)

| Sample location number | 1 | 2 | 3 | 4 | Standard deviation | | | |
|------------------------|------|-------|-------|------|--------------------|--------|--------|--------|
| | | | | | Loc. 1 | Loc. 2 | Loc. 3 | Loc. 4 |
| Number of samples | 12 | 12 | 12 | 12 | Loc. 1 | Loc. 2 | Loc. 3 | Loc. 4 |
| Average in total | 35,1 | 162,1 | 555,9 | 26,3 | 54,2 | 224,8 | 1290,7 | 22,5 |
| Group A | 2,7 | 11,6 | 23,4 | 3,4 | 3,7 | 17,1 | 36,2 | 2,5 |
| % of total | 7,6 | 7,2 | 4,2 | 13,0 | | | | |
| AB-FG | 25,1 | 120,0 | 384,0 | 16,1 | 41,9 | 165,4 | 889,8 | 17,3 |
| % of total | 71,6 | 74,1 | 69,1 | 61,2 | | | | |
| G-H | 7,0 | 27,5 | 144,4 | 4,8 | 10,4 | 41,3 | 371,5 | 6,3 |
| % of total | 19,9 | 16,9 | 26,0 | 18,2 | | | | |
| S-Empty | 0,4 | 3,0 | 4,2 | 2,0 | 0,7 | 4,3 | 7,3 | 6,3 |
| % of total | 1,2 | 1,8 | 0,8 | 7,6 | | | | |

(Average in total is 194.9 E.P.G.)

Table 3 shows that *Ascaris* eggs in location 3 [$555.9 \pm 1\ 290.7$ EPG. Minimum 13.6 EPG, maximum 4 756 EPG] which is from the corner between the dunging area, wooden divider and the neighbouring pen. The majority of eggs [69.1 %] were under development [group AB-FG] toward infective stadium. 26 % of the eggs had reached infective stadium [group G-H]. Only 4.2 % were still in group A [newly excreted or resting]. This may be due to excretion of the eggs within a few days or inhibition perhaps because of temperatures below 15 °C or relative humidity below 78 % [Nilsson 1987].

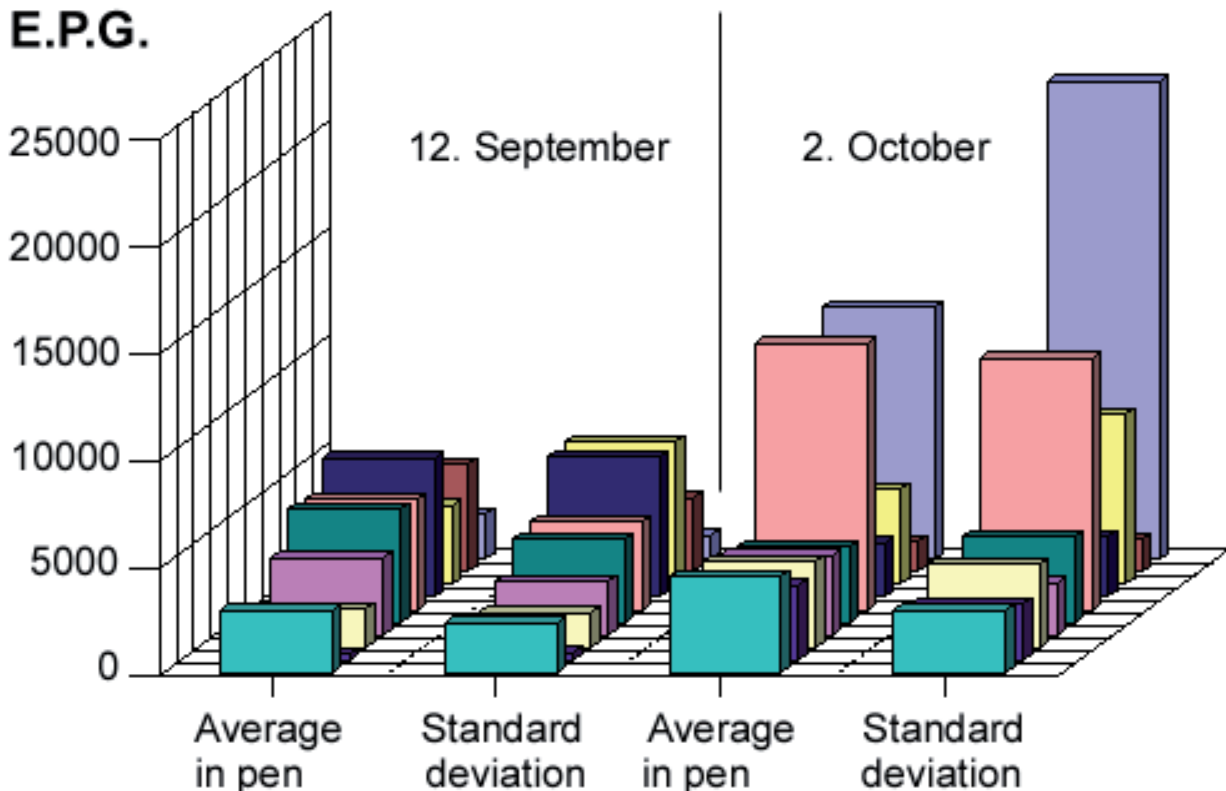
Unfortunately it was not possible to measure these quantities on the day of sampling but, in any case, the temperature was certainly well above 15 °C. Relative humidity in the sample material was evaluated at the time of sampling as 'moist' in 7 out of 12 and 'slightly moist' in 5 of 12 samples from location 3.

Table 4. Absolute and distribution percentages in groups of egg development [EPGs of all pens summed up] Finishing house, farm 1; Aug 1998. Samples taken from all 12 pens, 4 samples from each. Definition of egg stadia and groups: see figure 4 and also tables 3;7&8.

| | Eggs found | % of eggs found |
|---------------|----------------|-----------------|
| Group A | 465,1 | 5,0 |
| Group AB-FG | 6.566,3 | 70,2 |
| Group G-H | 2.212,7 | 23,7 |
| Group S-Empty | 108,9 | 1,2 |
| TOTAL | 9.353,0 | 100,0 |

One can see a large majority of developing eggs [AB-FG] in the finishing house taken as a whole. The average is 194.9 EPG.

