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Efficacy of Some Commercial Chemical Disinfectants on *Salmonella enterica Serovar Typhimurium*

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Abstract: Problem statement: Poultry industry is intensive and consistently applies an all-in, all-out system with the aim of minimizing infection pressure and targeting specific pathogens like *Salmonella* which remains one of the leading causes of food-borne illness, many questions regarding the introduction and persistence in animal production still remain. Therefore disinfection during production break is a routine part of the biosecurity programs of poultry houses. The correct usage of disinfectants is an important key of a successful biosecurity program in poultry farms and in-turn the role of the scientist was to evaluate the efficacy of these disinfection programs. **Approach:** In this study five commercial disinfectants [Green work (green non anionic surfactant), Sanidate RTU (hydrogen peroxide compound), Hi-yeild®consan 20® (phenolic compound), Tektrol® (quaternary ammonium compound) and Kreso®D (phenolic compound)] were evaluated against *Salmonella typhimurium* in two different experimental conditions. In Experiment I, *S. typhimurium* was inoculated into fresh poultry litter (aluminum trays L: 30 cm × W: 25 cm × D: 6 cm filled with wood shavings) by inoculums size of $\sim 10^7$ CFU mL^{-1} and then mixed with 100 g of fresh poultry droppings. Sample sizes of 3 g were obtained daily for the bacterial counts. Green work achieved100% killing of *S. typhimurium* by day 7 ($p \le 0.0001$); Sanidate RTU achieved100% killing by day 6 ($p \le 0.001$); Hi-yield® Consan®, Tektrol® and Kreso® D achieved100% killing by day 5 ($p \le 0.001$). Disinfectants were also compared to each other in their efficacy each day. At day 1, Green work was inferior to all other disinfectants at ($p \le 0.05$). On day 2, Kreso® D was significantly superior to Tektrol®, Hi-yield® Consan®, Sanidate RTU and Green work at $p \le 0.01$, $p \le 0.01$, $p \le 0.01$, $p \le 0.005$; respectively. At day 4 Kreso® D was significantly superior to Hi-yield® Consan® at $p \le 0.01$, Tektrol® was also significantly superior to Green work at $p \le 0.01$. In experiment II; MIC use-dilution test was used to evaluate the five disinfectants against *S. typhimurium* ($\sim 10^7$CFU mL^{-1}) in the absence of organic matter. **Results:** Hourly samples were collected for the bacterial counts. Maximum efficacy (100% killing efficacy against *S. typhimurium*) was achieved for Green Work after 16 h ($p \le 0.0001$), with Sanidate RTU after 8 h ($p \le 0.0001$), with Hi-yield® Consan® and Kreso® D after 2 h at ($p \le 0.0001$) and with Tektrol® after 4 h ($p \le 0.0001$). In presence of organic matter Green work and Sanidate RTUachieved 100% killing efficacy against *S. typhimurium* after 16 h ($p \le 0.0001$), Hi-yield® Consan® and Kreso® D after 2 h at ($p \le 0.0001$); Tektrol® after 8 h ($p \le 0.0001$). When disinfectants were compared to each other in relation to time; we found that there was no kind of significance between their efficacies. When compared to other tested disinfectants, Kreso® D which is a phenolic compound revealed superior activity against *Salmonella typhimurium* in the two experiments. **Conclusion:** The study showed that many disinfectants regardless to their constituents continues to give a very powerful efficacy against the most virulent bacterial strains, but the question remain can they be used in the presence of live birds. Further studies are required to explore the safety and the efficacy of these compounds when applied in poultry farms in the presence of live birds.

Key words: Disinfectants, *Salmonella typhimurium*, poultry, organic matter

INTRODUCTION

The objective of disinfection is to reduce microbial population[6]. Disinfectants act on microorganisms at several target sites resulting in membrane disruption, metabolic inhibition and lysis of the cell[4,14]. Removal of old litter followed by cleaning and disinfection of facilities helps reduce pathogen numbers and break

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disease cycles or at the minimum, keep pathogen numbers from reaching a level that can cause disease outbreaks. In addition, as live production becomes the target area of programs for the reduction of human pathogens such as *Salmonellae* on poultry carcasses, it will become necessary to document that sanitation procedures are effective. Unfortunately, poor sanitation procedures and/or increased soil moisture levels have been linked to increased or sustained bacteria levels[18,24].

