

Stalosan F

- Because drying is not enough



The logo for Stalosan F features a yellow circle on the left, partially overlapping a red horizontal bar. The text "Stalosan F" is written in white on the red bar. Below the red bar is a dark green horizontal bar.

Stalosan F

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Stalosan F

1. Setting a new standard

Stalosan F is an alternative and unique way to control bacteria, viruses, fungi, parasites, fly-larvae, ammonia, moisture and litter quality in animal housing.

Regular addition of Stalosan F stabilises the microbial flora and chemical balance of the litter and thereby creates a natural and healthy animal environment during the entire course of the animal production period.

Even hard to kill parasites including coccidia oocysts and round worm eggs can be controlled by regular addition of Stalosan F.

Stalosan F can be used as a treatment of existing problems with very good results, but the real cost beneficial value comes from continuous use as a prophylaxis.

Stalosan F has proved to be effective when it comes to keeping several diseases under control including lawsonia, diarrhoea, mastitis, salmonella infections etc.

2. Animals

Stalosan F can be used for all domestic animals and pets

Pigs

Poultry

Cattle

Sheep

Goats

Horses

Furred animals

Rabbits

Camels

Dogs

Cats

.....and many more!

Stalosan F

3. Effects

Contains highly reactive metals that eliminates pathogens.

Contains metals that inhibits metabolism and proliferation of pathogens.

Contains minerals that attaches bacteria, virus, fungi, parasites and fly-larvae leading to an inactivation of their life cycle. This binding is non specific and does not distinguish between types or strains of micro-organisms and therefore provides an ubiquitous antimicrobial effect.

Contains minerals that dehydrates parasites, larvae and worms leading to an kill.

Contains Acid-buffers with a high capacity that inhibits pathogens (sensitive towards pH values below 4 and has optimal living conditions at pH values of 7 to 9).

Absorbs, neutralizes and inhibits the formation of ammonia and water.

4. Side effects

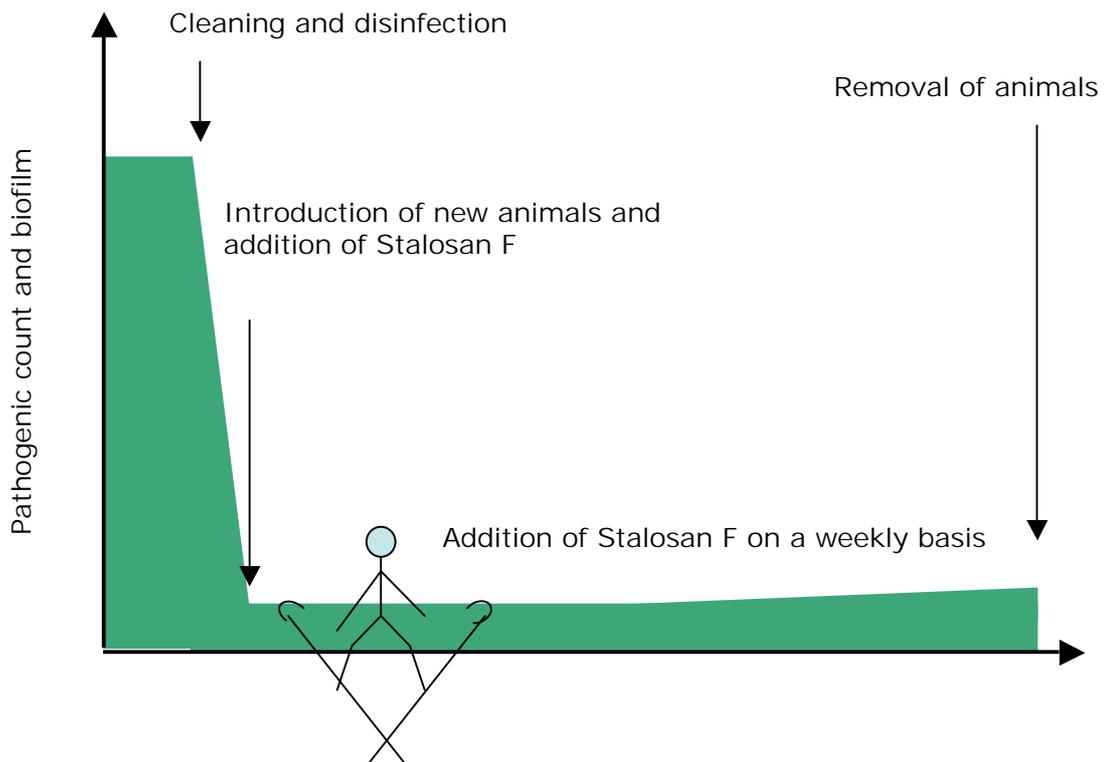
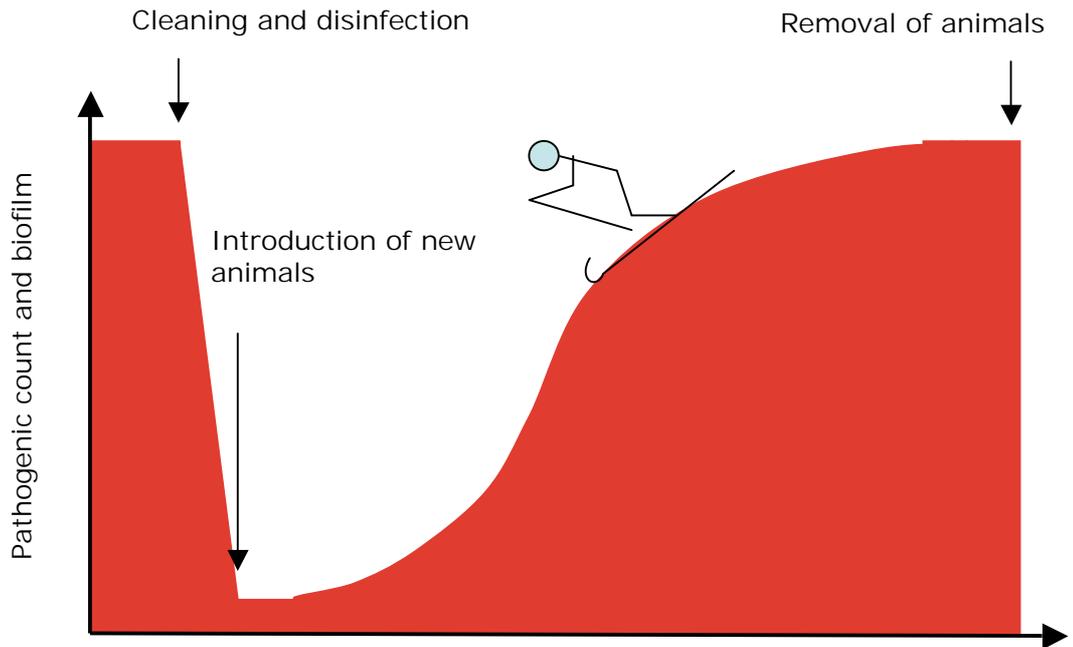
For 24 years, Stalosan has been under observation for side effects through quarterly reports from veterinarians in Denmark. Today, Stalosan has been on the market for more than 40 years and no side effects have ever been reported from the use of Stalosan throughout this period.

In this period, and subsequently, Stormøllen has not received any complaints related to Stalosan, not from Denmark nor from abroad. Therefore, the product may be characterised as being safe to use. Stalosan does not cause the development of resistant bacterial strains and can be used continuously.

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5. Prevention instead of Treatment

Figures illustrates levels of pathogens during a growth period with or without regular addition of Stalosan F





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6. Application

Basic application

In general, we recommend that Stalosan is added in rates of 50 g/m² once a day for the first three days. Hereafter, Stalosan should be used once a week with 50 g/m². In case of increased infection levels, the dosage should be increased to 2-3 applications per week. Stalosan F is spread directly on existing bedding especially along the edges where wet- and dampness occurs. Stalosan F can be added while the animals are present. For optimum effect, we recommend that Stalosan is spread as evenly as possible over the area the animals have access to.