Figure 5. Diagram showing egg excretion in rectal samples. Columns 1 & 3 represent the average EPG in the finishing house pens on farm 1. Columns 2 & 4 represent the matching standard deviations. Pen no. 2 is at the back. In front of it are pens 3, 4, 5, 6, 7, 8, 9, 10 & 11. Foremost is pen 12.



In figure 5 one can see that the average egg excretion in most pens was slightly larger in the second sample sequence. Pens 2 & 6 had much bigger excretion levels [11 716 and 12 387 EPG on 2/10 as against 2 120 and 5 137 EPG on 12/9]. The average for all pens on 12/9 was 3 569 EPG [minimum 20 EPG and maximum 19 040 EPG]. The average on 2/10 was 4 827 EPG for all pens [minimum 60 EPG and maximum 35 760 EPG.] Rise in the average is therefore $[4 827 - 3 569 / 3 569]$ i.e. 35 %.

The most likely explanation is that the pigs entered the finishing house in the first week of August and the shortest time from infection to excretion [prepatens time] is 6-8 weeks [Monrad & Nansen 1994]. The eggs excreted on 12/9 can only come from infective eggs ingested in the farrowing house.

The second rectal sample sequence was taken approximately 8 weeks after removal of the pigs to the finishing house. These samples contain *A.suum* eggs ingested both in the farrowing house and the finishing house.

Table 5 EPG before and after treatment of the cavities as well as the reduction in EPG as a percentage. Please notice sign of reduction! For each group of treatment average, standard deviation and parameters for determining if results are Gauss-distributed (Bell-distribution). All 20 samples were taken in the dunging area in pen no. 12, fam 1. See fig. 3 for exact locations of samples taken. Samples before treatment were taken 17. Nov. and after treatment 18. Nov. 1998.

| | Sample number | Before | After | Before-After | Reduction in % | Calculations |
|--------------------|---------------|--------|-------|--------------|----------------|-------------------------------------|
| Control | 1 | 441,1 | 280,4 | 160,7 | 36,4 | Average (before-after)= 1,6 |
| | 2 | 62,3 | 74,8 | -12,5 | -20,1 | Standard deviation(before)=144,9 |
| | 3 | 158,4 | 71,3 | 87,1 | 55,0 | Standard deviation(after)=94,5 |
| | 4 | 101,4 | 72,0 | 29,4 | 29,0 | KS-distance(before;after)=0,33;0,31 |
| | 5 | 126,3 | 227,6 | -101,4 | -80,3 | P(before) > 0,10 P(after) > 0,10 |
| | 6 | 51,2 | 212,7 | -161,5 | -315,4 | Gauss' distributed (5%-level) YES |
| Slaked lime | 7 | 169,5 | 109,9 | 59,6 | 35,2 | Average (before-after)= 93,4 |
| | 8 | 177,4 | 91,4 | 86,0 | 48,5 | Standard deviation(before)=78,2 |
| | 9 | 119,1 | 73,9 | 45,3 | 38,0 | Standard deviation(after)=40,0 |
| | 10 | 65,2 | 44,6 | 20,6 | 37,3 | |
| | 11 | 228,0 | 48,1 | 179,9 | 78,9 | KS-distance(before;after)=0,18;0,13 |
| | 12 | 240,4 | 13,3 | 227,1 | 94,5 | P(before) > 0,10 P(after) > 0,10 |
| | 13 | 35,1 | 0,0 | 35,1 | 100,0 | Gauss' distributed (5%-level) YES |
| Stalosan | 14 | 98,5 | 21,1 | 77,4 | 78,6 | Average (before-after)=109,6 |
| | 15 | 63,3 | 33,2 | 30,1 | 47,6 | Standard deviation(before)=84,9 |
| | 16 | 25,7 | 4,6 | 21,1 | 82,1 | Standard deviation(after)=9,5 |
| | 17 | 116,3 | 8,8 | 107,5 | 92,4 | |
| | 18 | 280,4 | 10,3 | 270,1 | 96,3 | KS-distance(before;after)=0,26;0,14 |
| | 19 | 189,5 | 17,9 | 171,7 | 90,6 | P(before) > 0,10 P(after) > 0,10 |
| | 20 | 103,4 | 14,0 | 89,4 | 86,5 | Gauss' distributed (5%-level)YES |

Relative humidity in all samples was 81.5-85.0 %. The temperature range was 13.5-15.3°C in the samples taken before treatment and 71.9 ± 10.5 °C after treatment with slaked lime. Temperature did not change after treatment with Stalosan F.

In table 5 one can see equal numbers of Ascaris eggs in the cavities for control both before and after treatment. In other words it made no difference that the samples were taken profoundly or superficially. After lime treatment the situation was different: egg numbers were reduced by, on average, 93.4 EPG. The difference was even more marked after Stalosan F treatment: 1 09.6 EPG on average.

Control [before treatment], control [after treatment], lime [before], lime [after], Stalosan F [before], Stalosan F [after], control [reduction], lime [reduction] and Stalosan F [reduction] were all shown to have Gauss's distributed EPGs. Another result worth noting is the extreme morphological changes to the Ascaris eggs after treatment. More about this later.

