The Effect of Antibiotic, Disinfectant and Formaldehyde gas on Hatchability of Broiler Eggs.

Thomas Junne Kaudia, MSc, Broiler Contract Manager, Kenchic Limited. P. O. Box 20052, Nairobi, Kenya.

Abstract

The present study tested: 1 The biocidal effectiveness of antibiotic, quarternary ammonium disinfectant and formaldehyde gas in disinfecting broiler hatching eggs. 2 Effectivemess of packaging eggs in polythene bags to reduce contamination and 3. Effectiveness in changing nesting materials in controlling egg shell contamination. Experimental broiler eggs were laid between 8.00 and 9.00 am. From these eggs, samples were collected hourly and disinfected using antibiotic, quarternary ammonium disinfectant and formaldehyde gas. Five egg collections were made. Antibioic and disinfectant were sprayed on the egg shell using a manual hand sprayer. Bacteriological analysis involved the determination of aerobic plate counts. Eggs were transported to the hatchery in two batches. Batch one was packed in polythene bags while batch two was left open. Saleable hatchability significantly increased to 60%, 80% and 75% for antibiotic, disinfectant and formaldehyde gas respectively compared to 6% for control that were not disinfected. There was a reduction in aerobic plate counts of 87%, 99.1% and 91.1% for antibiotic, quarternary ammonium disinfectant and formaldehyde gas respectively. Packaging in polythene bags reduced egg shell recontamination. Changing nesting materials daily did not significantly reduce egg shell contamination when eggs were collected within one hour after lay. Hazard Analysis Critical Control Points (HACCP) emerging from this study are; environment where egg is laid, collection and sanitation time after lay, method of sanitation and mode of conveying eggs from the farm to the hatchery. In conclusion, antibiotics and disinfectants have a lot of potential in sanitising hatching eggs in developing countries.

Key word: Hatching eggs, Sprayed. Disinfected, Packed in polythene bags, Hatchability, Aerobic Plate Count

Introduction

Hatching eggs are disinfected to kill micro-organisms on the surface of the shell. This allows the production of healthy chicks (Futura, 1981). Disinfection using formaldehyde gas is widespread (USDA, 1975; Futura and Sato, 1977). The study of hatching egg contamination, sanitation and hatchability has been a key issue in poultry development. Several studies have been undertaken in this respect. Futura and Sato in 1981, evaluated the effect of bacterial contamination on eggs during incubation and hatching of eggs from broiler breeders. Brax and Shelton, did some work on egg sanitation using quarternary ammonium egg sanitizer. These studies have however, concentrated only on general aspects of contamination. Very little attention has been paid to egg contamination aspects between the point of lay and the hatchery with the aim of identifying critical control points relevant to developing countries.

The average hatchability of broiler eggs in Kenya is low (68%) (Kenchic, 1994; 1994; ADS/NFU. 1978) despite using formaldehyde gas. This makes broiler day old chicks more expensive in Kenya than other countries. The present study was designed to; assess the effectiveness of antibiotics, quarternary ammonium disinfectant compared to formaldehyde gas disinfecting broiler hatching eggs, identify critical control points in handling hatching eggs between the point of lay and the hatchery. The hypohesis is that the biological effect of antibiotics and disinfectant are more or less the same as formaldehyde gas. H0 : r = 0 against HA : not all r = 0. VR being the test statistic where VR follows F distribution.

Material and Methods

Eggs

Experimental hatching eggs were laid by 37 weeks old commercial duel purpose brown broiler parents hens held at Kenchic Athi River breeding farm. The hens were reared under deep litter. They were a standard breeder ration (28%CP, 2800Kcal of ME per g). All the eggs were laid between 8.00 and 9.00 am in a nest box with fifteen laying point. The nesting material (wood shavings) was changed before laying started. The box was screened off after the laying period using an aluminium metal sheet. All the eggs were laid in the box for one hour after lay (9.00-10.00 am). Eggs were collected hourly and taken into a washed and disinfected room within the poltry shed. A total of five collections were done.

Dirty eggs with visible stains on the shell were discarded. Remaining nesting material on clean eggs shell were gently removed by hand.