Specific application

In case of increased risk of infection levels (*cases of stressful conditions such as new animals, before deliveries, in case of sheep and goats from six weeks before lambing, diarrhoea, inflammation of the udder, other infectious diseases, new staff, sudden changes in the animals' immediate environment (e.g. burst water pipe)*), Stalosan should be used several times during a week according to the demands. Hereafter, we recommend that you return to one dosage a week. If Stalosan is used in higher doses, it may be an advantage to target the dosage towards the critical areas such as damp places, areas with wasted feed, edges etc.

In general, Stormøllen recommends blowing out Stalosan rather than scattering, to ensure optimal spread of the product. Often, you will be able to reduce the consumption of Stalosan by changing to the blowing method. The reason is a tendency to overdose when using the scattering method.

Stalosan F

7. The blowing Method

Blowing Stalosan F, as an improved alternative to the traditional addition by hand, takes the practical use to a higher level. At the same time, the possibility of getting a uniform distribution increases several times. Consequently, the consumption decreases and the efficacy per used gram increases. Finally, the time spend by blowing Stalosan F will correspond to a few minutes per 100 m². This advantage is specially important in modern animal production, where houses reach larger sizes.

Blowing Stalosan F

1. Switch off any ventilation system before blowing out Stalosan F.
2. Pour an adequate amount of Stalosan F into a bucket. Taking into account the blower's range, we recommend a maximum quantity of 5 kg Stalosan F for each blowing session (5 kg/100 m²).
3. Place yourself safely, holding the blower in your left or right hand. The blower is designed for one hand operation.
4. Dip the suction pipe of the blower into the bucket containing Stalosan F, leaving a small opening for air inlet.
5. The exhaust end of the blower should point upward (15-45° from ground level) during blowing out and be moved from side to side. This procedure ensures optimum spread and distribution of Stalosan F. Tests show that a single spot allows a blowing range of about 100 m². One blow takes about 1-1½ minute. Under normal circumstances the blower can spread up to 8 kg Stalosan F per minute.
6. For animal housings larger than 100 m², we recommend to split the area into sections of an appropriate size. In the case of a 500 m² housing, five strategic positions are required for optimum spread.
7. To maintain the desired effect you need to blow out Stalosan F once a week.
8. After blowing out, empty the blower for Stalosan F.





Stalosan F

8. Laboratory Efficacy Studies

Most of the efficacy studies done for Stalosan F was performed by the well known Steins Laboratory, today called Eurofins, a world wide laboratory. The major advantages of those studies are the fact that manure or bedding samples from domestic animal production was used as a source of both pathogens and organic material. In the protocol, the samples first went through qualitative and quantitative analyses to reveal types of pathogens presented and the total pathogenic count. Hereafter all samples were incubated with Stalosan F in manure or bedding. Afterwards, samples were analysed once again, to detect the types of pathogens surviving and the surviving number, if any. In this way, we were able to get a useful efficacy profile that mimicked normal habitat surrounding domestic animals.

Also, Stalosan F has been tested on several international laboratories, including Central Veterinary Laboratory (England), Rostock University (Germany), Microchemlab (USA) and several other laboratories in countries worldwide.

The following page include many of the test microorganisms that has been exposed to Stalosan F in application rates corresponding to 50 g/m². All test results from efficacy studies on bacteria, virus and fungus showed a 99,9% kill within a few hours. Efficacy studies performed with parasites did not show a total kill, but went far beyond the levels normally seen for many biocides.

Stalosan F

Bacteria

Actinobacillus
Aerococcus
Bacillus subtilis
Clostridium perfringens
Clostridium tyrobutyricum
Coliforme bacteria
Eschericia coli
Eschericia coli O 149
Eschericia coli O 157
Enterobacter agglomerans
Enterobacter cloacal
Enterococcus faecium
Eschericia coli
Fusobacterium necrophorum
Haemophilus
Micrococcus varians
Pasteurella multocida
Proteus
Proteus mirabilis
Pseudomonas aeruginosa
Pseudomonas fluorescent
Pseudomonas paucimobilis
Salmonella dublin
Salmonella enteritidis
Salmonella typhimurium DT 104
Salmonella typhimurium
Serratia marcescens
Staphylococcus hyicus
Staphylococcus aureus
Staphylococcus epidermis
Staphylococcus
Streptococcus faecalis
Streptococcus pyogenes
Streptococcus uberis

Viruses

Canine Parvovirus
Newcastle Disease Virus
Porcine Parvovirus
Reo-Virus
Vaccinia-Virus

Fungi

Alternaria
Aspergillus
Aspergillus flavus
Candida ciferii
Candida lusitaniae
Candida parapsilosis
Candida pendotropicalis
Candida pseudotropicalis
Candida rogosa
Candida torulopsis
Cladosporium
Cladosporium herbarum
Cryptococcus laurentii
Verticillum cinnabarium
Fungi imperfecte
Fusarium
Heminthosporum
Maris torulopsis
Mucor
Mucor plumbens
Penicillium
Penicillium viridicatum
Pullularia
Rhodotorula slutinis
Saccharomyces cerevisiae
Trichoderma viride
Trichosporon beigellii
Verticillum cinnabarium

Parasites

Ascarida galli
Ascaris suum (Round worm)
Capillaria obsignata
E.acervulina (chicken coccidia)
Heterakis gallinarum

Fly-larvae

Stalosan F

Occurrence of *Ascaris suum* eggs in the Danish swine breeding
Med. Vet. A.E.W. Ibsen, Master Thesis, Centre for Experimental Parasitology, Institute of Veterinary Microbiology, Royal Veterinary University 1999

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.

Light slaked lime and Stalosan F strongly reduces the egg number and have a significant effect on *A. suum* eggs in the houses.

Farm 2 was described by the slaughterhouse as a 'well managed farm but with a high incidence of milk spots in the liver's. 6 houses were investigated [1: cleaning in progress, 2: 1 week after cleaning, 3: 6 weeks after cleaning, 4: 7-8 weeks after cleaning, 5: more than 16 weeks after cleaning, 6: daily cleaning]. The proportion of different egg stages was as found like in the finishing house on farm 1 but the egg counts were much lower [0.1-22.0 EPG]. 'Black' eggs were also found here.

Stalosan F

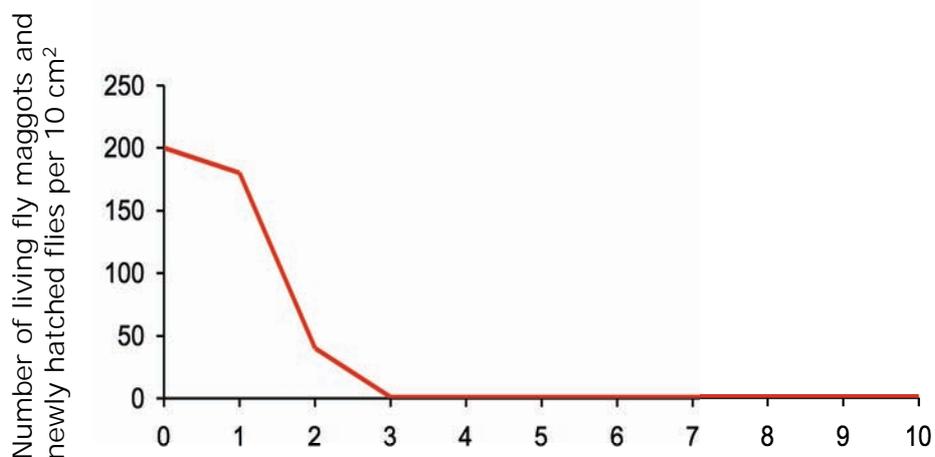
Fly-larvae

A Danish Study, performed by The Danish Food and Veterinary Research Centre, showed that during a normal day in a broiler house an average of 1000 flies enter through the ventilation system. More than 8% of the flies was infected with *Campylobacter*. By blocking the pathway for the flies to enter the house, it was shown that *Campylobacter* infected birds can be reduced by 99,9%. This suggest that flies play a major rule in the transmission of infections like *Campylobacter* entering broiler houses. Similar cases have been observed with other infections like *Salmonella*. Other important routes of transmission of infections is by contaminated people, poultry equipment and farm vehicles.