To test whether the differences between the 3 treatments [control, slaked lime and Stalosan F] were significant, a paired two-tailed T test was carried out. The result was that there was *no significant difference between control [before] and control [after]*. P = 0.995 t = 0.0006, df=5 95% confidence interval = {-12.46 to 125.2} r = 0.6
 There was *significant differences between slaked lime [before] and slaked lime [after]*. P=0.0205 t = 3.122 df=6 95% confidence interval = {20.2 to 166.5} r = 0.2. There was also a *significant difference between Stalosan F [before] and Stalosan F [after]*. P=0.0156 t = 3.340 df=6 95% confidence interval {29.3 to 189.9} r = -0.1

Table 6. ANOVA parameters to test differences between treatments [control, slaked lime and Stalosan F] before and after in farm 1, finishing house, pen 12, dunging area;Nov1998.

| ANOVA (before) | SS | df | MS | | Signifikans ? |
|---|----------------------|-----------|-----------|-------------------------|---------------|
| Treatments (between columns) | 3487 | 2 | 1.744 | | |
| Residual (in columns) | 184.800 | 17 | 10.870 | | |
| Total | 188.300 | 19 | | | |
| Tukey's multiple comparison test | | | | | |
| <i>Before treatment</i> | Differens in average | q | P-value | 95% confidence interval | |
| Control versus slaked lime | 8,963 | 0,2185 | P>0,05 | 139,9 to 157,8 | NO |
| Control versus Stalosan F | 31,46 | 0,7669 | P>0,05 | -117,4 to 180,3 | NO |
| Slaked lime versus Stalosan F | 22,50 | 0,5709 | P>0,05 | -120,5 to 165,5 | NO |
| ANOVA (after) | SS | df | MS | | |
| Treatments (between columns) | 67.150 | 2 | 33.580 | | |
| Residual (in columns) | 54.800 | 17 | 3.223 | | |
| Total | 121.900 | 19 | | | |
| <i>After treatment</i> | Differens in average | q | P-value | 95% confidence interval | |
| Controle versus slaked lime | 102,0 | 4,568 | P<0,05 | 21,00 to 183,1 | YES |
| Control versus Stalosan F | 140,8 | 6,303 | P<0,05 | 59,74 to 221,8 | YES |
| Slaked lime versus Stalosan F | 38,74 | 1,805 | P>0,05 | -39,11 to 116,6 | NO |

From table 6 one can see that there is no significant difference between samples taken before treatment. After treatment there is still no significant difference between slaked lime [after] versus Stalosan F [after] but there is a *significant difference* between control [after] v slaked lime [after] and control [after] v Stalosan F [after].

Table 7. Absolute counts and distribution in percentages differentiating Ascaris eggs by groups related to their development status before and after treatment [control, slaked lime and Stalosan F all together.] Finishing house, pen 12, farm 1; November 1998 [see also tables 3; 4 & 8]

| | Before treatment | | After treatment | |
|---------------|-------------------------|-----------------|------------------------|-----------------|
| | Egg amount | % of egg amount | Egg amount | % of egg amount |
| Group A | 66,8 | 2,3 | 45,9 | 3,2 |
| Group AB-FG | 1.399,0 | 48,5 | 652,3 | 45,6 |
| Group G-H | 381,5 | 13,2 | 129,0 | 9,0 |
| Group S-empty | 1.034,9 | 35,9 | 602,7 | 42,1 |
| TOTAL | 2.882,2 | 99,9 | 1.429,9 | 99,9 |

After treatment, approximately 50% less eggs were found than before treatment. In group A and group S-empty [presumed dead eggs] a few more eggs were found after treatment than before. The average number of eggs before treatment was 144.1 EPG and after treatment 71.5 EPG.

Table 8. Relativ distribution of the Ascaris eggs in development groups related to treatments. Farm 1, finishing house, pen 12, Nov. 1998. (see also table 3; 4 & 7)

| | CONTROLE % of number | | SLAKED LIME % of number | | STALOSAN F % of number | |
|---------------|--------------------------------|--------------|-----------------------------------|--------------|----------------------------------|--------------|
| | Before | After | Before | After | Before | After |
| Group A | 1,3 | 3,6 | 2,0 | 3,3 | 3,3 | 0,0 |
| Group AB-FG | 39,3 | 61,5 | 42,8 | 10,4 | 69,7 | 31,8 |
| Group G-H | 16,0 | 12,2 | 9,1 | 0,8 | 13,0 | 10,6 |
| Group S-empty | 43,4 | 22,7 | 46,2 | 85,5 | 14,1 | 57,6 |
| TOTAL | 100,0 | 100,0 | 100,1 | 100,0 | 100,1 | 100,0 |

In *table 8* one can see relatively more eggs in group S-empty [presumed dead eggs] after treatment, both with slaked lime and with Stalosan F. This indicates that both slaked lime and Stalosan F have a destructive effect on Ascaris eggs. By way of contrast, the percentage of group S-empty in the control samples [after] almost halved. The difference factor between control and slaked lime is $[43.4 / 22.7 : 46.2 / 85.5 =]$ 3.5 and between control and Stalosan F the difference factor is $[43.4 / 22.7 : 14.1 / 57.6 =]$ 7.8

Table 9. Percentage reduction in Ascaris eggs related to treatment. Farm 1, finishing house, pen 12, November 1998

| EPG per sample | CONTROLE | | SLAKED LIME | | STALOSAN F | |
|--------------------------|-----------------|------------|--------------------|-------------|-------------------|-------------|
| | Before | After | Before | After | Before | After |
| Number of samples | 6 | 6 | 7 | 7 | 7 | 7 |
| Average | 156,8 | 156,5 | 147,8 | 54,4 | 125,3 | 15,7 |
| Reduction in % | | 0,2 | | 63,2 | | 87,5 |

Table 9 shows that slaked lime reduces the total number of eggs by 63.2 % and Stalosan F by 87.5 %

FARM 2

Table 10. The average EPG in each of the 6 houses. Floor samples taken in finishing houses, farm 2; 14/10/98

| House No. | Cleaning status | EPG | Positive Samples | Total number Samples |
|-----------|-----------------------------------|------|------------------|----------------------|
| 1 | Soaking | 0,4 | 1 | 2 |
| 2 | 1 week after cleaning | 1,1 | 2 | 6 |
| 3 | 6 weeks after cleaning | 0,1 | 1 | 4 |
| 4 | 7-8 weeks after cleaning | 22,0 | 4 | 4 |
| 5 | More than 16 weeks after cleaning | 10,3 | 1 | 2 |
| 6 | Daily cleaning | 2,4 | 2 | 2 |

In table 10 it can be seen that there were more eggs per gramme from the floor samples in houses 4 and 5 than in the other houses. House 6 should be noted because of the relatively low EPG results combined with a concrete floor and daily cleaning. The reason for the low number of samples taken from farm 2 is pressure of business from the farm owner combined with owners' desire to participate in and supervise the sampling process.