Experiments

Surface Sanitation of Egg Shells

In the first experiment, lots of clean eggs were randomly arranged in clean plastic setting trays. The trays were divided into three lots of 36 eggs each. Lot 1 and 2 were sprayed with 101/l flumisol and 10ml/l quarternary ammonium disinfectant respectively. The spraying solutions were maintained at 35° C. The third lot was fumigated with formaldehyde gas. The fumigation was done at the farm next to the poultry shed in a 1m x 1m concrete chamber for 20 minutes. Formaldehyde was generated by mixing 40ml formalin (containig 372g formaldehyde/l) and 20g potassium permanganate/m^3 room capacity in a small cylindrical metal tin. The chamber had no mechanical exhaution system. At the end of the fumigation time the door was left open for five minutes before the eggs were removed. Flumisol is a synthetic anti-infective antibiotic belonging to the fluoroquonolone group, with a bactericidal activity on gram negative (*Staphylococcus*). It is used only in livestock. Spraying was done using a plastic hand sprayer.

The disinfectant used (TH4) is a third generation synergistic disinfectant for livestock farms. It is said to have virucidal, mycoplasmicidal, bacterial and fungicidal properties.

Spraying solutions were held at 35° C in one litre thermos flasks. Water used to mix the chemicals was previously boiled in an electric kettle and allowed to cool

35° C. This temperature was chosen to stop water being drawn into the egg through the pores. Spraying at the farm was done in a 1m x 2m cabin made of 1000 gauge transparent plastic sheet. Eggs were left in the cabin for 40 minutes before samples were drawn for bacterial analysis. Fumigated eggs were also kept in a similar cabin after fumigation. Surface treatment was done after fumigation. Surface treatment was done after each collection.

Transporting eggs to the Hatchery

Twenty eggs from the treated lots were randomly divided into two lots of ten eggs each. Lot one, eggs were individually packed in 9 x 15, 150 gauge polythene bags and held until the end of the experimentation.

The second lot eggs were left in the sitting trays. Both lots were transported to the hatchery at 3.00 pm.

Change of nest bedding materials

A laying nest box with fiteen laying points was used. The laying points were divided into two lots of seven each. All nesting materials were removed from the two lots and replaced with fresh clean wood shavings. Birds were allowed to lay eggs randomly in these two lots. The experiment was done for five days. Each day, nesting material was changed in the control kit by scooping off to layer of wood shavings and replacing with clean shavings.

Ten eggs were collected at random from each lot for bacterialogical analysis within 20 minutes after lay.

Bacteriological analysis

For bacteriological anaylis whole egg washing technique (Brake and Sheldon, 1990; Gentry and Quarles, 1972) was used to recover bacteria on the surface of the egg shell. Eggs were aseptically placed in sterile 10 x 20, 250 gauge polythene bags and rinsed with 20 ml 0.1% peptone water. Eggs at this stage were handled with disposable surgical hand gloves. The rinse from each egg was serially diluted in 0.1% peptone water. All counts were reported per egg by multiplying the counts per ml of rinse by 20. Eggs were sampled pre and 40 minutes post surface treatment.

Bacteriological analysis done were: General viable count plater count agar, 37°C 48 hours), presumptive coliform (Violet red bile, 37°C, 18 hours) and yeast and molds (Davis yeast salt agar acidified to a pH of 3.5, 25°C, 72 hours).

Incubation at the Hatchery

Eggs were set in a setter model Chick Master running at 37.5°C with 50-55% RH. The eggs remained in the setter for 19 days. At the end of 19th day, eggs were transferred to the hatcher model Chick Master running at 36.5°C with 65-75% RH. Hatching was done on the 21st day.

Unhatched egg analysis

Analysis of unhatched eggs was done using the Chick Embryo Development Chart (Tad Pharmazeutisches werk GMBH D-2190 Cuxhaven P.O Box 720. West Germany).

Statistical analysis

Analysis of variance (ANOVA) statistical method was used to compare the sample means for test significance on a randomized complete block design (Snedecor, G.W., and Cochran, W.G. (1967); Raghuramulu, et al. (1983).

Results and Discussion

Hatchability

Application of 10ml/1 flumisol, 10ml/1 quaternary ammonium disinfectant and formaldehyde gas significantly increased the saleability hatchability of eggs in the 37 week old flocks to 60%, 80% and 74% respectively compared to 6% for control (**Table 1**). These result contrust those Brake and Sheldon, 1990 in which control egg samples hatched between 80% and 90%.

A drop in hatchability was noted in eggs disinfected 3, 5 and 2 hours of anibiotic, disinfectant and formaldehyde gas respectively **(Table 1)**. The drop in hatchability was more gradual in fumigated eggs than in surface sprayed eggs. Hatching eggs should be collected and disinfected within two hours of lay for maximum hatchability.