In vivo test below performed by Steins Laboratory

Examination of the effect of Stalosan on fly-larvae.

The test was carried out in a calf stable where the calves are in separate stalls.



The larvae which are in contact with Stalosan F dies, and so do the newly hatched flies.

In the barn where there is a deep layer of straw, it is necessary to spread Stalosan F every day. Dosage of Stalosan F is 50 g/m².

Technical staff: Randi Rasmussen, Steins Laboratory

Stalosan F

Effects of Stalosan F on Unsporulated Oocysts.

Janet Catchpole, BSc, CENTRAL VETERINARY LABORATORY (MAFF)

Parasitology Department, Central Veterinary Laboratory, New Haw, Weybridge.

The unsporulated oocysts exposed to Stalosan F showed reduced sporulation rates compared both to the silver sand control material and to untreated oocysts. The reduction in sporulation rate increased with exposure time. Stalosan F would appear to have activity against unsporulated oocysts and as this is the stage excreted by the host animal there may be a role for this product in reducing the initial environmental challenge.

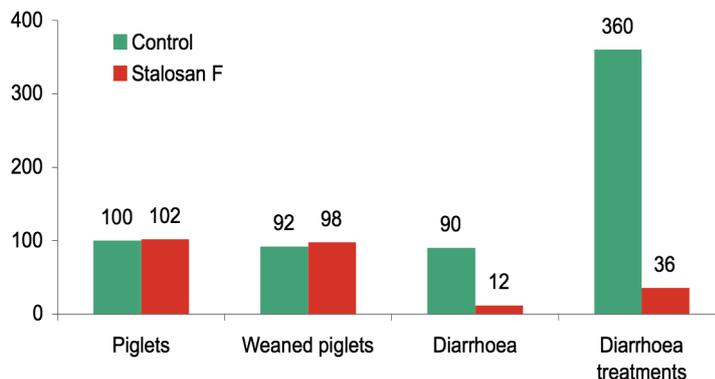
Table. Reduction in sporulation rate of treated oocysts

The sporulation rates of oocysts treated with Stalosan F are reduced in comparison to the oocysts treated with silver sand and also markedly reduced when compared to the 80% seen with untreated oocysts shown in the table below.

| Powder | Concentration | Time | Reduction of sporulation compared to untreated sporulation number |
|------------|---------------------|------|---|
| Stalosan F | 30 g/m ² | 24 h | 14% |
| Stalosan F | 50 g/m ² | 24 h | 50% |
| Stalosan F | 80 g/m ² | 24 h | 56% |
| Silversand | 30 g/m ² | 24 h | 1% |
| Silversand | 50 g/m ² | 24 h | 4% |
| Silversand | 80 g/m ² | 24 h | 6% |

Trial Farrowing House, 1999

Unit had previously experienced scour especially Coccidiosis and higher desirable mortality. Conducted on 20 sows and litters - 10 treated with Stalosan (days 1, 2, 3, 7, 11, 15, 19, 23, 27) - 10 untreated. All piglets given Mediceal 4 and 8 days. Any scoured pigs was given 2 ml daily of Neftin until recovered. All weaned at 28 days.



| Treatment | Mortality |
|------------|-----------|
| Control | 8% |
| Stalosan F | 3,9% |

Stalosan F

9. Farm Trials

Danish and Swedish Trial on broilers (Two years trial period)

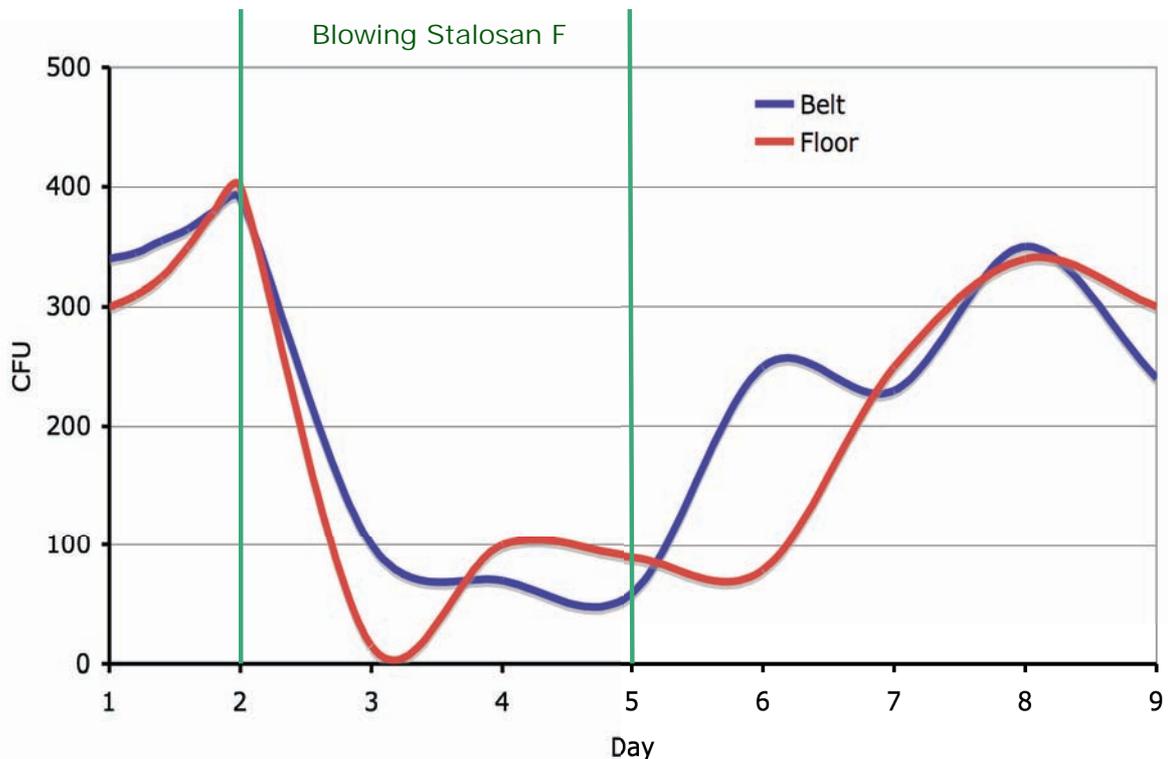
Aim of the trial: To evaluate the potential financial advantages of Stalosan F in broiler houses. This was done by comparing the gross profit with and without Stalosan F. 100.000 birds from three different houses were included in the trial.

Conclusion: By using Stalosan F, the gross profit was increased by 10 %, which included the cost of Stalosan F.

Coliforme bacteria in egg layer housing

Stalosan was sprayed between week 2 and week 5. Swabs were taken before, during and after spraying. The Coliform pressure was reduced during blowing of Stalosan F, but eventually returned to the initial number after the blowing was stopped.

