Biosecurity, Waste Disposal, and Sustainability Measures in Some Commercial Poultry Hatcheries

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Abstract

Hatcheries are a crucial part of the poultry production chain, acting as the vital link between breeder operations and commercial farms. While they play an essential role in ensuring flock productivity, hatcheries can also serve as significant sources of microbial contamination and pathogen transmission. This can negatively impact hatchability and chicks' quality. This study assessed the biosecurity practices and waste disposal protocols in some commercial poultry hatcheries and their impact on hatching outcomes. For this purpose, bacteriological and mycological examination of air, personnel hands, hatching eggs, environmental surfaces, and chick navels samples collected from four commercial poultry hatcheries were performed. The collected samples were examined before and after decontamination procedures were applied according to the bio-security program of these premises. Log_{10} values were calculated and statistically compared.

The results obtained indicated that the biosecurity and hygiene practices across the four hatcheries revealed inconsistent adherence to standard protocols. All the collected samples harbor considerable levels of microbial load and showed variable degrees of resistance to the used chemicals and disinfectants. Floors, as well as corners of walls and floors, emerged as focal points of contamination. Chicks' navels and personnel hands emerged as critical contamination points in all examined hatcheries. Among the hatcheries visited, 75% stored waste in designated waste rooms outside of production areas, while one hatchery utilized a pressurized tank system. Hundred percent of hatcheries recycled egg cartons and sold certain waste. Only two hatcheries demonstrated relatively acceptable fertility and hatchability rates, along with comparatively lower embryonic mortality and cull percentages.

Hatchability assessment across the four hatcheries provided insights into how biosecurity and managemental practices impact hatchery performance, including hatching rates and chick quality. Various strategies, such as using single-stage (SS) or multi-stage (MS) incubation systems, are employed to enhance production. Conclusively, a comprehensive and well-enforced hatchery biosecurity program that includes effective decontamination methods, incubation system design, and sustainable waste management can significantly reduce the microbial load and enhance hatchability and chick viability, contributing to safer and more efficient poultry production environment.

Integrating targeted sanitation with structured, sustainable waste management is essential for improving hatchery hygiene and minimizing environmental impact.

Keywords: Hatcheries, Biosecurity, Hatchability, Hatchery Hygiene, Hatching Eggs and Disinfectants.

Introduction

Poultry is a high-quality protein source for people worldwide ***(Mottet & Tempio, 2017)***. Poultry has grown by over 108% over the past 20 years, with a 36% increase in its participation in total meat production ***(Mitrović et al., 2018)***. To meet the increasing requirement for poultry products, hatcheries have to increase chick production through incubation of more fertilized eggs, hatchability of healthy chicks with high survival rates, in addition to the full expression of their genetic growth potential under all field conditions ***(Boleli et al., 2016)***.

Hatchery plays a crucial role in poultry breeding; it connects breeder farms to production houses ***(Wideman, 2016)***. Production of high-quality chicks depends on multiple factors, including; healthy breeder flock, proper hatchery hygiene, and management ***(Van Limbergen et al., 2020)***. Hatchery hygiene depends on proper cleaning and sanitation of the hatchery as well as hatching eggs ***(Mohammed, 2024)***. Hatcheries allow microbes to spread from breeders to broiler farms, contaminating both the eggs and the hatchery. Hatcheries receive eggs from breeder farms; these eggs might have high bacterial burdens despite their clean appearance ***(Wilson, 1949)***.

Hatcheries are susceptible to infectious agents that can arrive on or inside eggs, on personnel, on equipment like trolleys and trays, or as airborne pathogens ***(McMullin, 2009)***. In some instances, eggs may already be internally infected, leading to vertical transmission of diseases to the offspring ***(McMullin, 2009)***. Pseudo-vertical transmission may occur, where microbes are

initially found on the egg's outer surface but can penetrate through the shell pores ***(Cox et al., 2000)***. Another challenge in hatchery infection control is the potential for horizontal transmission of infectious agents between eggs and chicks, with the hatchery possibly acting as a reservoir and amplifier of microbes ***(McMullin, 2009)***.

