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HYGIENIC CONTROL OF SALMONELLA IN ARTIFICIALLY CONTAMINATED FEED

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SUMMARY

Experiments were carried out to evaluate the antibacterial effect of formaldehyde solution 40%, two commercial formic-propionic acid mixture, antibiotic preparation and physical treatment against salmonella microorganism in artificially contaminated feed.

Formaldehyde solution 40% gave the best results of reduction (100% reduction) after one hour of treatment (10L/ton) and 3 hours of treatment for a concentration of (5L/ton). After 48 hours of treatment, the results obtained were relatively close to each other (98.1% for antibiotic preparation, 98.7&99.3% for organic acid preparations and 99.4% for physical treatment). However, the four treatments gave a complete decontamination of feed after 72 hours.

INTRODUCTION

Many years ago, It had been recognized that the three major sources of salmonella infection for poultry are; the introduction of infected stock, the environment and contaminated feed (Gordon and Jordan, 1982, Kampelmacher, 1987 and Veldman et al.,1995). Considerable interest had consequently developed in methods to reduce feed as a potential vehicle of infection (Williams, 1981). So different approaches had been used to reduce the contamination of feed materials as well as the finished feed (Haggblom, 1993 and Renggli, 1996). Currently acceptable methods include pelleting finished feed at high temperature which reduces, but does not eliminate, the bacterial load and which does not prevent reinfection after the pelleting process (Iba and Berchier, 1995).

Fumigation of feed with formaldehyde, volatile fatty acids or other organic acids had also been

investigated and the subsequent studies showed that formic acid was the only organic acid sufficiently antibacterial to be of use for this purpose (Chermington et al., 1991).

However, other methods for feed decontamination were tried with variable degrees of success such as the use of other organic acids and air conditioners at which the feed is to be treated by the exposure to high temperature (over 85°C) then to be cooled within few minutes to a temperature below 20°C (McCapes et al., 1989).

The present study was planned to evaluate the efficiency of some available methods and products currently used in controlling salmonella contamination in poultry feed.

MATERIAL AND METHODS

***The used feed material:**

Seven finished samples, each of 10 kg were prepared and sterilized by autoclaving at 121°C for 20 minutes. Six of these samples were used for the different decontamination trials, while the seventh one was left untreated following the artificial contamination to be the control. The used feed was a commercial starter poultry feed, which was obtained from Pyramids Poultry Co., El-Mansuria, Giza.

***Bacterial strains:**

Pure strain of Salmonella typhimurium organism was obtained from Department of Bacteriology, Faculty of Veterinary Medicine, Cairo University.

***Artificial contamination of feed:**

It was carried out in a way similar to that of Ibe and Berchieri, 1995 where ten milliliters of an overnight broth culture of the Salmonella typhimurium were mixed thoroughly with 50 grams of the previously treated commercial poultry feed for five minutes in a sterile container. This was then mixed again with the previously sterilized 10 kg of feed in a large plastic bag by inverting the contents continuously for ten minutes.

Bacterial numbers per gram were determined on several samples taken from different parts of each batch to check for a homogenous distribution of bacteria and the average was obtained.

***Feed treatment:**

***Trial (1):**

40% formaldehyde solution (A product of Helliopolis Co. for pharmaceuticals chemicals. Egypt) was added to batch number 1 and 2 in a concentration of 50-ml/10 kg (trial1-1) and 100ml/10kg (trial 1-2). These amounts are equivalent to 5 and 10 L/ton respectively.

***Trial (2):**

An antibiotic preparation of Phizer Co. Animal health Department, Egypt (oxytetracycline 40%) was used in a concentration of 20g/10kg (equivalent to 2 kg/ton).

***Trial (3):**

Two commercial products of organic acids were used in this trial. The first product is salmnil (Trial 3-1) (A product of Nutri-AD international).

***Salm-Nil**

Salts of propionic acids
Formic and citric acid
Protein meals
Fish meal

The preparation was used in a concentration of 30g/10kg (equivalent to 3kg/ton).

* The second preparation was Germicin (A product of IQF-Spain) trial 3-2.

***Germicin:**

Propionic acid	36.0%
Formic acid	28.5%
Ammonia	25.7%

The preparation was used in a concentration of 10g/10kg (equivalent to 1kg/ton).

***Trial 4:**

In this trial feed batch was treated physically through heating in hot air oven at 87∫C for ten minutes, then cooled to the room temperature within 20 minutes.

***Bacterial enumeration:**

At intervals of 1,2,3,24,48 and 72 hours, 25 grams from each batch were aseptically weighed and suspended in 225 ml of sterile distilled water for about 10 minutes to obtain a dilution of 10^{-1}, then 0.1 ml of each suspension was then plated on MacConkey agar and salmonella shigella agar (Oxoid). Plates were then incubated for 18-24 hours at 37°C before counting and recording results.

RESULTS

The effect of different feed treatments on the number of salmonella organism is given in the following table:

Effect of different methods of feed treatment on Salmonella count.