Egg Contamination

Table 2 shows average number of bacteria aerobic plate count) on clean egg shell surfaces. Contamination was in the tune of 10^3 to 10^4 bacterial colonies. Furuta and Murayama, 1980 isolated 10^3 and 10^4 bacterial colonies from clean and dirty eggs respectively. Contamination levels increased with time after lay. According to Board (1966) a contamination levels between 10^4 and 10^5 is not usuall. Contaminations however, depends on which the eggs are produced. Carter et al (1971, 1973) observed different levels of contamination between eggs produced on litter and those produced on wire floors. In view of the increase in contamination after lay, hatching eggs be disinfected

Table 1. Hatchability of control and sanitised eggs

Treatment	Hatchabiliity (out of 10 eggs) based on collection tiome (Hours)					Total hatchanalysis			
	1	2	3	4	5	No	%	Cull	%Sh
Control	2	2	1	1	0	6	12	3	6
Antibiotic	10	10	7	4	2	33	66	3	60
Disinfectant	10	10	10	10	5	45	90	5	80
Formaldehyde	10	8	10	7	7	42	84	5	74

SH- Saleable Hatch

Table 2. Aerobic plate count on control samples

Microbiological test hours	Aerobic plate counts based on sampling time in				
	1	2	3	4	5
Viable counts	1202	2100	4000	5760	6080
Presumptive coliform	6160	7200	9600	10408	14080
Yeast and mold	0	0	60	20	0

within two hours after lay.

Biocidal effect

Antibiotic, quaternary ammonium disinfectant and formaldehyde treatments reduced aerobic contamination levels by 87.7%, 99.8% and 91.1% respectively as indicated by viable count results (**Table 3 and Table 4**). The results agree with the findings of Brake and Sheldon, 1990, in a study using quartenary ammonium sanitizer for hatching eggs. Both researchers achieved a significant reduction of aerobic plate counts within 30 minutes of application.

The effect of the treatments on presumptive coliform are summarised in (Table 4.) Coliform were in general 60-85% of the total aerobic plate count. The counts however, deceased to the levels of 2.6%,0% and 1.2% after treatments with antibiotics, quartenary ammonium compound and formaldehyde gas respectively.

The pattern of yeast and molds contamination of egg shell surfaces could not be established from these studies (**Table 2**). Yeast and mold are not major causes of egg spoilage (Fraizier and Westhoff, 1988).

Packaging in polythene bags

Packaging in polythene bags reduced recontamination of egg shells after sanitation. Shells of unpacked eggs were - thus heavily re-contaminated after sanitation (**Table 5**). The levels of recontamination was higher with both antibiotic and disinfectant compared to formaldehyde gas (**Table 5**). This is due to wetness of the surface due to spraying making dust stick on the egg shell. Bacterial contamination compared to level 40 minutes after disinfection (**Table 5**). This indicates that residual antibacterial effect of the treatments continued in packed samples. This is another advantage of packaging hatching eggs in polythene bags as it not only stops recontamination, but also creates a mico-environment in which bacterial activity dis-continues. Hatching eggs should therefore be packed in polythene bags while on transit form the farm to the hatchery.

Change of nesting material (Wood Shaving)

Table 3. Biocidal effect of sanitation on viable counts

Treatment	Reduction in viable counts per sampling time				
	1	2	3	4	5
Control	1202	2080	4000	5760	6080
Antibiotic	70 93.8%	75 96.4%	200 95%	802 86.1%	1200 80.4%
Disinfectant	0 100%	0 100%	0 100%	0 100%	60 99.6%
Formaldehyde gas	66 94.6%	80 96.2%	150 96.3%	591 97.4%	800 86.8%

Table 4. Biocidal effect on coliform counts

Treatment	Reduction in coliform counts per sampling time				
	1	2	3	4	5
Control	6160	7200	9600	10400	14080
Antibiotic	0 100%	0 100%	0 100%	0 100%	60 80.4%
Disinfectant	0 100%	0 100%	0 100%	0 100%	0 100%
Formaldehyde gas	0 100%	0 100%	0 100%	0 100%	10 99.9%

Table 5. Changes in aerobic viable counts due to packaging

Treatment	Viable counts per sampling time (hrs)				
	1	2	3	4	5
Control	1202	2100	4000	5760	6080
Antibiotic	70	75	200	802	1200
Open	*3600	*1600	*1100	*100	*60
packed	10	30	35	60	80
Disinfectant	0	0	0	20	20
Open	*1200	*800	*800	*100	*110
Packed10	0	0	20	80	
Formaldehyde	60	80	150	600	800
Open	*800	*400	*260	*388	*400
packed	10	40	80	70	30