Due to condensed production, different hatchery compartments can harbor pathogenic organisms. Examples of these organisms are *Escherichia coli*, *Staphylococcus* spp., *Klebsiella* spp., *Pseudomonas* spp., *Micrococcus* spp., *Proteus* spp., *Enterobacter* sp., *Streptococcus* spp., *Clostridium* spp., *Bacillus cereus*, *Salmonella typhimurium*, *Enterococcus*, and *Salmonella enteritidis.* All were recovered from hatching eggs and proved to penetrate the eggshell, causing yolk sac infection, and embryonic death ***(Rezaee et al., 2021)***. These organisms can infect chicks and result in omphalitis, salmonellosis, chondronecrosis with osteomyelitis, and death by 7 days of age ***(Wilson, 1949)***. Exploders, bangers, and bombs are terms used to refer to contaminated eggs that burst during the incubation phase. Typically, exploding eggs are caused by *Pseudomonas* spp., a bacterium that produces gas. When an egg bursts, it expels its contents and bacteria into the air as an aerosol that spreads throughout the incubator. This affects the hatching chicks, resulting in high mortality when it increases ***(Jordan, 2019; Eraky et al., 2020)***. *Aspergillus* spp. are examples of fungal species contaminating hatcheries. Owing to their small size, *Aspergillus fumigatus* spores can penetrate physical barriers, causing rashes or infection of the young chicks' lungs and air sacs ***(Fedde, 1998)***.

Biosecurity measures have been established to reduce the risk of spreading infectious pathogens on poultry premises. Hatchery hygiene and proper sanitation are recognized as important factors in healthy poultry production and reducing the spread of fungal and bacterial infectious diseases ***(Lazarov et al., 2018; Rodgers et al., 2001)***. It can consequently improve embryonic health, reduce chick mortality, and prevent eggshell contamination ***(Oliveira et al., 2022)***. Chemical disinfectants, including formaldehyde, ozone, halogen solutions, aldehydes, quaternary ammonium, alcohols, phenols, and hydrogen peroxide, are commonly used for effective bacterial disinfection in poultry hatcheries ***(Moustafa, 2009)***. Some disinfectants can be used to sanitize hatching eggs using variable techniques including spraying, wiping, micro-aerosol fogging, injection, dipping, and washing ***(Berrang et al., 2000; Mohammadi-Aragh et al., 2022)***. Formaldehyde is commonly used as a hatching egg sanitizer in European, Brazilian, and Egyptian poultry farms. However, it is highly poisonous, irritating, and

carcinogenic to poultry producers and chicken embryos ***(Oliveira et al., 2022)***. So, more research is required to find safe alternatives. Other hatchery disinfectants are organic peroxides (such as peracetic acid), quaternary ammonia, or chlorine. Each chemical works differently to inactivate microorganisms. Quaternary ammonium compounds were very successful in lowering the levels of coliforms, general aerobic bacteria, *Staphylococcus aureus*, molds, and yeasts when it was tested in hatchery conditions ***(Brake & Sheldon, 1990; Rodgers et al., 2001)***. Chlorine solutions achieved a considerable reduction in aerobic, coliform, and fungal burdens on the egg surface. However, peroxide preparations could achieve total elimination of egg surface contaminants ***(Moustafa, 2009)***.

Poultry hatchery generates a massive amount of solid and liquid waste. Solid wastes include empty shells of hatched eggs, dead embryos, infertile eggs, deformed chicks, late hatchings, and dead chickens, and a viscous liquid from eggs and decaying tissue. The wastewater is generated after washing the hatchery incubators, hatchers, and chick handling rooms. Hatchery waste can be converted into protein feedstuffs, other value-added products, or utilised as an organic fertiliser after appropriate treatment. Solid hatchery waste is typically disposed of by landfilling, composting, rendering, or incineration; however, wastewater can be disposed of by land filling, irrigation, disposing it directly into the sewer or a wastewater lagoon, or using a wastewater treatment system ***(Das et al., 2002; Glatz et al., 2011)***. So, the present study aimed to evaluate the impact of biosecurity practices and waste disposal and handling on hatchery sanitation and hatching results in some commercial poultry hatcheries.