Several studies were carried out on disinfectants and many of these disinfectants are not considered to be environmentally safe e.g., gluteraldhyde, formaldehyde to show their effectiveness against *Salmonella*[7,8,21]. Further, poultry houses have inaccessible equipment and considerable amounts of organic matter and high contents of protective compounds (fats, carbohydrates and proteins) from which *Salmonella* are difficult to remove[8]. On the other hand, water hardness, low temperature and biofilm development also decrease efficacy of disinfectants[8,12,25].

Disinfectant efficacy is often tested against laboratory bacterial suspensions[1,16]. However, this approach may not always prove to simulate commercial production conditions, thus, making it difficult to determine the true effectiveness of the disinfectant. Disinfectants that are effective against bacterial suspensions may have a reduced effect against bacteria that adhere to surfaces[15].

The main objective of this study was to compare the efficacy of some new commercial disinfectants against *S. typhimurium*, in the presence or absence of organic matter as an extra-challenge for the disinfectants.

MATERIALS AND METHODS

Propagation of *Salmonella typhimurium*: *S. typhimurium* (ATCC 1331) genomic DNA strain NCTC74 was propagated and counted using drop plate technique, Zelver *et al*.[27] and Herigstad *et al*.[10]. The procedures were carried out by pipetting 1 mL of bacterial suspension into a dilution tube containing 9 mL of tetrathionate broth; making dilution 10^1. Tenfold serial dilutions were made to obtain dilutions of 10^2,10^3, 10^4, 10^5, 10^6, 10^7 and 10^8 mL^{-1}. Bacterial count in each dilution was obtained by inoculating on CHROMagar *Salmonella* plates (Becton-Dickinson, VMR Int.) The plates were incubated overnight for 17-20 h at 35-37°C. Viable cell counts were expressed as CFU/surface area. The calculation was carried out using the following formula:

Log (average CFU/drop vol.) (dilution factor) (Vol. scrapped into/surface area)

Biochemical identification: The biochemical identification of *Salmonella* was carried out using automated method (MICROSCAN auto SCAN4, Dade Behring), which confirmed that the suspension was positive for Glucose, Lysine, Citrate utilization, Rafinose, Hydrogen Sulphid, Sorbitol, Arabinose, Meltonin and Ornithin.

Experiment I: Efficacy of chemical disinfectants against Salmonella typhimurium under conditions simulating naturally ventilated poultry houses.

Inoculation of the litter with *Salmonella typhimurium*: Six trays of aluminum foil (L: 30 cm × W: 25 cm× D: 6 cm) were filled with litter (wood shavings). All the trays were sterilized by autoclaving at 121°C for 1 h. Sterilization was confirmed by placing 25 g of autoclaved litter into 225 mL of buffered peptone water (BPW; Oxoid, Fisher Scientific Int.) and incubated in rotatory incubator for 3 h; followed by spread plating on CHROMagar *Salmonella* plates (BD, VMR Int.). All trays were incubated at 37°C for 20-24 h and then the colonies were enumerated.

Autoclaved fresh poultry droppings were added to the trays as a challenge to the chemical disinfectants at rate of 100 g tray^{-1}. The trays were labeled, one for each disinfectant, then the six trays were inoculated with *S. typhimurium* suspension (4.8×10^7 CFU mL^{-1}), five trays were used for treating with the disinfectants and the last one was used as control.

Application of the chemical disinfectants:
The following disinfectants were chosen to be tested against *S. typhimurium*: Hi-Yield® Consan20® (Parkway Research Corp., Houston, TX) a mixture of quaternary ammonium compounds as 20 with 80% inert ingredients. The recommended dilution rate was 1 floz per 30 gallon water (1.85 mL of the disinfectant was added to 483 mL of distilled water, pH 9.22).

KRESO®D (Elmwood Park Station, Omaha) a mixture of coal tar neutral oil, coal tar phenols, soap and water. The recommended dilution rate was 1 part of KRESO®D to 72 parts warm water (5 mL of the disinfectant was added to 360 mL distilled water, pH 9.57).

TekTrol® (ABC Compounding Co, Int., Atlanta, GA) is a mixture of Paratertiary Amylphenol (40%), Orthobenzyl Para Cholorophenol (10%), Orthophenyl phenol (12%) and inert ingredients (74%). The recommended dilution rate was 15 cc per one gallon water (1ml of the disinfectant was added to 258 mL distilled water, pH 10.20).