Study performed by Prof. Bud Harmon, Perdue University, USA



Stalosan F

English Turkey Brooders Trial

Aim of the trial

To evaluate the effect of Stalosan F on bird's conformity, high quality clean carcass meat, financial benefits

Results

Reduction in larvae of *Alphitobius diaperinus* (meal worm beetle). Reduced mortality (See graph 1)

Improved litter quality (See graph 2)

Significant reduction in ammonia levels (See graph 3)

Reduction in secondary bacterial infection

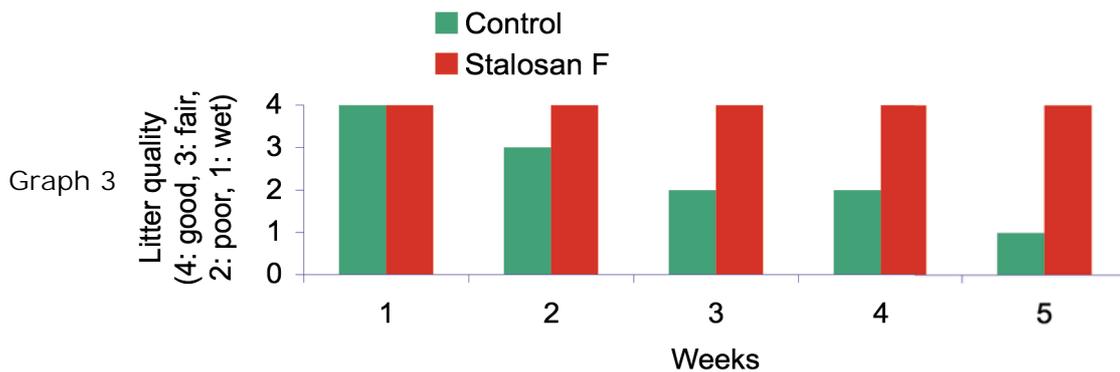
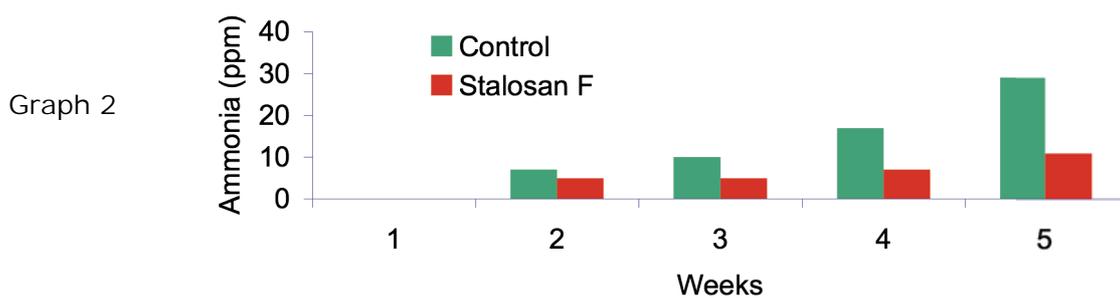
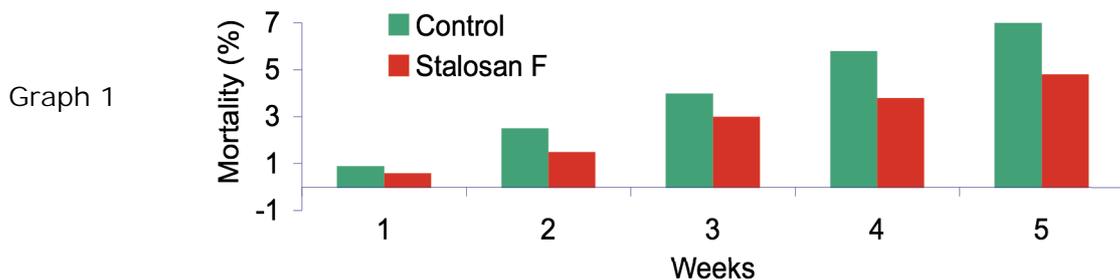
Improved liveability and health status

Reduction in drug treatment of up to 80 %

Reduction of overall feed consumption of over 1 %

Savings in veterinarian cost and larvicidal spray of over €4,20 per bird

Increase in feed conversion rate (FCR)





Stalosan F

Ileitis Farm Trial, 2001

Ileitis is caused by *Lawsonia intracellularis*, a bacteria. During stressful periods can it make the pigs suffer from especially diarrhoea and enteritis. Reduced growth, benefit of feed and condition are effects, the farmer is not always aware of. Never-the-less a lot of welfare and money is lost because of this bacteria.

Eliminating *Lawsonia intracellularis* from the farms have been considered as practically impossible. Guidelines for that tell the farmer to medicate the breeding animals for 14 days, move the animals to newly cleaned and disinfected and/or fumigated sties. After this the animals should be medicated again for 14 days. No new animals can be introduced from other farms because they will bring the bacteria into the sties again. The bacteria can survive 14 days on things used in the sties (boots, clothing, broom, shovel etc). These should be cleaned thoroughly or removed permanently. Attention to and from the staff is also essential. The bacteria is easily spread after just one oversight. Rodents, birds and even insects can probably also spread the bacteria.

Stalosan F was spread in 4 farms for 3 consecutive days and then once a week (50 g/m²) for 3 months. In addition to this Stalosan F was put on suspiciously looking faeces (diarrhoea). To find a possible effect floor-samples were taken at the start before Stalosan F was used the first time and then after using Stalosan F for 3 months.

These 4 farms paid a lot of attention to *Lawsonia intracellularis* and was chosen to this trial by their vet. Torben Svendsen and Leon Lau, (certificate holders in swine diseases). I visited the farms at trial-start, once monthly till the floor-samples were taken after 3 months. All the samples were PCR-tested, which is a very.

| Test-results floor-samples | Before Stalosan F | | After Stalosan F | |
|----------------------------|-------------------|-------------|------------------|-------------|
| | Lawsonia | No Lawsonia | Lawsonia | No Lawsonia |
| Brinkmann, Farm 1 | 9 | 1 | 0 | 10 |
| Sæderup, Farm 2 | 9 | 5 | 0 | 13 |
| Drost, Farm 3 | 1 | 9 | 0 | 10 |
| Jørgensen, Farm 4 | 6 | 22 | 0 | 29 |
| Total | 25 | 37 | 0 | 62 |

Stalosan F

Salmonella Farm Trial, 2004

In Denmark, the slaughter house divide the level of salmonella infection in incoming pigs from 1 to 4, where 1 is an insignificant infection level and 4 is maximum Salmonella infection. Depending on the level of infection, the pig producer will get a significant money deduction.

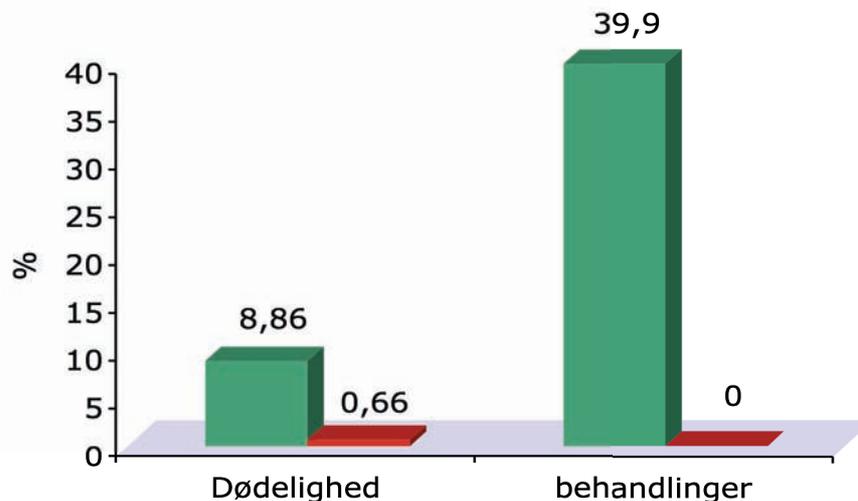
| Name | Production Pigs | Salmonella Level | Start Stalosan F | Salmonella Level 1 | Stalosan F Treatment |
|------------|-----------------|------------------|------------------|--------------------|----------------------|
| Krag | 5000 | 3 | January 2005 | March 2005 | 2,5 month |
| Kristensen | 1400 | 2 | December 2004 | February 2005 | 2 months |
| Andersen | 4000 | 3 | July 2004 | August 2004 | 1,5 month |
| Korsgaard | 4400 | 3 | October 2004 | January 2005 | 3,5 month |
| Thomsen | 5000 | 3 | July 2004 | September 2004 | 2 months |

Pig Breeder trial, UK

A 15 day test was carried out inspecting any advantages of Stalosan F treatment in farrowing houses. Two houses were selected each having 150 piglets. All conditions in the two houses were the same apart from the Stalosan F treatment. Application of Stalosan F day 1,2,3 and 7.