Table 11. Percentage of development groups of Ascaris eggs. Farm 2, finishing pens [all together] 14/10/98. See definition of development groups in figure 4.

| | Amount of eggs | % of amount |
|---------------|----------------|--------------|
| Group A | 3,4 | 2,8 |
| Group AB-FG | 79,9 | 65,8 |
| Group G-H | 30,7 | 25,3 |
| Group S-empty | 7,4 | 6,1 |
| TOTAL | 121,4 | 100,0 |

It can be seen from table 11 that the majority of Ascaris eggs are in the developing stadia [group AB-FG] and, after that, most eggs are in group G-H [quite or almost infected eggs]. This is exactly the same result as in farm 1 [see tables 3; 4; 7 & 8].

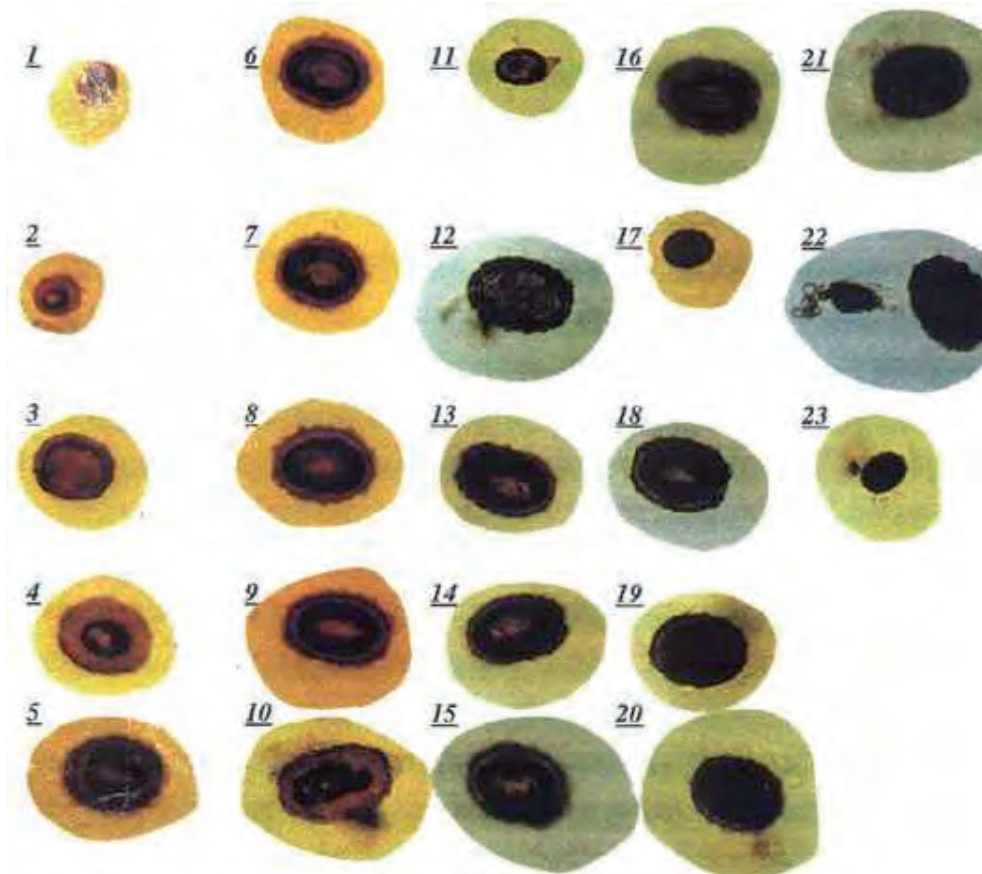
Table 12. Average of EPG and distribution in developing groups in finishing houses, farm 2; 14/10/98. {Values are average EPG in each house individually.} See definition of developing groups in figure 4.

| Cleaning status | No. | Total | Group A | | Group AB-FG | | Group G-H | | Group S-empty | |
|-----------------------------------|-----|-------|----------|----|-------------|----|-----------|----|---------------|-----|
| | | | Abs. EPG | % | Abs. EPG | % | Abs. EPG | % | Abs. EPG | % |
| Soaking | 2 | 0,4 | 0,2 | 50 | 0,2 | 50 | 0 | 0 | 0 | 0 |
| 1 week after cleaning | 6 | 0,8 | 0 | 0 | 0,7 | 88 | 0 | 0 | 0,1 | 13 |
| 6 weeks after cleaning | 4 | 0,1 | 0 | 0 | 0 | 0 | 0 | 0 | 0,1 | 100 |
| 7-8 weeks after cleaning | 4 | 22,0 | 0,7 | 3 | 13,7 | 62 | 6,7 | 30 | 0,9 | 4 |
| More than 16 weeks after cleaning | 2 | 10,3 | 0 | 0 | 8,2 | 80 | 1,4 | 14 | 0,7 | 7 |
| Daily cleaning | 2 | 2,5 | 0,2 | 8 | 1,2 | 48 | 0,6 | 24 | 0,5 | 20 |

Table 12 shows that, despite the very small number of samples and the very small number of eggs in each sample, there was a clear tendency toward an increasing percentage of more developed Ascaris eggs with increasing time between cleaning and sampling. It may appear serious but the numbers represent 2 black eggs found in 4 samples all together, in other words only 2 eggs were found in house 3.

