

Determination of Most Effective, Chemical Disinfectant for *Salmonella Gallinarum* in Poultry Industry

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Introduction

Poultry industry has been fully integrated in Sri Lanka during last two decades and the overall production and consumption has increased. With the improvement of the industry, occurrence of diseases has increased and Salmonellosis is reported as one of the prevalent diseases. Fowl Typhoid (FT), also known as Klein's disease, is caused by *Salmonella gallinarum*, and is host specific for chicken and turkey. FT creates serious economic losses to producers, since it affects on any stage of chickens. Chicks infected through trans-ovarian transmission experience to high mortality. Vaccination against FT is only allowed for commercial layers, in Sri Lanka, but with strict post vaccination monitoring. Hence, poultry producers are on constant look out for better and innovative remedy. The bacterium is fairly resistant to normal climate, surviving for months, but is susceptible to normal disinfectants (McMullin, 2004). Therefore, prevention of microbial contamination is one possible solution and can be achieved through proper disinfection which is one of the important practices in every unit of poultry industry. Most importantly, disinfection offers long-term benefits and have combined properties of detergents and sanitizers. Disinfectant chemicals act to disrupt cellular structures or processes in order to kill or eliminate microorganisms. A number of different chemicals have been commercialized as disinfectants and those can be grouped as alcohols, halogens, quaternary ammonium compounds, phenolics, aldehydes, oxidizing agents and other. Not all disinfectants are suited for every situation. Therefore, evaluation of different disinfectants against infective agents may result benefits. Many disinfectants of similar composition are sold under different trade names (Saif, 2003). Therefore, five farm-use-disinfectants which represent five chemical groups were selected and tested to identify most effective concentration-contact time combination of highest effective disinfectant, for *Salmonella gallinarum*.

Methodology

The study was carried out at the Microbiology Laboratory of Quality Control Department, at Ceylon Grain Elevators Limited, Colombo 15, Sri Lanka.

Tested disinfectants are Chlorine releasing compound (Sodium-N Chloro-P Toluene sulphonchloramide 100% powder), Iodine compound (2.5% titrable Iodine), Quaternary Ammoniums 12.5% + Gluteraldehyde 6.25% compounds, Quaternary Ammonium compound (80% active QAC) and an Oxidizing agent (Di potassium peroxidisulphate). Stock culture sample of *Salmonella gallinarum*, which had been stored at microbiology laboratory were used as the source of bacterial organism. Young cultures were prepared from stock cultures and confirmation test was performed for the test organism. Tryptone Soya Agar (TSA) was used as culture media and Buffered Peptone (BP) broth was used for preparation of primary inoculums. 0.87% saline was used to prepare bacterial suspensions.

Bacterial suspensions were prepared similar to McFarland Standards 1 ($=3 \times 10^8$ CFU/mL) and 1 mL was introduced into 9 mL BP to prepare primary inoculum. Six primary inoculums were prepared to test five disinfectants and for the control. The test was carried out in two steps. In the first step, manufacturer recommended concentrations of disinfectants were prepared and tested for four contact times as 2, 8, 18 and 28 minutes. For each treatment, three replicates were used. Disinfectants, which eliminated *Salmonella gallinarum* completely from culture plates were selected for the second step. In the second step, three concentrations; Manufacturer recommended concentration, Half of manufacturer recommended concentration and Twice of manufacturer recommended concentration were prepared for each disinfectant and those were tested for three contact times as 2, 10 and 18 minutes. Surviving bacterial number was enumerated by total bacterial colony count in incubated culture plates. Statistical analysis was performed by using Two Factor Factorial Design and Descriptive Statistics (MINITAB 14) to compare treatments to evaluate most effective concentration-contact time combination.

Results and Discussion

As observed in the first step, Quaternary Ammoniums 12.5% + Gluteraldehyde 6.25% compound and Quaternary Ammonium compound (80% QAC) have completely eliminated *Salmonella gallinarum* in culture plates, while other compounds remained with significantly higher number of surviving bacterial counts in the cultured plates. Therefore, those two disinfectants were used for further testing.

Table 1: Results of step 02

Disinfectant type	Actual concentrations of disinfectants	Means of TBC counts with tested contact times		
		Contact time (minutes)		
		2	10	18
1	0.002 v/v	194	0	0.67
1	0.001 v/v	233.67	0	0
1	0.0005 v/v	250	68.33	0**
2*	0.01 v/v*	0	0	0
2	0.005 v/v	1	0	0.33
2	0.0025 v/v	110.67	24	0.33**

(Disinfectant 1 = Quaternary Ammonium compound; Disinfectant 2 = Aldehydes + Quaternary Ammonium compounds) (*highest effective disinfectant, **contact times for low cost combinations)

According to the descriptive statistics, twice of manufacturer recommended concentration of Quaternary Ammoniums 12.5% + Gluteraldehyde 6.25% compound (0.01 v/v) has recorded the highest effective concentration. The higher effectiveness of this disinfectant compared to 'Disinfectant 1' may have resulted by the combined effect of both active ingredients, on eliminating *Salmonella gallinarum*. As revealed by the second experiment, 2 minutes of contact time of the above concentration was successful in complete elimination of *Salmonella gallinarum*. Therefore, 2 minutes contact time was taken as the highest effective contact time for that concentration. Additionally, according to the statistical analysis, lowest tested concentrations of 80% Quaternary Ammonium compound (0.0005 v/v) and Quaternary Ammoniums 12.5% + Gluteraldehyde 6.25% compound (0.0025 v/v) have effectively reduced Colony Forming Units in the contact time of 18 minutes and those indicate most cost effective combinations for users.

Conclusion

The disinfectant, which contains Quaternary Ammoniums 12.5% + Gluteraldehyde 6.25% compounds as active ingredients are found to be the highest effective disinfectant and the most effective concentration-contact time combination for completely eliminate *Salmonella gallinarum* was reported as 0.01 v/v and 2 minutes contact time.

References

- McMullin, P., 2004. A Pocket Guide to Poultry Health and Diseases, 1st edition. 5M Enterprises Limited, Sheffield, United Kingdom. 182-183P.
- Saif, Y.M., 2003. Diseases of poultry, 11th edition. Blackwell Publishing, Ames, Iowa. 39-43P.