■ Conditions before adding Stalosan F

■ Conditions after 4 Stalosan F applications in 15 days



Stalosan F

Fattening pig trial

Objectives was to conduct a trial for 11 weeks to evaluate the benefits of Stalosan in a fattening house. Trial was held on an East Anglian unit fattening over 2300 hybrid pigs, drawn from local established breeder-weaner sources. The unit was selected for its relatively high susceptibility to swine dysentery outbreaks. Pigs were fattened to 80kg plus. Stalosan was added on a weekly basis of 50 g/m². Medication: Tiamutin, Tivettrin/Tylan, Lincospectin and Amoxisol.

Pigs treated with Stalosan showed the following benefits:

Lower mortality, lower vet/med costs, reduction in bacterial scours, improved growing environment, reduced levels of cross infection, quicker recovery from infection, improved liveability and health status

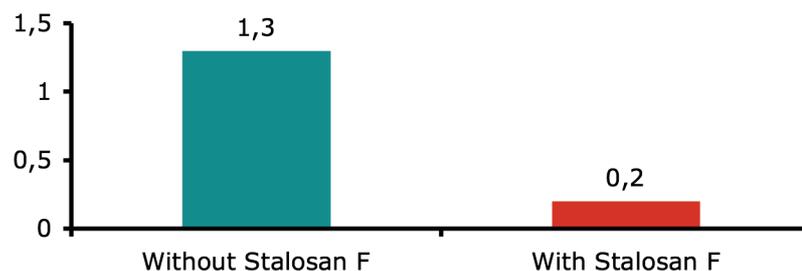
Cost of mortality: Value piglet: €75
Control: 16 deaths
Stalosan F: 9 deaths
Gain: €525

Cost of medication: €0,74
Control: 1892 doses
Stalosan F: 233 doses
Gain: €1228

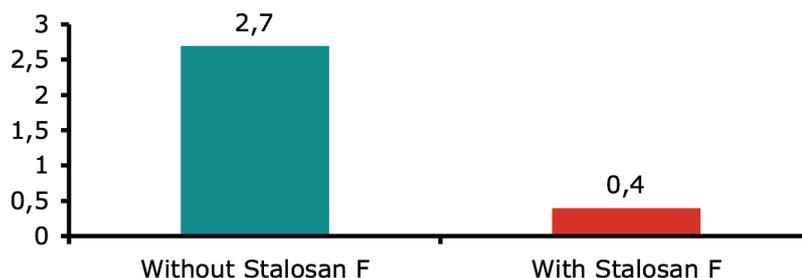
Cost of Stalosan F: €1/kg, used 176 kg ⇒ €176

Saving per house: €1577

Death pigs due to dysenteri (%)



Treatments per pig



Stalosan F

10. Ammonia Control

By attaching from several different points, Stalosan F manages to control ammonia, significantly.

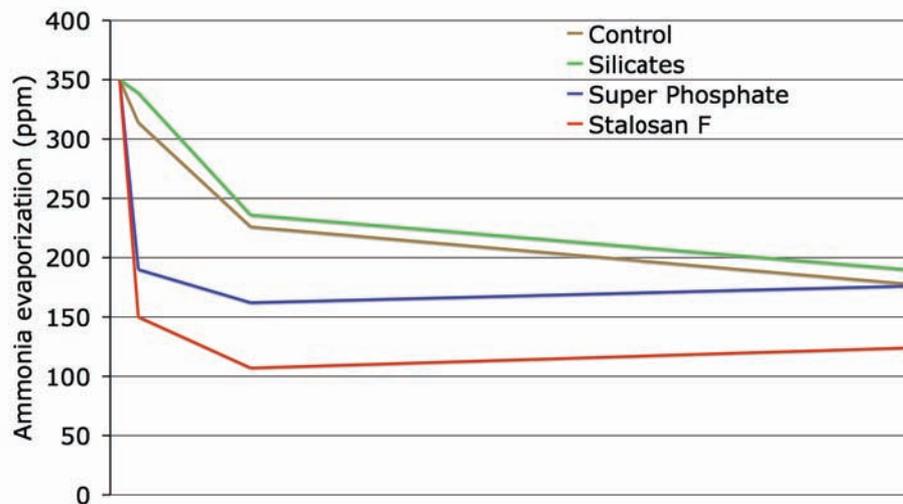
Stalosan F inhibits the enzyme urease, which is produced by many pathogens, to catalyze the formation of ammonia (ammonia creates a chemical suitable pathogenic environment and aids in the infection of hosts by dissolving skin and soft tissue).

Stalosan F functions as a buffer that chemically controls the levels of moisture, and ammonia and other toxic gases. The process is dependent of all ingredients.

Stalosan F lowers the pH value in micro environments, leading to decreased ammonia formation and evaporation.

Performance of Additives in Reducing Ammonia Emission in Slurry
Mats Andersson, Inst. of Environmental Technology, Lund University

The study took place over 42 days and measurement of ammonia emission were taken on day 1, 7 and 42. All products were only added once at day 0.