Sanidate (RTU) a ready to use suspension that clean and sanitize with a green chemistry and chlorine alternative that is effective and works on contact. This disinfectant utilizes activated peroxygen chemistry, which is highly stable chemical technology utilizing hydrogen dioxide, peroxyacetic acid and proprietary stabilizers and buffers, pH 4.75.

Green Works (Clorox Comp., Oakland, CA) is a non anionic surfactant (alkyl polyglucoside) and some essential oils with citric and lactic acid. It has an extraordinary cleaning power without harsh chemical fumes or residues. It was diluted at the rate of 1:1 (200 mL of the disinfectant was added to 200 mL distilled water, pH 2.40).

All the disinfectants were applied using a sprayer bottle of 200 mL capacity; 50 mL of each disinfectant was sprayed on its specific litter tray. The time required by each disinfectant to kill *S. typhimurium* population in the presence of organic matter was measured by following the decline in *S. typhimurium* count.

Collection of the litter samples: Three samples each of three grams were collected from each inoculated aluminum tray and the control tray through the whole depth of the litter on a daily basis. The samples were added to 27 mL Phosphate Buffered Saline (PBS). They were allowed to stand for 20-25 min and then the mixture was filtered using filter paper (7 cm diameter)[26]. The ambient temperature, relative humidity percentage of the atmosphere and litter pH were recorded daily throughout the length of the experiment.

Salmonella Typhimurium count: The filtrate was used for obtaining bacterial count using the drop plate technique with CHROMagar *Salmonella* plates[25] and Herigstad *et al*.[10] as described previously. Viable cell counts were expressed as CFU/surface area. The calculation was carried out using the following formula:

Log (average CFU/drop vol.) (dilution factor) (Vol. scrapped into/surface area)

Experiment II: Laboratory evaluation of the chemical disinfectants using modified use-dilution test Robinson *et al*.[22]. The test was repeated twice; once in the presence of organic matter and the second time in the absence of the organic matter.

Evaluation of the efficacy of the chemical disinfectants in the presence of organic matter: Tenfold serial dilutions were carried out for each disinfectant in 15 mL tubes using PBS to obtain the dilutions of 10^1, 10^2, 10^3, 10^4 and 10^5 mL^{-1}. About 0.5 mL^{-1} of the bacterial suspension (4.8×10^7 mL^{-1}) was added to 4.5 mL organic matter suspensions (fecal droppings 5%). One mL of the bacterial-organic matter suspension was added to the tubes containing 9 mL of each of the disinfectant dilutions. The treated tubes were votexed. One mL was transferred from the bacterial/organic matter-disinfectant tubes to 15 mL tubes containing 9 mL glycine 1.2% (neutralizer) at 2, 4, 8 and 16 h from the zero time of bacterial-OM mixture inoculation to the disinfectant dilutions. *Salmonella typhimurium* was counted on chromagar *Salmonella* plates using plate drop techniques Zelver *et al*.[25] and Herigstad *et al*.[10] as described previously. Viable cell counts were expressed as CFU/surface area. The calculation was carried out using the following formula:

Log (average CFU/drop vol.) (dilution factor) (Vol. scrapped into/surface area)

Evaluation of the efficacy of the chemical disinfectants in the absence of organic matter: Tenfold serial dilution was carried out for each disinfectant in 15ml tubes using PBS to obtain the dilutions of 10^1, 10^2, 10^3, 10^4 and 10^5 cfu mL^{-1} 0.1 mL (100 µL) of the bacterial suspension (4.8×10^7cfu mL^{-1}) was added to the tubes containing 10 mL of the disinfectant dilutions. The treated tubes were votexed. One mL was transferred from the bacterial-disinfectant tubes to 15 mL tubes containing 9 mL glycine 1.2% (neutralizer) at 2, 4, 8 and 16 h from the zero time of bacterial-OM mixture inoculation to the disinfectant dilutions.

Salmonella typhimurium was counted on CHROMagar *Salmonella* plates using plate drop techniques Zelver *et al*.[25] and Herigstad *et al*.[10] as described previously. Viable cell counts were expressed as CFU/surface area. The calculation was carried out using the following formula:

Log (average CFU/drop vol.) (dilution factor) (Vol. scrapped into/surface area)

Statistical analysis: The statistical analysis was carried out by performing Analysis Of Variance (ANOVA, GLM, MIXED) using SAS 9.2.0 software.

RESULTS

Efficacy of the tested chemical disinfectants on the survival of *Salmonella Typhimurium* in poultry litter: The objective of this study was to evaluate the efficacy of some new commercial disinfectants some of which are considered environmentally safe and possibly could be used in poultry houses while the birds are still present.

