A New Approach to Evaluate the Hygienic Condition of Commercial Hatcheries

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Abstract: Hatchery hygiene was evaluated in two commercial broiler hatcheries using open-plate method, surface swabbing and microbiological examination of hatchery fluff. Air samples and surface swabs were collected from: inside the hatch unit, the corridor outside the hatch unit and the chick processing room. After cleaning and disinfection process of the hatch, four commercial disinfectants were evaluated for their effectiveness for controlling the hatchery contaminants, TH4® (combination of glutaraldehyde + quaternary ammonium compound), Virucidal extra® (chlorine preparation), Advantage 256® (phenolics) and Perasan® (per-acetic acid). The obtained results indicated that surface swabbing and microbiological examination of hatch fluff could detect higher degree of contamination than open-plate method in the two investigated hatcheries. Per-acetic acid preparations and (glutaraldehyde + quaternary ammonium compound) could reduce completely hatchery contaminants after 30 min of application. Conclusively, open-plate method is easy to perform and inexpensive but may give false indication that the air is clean when it is not. So, surface swabbing and microbiological examination of hatch fluff are more reliable methods for evaluating the hygienic status of a hatchery. Moreover, Surface swabbing method is more accurate than open-plate method in evaluating the decontamination process of the hatch. Hatchery sanitation and the proper use of effective disinfectant are essential for successful operation of any commercial poultry hatchery. Per-acetic acid and (glutaraldehyde + quaternary ammonium compound) proved their efficiency in controlling hatchery contaminants and can be used as safe alternatives to formalin in poultry hatcheries.

Key words: Hatchery hygiene, open-plate methods, fluff, TH4, perasan

INTRODUCTION
Hatchery hygiene is recognized as an important factor in healthy poultry production (Rodgers et al., 2003). Poor standards of hatchery hygiene may lead ultimately to an explosion of pathogenic organisms resulting in severe economic loss. The environment of a poultry hatchery is very susceptible to contamination by microorganisms which can adversely affect hatchability of the eggs and can result in embryonic deaths. Typical microorganisms which are believed to adversely affect chick quality and cause embryonic deaths include E. coli, Staphylococci species, Streptococci species and Aspergillus fumigatus (Sheldon and Brake, 1991).

Therefore, the development and maintenance of an effective hatchery sanitation program is essential for the successful operation of a poultry hatchery. Investigations have revealed large microbial populations in many hatcheries despite the application of various sanitation measures. The degree of contamination was first measured numerically by the microbiological examination of hatchery fluff, a method developed by (Nichols et al., 1967; Furuta and Maruyama, 1981; Chen et al., 2002) and later by the air sampling technique which is used extensively in the poultry industry to monitor bacterial and fungal levels in air and to evaluate the efficiency of decontamination (Chute and Gershman, 1961; Gentry et al., 1962; Ernst, 1987; Rodgers et al., 2003; Moubarak, 2007). Although these tests reveal the magnitude of the contamination in the hatchery environment, they do not indicate where the organisms come from, how they reach the hatcher or where they multiply. This information can only be gained by periodically surveying the microbial populations of the many objects and surfaces which may harbor organisms in the hatchery.

Hatchery sanitation programs should include the use of one or more disinfectant to inhibit the growth of microorganisms and maintain a desirable level of hatchability of fertile hatchery eggs. Traditionally, formaldehyde has been utilized as the fumigant or disinfectant in many hatcheries in order to control the unwanted spread of microorganisms. However, other moderately effective disinfectants such as quaternary ammonium compounds, peroxides, glutaraldehyde and phenolics are also currently utilized in the poultry industry.

In normal use, formaldehyde gas is generated and released in hatchers by mixing formalin and potassium permanganate in specific ratios. This technique requires the handling of potentially hazardous chemicals by hatchery workers and possible exposure of the workers to the gas when initiating the chemical reaction. The use...
of formaldehyde as disinfectant has further
disadvantage, that formaldehyde has been suspected of
being carcinogenic and hence faces possible further
governmental regulation of its use (Sheldon and Brake,
1991). A need therefore exists for safe and effective
disinfectants for use in hatchery sanitation programs
which have the ability to inhibit the growth of
microorganisms and maintain an acceptable level of
hatchability of the eggs treated therewith. A need also
exists for a disinfectant that is convenient to use and can
minimize the time required for satisfactory sanitization.
So, the objectives of the present study are:

C To evaluate the sanitary condition of two
commercial broiler hatcheries using, open-plate
method, surfaces swab and microbiological
examination of fluff.
C To investigate the bactericidal and fungicidal
efficiency of some available disinfectants other than
formaldehyde in a trial to evaluate their effect in
controlling contaminants of commercial hatcheries.