BLACK Ascaris EGGS

In the following microphotos damaged Ascaris eggs are shown. Black Ascaris eggs were found on both farms. Magnification is 100x and 200x



These 23 photographs show Ascaris eggs initially with small alterations [photo 1] but eventually with drastic alterations [photos 21;22 & 23] making the egg interior totally black. No visible difference was observed regardless of whether the samples had been treated with slaked lime or Stalosan F. Some black Ascaris eggs were also found in samples taken before any treatment. These samples did contain fewer black eggs.

MILKSPOTS IN THE LIVERS

Unfortunately only partial results were obtained for the finishers on farm 1. At the last moment [when I telephoned the slaughterhouse for the results] 8 finishers from farm 1 were dissected and 2-10 milkspots were found in each of their livers. Farm 2 did not wish to participate in this aspect of the investigation.

DISCUSSION

The negative results from the lactating sow rectal samples in Farm 1 harmonise with investigations done by Urban et al. [1984] who found that natural infection of *A.suum* induced > 99 % resistance to new infection. At the same time it must be remembered that the sows on this farm are treated with anthelmintics one month before farrowing, in this case, early April 98. Nilsson [1982] describes that, from 72 lactating sows, none of them was excreting *A.suum* eggs. Regular faecal samples show that excretion from sows of *A.suum* is of short duration. His comment is that the life span of *A.suum* can be short in adult pigs or that the worms' fertile period is extremely short. After this he notes that many scientists have observed that the natural immune system of the adult pig appears to suppress larval migration and the fertility of the adult worm.

Roepstorff et al. [1997] write that there is no indication that different levels of infection relate to the number of established patent worms. On the contrary, in 1991 Roepstorff found that 46 % of sows on Farm 1 were excreting *A.suum* eggs. In total 8 farms were investigated at that time and between 0-46 % of sows were found to be excreting eggs over 2 breeding cycles including lactation. In that investigation, no anthelmintics were used. In the present investigation the sows were treated one month before farrowing with Panacur® [Fenbendazole]. Rectal samples were taken 24/6/98.

An investigation by Ryundiger [1996] demonstrated that sows treated with Panacur® produced piglets 3.2 % heavier than untreated sows. Roepstorff [1997] found that after treating sows with a range of anthelmintics, a lasting effect was variable and not specific to the anthelmintics. Eriksen et al. [1992] found that both experimental and natural infection *A.suum* results in 'earned' immunity. The most significant immunity is gained by repeated ingestion of infective eggs. Immunity is effected as the parasite is eliminated on its migratory route through the body also from the gut. She also writes that this earned immunity may be giving effect in the intestinal loci before the larval migration begins. Eriksen et al. [1992] found that the naturally exposed sows had a strong immunity which showed no or just a few milk spots in their livers when slaughtered.

Muff et al. [1984] investigated the presence of *A.suum* eggs in the pig housing and in that article a sample was considered positive if just one single egg in one of the group samples [up to 25 samples in each group] was found. Unfortunately this fact is not discussed. It is reported that only 3 % of animals more than 12 months old have positive egg counts in faecal samples. This is in harmony with my own investigations [table 1]

Whether the anthelmintic treatment or the earned and retained immunity [which the small number of identified eggs from floor samples in the farrowing pens indicate] is the reason for the negative faecal samples, it cannot be hypothesised from this investigation.

As shown in **table 1** only a few eggs were found in the farrowing pen at Farm 1. Only the sample areas in corners between resting and dung areas and close to the feed troughs and water supply were positive. These samples were all damp to wet. None of the dry samples contained eggs. Unfortunately the equipment for measuring relative humidity was not available until November therefore descriptions are subjective.

Table 2 shows that in floor samples from the farrowing house [Farm 1] 80 % of *A.suum* eggs were in Group A and this differs from all other samples [see tables 3;4;7;8;11 & 12] both on Farms 1 and 2 and taken before treatment. These others only had 1.3-5.0 % in Group A. The samples taken after treatment had 0-3.6 % in Group A. This indicates that the sample from the farrowing house either mainly contained newly excreted eggs or that the development of eggs was suppressed, probably because of low relative humidity.

The same tables [3;4;7;8;11 & 12] show that between 39.3-69.9 % of the eggs were in Group AB-FG with the exception of the farrowing house where 16.3 % were in this group.

In the farrowing house 3.3 % of eggs and in all other houses [both Farms 1 & 2] 9.1-25.3% were in Group G-H [developed larva in eggs].

This indicates that, despite the limited number of samples taken, the *Ascaris* eggs were probably effectively removed by cleaning the farrowing house before the sows were introduced [in Farm 1]. It also indicates that there was a new excretion of eggs from the sows [despite anthelmintic treatment which the farmer wasn't sure he had administered].

From **table 3** one can see there is a big difference in the mean EPG between the different sample locations but the percentage of the egg groups is approximately the same size in every location. In the floor samples [finishing pen Farm 1] from August the majority most significant in location 3 which had 69 % developing Ascaris eggs [group AB-FG] and 26.0% in Group G-H. Only 4.2 % in this location was newly excreted or resting [Group A] which happens when the temperature is beyond 15°C or the relative humidity in the substance that surrounds the eggs is beyond 78 % [Nilsson 1982]. Unfortunately it was not possible to measure these details because of problems with delivery of equipment for this purpose. My subjective impression was that air temperature in this house exceeded 15 °C pretty much. This makes me believe that the samples on locations exceeded 15 °C as well. The relative humidity in 7 of 12 samples was evaluated to be 'moist' 5 of 12 samples from location 3 were evaluated to be 'slightly moist'. This goes as well for the 3 x 12 samples from the other sample locations [locations 1;2 &4].

As early as 1935 Allocate [1935] was able to show/monitor that Ascaris eggs need some time after excretion until before they can become infective. He investigated it in the following range of temperatures: 22-24 °C and 32-34 °C and concluded that the development is 16 days at 32-34 °C and 28 days at 22-24 °C.