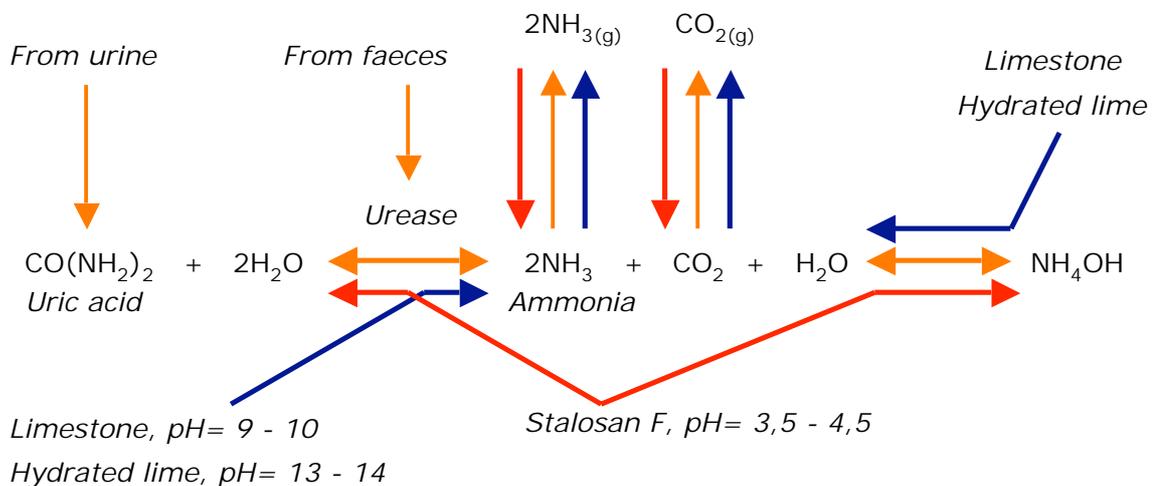
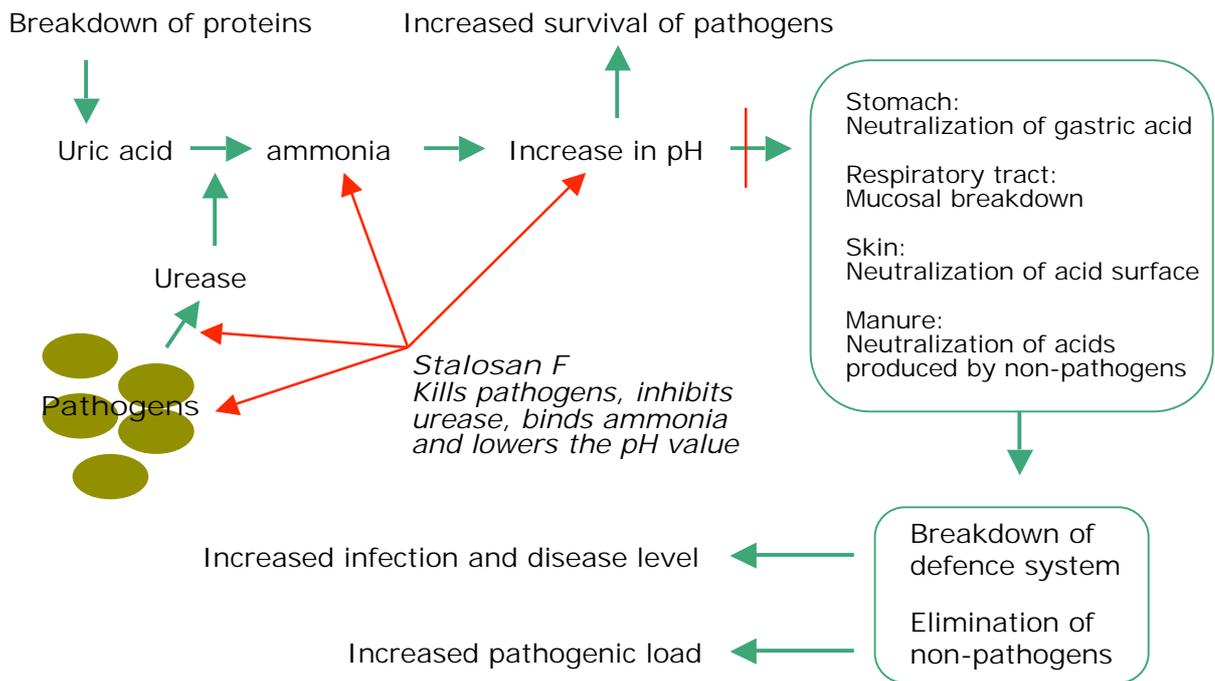


"6 weeks after application of the additives, Stalosan F still reduced the ammonia emission compared with the emission from the untreated slurries ($p < 0.05$).

The approximate reduction was 30%. All other slurries treated with the different additives emitted ammonia at the same rate as the control",
Comments: Mats Andersson.

Stalosan F

The equilibrium of ammonia and water in bedding/manure on the floor in animal houses are affected by many different factors and subsequently can be controlled by altering those conditions. Below is an illustration of the chemical equilibrium in material layers on the floor in animal buildings.



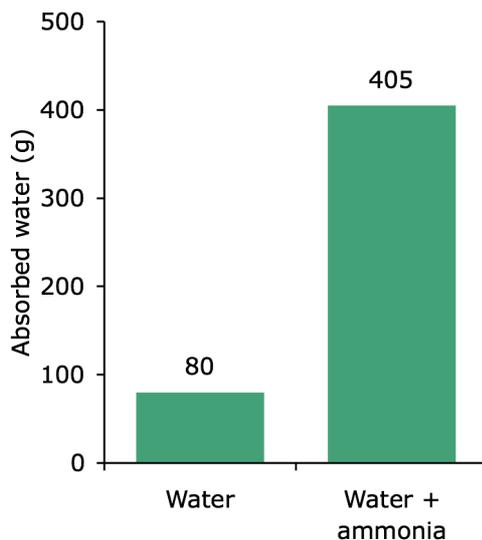
Stalosan F

11. Water binding capacity



Results from a laboratory test which shows the water binding capacity of Stalosan F with ammonia to the right and without ammonia on the left. The same amount of Stalosan F has been used in both glasses but the glass on the right has been added less ammonia (approx. 1 per cent solution.)

Stormøllen, 2004



Graphic display of the above test. 100 g Stalosan F binds up to 400 g water if ammonia is present.



Stalosan F

12. EPA Toxicity studies.

Product Safety Laboratories, USA, 2005

The acute toxicity studies are performed with 5 g of Stalosan F per kg body weight. If those figures are projected to an adult human with a body weight of 80 kg, he can be exposed to 0.40 kg of Stalosan F either on the skin, through the gastro intestinal tract or through the respiratory system without being affected in any way. Further, he will avoid any irritation or sensitization to the skin caused by exposure to Stalosan F. Caution should only be taken with contact to the eye, due to the mildly irritant action, when Stalosan F is applied directly in the eye.

Due to the EPA approved toxicity, irritant and sensitization study, Stalosan F can be recognized as being a well documented product with few or no side effects. Still, our recommendation is to follow the guidelines from the safety datasheet of Stalosan F.

| Test Methods and Aim of Study | Results | | | Conclusion |
|--|--------------------|---|--|---|
| | Body weight | Necropsy | General evaluation | |
| Acute Oral Toxicity To provide information on health hazards likely to arise from a short term exposure to Stalosan F by the oral route. | gained body weight | No gross Abnormalities were noted for any of the animals. | All animals survived and appeared active and healthy. There were no signs of gross toxicity, dermal irritation, adverse pharmacologic effects or abnormal behaviour. | The acute oral LD ₅₀ of Stalosan F is greater than 5,000 mg/kg of body weight. |
| Acute Dermal Toxicity Study To provide information on health hazards likely to arise from a short term exposure to Stalosan F by the dermal route. | gained body weight | No gross Abnormalities were noted for any of the animals. | All animals survived and appeared active and healthy. There were no signs of gross toxicity, dermal irritation, adverse pharmacologic effects or abnormal behaviour. | The single dose acute dermal LD ₅₀ of Stalosan F is greater than 5,000 mg/kg of body weight. |
| Acute Inhalation Toxicity Study To provide information on health hazards likely to arise from a short term exposure to Stalosan F by the inhalation route. | gained body weight | No gross Abnormalities were noted for any of the animals. | All animals survived and appeared active and healthy. There were no signs of gross toxicity, dermal irritation, adverse pharmacologic effects or abnormal behaviour. | The single dose acute dermal LD ₅₀ of Stalosan F is greater than 5,000 mg/kg of body weight. |
| Primary Eye Irritation Study To provide information on the irritation likely to arise from an instillation of Stalosan F into the eye. | gained body weight | No gross Abnormalities were noted for any of the animals. | All animals survived and appeared active and healthy. There were no signs of gross toxicity, dermal irritation, adverse pharmacologic effects or abnormal behaviour. | Stalosan F is classified as mildly irritating to the eye. |
| Primary Skin Irritation Study To provide information on the skin irritation likely to arise from a single topical exposure to Stalosan F. | gained body weight | Not relevant | All animals survived and appeared active and healthy. There were no signs of gross toxicity, dermal irritation, adverse pharmacologic effects or abnormal behaviour. | Stalosan F is classified as non irritating to the skin. |
| Dermal Sensitization Study To determine the potential for Stalosan F to elicit a skin sensitization reaction. | gained body weight | Not relevant | All animals survived and appeared active and healthy. There were no signs of gross toxicity, dermal irritation, adverse pharmacologic effects or abnormal behaviour. | Stalosan F is not considered to be a Contact sensitizer. |

Stalosan F

13. Aquatic Toxicity Studies

Algal Growth Inhibition Test

M.Sc. Jane Pors, Eurofins, 2004

The algae growth test was performed with a range of test concentrations of Stalosan F. The inhibition values did not indicate a toxicity relationship between the growth of *Chlorella vulgaris* and Stalosan F.