Materials and methods

Ethical approval

Ethical approval to conduct this study was obtained from the Institutional Animal Care and Use Committee, Faculty of Veterinary Medicine at Cairo University, Egypt, with Approval Reference No. (Vet CU110520251133).

Investigated hatcheries

The experimental work was carried out in four commercial broiler hatcheries. Hatcheries 1, 2, and 3 were in El Behera, while Hatchery 4 was at Giza. All hatcheries were away from poultry premises and 300 meters from the main road. Two separate visits, 21 days apart, were applied to each hatchery under investigation during the study period. The first one was on the day of hatching-egg receival and traying, the second was as hatching chicks were being processed, and after the cleaning and disinfection process of the hatchery had been completed. On each visit, air samples, hatching egg swabs, chick's navel swabs, and surface swabs were collected from

egg receiving rooms, setters, hatchers, and the chick processing rooms.

Two questionnaires were collected from each hatchery to assess the biosecurity measures applied and waste handling and disposal methods. The biosecurity questionnaire involved biosecurity procedures, management strategies used, disinfection programs, and hygiene practices. The waste handling and disposal questionnaire focused on inquiries into the amount of waste produced, waste separation, pre-treatment processes, disposal method, and waste reuse. Additionally, face-to-face interviews were conducted with hatchery operators during the data collection process ***(Swai et al., 2013)***.

The used chemical disinfectants

Seven commercial chemical disinfectants were used according to the biosecurity programmes of the four investigated hatcheries, either for disinfection of hatching eggs or for different hatchery surfaces. The active ingredients are shown in **Table1**, while the dilution rate and methods of application are shown in **Table 2**.

Sampling

1. Air

The settle plate method was applied for sampling air according to the method described by ***(Wright & Epps, 1958; Berrang et al., 1999)***. Briefly, previously prepared sterile petri dishes of plate count agar (HIMEDIA®, India), MacConkey agar (HIMEDIA®, India), and Sabouraud dextrose agar (HIMEDIA®, India) media were placed uncovered at a height of one meter from the floor surface in the different hatchery compartments for 10 min then covered, inverted, and sent to the laboratory.

2. Personnel hands

Five workers' hands were swabbed during each hatchery visit using cotton swabs pre-moistened with sterile normal saline according to the method described by ***(Genigeorgis et al., 1989)***.

3. Hatching eggs

For sampling hatching eggs, before sanitization, the eggshell surface of each egg was totally swabbed using a sterile cotton swab moistened with sterile normal saline and then received into a marked sterile test tube containing 5 ml of sterile normal saline ***(Schmaltz et al., 2006)***.

4. Surfaces

Swabs moistened with sterile saline were used to sample walls, floors, egg trays, hatch, and transport boxes of different hatchery compartments. Surface area of 25 cm^2 was swabbed from each site, determined using a 5 × 5 cm sterile template. Swabs were then received into sterile test tubes containing sterile normal saline ***(Swai et al., 2013)***.

5. Hatched chicks

A total of 16 navels from randomly selected newly hatched chicks were swabbed using sterile cotton swabs moistened with sterile normal saline. The swabs were then placed into

labelled sterile test tubes containing 5 ml of sterile normal saline ***(Shahjada et al., 2017)***.

6. Disinfectant neutralizing solution

Following egg sanitation and disinfection of hatchery compartments, hatching eggshells and surface swabs, excluding navel swabs, were received into sterile test tubes, each containing 5 ml of a disinfectant neutralizing solution. This solution consisted of 0.5% sodium thiosulfate, 0.3% lecithin, 3% Tween 80, 1% histidine, and 3% saponin. Its purpose was to neutralize any residual disinfectants in the recovery medium after the designated contact time ***(Espigares et al., 2003)***. Collected samples were sent to the laboratory in an ice tank with minimum delay to undergo microbiological examination ***(Cortés et al., 2004)***.