MATERIALS AND METHODS
The experimental work was carried out in two
commercial broiler hatcheries located in Giza
governorate. Samples were collected on four separate
dates, as hatching chicks were being processed and
after cleaning and disinfection process of the hatchery
had been completed. On each visit, air samples and
surface swabbing were collected from: inside the hatch
unit, the corridor outside the hatch unit and the chick
processing room, while, fluff samples were collecte
dafter hatching from the hatch chamber and from the
chick processing room.

Open-plate method: At each sampling site, sterile Petri
plates containing either plate count agar (for total
bacterial count), Sabaurd’s dextrose agar (for total
fungal counts) or, MacConkey’s agar (for total coliform
count) were placed uncovered for 10 min at a height of
one meter from the floor surface (Berrang et al., 1995).

Surfaces swab: Sterile moistened swabs with sterile
normal saline were used to swab walls and floors of
each sampling site. Swabs were received in sterile test
tubes containing sterile normal saline and then, were
transferred to the laboratory in ice box where 0.1 ml of
each sample was plated on sterile plates of plate count,
MacConkey’s and Sabaurd’s dextrose agar (Willinghan
et et al., 1996).

Fluff testing: After hatching, fluff was collected from the
surfaces of racks, hatch baskets; corners of hatches and
chick processing room then, samples were placed in
clean sealed plastic sampling bags and were carried
back to the laboratory in ice box. Under aseptic
conditions 0.5 gram of the fluff was placed into 50 ml of
sterile normal saline, mixed well, then 0.1 ml was
inoculated onto each of sterile plates of plate count agar,
MaConkey’s agar and Sabaurd’s dextrose agar (Chen
et al., 2002).
All the inoculated plates and air sampling plates of plate
count agar and MaConkey’s agar were incubated at
37°C for 24-48 h while those of Sabaurd’s dextrose
agar were incubated at 25°C for 3-5 days and were then
e numerated. Microbial levels were expressed as Colony
Forming Units (CFU) per 10 cm diameter plate. Isolation
and identification of the suspected colonies was done
according to MacFaddin (1980). Results are recorded in
Tables (2-6).

Disinfection of the inner chamber of the hatchery
machine: The inner chamber of the hatch machine was
sprayed with one of four commercial disinfectants using
the concentrations recommended by the manufacturers
as shown in (Table 1). After 30 min of each treatment,
aerial microorganisms were tested using open-plate
method. Also walls and floors were swabbed as
explained to judge the effectiveness of the disinfection
process before.

RESULTS AND DISCUSSION
Hatchery sanitation was evaluated in two commercial
broiler hatcheries designated as hatchery I and II, using

<table>
<thead>
<tr>
<th>Disinfectant</th>
<th>Supplier</th>
<th>Composition</th>
<th>Used dilutions</th>
</tr>
</thead>
<tbody>
<tr>
<td>TH4+®</td>
<td>SOGEVAL (France)</td>
<td>Glutaraldehyde 6.25% 0.5%</td>
<td>0.5%</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Quaternary ammonium compound 12.5%</td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
<td>Terpine derivatives 4.0%</td>
<td></td>
</tr>
<tr>
<td>Virucidal extra®</td>
<td>AVS Ltd. Northern Ireland UK</td>
<td>Potassium monopersulphate 23.0% 1%</td>
<td>1%</td>
</tr>
<tr>
<td>Advantage 256®</td>
<td>Preserve international USA</td>
<td>Sodium dichloro-s-triazinetriol 5.0%</td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
<td>Ortho-phenylphenol 11.0% 0.4%</td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
<td>Ortho-Benzyl-para-chlorophenol 6.0%</td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
<td>Para-Tertiary-Amylphenol 4.0%</td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
<td>Inert engredient 79.0%</td>
<td></td>
</tr>
<tr>
<td>Perasan®</td>
<td>Henkel (Germany)</td>
<td>Peracetic acid 5.0% 1%</td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
<td>H2O2 20.0%</td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
<td>Acetic acid 10.0%</td>
<td></td>
</tr>
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Table 2: Total bacterial count, fungal and coliform counts recorded using open-plate method, surface swab and fluff testing methods