Nilsson [1982] describes that eggs do not develop in relative humidities beyond 78.5 % and beyond 15 °C. He found that the shortest time from excretion until infectivity is 56 days in housing environment in Sweden.

In conclusion I would say that the finishing house, Farm 1, had a favourable environment for developing eggs in August 1998.

From **fig. 5** one can see in the majority of pens in finishing house, Farm 1, the mean average excretion in the second rectal [faecal] samples exceeded the first excretion. Pens 2 & 6 showed much higher EPG counts at the two samples [2/10/98] The average for all 12 pens 12/9/98 was 3 569 EPG with minimum 20 EPG and maximum 19 040 EPG. The 2/10/98 average was 4 827 EPG minimum 60 EPG and maximum 35 760 EPG. The pigs entered the finishing pens during the first week of August 1998 and when [as mentioned before] the shortest time from excretion until infective state can be reached is 6-8 weeks [Monrad and Nansen 1994]. First rectal [faecal] samples were taken 5 weeks after the pigs were put into the finishing pens. This makes me sure that the egg excretion could only come from infection sources in the farrowing house. The 35 % rise from September to October could only depend on the fact that the infection picked up in the finishing pens had become patent.

Table 4 shows 1.2 % of all eggs found in floor samples [finishing house; Farm 1; August 1998] belong to Group S-Empty which are, presumably, dead eggs. Further discussion of this point later [table 8].

Table 5. There is no difference whether the samples from the slats are taken superficially or profound in the cavities. The samples of controls had same size of E.P.G. on average before and after 'treatment' [which was to do nothing] After lime treatment E.P.G. was, on average, 93.4 EPG lower than before lime treatment of the dung in the cavities. Even more marked was the effect of Stalosan F. After Stalosan F treatment 109.6 EPG less than average was found than before the treatment. THESE RESULTS ARE ASTONISHING. I especially did not expect to find a reduction of EPG after treatment with Stalosan F which I actually chose because I had not thought it could harm Ascaris eggs. This opinion was based on a lot of published articles which described the chemicals as having no effect on Ascaris eggs.

In addition it was noted that Stalosan F contained no aggressive substances [see Stalosan F contents pages 15]. For instance an investigation by van den Burg et al. [1987] showed that none of the 11 disinfectants containing the following active chemicals: aggressive chlorine, phenol, cresol, sodium hydroxide, potassium hydroxide, quaternary ammonium compounds, glutaraldehyde and para-glutar aldehyde was found to be effective toward/ against Ascaris eggs. This investigation administered both the recommended and double the recommended doses of those disinfectants. Volkov [1993] found no effect on Ascaris eggs after treatment with ammonium-chloride and ammonium-poly-sulphide.

Juris and Breza [1988] showed that Orsanol BF12, potassium hydrochloride, sodium hydrochloride, Jodonal B and liquid Jodisol only affected Ascaris eggs slightly.

Hurley and Sommerville [1982] monitored reversible inhibition of developed Ascaris eggs hatching. Hydrogen sulphide is able to reconstruct the ability of hatching.

Of course Ascaris eggs can be killed by chemicals but very strong and concentrated poisons are needed. This is NOT ACCEPTABLE for the environment nor farmers/animals. DDT and Carbathio kill more than 60 % of A.suum eggs in soil samples treated with 3 % solutions for 24 hours according to Volkov [1973].

A better way of Ascaris control is heat treatment. Persson [1973] says that after aerobic decomposition in liquid dung, all Ascaris eggs died after 6 days at 56 °C or 8 days at 50°C. Birbaum and Eckert [1985] write that all A.suum eggs were destroyed after 10-110 minutes at 50-56°C or [if the eggs were in sludge] 30 minutes at 65-70°C and only 10 minutes at 80°C. According to Grebenkin [1982] relative humidities beyond 60 % are lethal for A.suum eggs [they died within 7 days] which makes drying out another possibility in Ascaris control. Ultrasound [50 w/cm², 24 kHz] for 10 minutes kills 98.7 % of Ascaris eggs kept in ferro-magnetic powder which is at waste after manufacturing engine tools according to Sokhrokov [1979]. Other existing methods for controlling Ascaris are for instance growing spores of fungi [mycelium] in samples with Ascaris eggs. Sobenina [1975] found reducing effect of survival of Ascaris eggs after growing *Aspergillus nidulans* some *Penicillium*-species and *Catenaria angillulae* the reduction is respectively 72 %, 61 % and 20 %. Lysec [1979] investigated *Verticillium clamydosporum* and concluded that it is a strong natural regulator towards *Ascaris lumbricoides*.

Certain plantroots [rhizosphers] also can be ovicide. Volkov [1976] found that *Tagetes*, *Calendula* and barley but not oat and wheat can destroy Ascaris eggs. Oats and wheat however inhibit the development of Ascaris eggs. Simon and Volkov [1973] described ovicide effect [50-80 % killed eggs] after growing *Pisum*, *Vicia*, *Panicum miliaceum* and *Calendula* in samples with A.suum eggs. This should be compared to DDT which kills 60 % after 24 hours [Volkov 1973].

On the other hand it has no ovicide effect when earth worms eat Ascaris eggs [Smirnov 1975] [Schumakovitch 1976] or when adult flies [*Musca domestica*] or larvae from these eat Ascaris eggs [Dipecoln, 1982 & Koltypin, et al. 1975].

In short it was ASTONISHING to find this large ovicide effect from lime and especially from Stalosan F.

Table 7. After treatment was found approximately half as many eggs as before treatment when all treatments [controls, lime, Stalosan F] are put together. This goes as well in total and for the four groups [A, AB-FG, G-H and S-Empty].