Microbiological examination

At the laboratory, air sampling plates containing Plate Count Agar and MacConkey's Agar were incubated at 37°C for 24–48 hours, while those containing Sabouraud Dextrose Agar were incubated at 25°C for 3–5 days, then colony counts were expressed as $\log_{10}$ colony-forming units (CFU) per 10 cm diameter plate. For the collected swabs, to determine total bacterial count (TBC), total coliform count (TCC), and total fungal count (TFC), 0.1 ml aliquots from each dilution were spread onto sterile plates of Plate Count Agar, MacConkey's Agar, and Sabouraud Dextrose Agar, respectively. The bacterial and coliform plates were incubated at 37°C for 24–48 hours, while fungal plates were incubated at 25°C for 3–5 days. After incubation, colonies were enumerated and reported as $\log_{10}$ CFU per eggshell (CFU/eggshell) or per 25 cm^2 for surface samples (CFU/25 cm^2) ***(Willinghan et al., 1996)***.

Statistical analysis

The results are presented as means ± standard error of the mean (SEM). Data analysis was conducted using the independent sample *t*-test and one-way ANOVA, followed by post-hoc comparisons with the Tukey test. Comparisons between results before and after disinfection were done using the paired sample *t*-test. Statistical analyses were performed using PASW Statistics Software (SPSS Inc., Chicago, IL, USA, Version 18.0). A significance level of $P \leq 0.05$ was set for all tests. Boxplots were generated with the *ggplot2* package ***(Kassambara, 2023; Wickham et al., 2025)*** in R software (Version 4.4.3).

Table 1. *Active ingredients of disinfectants used in hatchery sanitation programs.*

Disinfectant	Active ingredients
A	Quaternary ammonium compounds (Didecyl dimethyl ammonium chloride 9.2%, Alkyl dimethyl benzyl ammonium chloride 9.2%, Alkyl benzyl ammonium chloride 4.6%)
B	Quaternary ammonium compounds (benzyl alkyl dimethyl chlorides 15 – 30%, Didecyl dimethyl ammonium chloride 5-15%) and Glutaraldehyde 5-15%
C	Hydroxy acetic acid (Glycolic acid 4%)
D	Hydrogen peroxide and silver ions
E	Paraformaldehyde
F	Hydrogen peroxide 20% and Peracetic acid 5%
G	Dichloroisocyanurate (NaDCC) 25 ppm
H	45% Formaldehyde, 45% Glutaraldehyde, 5% Quaternary Ammonium Compounds (QACs) and 5% Excipients
I	Phenol 25%
J	Quaternary ammonium compounds (Didecyldimethylammonium chloride 1.875%, dioctyldimethylammonuim chloride 1.875%, octyldecdimethylammonium chloride 3.75%, Alkyl dimethyl ammonium chloride 5%) and Glutaraldehyde 6.2%
K	Iodine nonoxynol 2.8% and Orthophosphoric acid 17%

Table 2. *Evaluation of biosecurity implementation and hygienic management in the selected hatcheries.*

Items	*POC	Hatchery-1	Hatchery-2	Hatchery-3	Hatchery-4	Yes/Total (%)
Site and Entrance	Biosecurity signs posted warning people not to enter any of the buildings on the premises	No	Yes	Yes	No	2/4 (50)
	Presence of fence surrounding the premises	Yes	Yes	Yes	Yes	4/4 (100)
	Footwear disinfection stations	No	Yes	Yes	No	2/4 (50)
	A visitor record	No	Yes	Yes	No	2/4 (50)
	The farm policy requires that employees and visitors/contract workers do not own other birds	No	Yes	Yes	No	2/4 (50)
	Visitors change into dedicated clothing and boots	No	Yes	Yes	No	2/4 (50)
	truck drivers are prohibited from entering the hatchery	Yes	Yes	Yes	Yes	4/4 (100)
	Presence of shower area	No	Yes	Yes	No	2/4 (50)
	sharing equipment or supplies with other farms or hatcheries	No	No	No	No	0/4
	If tools or equipment must be brought in, are they cleaned and disinfected as they enter	Yes	Yes	Yes	Yes	4/4 (100)
	Car station	Yes	Yes	Yes	Yes	4/4 (100)
	Sprayed disinfectant	Disinfectant (K) 20 ml/L	Disinfectant (J) 5 ml/L	Disinfectant (H) 5 ml/L	Disinfectant (B) 5 ml/L	