Table 1: Efficacy of chemical disinfectants against *S. typhimurium* in poultry litter ($\log_1$ CFU g^{-1} and killing percent during each day of the 7 days experimental period) in experiment I

Days	Parameter	Green work	Sanidate	Consan 20	Tektrol	Kreso D	Control
1	Log_1 count	7.50^{aA}	6.45^{aB*}	6.25^{aB}	5.46^{aB}	5.44^{aB}	7.59^{aB}
	Killing (%)	32.50	94.04	96.18	99.41	99.45	10.41
2	Log_1 count	7.09^{aA}	6.12^{abA}	5.75^{abA}	5.30^{aA}	5.04^{bc*B*}	7.58^{abB}
	Killing (%)	74.79	97.18	98.82	99.69	99.76	10.41
3	Log_1 count	6.72^{abA}	5.72^{abcA}	5.52^{abA}	4.81^{abA}	4.40^{bcA}	7.59^{abA}
	Killing (%)	88.18	98.88	99.30	99.85	99.92	14.58
4	Log_1 count	6.45^{abA}	5.36^{bcd*AB}	5.20^{bc*AB}	4.58^{bc*BC*}	4.33^{cd*C*}	7.51^{bc*C}
	Killing (%)	94.04	99.51	99.66	99.91	99.95	20.83
5	Log_1 count	5.86^{b**A}	5.08^{cde**A}	0.00^{cd**A}	0.00^{cd*A}	0.00^{de*A}	7.49^{cd**A}
	Killing (%)	98.48	99.74	100.00	100.00	100.00	25.00
6	Log_1 count	4.68^{bcA}	0.00^{e**A}	0.00^{d**A}	0.00^{d**A}	0.00^{e**A}	7.49^{cdA}
	Killing (%)	99.89	100.00	100.00	100.00	100.00	27.08
7	Log_1 count	0.00^{c**A}	0.00^{e**A}	0.00^{d**A}	0.00^{d**A}	0.00^{eA}	7.47^{d*A}
	Killing(%)	100.00	100.00	100.00	100.00	100.00	

*: Represents values with significance at $p \le 0.001$; **: Represents values with highly significance at $p \le 0.0001$; $^{a,\ b,\ c,\ d\ and\ e}$: Represented the significance in each disinfectant through 7 days. $^{A,\ B\ and\ C}$: Represented the significance between the five disinfectants in each day through 7 days

Table 2: pH of the litter treated with each chemical disinfectant during the 7 day experimental period in experiment I

Days	Kreso D	Tektrol	Consan 20	Sanidate	Green work	Control
1	8.07	8.58	8.50	8.90	7.95	8.07
2	8.07	8.31	7.68	8.15	8.05	8.00
3	7.86	8.23	7.81	7.89	7.81	7.86
4	7.45	7.68	7.39	7.71	7.56	7.51
5	7.35	7.57	7.24	7.35	6.39	6.68
6	7.22	7.38	7.03	7.19	6.20	6.46
7	7.09	7.21	6.92	6.99	6.13	6.31

Table 3: Efficacy of chemical disinfectants against *S. typhimurium* in a laboratory trial in the absence of organic matter ($\log_1$ CFU g^{-1} and killing percent during 24 h experimental period)

		Time (h)			
Disinfectants	Parameters	2	4	8	16
Green work	Log_1 count	6.05^{a**A}	5.84^{b**A}	4.71^{c**A}	0^{d**A}
	Killing (%)	97.50	98.66	99.88	100
Sanidate	Log_1 count	5.18^{a**A}	4.64^{b**A}	0^{c**A}	0^{cA}
	Killing (%)	99.62	99.90	100	100
Consan 20	Log_1 count	0.00^{a**A}	0.00^{aA}	0^{aA}	0^{aA}
	Killing (%)	100.00	100.00	100	100
Tektrol	Log_1 count	4.55^{aA}	0.00^{b**A}	0^{bA}	0^{bA}
	Killing (%)	99.97	100.00	100	100
Kreso D	Log_1 count	0.00^{a**A}	0.00^{aA}	0^{aA}	0^{aA}
	Killing (%)	100.00	100.00	100	100

*: Represents values with significance at $p \le 0.001$; **: Represents values with highly significance at $p \le 0.0001$; $^{a,\ b,\ c\ and\ d}$: Represented the significance in each disinfectant through 7 days. ^{A}: Represented the significance between the five disinfectants in each day

In this experiment (experiment I), the object was to compare the efficacy of these disinfectants in poultry litter with organic matter during 7 days of the experiment. The data are shown in Table 1 as CFU g^{-1}.