<table>
<thead>
<tr>
<th>Sampling site</th>
<th>Hatchery</th>
<th>T.B.C</th>
<th>T.F.C</th>
<th>T.C.C</th>
<th>T.B.C</th>
<th>T.F.C</th>
<th>T.C.C</th>
<th>T.B.C</th>
<th>T.F.C</th>
<th>T.C.C</th>
</tr>
</thead>
<tbody>
<tr>
<td>Inside hatch unit</td>
<td>I</td>
<td>75</td>
<td>14</td>
<td>45</td>
<td>50x10^2</td>
<td>40x10</td>
<td>30x10^2</td>
<td>80x10^2</td>
<td>70x10</td>
<td>30x10^2</td>
</tr>
<tr>
<td></td>
<td>II</td>
<td>60</td>
<td>40</td>
<td>30</td>
<td>35x10^2</td>
<td>20x10</td>
<td>20x10^2</td>
<td>60x10^2</td>
<td>55x10</td>
<td>22x10</td>
</tr>
<tr>
<td>Corridor outside hatch unit</td>
<td>I</td>
<td>30</td>
<td>8</td>
<td>10</td>
<td>20x10^2</td>
<td>25x10</td>
<td>10x10^2</td>
<td>80x10^2</td>
<td>13x10</td>
<td>28x10^2</td>
</tr>
<tr>
<td></td>
<td>II</td>
<td>22</td>
<td>8</td>
<td>5</td>
<td>10^3</td>
<td>10x10</td>
<td>nil</td>
<td>60x10^2</td>
<td>nil</td>
<td>22x10</td>
</tr>
<tr>
<td>Chick processing room</td>
<td>I</td>
<td>128</td>
<td>17</td>
<td>35</td>
<td>50x10^2</td>
<td>10x10</td>
<td>10x10^2</td>
<td>68x10^2</td>
<td>90x10</td>
<td>23x10^2</td>
</tr>
<tr>
<td></td>
<td>II</td>
<td>90</td>
<td>14</td>
<td>35</td>
<td>50x10^2</td>
<td>80x10</td>
<td>18x10^2</td>
<td>68x10^2</td>
<td>90x10</td>
<td>23x10^2</td>
</tr>
</tbody>
</table>

Table 3: Incidence of bacterial isolates recovered from the investigated hatcheries using open-plate method, surface swabs and fluff testing

<table>
<thead>
<tr>
<th>Methods of sampling</th>
<th>Hatchery</th>
<th>No. of samples</th>
<th>No. (%)</th>
<th>No. (%)</th>
<th>No. (%)</th>
<th>No. (%)</th>
<th>No. (%)</th>
<th>No. (%)</th>
<th>No. (%)</th>
<th>No. (%)</th>
<th>No. (%)</th>
<th>No. (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Open-plate</td>
<td>I</td>
<td>40</td>
<td>nil</td>
<td>nil</td>
<td>nil</td>
<td>4</td>
<td>10</td>
<td>nil</td>
<td>nil</td>
<td>2</td>
<td>5</td>
<td>nil</td>
</tr>
<tr>
<td></td>
<td>II</td>
<td>40</td>
<td>nil</td>
<td>nil</td>
<td>nil</td>
<td>3</td>
<td>7.5</td>
<td>nil</td>
<td>nil</td>
<td>1</td>
<td>2.5</td>
<td>nil</td>
</tr>
<tr>
<td>Surface swabs</td>
<td>I</td>
<td>64</td>
<td>4</td>
<td>6.25</td>
<td>5</td>
<td>7.81</td>
<td>5</td>
<td>7.81</td>
<td>3</td>
<td>4.68</td>
<td>4</td>
<td>6.25</td>
</tr>
<tr>
<td></td>
<td>II</td>
<td>64</td>
<td>2</td>
<td>3.12</td>
<td>4</td>
<td>6.25</td>
<td>3</td>
<td>4.68</td>
<td>1</td>
<td>1.56</td>
<td>2</td>
<td>3.12</td>
</tr>
<tr>
<td>Fluff testing</td>
<td>I</td>
<td>16</td>
<td>2</td>
<td>12.5</td>
<td>1</td>
<td>6.25</td>
<td>1</td>
<td>6.25</td>
<td>2</td>
<td>12.5</td>
<td>3</td>
<td>18.75</td>
</tr>
<tr>
<td></td>
<td>II</td>
<td>16</td>
<td>1</td>
<td>6.25</td>
<td>1</td>
<td>6.25</td>
<td>nil</td>
<td>nil</td>
<td>1</td>
<td>6.25</td>
<td>2</td>
<td>12.5</td>
</tr>
</tbody>
</table>

Table 4: Incidence of fungal isolates recovered from the investigated hatcheries using open-plate, surface swabs and fluff testing method