Key: *= Control

Table 6. Effects of nest materials on aerobic plate counts

	Aerobic plate count per sampling time in days				
	1	2	3	4	5
Viable counts	*1080 1080	*870 1120	*960 1144	*1100 1357	*1030 1560
Presumptive Coliform	*5800 5800	*4870 6680	*4620 9800	*6000 11200	*5860 12600
Yeast and Mold	*0 0	*10 5	*5 0	*0 20	*0 36

Key: * = Control

Table 6 shows that the level of bacterial contamination increase when the nesting material is not changed. The level of contamination is however, lower than eggs left longer before collection and disinfection. This further emphasises the need to collect and disinfect hatching eggs immediately after lay. This observation disagrees with the findings of Kirk *et al.*, 1980 who indicated that collecting eggs hourly rather than after five hours after lay reduces hatchability.

Conclusion

The following conclusions can be drawn from this study.

1. Hatching eggs should be collected and disinfected hourly after lay.

2. Leaving hatching eggs uncollected in the nest boxes leads to increase in bacterial contamination resulting in low hatchability.

3. Hatching eggs be packed in polythene bags before transporting to the hatchery from the farms.

4. Where it is not possible to change nest materials once per week, hatching eggs be collected and disinfected hourly.

5. Quarternary ammonium disinfectants can be used to disinfect eggs instead of formaldehyde gas.

6. Use of antibiotics in disinfecting hatching eggs require further investigations.

7. Four Key Hazard Analysis Critical Control Points have emerged from this study.

These are:

(a) Hatching egg collection time
(b) Hatching egg disinfection time
(c) Packaging hatching eggs in polythene bags during transportation.
(d) Training and capacity building for personnel handling hatching eggs.

References

Animal Industry Bureau. 1975 *The Guidance of Hatcher Sanitation*. Tokyo, The Japanese Ministry of Agriculture and Forestry.

Brake, J. and Cheldo, B. W. 19990. Effects of a quarternary ammonium sanitizer for hatching eggs on their contamination, permeability, water loss and hatchability. *Poultry Science*, 69: 517-525.

Chemical Engineering News, 1984.

Ernst, R. A Shroeder, J. P, Pfost, R. E. and Holte, R. J. A 1984. Field studies to evaluate commercial disinfectants for turkey hatcjing eggs sanitation. *Poultry Science*, 53: 149-156.

Frazier W. C., and Westhoff, D. C. 1998. *Food Microbiology 4th edition* McGraw-Hill Book company, Singapore.

Futura, K., and Sato S. 1977. Studies on the disinfection of hatching during incubation and hatching, and fluffs of newly-hatched chicks. *British Poultry Science*, 22: 247-254

Futura, K and Sato S., 1977. Studies on the disinfection of hatching eggs 2nd Edition. The effect of formaldehyde fumigation on bacteria contaminating the egg shell surface. *Japanese Poultry Science*, 14:27-32.

Gentry, R. F. and Quarles, C.L. 1972. The measurement of biological contamination on egg shells. *Poultry Science*, 51: 930-933.

Kenchic Ltd. 1984. Hatchery management report. December, 1984.

Kirk, S., Emmans, G. C., McDonald R., and arnot, D., 1980. Factors affecting hatchability of eggs from broiler breeder. *British Poultry Science*, 21: 37-53.

Raghuramulu, N., Nair, K. M., and Kalyanasundaram, S., 1983. *A manual of Laboratory Techniques*. National Institute of Nutrition. India Council of Medical Research, Jamai-Osmania, hyderabad-500 007 A. P. India

Snedecor, G. W., and Cochran, W. G. 1967. *Statistical Methods-6th edition*, Oxford and I.B.H Publishing Co.

Tad Phrmazeutisches werk GMBH D-2190 cuxhaven P. O. Box 720. Western Germany

Taylor, T. G. 1970. How an egg is made. *Sci. Am*. 222:88-95.

United States Department of Agriculture, 1975. The National Poultry Improvement Plan and Auxiliary Provisions. Washington D.C.

Walker, J. R. 1944. *Formaldehyde*. Ronald Publishing Corp., New York, NY.

Williams, J. E. 1970. Effect of high level formaldeyde fumigation on bacterial population on the surface.