	Dipping wheels disinfectant	Disinfectant (I) 20 ml/L	Disinfectant (I) 5 ml/L	-	-	
	Workers stay in hatchery	Yes	Yes	Yes	Yes	4/4 (100)
	Presence of Foot dip	Yes	Yes	Yes	Yes	4/4 (100)
	Disinfectant used in foot dip	Disinfectant (I) 20 ml/L	Disinfectant (A) 5 ml/L	Disinfectant (H) 5 ml/L	Disinfectant (B) 5 ml/L	
	Changing time	Daily	Daily	Daily	Daily	
Management strategies	Annual setting capacity (Million Eggs)	12	50	18	16	
	Incubation system	Multistage trolley loading	Single stage trolley loading	Multistage trolley loading	Multistage trolley loading	
	Number of setters	6	24	9	8	
	Number of hatchers	6	24	9	8	
	Egg Storage period / days	7	7-14	2 – 12	3 – 35	
	Egg Storage Temperature (°C)	15 – 20	15 – 20	15 – 20	15 – 20	
	Egg storage RH (%)	75-80	75-80	65-75	70-75	
	A record for the production data	Yes	Yes	Yes	Yes	4/4 (100)
	**High-risk procedure	No	Yes	Yes	No	2/4 (50)
	Biosecurity training courses for all employees	No	Yes	Yes	No	2/4 (50)
	Pest and rodent control	No	Yes	Yes	No	2/4 (50)
	Microbial testing	Yes	Yes	Yes	No	3/4 (75)
	Time of microbial test (week/ month)	Every 3 months (chick)	Every 15 days	Every 15 days	-	
Workers	Showers at the beginning of the workday	Yes	Yes	Yes	Yes	4/4 (100)
	Wearing protective gloves & masks	Yes	Yes	Yes	Yes	4/4 (100)
	Hand wash and sanitizers	Yes Alcohol & Dettol	Yes Alcohol & Dettol	Yes Alcohol & Dettol	Yes Alcohol & Dettol	4/4 (100)
	***Restrict Movement	No	Yes	Yes	No	2/4 (50)
Disinfection program	****Detergent for cleaning surfaces	No	Yes	Yes	Yes	3/4 (75)
	Name & dilution rate	-	Sodium hydroxide (foam) 1:50 - 1:150 parts of water	Sodium hypochlorite (non-foaming) 0.5-1% (1:200 - 1:100)	Sodium hypochlorite (non-foaming) 0.5-1% (1:200 - 1:100)	
	Surface disinfectant	Yes	Yes	Yes	Yes	4/4 (100)
	Disinfectant & the dilution rate	Disinfectant (A) 5 ml/L	Disinfectant (A) 5ml/L	Disinfectant (B) 5ml/ L	Disinfectant (B) 7 ml/L	
	Application method	Spray	Spray	Spray	Spray	
	Aerial disinfection	Yes	Yes	Yes	Yes	4/4 (100)
	Disinfectant & the dilution rate	Disinfectant (A) 1.5 ml/L/113m^3	Disinfectant (C) Hydroxy acetic acid (HA) 1.2 g/m^3	Disinfectant (B) 1-2L/3 L water/1000 m^3	Disinfectant (B) 1-2L/3 L water/1000 m^3	
	Application method	Thermal fogging	Fumigation	Thermal fogging	Thermal fogging	