The effect concentration, EC20, could not be assessed on the test data given, and therefore the EC20 was assessed to the following: EC50-72h > 1600 mg/l.

Daphnia, acute Toxicity Test

M.Sc. Jane Pors, Eurofins, 2004

The acute test with the *Daphnia magna* was performed with two concentrations of Stalosan F. The test was performed as a limit test and effect concentrations can therefore not be determined using statistical calculations. However, the effect concentrations can be assessed to be the following:

EC20-24h > 1000 mg/l, EC50-24h > 1000 mg/l

EC20-48h > 1000 mg/l, EC50-48h > 1000 mg/l

Fish (*Poecilia reticulata*), acute Toxicity Test

M.Sc. Jane Pors, Eurofins, 2004

The acute test with the fish (*Poecilia reticulata*) was performed with two concentrations of Stalosan F. The test was performed as a limit test and effect concentrations can therefore not be determined using statistical calculations. However, the effect concentrations can be assessed to be the following:

EC20-96h > 1000 mg/l, EC50-96h > 1000 mg/l



Stalosan F

14. An environmental study of a hygiene substance used in animal buildings

D.Sc. Finn Eiland (Head of department), Danish Institute of Agricultural Sciences

A field experiment was initiated at Research Centre Foulum in 1997. The purpose was to measure possible effects of Stalosan F on the chemical and microbiological parameters in soil and in the slurry used for the field experiment. The yields of ryegrass and crop uptake of copper in the crop were also measured. The measurements in the field experiment will be continued for a further two years period with a crop of winter wheat and with similar amounts of slurry and Stalosan F. After two years with yearly loading of slurry containing different concentrations of Stalosan F, the following results were obtained:

Stalosan F in slurry even at 10 times the normal applied amount does not reduce the number and the activity of anaerobic microorganisms in the slurry to an unacceptably low level. It is therefore expected that Stalosan F in slurry can be used in e.g. biogas production without any negative effects.

Potential nitrification activity (PNA) was examined in the soil with the different additions of slurry and Stalosan F (August 1998).

The addition of a normal amount of Stalosan F in 30 T slurry per ha did not affect the activity of the nitrifying bacteria at the 0-5 and 5-20 cm depth. There was a significantly increase of PNA at both 0-5 and 5-20 cm depths, when 10 x the normal amount had been applied.

Adding 90 T slurry per ha with normal and 10 x normal amounts of Stalosan F resulted in a decrease of PNA in the surface soil layer with increasing amounts of Stalosan F compared with the reference soil with addition of 90 T slurry per ha and 0 kg Stalosan F. However, PNA was not lower than the corresponding treatments receiving Stalosan F directly to the soil.

PNA at the 5-20 cm depth in treatments receiving 90 T slurry per ha and Stalosan F increased, showing that the nitrifying bacteria were stimulated by Stalosan F below the surface layer. Therefore it is most likely that the inhibition in the surface layer was of a temporary nature, maybe due to an accumulation of residues of Stalosan F and slurry after two years with ryegrass, where no soil cultivation took place.

The overall picture of measurements of microbial biomass and activity showed that Stalosan F, both in slurry and added directly to the soil, often improves soil conditions for microbial life, resulting in a stimulation of the microorganisms. Stalosan F added in very high concentrations may sometimes cause a temporary inhibition of the organisms and their activity. However, an inhibition followed by a stimulation of the microorganisms is commonly observed in soils, when high amounts of organic matter were added to the soil. The occasionally observed inhibition was not found to an extend, where it could be critically for the microorganisms.

A positive effect was found on the yields of ryegrass in 1997 and 1998 with Stalosan F in 30 T slurry per ha. The surplus yields were 570 and 260 kg dry weight per ha, respectively. This was also the case, when Stalosan F was added directly to land together with inorganic N, where an increase of 600 kg dry weight per ha was found in 1998. There could be several explanations for the increased yields. One possibility is improved conditions for the soil microorganisms or an increased content of sulphur in soil originating from Stalosan F.

By the application of Stalosan F in animal housings at the levels suggested by the producer, gives only a moderate increase of copper at about 60 g copper per ha per year. The main part of copper applied to the soil originated from pig feed for sows and piglets. The copper content in slurry without Stalosan F, equivalent to 30 T slurry per ha, was 620 g copper.

Most crops are expected to take up 20-50 g copper per ha per year, which is in the order of magnitude of the added copper by Stalosan F. In the Foulum experiment with ryegrass and three cuts, 57 g copper per ha was removed after applying of 30 T slurry per ha in 1998. The amount is dependent on Cu content in the soil and the crop yields.

After two years study of possible effects of the hygiene substance Stalosan F on the chemical and microbiological conditions in slurry and in soil, the main conclusion is that the product is very useful and it has no observed harmful effects on the soil environment, when it is used in the amounts suggested by the producer. Occasionally, the product even stimulated the soil microorganisms and raised the yields of ryegrass.

Stalosan F

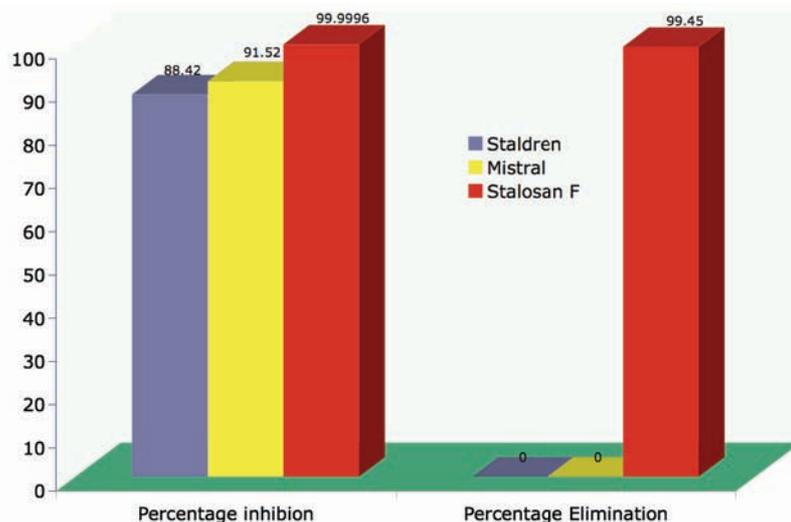
15. EPA Registration

| | | |
|---|--|--|
|  <p>U.S. ENVIRONMENTAL PROTECTION AGENCY Office of Pesticide Programs Antimicrobials Division (7510C) 1200 Pennsylvania Avenue NW Washington, D.C. 20460</p> <p>NOTICE OF PESTICIDE: <input checked="" type="checkbox"/> Registration <input type="checkbox"/> Reregistration <small>(under FIFRA, as amended)</small></p> | <p>EPA Reg. Number: 75613-1</p> | <p>Date of Issuance: JAN 31 2007</p> |
| | <p>Term of Issuance:</p> | |
| | <p>Name of Pesticide Product: Stormoellen A/S - Stalosan F</p> | |
| <p>Name and Address of Registrant (include ZIP Code): Stormoellen A/S Ringsbjergvej 16 DENMARK 4682 Tureby</p> | | |

Comparison Study

Performed by the EPA approved laboratory, MicroChem Laboratory, USA. A general inhibition of bacteria by a dry powder product without any elimination action is considered to be caused by binding water only. The effect is fragile and insignificant in cases of an excess of water, seen on most floors in animal houses.

On the other hand, if the inhibition is close to 100% and followed by a significant elimination rate, like seen for Stalosan F, there exist a solid base for antimicrobial control.