The results showed superiority in action that was obvious in case of Kreso®D, Tektrol® and Hi-yield® Consan® when compared with the other disinfectants that were used in the experiment. pH of the litter didn't show any significant level which can be reflected to thr effect of the chemical disinfectants of *Salmonella Typhimurium*, (Table 2).

Table 4: Efficacy of chemical disinfectants against *S. typhimurium* in a laboratory trial in the presence of organic matter ($\log_1$ CFU g^{-1} and killing percent during 24 h experimental period)

		Time (h)			
Disinfectants	Parameters	2	4	8	16
Green Work	Log_1 count	6.46^{a**A}	6.08^{b**A}	4.21^{cA}	0^{d**A}
	Killing (%)	93.75	97.66	99.62	100
Sanidate	Log_1 count	6.47^{a**A}	5.90^{b**A}	5.11^{c**A}	0^{d*A}
	Killing (%)	94.58	98.33	99.73	100
Consan 20	Log_1 count	0.00^{a**A}	0.00^{aA}	0.00^{aA}	0^{aA}
	Killing (%)	100.00	100.00	100.00	100
Tektrol	Log_1 count	4.93^{aA}	4.50^{b**A}	0.00^{bA}	0^{bA}
	Killing (%)	99.81	99.92	100.00	100
Kreso D	Log_1 count	0.00^{a**A}	0.00^{aA}	0.00^{aA}	0^{aA}
	Killing (%)	100.00	100.00	100.00	100

*: Represents values with significance at $p \le 0.001$: **: Represents values with highly significance at $p \le 0.0001$; $^{a,\ b,\ c\ adn\ d}$: Represented the significance in each disinfectant through 7 days. ^{A}: Represented the significance between the five disinfectants in each day

Efficacy of the chemical disinfectants against *Salmonella Typhimurium* in the laboratory in the presence and absence of organic matter: In this experiment (experiment II), the object was to compare the efficacy of these disinfectants in the laboratory without poultry litter and with or without organic matter for 24 h. The data are presented in Table 3 and 4 as CFU g^{-1}. The data reflect nearly the same effect for the superiority of the phenolic and quaternary ammonium compounds by killing *Salmonella Typhimurium* in faster time and higher significant when compared with other tested chemical disinfecnats.

DISCUSSION

Commercially available disinfectants are not classified as broad spectrum agents. Multiple factors should be considered when a disinfectant is chosen, such as organic matter on the surface to be treated, presence of organic matter in the diluents, quality of water, corrosiveness or toxicity of the product, application method, temperature, porosity of the surface being treated, length of contact time, infectious organisms targeted, susceptibility of the infectious organisms and correct dilution[17,19].

It is well known that elimination of *Salmonella* from poultry houses is a difficult task[3,9]. The main risk of *Salmonella* contamination of poultry flocks are the *Salmonella* status of the previous flock[23] *Salmonella* status of day old chicks[2] contaminated litter, feed and water[11] presence of contaminated carriers[3]; rodents, flies and beetles and inadequate disinfection of abattoir trucks[21].

Data showed that Green work was effective against *S. typhimurium* starting at day 5 ($p\leq0.001$) with killing ratio of 98.48% and showed 100% killing efficacy on day 7 ($p\leq0.0001$). Sanidate RTU was effective starting at day 4 ($p\leq0.05$) with killing ratio of 99.51 and showed 100% killing efficacy starting at day 6 ($p\leq0.001$) and day 7 ($p\leq0.0001$). Hi-yield® Consan® was effective starting at day 4 ($p\leq0.05$) with killing ratio of 99.66% and showed 100% killing efficacy at day 5 ($p\leq0.001$), day 6 ($p\leq0.0005$) and day 7 ($p\leq0.0001$). Tektrol® was effective starting at day 4 ($p\leq0.05$) with killing ratio of 99.91% and showed 100% killing efficacy at day 5 ($p\leq0.001$), day 6 ($p\leq0.0001$) and day 7 ($p\leq0.0001$). Kreso® D was effective starting at day 2 ($p\leq0.05$) with killing ratio of 99.76% and showed 100% killing efficacy at day 5 ($p\leq0.001$), day 6 ($p\leq0.0005$) and day 7 ($p\leq0.0001$).