Table 8 is itemised account, specifying this, and shows that the proportions between group S-Empty is markedly different after the treatments [controls, lime and Stalosan F] in control samples the reduction of presumed dead eggs [S-Empty] goes from 43.4 % before treatment to 22.7 % after treatment . In lime samples this proportion is 46.2 % before treatment and 85.5 % after treatment. In Stalosan F samples before treatment 14.1 % presumed dead eggs were found and 57.6 % after treatment. Comparing these findings one finds a difference from control [after] to lime [after] on factor 3.5. The difference from control [after] to Stalosan F [after] is found to be factor 7.8. It must be pointed out that one half of the presumed dead eggs in the control samples [after] can be due to the deeper and more protected [more profound] location in the cavities. This goes naturally also for the treatment with lime and Stalosan F. Therefore the destructive effect on A.suum eggs of these treatments is even stronger. The reason for the rather big proportion of presumed dead eggs before the treatment is probably found in the fact that the environment influences the Ascaris eggs over time. Table 4 showed 1.2 % presumed dead eggs. The rise of presumed dead eggs has risen markedly.

In autumn temperatures fall to suboptimal values [A.suum eggs: temperature optimum 30-33 °C, Seamster 1950. Arene [1986] monitored egg development between 16 ± 1 °C and 31 ± 1 °C and concluded that temperature optimum to be 22 ± 1 °C for development of infective larvae.

As to whether or not suboptimal temperature can damage the Ascaris eggs, I did not find support for anywhere but keeping A.suum eggs in a refrigerator did not seem to change the morphology of A.suum eggs [personal observation].

Before lime and Stalosan F treatments the temperature in the present investigation was measured to 13.5-15.3 °C. Urine from the pigs can damage the eggs [Nilsson 1982]. Despite the slat floor, the dung area the substances deposited/collected in the cavities [see fig.3] can absorb humidity from air and from pig urine. Relative humidity before treatments was 81.5-85.0 %. Unfortunately I do not know how much of this was urine.

The morphological changes of the A.suum eggs also could be due to drying out. Relative humidity below 60 % for more than 7 days is, according to Grebenkin [1982], lethal to Ascaris eggs.

After treatment with lime, especially eggs from group G-H were damaged [9.1 to 0.8 %] also the proportion of eggs in development [group AB-FG] was strongly reduced [42.8 % to 10.4 %]. The number of eggs from group A [no development started yet] on the other hand rose from 2.0 % to 3.3 %. A predictable consequence of at least the lime treatment which produced a lot of heat in the samples themselves [the maximal temperature in these samples were, on average, 71.9 ± 10.5 °C] was changes in the centre of the eggs. According to Wharton [1980] temperatures beyond 30 °C cause a loss of water. This loss follows the rising temperature in an exponential way. At the same time a graduated melting or transition of the complexes of the components which form the lipid layer of the A.suum shell. Wharton [1979] concluded that the permeability barrier of the eggs to be totally destroyed at 63-65 °C.

The reason for the 14.5 % morphologically undamaged eggs after the temperatures achieved during the lime treatment is not known. Unequal heat distribution is one possible reason. Another could be not all damage could be seen for sure in a microscope [magnification 100 x].

After Stalosan F treatment the situation was different; all the eggs in group A [not yet developing] were gone [27 observed in group A before Stalosan F treatment. Total observed eggs before treatment 691]. The proportion of group AB-FG was reduced from 69.7 % to 31.8 %. The number of almost / quite infective eggs [group G-H] was only reduced from 13.0 % to 10.6 %. This indicates Stalosan F can particularly effect eggs in the early stages of development. No explanation was found for the effect of Stalosan F.

The qualitative changes after these treatments [lime and Stalosan F] seem to be equal. No references on this issue were found. Further investigations must be undertaken to evaluate if lime and Stalosan F treatments one after the other could produce better disinfection.

Egg survival changes after lime and Stalosan F treatments respectively and investigating if the black eggs are dead eggs seem to be interesting questions.

Table 9 describes absolute egg counts estimates from EPG in the group controls, lime and Stalosan F treatment. Lime treatment reduced the absolute egg count by 63.2 % and Stalosan F treatment by 87.5 %. These results are surprising especially when one considers that these treatments were disinfections directly in organic substances [partly composted faeces].

Table 10 is monitoring EPG according to cleaning status. As expected, the egg counts are lower in houses 1; 2 and 3 which are cleaned on the day of sampling, 1 week and 6 weeks before respectively, that is 0.4 EPG; 1.1 EPG and 0.1 EPG. This agrees with *A.suum* eggs taking on average 6-8 weeks to become patent [Monrad & Nansen 1994]. In houses 4 and 5 [cleaned 7-8 weeks and more than 16 weeks before sampling] 22.0 EPG and 10.3 EPG were found on average.

It was surprising to find only 2.4 EPG on average in the 'old fashioned' house. This indicates effective cleaning. By comparison with farm 1, all the EPG's were low in farm 2. The average in farm 1 was 125.3 - 194.9 EPG [table 6: Nov 1998 before treatment and table 3: Aug 1998] in the finishing house. The highest egg count in farm 2 was 22.0 EPG. This difference may be due to differences in management.

In the farrowing house in farm 1 [concrete floor like the old fashioned house in farm 2] the highest egg count found was 3.0 EPG but, as mentioned in the discussion of rectal [faecal] samples of lactating sows, adult swine only excrete a few eggs because of the earned and retained immunity.

Taken together, the results in table 10 indicate careful and effective cleaning in farm 2. Anthelmintic treatment of bought pigs before transportation to farm 2 could reduce the problems with milkspots in the livers of the finishers [which was the criterion for choosing farm 2].

Table 11. One can see that a majority of eggs are under development [group AB-FG] in farm 2. Despite extensive efforts to limit *A.suum* [among other microbes] there is a favourable environment which makes it possible for the small number of *A.suum* eggs left to develop to the infective state [group G-H].

Table 12 shows a fast development after end of prepatent periode [6-8 weeks after ingestion of infective *A.suum* eggs; Monrad & Nansen 1994]. It should be noted that October temperatures can be low and therefore inhibit egg development. Unfortunately no temperature measurements were available but the impression gained was of 18-20 °C in all houses in farm 2.