<table>
<thead>
<tr>
<th>Methods of sampling</th>
<th>Hatchery</th>
<th>No. of samples</th>
<th>No. (%)</th>
<th>No. (%)</th>
<th>No. (%)</th>
<th>No. (%)</th>
<th>No. (%)</th>
<th>No. (%)</th>
<th>No. (%)</th>
<th>No. (%)</th>
<th>No. (%)</th>
<th>No. (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Open-plate</td>
<td>I</td>
<td>40</td>
<td>9</td>
<td>22.50</td>
<td>8</td>
<td>20.00</td>
<td>8</td>
<td>20.00</td>
<td>1</td>
<td>2.5</td>
<td>7</td>
<td>17.5</td>
</tr>
<tr>
<td></td>
<td>II</td>
<td>40</td>
<td>5</td>
<td>12.50</td>
<td>4</td>
<td>10.00</td>
<td>4</td>
<td>10.00</td>
<td>3</td>
<td>7.5</td>
<td>2</td>
<td>5.00</td>
</tr>
<tr>
<td>Surface swabs</td>
<td>I</td>
<td>64</td>
<td>15</td>
<td>23.43</td>
<td>11</td>
<td>17.18</td>
<td>14</td>
<td>21.87</td>
<td>6</td>
<td>9.37</td>
<td>6</td>
<td>9.37</td>
</tr>
<tr>
<td></td>
<td>II</td>
<td>64</td>
<td>8</td>
<td>12.50</td>
<td>7</td>
<td>10.93</td>
<td>5</td>
<td>7.94</td>
<td>2</td>
<td>3.12</td>
<td>2</td>
<td>3.12</td>
</tr>
</tbody>
</table>
method for evaluating the hygienic status in a hatchery. However, Berrang et al. (1995) found that swab samples may be negative for salmonella while hatching air samples may not be negative so, he concluded that environmental samples do not necessarily reflect the contamination in the air. From the recorded results in Tables 2-4, it could be noticed that the sanitary condition of hatchery II is better than that of hatchery I. According to literature this should result in lower degree of contamination of hatching eggs and consequently enhance the hatchability and improve the chick quality. Effect of different disinfectants on T.B.C&T.C.C and T.F.C are shown in Tables 5-6. Both TH4® (glutaraldehyde + quaternary ammonium compound) and perasan® (per acetic-acid) preparations recorded satisfactory results in controlling hatchery contaminant. The obtained results are agreeable with those of Suweify, 1999 who found that TH4® was effective against S. pullorum, E. coli, St. aureus, A. fumigatus and Candida albicans in 15 min (McDonnell and Russell, 2001) who found that glutaraldehyde, has a broad spectrum activity against bacteria and their spores, fungi and viruses and Deeba et al. (2003) who proved that S. pullorum, Proteus mirabilis, Klebsiella pneumoniae and Streptococcus faecalis were 100% susceptible for TH4® and Soliman et al. (2009) who proved that TH4® is the most powerful disinfectants because of the synergistic action of the quaternary ammonium and glutaraldehyde bases.

However, Rodgers et al. (2001), Kaskova et al. (2007), Bauermeister et al. (2008) proved that per-acetic acid is an effective antibacterial agent in hatcheries, poultry houses and poultry processing plants. Virucidal extra® (chlorine preparation) could reduce the total bacterial and coliform count completely but can not reduce the total fungal count. These results are comparable to those of Ramesh et al. (2002) who found that chlorine based disinfectant was effective in controlling bacterial contaminants. However, advantage 256® (phenol) could not reduce both the total bacterial and fungal count after such contact time (Table 5); may be it needs a longer time or a higher concentration. This result is agreeable with those of Sander et al., 2002) who recorded variable degrees of bacterial resistance to advantage 256®. Also, from (Table 5 and 6) it can be noticed that, surfaces swabbing is more accurate than open-plate method in evaluating the decontamination process of the hatcher.

**Conclusion:** In conclusion, air sampling to monitor hatchery sanitation is an easy method to perform and inexpensive but it may give a false impression that the air is clean when it is not. So, surface swabbing and microbiological examination of fluff are more reliable methods for evaluating the hygienic status of a hatchery. Fluff from chicks can be send by mail to the laboratory for evaluation of the hygienic status in case of a distant hatchery. Otherwise, open-plate can be taken as a useful tool but, if counts continue to be above the expected levels, it would be necessary to sample surfaces. On the other hand, many available disinfectants in the Egyptian market as combination of per-acetic acid and (glutaraldehyde + quaternary ammonium compound) and are effective in controlling hatchery contaminations and safe than formaldehyde.

**REFERENCES**