The disinfectants were compared to each other in their efficacy each day and the results showed that on day 1 there was a significant difference between Sanidate RTU and Green work ($p\leq0.05$). On day 2 there was a significant difference between Kreso® D and Tektrol®, Hi-yield® Consan®, Sanidate RTU, Green work ($p\leq0.01$), ($p\leq0.01$), ($p\leq0.01$), ($p\leq0.005$), respectively. On day 4 there was a significant difference between Kreso® D and Hi-yield® Consan® ($p\leq0.01$), Tektrol® and Green work ($p\leq0.01$). On day 3, 5, 6 and 7 there were no significant differences between the efficacy of the disinfectants and each others.

The intention of disinfectant programs in poultry facilities is to reduce the pathogenic micro-organisms. However, if disinfectants are used without properly cleaning the facility prior to application, then the effectiveness of the disinfectant may be compromised. Organic matter provides a physical barrier that protects microorganisms from contact with the disinfectants[5]. In this study both Green work and Sanidate RTU showed delayed action. Although they are environmentally safe their action seems to be affected by the presence of organic matter. It would be prudent to study their efficacy at higher concentrations.

The pH of the litter did not show any obvious influence on the activity of the disinfectants, although it is well known that each disinfectant has its own favorable pH range to act. *Salmonella* is known to survive in pH range of up to 5.3. In this study the pH values broadly were ranged from 8.90-6.13 which is considered to be within the working range pH for the optimum action of all the disinfectants as well as optimum range for the growth of *Salmonella*. It is to be mentioned that Sanidate RTU and green work when diluted to the recommended concentrations, pH was highly acidic 4.75 and 2.40, respectively but when mixed with the litter the pH was in same range of the litter treated with other disinfectants (Table 2).

These data showed that in the absence of organic matter Green work started to show high efficacy after 2 h ($p\leq0.0001$) with killing ratio of 97.50% and showed 100% killing efficacy after 16 h ($p\leq0.0001$). Sanidate RTU started to show high efficacy after 2 h ($p\leq0.0001$) with killing ratio of 99.62% and showed 100% killing efficacy after 8 h ($p\leq0.0001$).

Hi-yield® Consan® and Kreso® D showed 100% killing efficacy after 2 h ($p\leq0.0001$). Tektrol® started to show high efficacy after 2 h ($p\leq0.0001$) with killing ratio of 99.97% and showed 100% killing efficacy after 4 h ($p\leq0.0001$) (Table 3).

In the presence of organic matter Green work and Sanidate RTU, both starting to show high efficacy after 2 h ($p\leq0.0001$) with killing ratio of 93.75 and 94.58%, respectively and showed 100% killing efficacy after 16 h ($p\leq0.0001$). Hi-yield®Consan® and Kreso®D showed 100% killing efficacy after 2 h ($p\leq0.0001$). Tektrol® starting to show high efficacy after 2 h ($p\leq0.0001$) with killing ratio of 99.81% and showed 100% killing efficacy after 8 h at ($p\leq0.0001$), (Table 4).

Latasa *et al.*[13], reported that life in a biofilm state protects the bacteria against environment insults like chemical sanitizers which are generally unable to eliminate most biofilm-associated bacteria.

The disinfectants were compared to each other in their efficacy at each specific time by taking samples from the disinfectant/bacterial mixture. There were no significant differences between Green work, Sanidate RTU, Hi-yield® Consan®, Tektrol® and Kreso® D.

Quinn and Markey[20] suggested that phenolic compounds should be used for any application where excessive organic matter may be present, due to their efficacy even in the presence of organic matter.

CONCLUSION

In summary both of Kreso® D and Hi-yield® Consan 20® which are phenolic compounds have shown higher efficacy against *S. typhimurium* compared with the other compounds. In experiment I in the presence of organic matter in the litter, they achieved 100% lethal effect by day 5 ($p \leq 0.0001$) (Table 1) and in experiment II they achieved 100% lethal activity after 2 h ($p \leq 0.0001$) (Table 3 and 4) irrespective of the absence or the presence of organic matter.

Green work and Sanidate RTU are considered environmentally safe disinfectants. Although their efficacy was less compared to Kreso® D in this study, future experiments are necessary to see if they would be effective at higher concentrations. Future studies are also needed to study the efficacy of environmentally safe disinfectants while the birds are present in poultry houses.

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