Method of treatment	Sslmonella count/gm before treatment	Salmonella count/gm following different contact times											
		One hour		*Two hour*		*Three hour*		*24hour*		*48hour*		*72hour*	
		Count	*Red.%*	*Count*	*Red.%*	*Count*	*Red.%*	*Count*	*Red.%*	*Count*	*Red.%*	*Count*	*Red.%*
Trial 1-1	150×10^2	$9x10^2$	94.0	$1x10^2$	99.3	Nil	100.0	Nil	100.0	Nil	100.0	Nil	100.0
Trial 1-2	$155x10^2$	Nil	100.0	Nil	100.0	Nil	100.0	Nil	100.0	Nil	100.0	Nil	100.0
Trial 2	$160x10^2$	$46x10^2$	71.2	$9x10^2$	94.4	$9x10^2$	94.4	$7x10^2$	95.6	$3x10^2$	98.1	Vil	100.0
Trial 3-1	$160x10^2$	$80x10^2$	50.0	$30x10^2$	81.2	$16x10^2$	90.0	$8x10^2$	95.0	$2x10^2$	98.7	Nil	100.0
Trial 3-2	$150x10^2$	$3x10^2$	98.0	$3x10^2$	98.0	$1x10^2$	99.3	$1x10^2$	99.3	$1x10^2$	99.3	Nil	100.0
Trial 4	$170x10^2$	$20x10^2$	88.2	$12x10^2$	92.9	$4x10^2$	97.6	$3x10^2$	98.2	$1x10^2$	99.4	Nil	100.0
Control	$165x10^2$	$165x10^2$	0.0	$165x10^2$	0.0	$160x10^2$	3.0	$140x10^2$	15.1	$125x10^2$	24.2	$100x10^2$	39.4

DISCUSSION

The results obtained in the present study shows that the addition of formaldehyde 40% solution at a rate of 10L/ton was highly effective even with such high level of salmonella contamination of feed as complete reduction was achieved within the first hour of treatment and that was maintained till the end of the experimental period. The effect of the same product (formaldehyde solution 40%) did not demonstrate such a quick result when added to the feed at a rate of 5L /ton. With such concentration a reduction of 94% was obtained after one hour of exposure time, 99.3% after two hours but complete reduction of salmonella count was only possible in the sample obtained after 3 hours contact time. Duncan and Adams, 1972 used formalin fumigation as a method of decontamination but they suggested that regulatory problems might limit the use of formalin as a feed additive.

When the antibiotic preparation was used for feed decontamination in a rate of 2kg /ton it was observed that the number of salmonella organism was reduced rapidly through the first 24 hours (94.4% reduction) but more slowly thereafter. Complete reduction was obtained after 72 hours of treatment. These results are in agreement with Swezey et al., 1981 who recommended the use of oxytetracycline as a turkey feed additive for controlling salmonella, Williams, 1985 who found that the incidence of salmonella was lower in chickens fed a combination of oxytetracycline and neomycin sulfate, Barrow, 1992 and Ebner and Mathew, 2000 who recommended the use of oxytetracycline as a feed additive to reduce the fecal shedding of Salmonella typhimurium.

Regarding the use of organic acids (Formic and propionic acids) the results obtained in (trial3-1) revealed a 95% reduction within the first 24 of treatment but 72 hours were required to achieve the total decontamination of feed.

For Trial (3-2) there was a rapid drop in the number of salmonella organism after one hour of treatment, the reduction percentage was 99.3, which could be considered a relatively high percentage of reduction when compared with trial 2,3-1 and 4. Complete reduction was obtained after 72 hours of treatment. The obtained results are not agreeable with those of Hinton and Linton, 1988 who found that the same preparation had little effect on the ability to isolate very low numbers of salmonella from feed. Also, other authors had found little effect on salmonella viability in feed (Khan and Katamy, 1969;Duncan and Adams, 1972,and Vanderwal, 1979) when similar products were used for feed decontamination.

Since these authors all used naturally, as opposed to experimentally contaminated feed, this difference may account for discrepancy. However, these results are in agreement with Rouse et al., 1988 who found that addition of organic acids to

artificially contaminated feed resulted in the elimination of detectable salmonella from heavily contaminated feed within 72 hours, Iba and Berchieri, 1995 who proved that a commercial formic acid-propionic acid preparation was highly antibacterial even with high numbers of salmonella organisms in feed. They reported that the number dropped rapidly within the first two days of treatment and more slowly thereafter, although viable organisms were still detectable after 7 days.

Also, other authors had recommended the use of organic acids as a feed additive to control salmonella in poultry feed as Hinton et al., 1985,Humphery and Lanning 1988, Cherrington et al., 1991, Hinton and Mead 1991, Hafez et al., 1992, Berchieri and Barrow 1996, Thompson and Hinton 1997 and Hafez 1999.

Physical treatment of feed showed relatively satisfactory results, as there was a rapid reduction during the first 3 hours of treatment (97.6%), while complete reduction was obtained after 72 hours.

Physical treatment had been suggested by many authors as (VanCauwenberg et al., 1981, Burdick et al., 1983, Haggblom 1993, Himathongkham et al., 1996, Renggli, 1996, Matlho et al., 1997 and Archer et al., 1998) to reduce salmonella contamination of feed materials as well as the finished feed.

Regarding the control group, a small reduction in the number of organism was observed probably caused by a loss of viability due to desiccation as reported by Iba and Berchieri 1995 and Archer et al., 1998.

From the obtained results, it was clear that the use of formaldehyde solution 40% in either 5L or 10L/ton for the decontamination of feed is recommended for economical and efficiency reasons, whenever there is no legal limitations for the use of such product. On the other hand physical treatment of feed and addition of organic acids gave a relatively satisfactory results of decontamination.

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