Stalosan F

16. Evaluation of dry-powder product for treatment of floorings in animal housing

Water binding or antimicrobial

When choosing a dry-powder product for animal housing, it is important first to decide, if the goal is simple water control or if the demands are more extended towards direct antimicrobial and environmental control. None of the goals are practically possible to a level of 100%, unless an unrealistic large amount of product is used on a frequent basis. Still there is good arguments for using dry-powder products and also to take a closer look at the different types of product, before making any decisions, mainly because of large variations in efficacy and price.

| Product | Ingredients | Approx. price €/kg |
|--------------------|--|-----------------------|
| Limestone | Calcium carbonate | 0.05 |
| Diatomaceous Earth | Silicates, clay | 0.19 |
| EcoDry | Silicates, clay | 0.68 |
| Actilith Plus | Silicates, clay | 0.74 |
| Staldren | Calcium carbonate, silicates, oils, blue vitriol | 0.47 |
| Mistral | Silicates, clay, calcium carbonate, oils | 0.91 |
| Hydrated Lime | Calcium hydroxide | 0.26 |
| Stalosan F | Phosphates, sulphates, silicates, iron salts, copper salts, oils | 0.95 |

Calcium carbonate, silicates and clay

Raw materials like calcium carbonate, silicates and clay has, from a practical point of view in animal housing, primarily a drying effect. The three raw materials can be placed in the following order with the highest water binding capacity first: Silicates, clay and calcium carbonate, with calcium carbonate showing an insufficient capacity. Danish veterinarian recommend not to use calcium carbonate in animal production, because of the slippery flooring caused by the product that can lead to broken limbs. From a theoretical point of view, silicates and clays can absorb anything from water, ammonia and other chemical components onto organic material including oils and microorganisms. A disadvantages is the fact that silicates and clays will be saturated by the same components within a short period of time, leaving a need for daily addition of products. The major disadvantages, though, is the survival of absorbed microorganisms that will be released and grow high in number as soon as water is added to the environment. This means that as long as the material layer is moist, there will be no inhibition of microorganisms with silicates and clays. None of the three raw materials has any antimicrobial or biocidal effect.

Stalosan F

Hydrated Lime

Strong bases, like hydrated lime, are very efficient in removing biofilm and organic remains from the washing and cleaning, because of its highly corrosive effect on any organic material including oils, microorganisms, animals and humans. However, Hydrated lime is not recommended for use in the presence of animals, because of this corrosive effect. Especially the skin of animals will be broken down in contact with the product, leaving the surface of animals open for infection. The respiratory tract, eyes and stomach will also suffer if there is a direct contact with hydrated lime. Therefore, all hydrated lime, if used, should be washed away before the introduction of animals. Be aware of the greasy consistency of hydrated lime that makes it difficult to wash away.

Phosphates/Sulphates

In general, these materials has a pH value of 3 to 4 and is therefore considered a weak acid. Also, they are well known acid buffers that can maintain a low pH value at high base pressures. Pathogenic microorganisms are very sensitive towards pH values below 4 and will often die under such conditions. Only certain spores of bacteria etc. Will survive such an environment. Above pH 4 and below 7, the pathogens will be more or less inhibited, unable to grow in number. Pathogens prefer a higher pH value between 8 and 9 for optimal living conditions and growth. This is in contrast to many harmless soil and internal bacteria that grows well at pH values around 4 to 5. Lactobacillus, which is considered an beneficial bacteria, produces lactic acid as a waste product. The production of lactic acid makes its environment acidic which inhibits the growth of many harmful bacteria. Both human and animals produce acids as a first line defence against many pathogens. That is why, in humans, the pH value of gastric juice and skin is 0 and 5,5, respectively.

If this pH dependent reality is to be transferred to the material layer in animal houses, it is quite obviously that acids should be used for bedding control instead of bases.



Stalosan F

Copper and Iron

Iron and specially Copper is well known for its antimicrobial effect towards bacteria, fungi and viruses. Beside the antimicrobial effect, both metals are crucial micro minerals essential for all living creatures. Therefore, the concentration and source of Copper and Iron, is responsible for the profile as a micro mineral or antimicrobial towards pathogens.

When Copper is used as an antimicrobial, it is important to choose a highly reactive source to obtain a high efficacy from a fairly small amount of product. In this case, Blue Vitriol would be less active product, because of its relatively low reactivity and high solubility in ammonia containing bedding. Instead, Copper from a highly reactive source and with a higher stability under alkaline conditions would be much more suitable for the bedding in animal housing. As an extra stabilising effect, it is highly recommended to always use Copper in combination with acid buffers, because studies show that the efficacy can be increased by a tenfold in combination with acid compared to bases like lime stone.

| Product | Real effects |
|-------------------------|---|
| Diatomaceous Earth | Absorbs water and small amounts of organic material |
| Calcium carbonate | Absorbs small amounts of water |
| Hydrated lime | Kills most microorganisms excluding spores of bacteria |
| Copper based components | Kills bacteria, fungi and viruses. The efficacy is highly dependent upon the Copper source and also the pH value of the nucleus in the end product. |
| Iron based components | Kills fungi and controls bacteria and viruses |
| Phosphates/sulphates | Binds water and neutralizes ammonia and holds a high buffer capacity. Lowers the pH value that will lead to a inhibition of pathogens. Increases the biocidal efficacy of copper and iron in 10-fold. |
| Oils | Primarily used as fragrance. Secondary used as repellent. |



Stalosan F

Conclusion

If the idea is to bind water only, it is recommended to invest in Diatomaceous earth instead of spending extra money on products like Staldren, Mistral, Actilith Plus that all have a composition either very close to Diatomaceous Earth or less effective. It seems completely wrong to pay €0.9/kg of Mistral which is a mixture of Diatomaceous earth and lime stone, when the price for Diatomaceous earth is €0.19/kg and, at the same time, is a better choice, because of the lack of lime stone.

Hydrated lime should only be used in empty animal houses and an intensive washing is needed before the introduction of new animals.

If the goal is to obtain direct antimicrobial and environmental control, the main nucleus of a dry-powder product should be acidic. Phosphates, sulphates or any other acid buffer with a low solubility and a pH value below 4 is an effective choice. In the bedding, the pH value is often much too high because of the large production of ammonia (pH=13), caused by bacteria. This lets both animals and beneficial microorganisms suffer and the former even die. Animal resistance towards diseases will decrease due to the increase in pH value on any animal tissue surface. Again, it is important to control the production of ammonia, decrease the pH value and restore the natural environment.

Active metals like Copper and Iron are excellent choices for antimicrobial control in bedding. Also Copper is a very potent inhibitor of the enzyme Urease that is responsible for the production of ammonia in animal houses.

Stalosan F

17. Liquid Disinfectants and Stalosan F

Liquid disinfectants kills pathogens in a clean laboratory, but are quickly inactivated by the organic matter in animal houses. Stalosan F also kills pathogens, and is not affected by organic matter and has a long term effect.

Liquid disinfectants has no effect on the diseases entering animal houses with the newly introduced animals. Because Stalosan F can be added, while animals are present, you will have a consistent disease control.

| Disinfectant | Detergent action | Effective against | | | | |
|---------------------------|------------------|-------------------|--------|-------|--------|-----------|
| | | Bacteria | Spores | Virus | Fungus | Parasites |
| Strong bases | yes | ++ | (+) | +++ | +++ | ++ |
| Aldehydes | no | +++ | +++ | +++ | +++ | ++ |
| Chloride products | no | +++ | - | +++ | ++ | + |
| Quaternary ammonium salts | yes | +(+) | - | - | - | - |
| Phenols | yes | ++(+) | - | (+) | ++ | - |
| Oxidizing agents | no | +++ | ++ | +++ | ++ | - |
| Stalosan F | no | +++ | ++ | +++ | +++ | +++ |

| Disinfectant | Fast action | Inhibited by organic material |
|-------------------------------|-------------|-------------------------------|
| Strong bases | + | - |
| Aldehydes | - | - |
| Chloride products | +++ | +++ |
| Quaternary ammonium compounds | +++ | +++ |
| Phenols | +++ | +++ |
| Oxidizing agent | +++ | ++ |
| Stalosan F | - | - |