In house 2, 1 week after cleaning, 88 % of eggs found belonged to group AB-FG [developing] but none in group G-H [infective]. In houses 4 and 5 [7-8 weeks and more than 16 weeks after cleaning] 30 % and 14 % respectively in group G-H were found. In house 6 [daily cleaning] the distribution of *A.suum* eggs corresponds to the distribution in farm 1 [see tables 2; 3; 4; 7 & 8 - before treatment]. Apparently the thoroughness of cleaning is different as between farms 1 and 2 accounting for the difference in EPG.

Black *A.suum* eggs were found both in farms 1 and 2. The black eggs appear damaged but in several instances the inside structure of black eggs could be seen clearly. For example microphoto no. 16 shows an egg containing a larva almost developed to infective stadium [G-H]. I wonder whether the blackening could be due to previous cleanings using a high pressure hose and hot water or to the use of certain disinfectants but could find no answer. Neither extensive research in written material no enquiry of scientists and laboratory technicians revealed any information about the black *A.suum* eggs. In fact no-one at C.E.P. had ever observed similar black eggs. [A colour photo can be ordered from Stormollen A/S, Ringsbjergvej 16, 4682 Tureby, Denmark.]

Unfortunately only partial results were obtained concerning milk spots in the liver of finishers on farm 1. The slaughterhouse found 2-10 milkspots in livers from 8 finishers. It can be presumed that pigs on farm 1 had been exposed to *Ascaris* infection throughout their lives. This would explain the small number of milkspots in their livers.

Eriksen et al. [1992] concluded that the highest number of milk spots were found in growers (average 86 in 1 liver) decreasing to 71 in finishers [due to earned immunity] and the lowest number in adult sows/boars (average 3 in 1 liver). This also applies to naturally exposed animals which in an experiment were exposed to controlled *A.suum* infection.

The finishers in this present investigation were not exposed to experimental controlled *A.suum* infection. The

earned and retained immunity theory [supported by the finding of infective eggs in the farrowing pens as well as in the finishing pens] must be considered the explanation for the low number of milkspots in the livers of the finishers from farm 1.

Milkspots [interstitial hepatitis] develop in the liver 8 days after ingestion of infective *A.suum* eggs in unimmunised pigs and disappear again approximately 85 days later [Mehl et al. 1983] Eriksen et al. [1981] found milkspots in 43.6 % of 119 038 livers in finishers. Among these 43.6 %, 73.2 % were approved fit for human consumption after local cleansing. Wismer- Pedersen et al. [1990] describe problems with the quality of fried liver because of excess connective tissue in the livers after recovery from *A.suum* larva migration. At the same time they argued for better anthelmintic control and, with that, better quality livers for consumption.

Frank [1995] showed that in pigs *Ascaris* can reduce levels of nutrition, cause vomiting and anorexia, increase intake of food, reduce body weight and number of piglets born together with decreased immunity to other pathogens. Andersen [1976] found that the growth rate in finishers was 20 % lower in finishers not previously exposed to *Ascaris*. Among slightly immunised finishers the growth rate was reduced by 8 %.

How much farm 2 could gain from better anthelmintic control is not known because regular E-control reports [economic control reports] or similar information was not available. Considering the very low price of pork at present, even a slight improvement in growth rate, feed utilisation and immunity would make better anthelmintic control a good bargain.

CONCLUSION

A.suum eggs develop in the floor material mixed with condensed water and/or swine urine. The cavities within the dung area provide the/an ideal environment for egg development. Light slaked lime and Stalosan F reduce numbers very much and, significantly, *A.suum* eggs in the house. Lime especially attacks eggs in groups AB-FG and G-H [the more significant]. Stalosan F especially reduces egg numbers in group A and less significantly in group AB-GH. That is to say both light slaked lime and Stalosan F are suitable in conventional systems. From a health and safety point of view Stalosan F is preferable.

In addition it can be said that the Organic-Soil Association [LØJ] in Denmark does not accept either lime or Stalosan F in organic systems.

SUMMARY

Traditional farm [Farm 1] is known as 'a good place to find *Ascaris*' with 12 sows producing approximately 225 finishes per annum. This farm was investigated for occurrence of *A.suum* in the housing environment. The sows were treated with anthelmintics. Faecal samples were taken after 2.5 months and all 12 were negative. In the farrowing pens where the sows and piglets were housed floor samples contained a mean of 2.1 EPG [eggs per gram] mainly type group A [newly excreted].

Earned immunity seems to be present in the finishers; amongst 8 randomly selected finishers all were found to have 2-10 milk spots of the liver when slaughtered. Developing eggs were found in the floor samples which indicate immunity is sustained through repeated ingestion of infected eggs from the environment

Floor samples were collected from the floor of the finishing house mid Aug 98. Most of these eggs fell into group AB-FG [developing eggs] which indicates a favourable environment for *A.suum* [relative humidity >78 % and temperature 15 °C in the floor substances]. Not every part of the floor is suitable for egg development but especially damp areas with fissures or cavities which could harbour very small amounts of dung contained eggs at all stages of the cycle [A-H]. Dung that had fully composted [musty] and smelled like forest contained up to 4 856.4 eggs/gm of which 4 829.1 E.P.G. was recognised as living under the microscope [x 100]. The mean value in 4 locations was 26.3; 35.1; 162.1 and 555.9 EPG.

Faecal samples from the finishers 12/10 1998 and 2/11 1998 showed that the egg excretion had increased by 35 % between those dates. One may conclude that the rise in egg excretion was due to ingestion of eggs in the finishing pen completing a full cycle. This infection had become patent

After lime treatment in 7 cavities in the dung area between the slats and supports at each end the temperature rose to 71.9 ± 10.5 °C. The number of developing eggs was reduced to a level 63.2 % lower than the control.

After Stalosan F treatment in 7 other cavities the level was reduced to 87.5 % below the control.

After these two treatments, the percentage of 'black' [presumed dead] eggs was significantly increased: Lime from 46.2-85.5 % of the total EPG and Stalosan F 14.1-57.6 % of total EPG

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