Egg disinfection	Primary egg disinfection	Yes	Yes	Yes	Yes	4/4 (100)
	Disinfectant & the dilution rate	Disinfectant (D) 5 ml/L	Disinfectant (E) 4 g/m^3	Disinfectant (E) 7g/m^3	Disinfectant (E) 4 g/m^3	
	Application method	Spraying	Fumigation	Fumigation	Fumigation	
	2^{ry} Hatching egg disinfection at the hatchery	Yes	Yes	Yes	Yes	4/4 (100)
	Disinfectant & the dilution rate	Disinfectant (D) 5 ml/L	Disinfectant (F) 5 ml/L	Disinfectant (G) 1 tablet/10L	Disinfectant (F) 5 ml/L	
	Application method	Spraying	Spraying	Spraying	Spraying	
	Contact time (minutes)	20 to 30	20 to 30	20 to 30	20 to 30	
Chick processing room disinfection	Chick processing room disinfection	Yes	Yes	Yes	Yes	4/4 (100)
	Disinfectant & Application method	Disinfectant (B) Spraying	Disinfectant (B) Spraying	Disinfectant (B) Spraying	Disinfectant (B) Spraying	
	Transport boxes disinfection	Yes	Yes	Yes	Yes	4/4 (100)
	Disinfectant & the dilution rate	Only disinfectant (A) 5 ml/L without detergent	Disinfectant (B) 5 ml/L + Sodium hypochlorite	Disinfectant (B) + 5 ml/L Sodium hypochlorite	Disinfectant (B) 5 ml/L + Sodium hypochlorite	
	Application method	Spraying	Spraying	Spraying	Spraying	

RH: Relative humidity
*POC: Point of Comparison
**: In the event of a high-risk biosecurity issue, a standard operating procedure should be in place to contain any infectious materials. Procedures should include methods for the containment and decontamination of infectious and potentially infectious materials.
***: Keep those who work with eggs and those who work with chicks separated. Prevent personnel who work in dirty areas or areas that encounter chicks from accessing clean areas. Using color-coded uniforms can help restrict personnel movement.
****Main Active principle for detergent

Results

1. Biosecurity and hygiene measures in the investigated hatcheries

The assessment of biosecurity measures and hygiene practices across the four investigated hatcheries revealed variable levels of compliance with standard biosecurity protocols (**Table 2**). Only two of the four hatcheries (50%) displayed biosecurity signage and maintained visitor records, while all hatcheries (100%) were surrounded by fences and had designated car stations. Footwear disinfection stations, visitor clothing protocols, and the requirement that employees and visitors do not own other birds were implemented by 50% of the hatcheries. Notably, none of the hatcheries shared equipment with other farms, and all followed proper disinfection of tools upon

entry. All the investigated hatcheries employed disinfectants at various entry and operational points, with differences in types and concentrations. Disinfectants used for spraying and foot dips varied between hatcheries, with products such as A (QACs), B (QACs + glutaraldehyde), H (QACs + aldehydes), K (iodophor), and I (phenol) utilized at different dilutions. Workers in all hatcheries were housed on-site, used foot dips, and took daily showers, indicating consistent adherence to personal hygiene practices.

Regarding management strategies and microbial monitoring, the hatcheries varied in scale, with annual egg-setting capacities ranging from 12 to 50 million eggs. Incubation systems differed, with Hatchery-2 employing a single-stage system, while the others used multi-stage systems. Egg storage durations and conditions were consistent, although relative humidity levels varied slightly. The four hatcheries maintained production records; however, only two (50%) had designated biosecurity training programs and pest control measures in place. Microbial testing was performed in three of the four hatcheries (75%), with testing frequencies ranging from every 15 days to once every three months, depending on whether samples were from environment, eggs, or chicks.

For worker hygiene and disinfection protocols, all hatcheries provided showers at the beginning of work shifts and required protective gloves and masks. Hand washing and sanitizer use were consistent across facilities, with alcohol and Dettol as common agents. Movement restrictions within the hatchery were implemented in 50% of the facilities. Personnel movement was limited, keeping those handling eggs separate from those dealing with chicks in Hatcheries 2 and 3, but this was not followed in Hatcheries 1 and 4.

Hatchery disinfection protocols included the use of detergents (sodium hypochlorite) for surface cleaning in three of the four hatcheries (75%), with varying active ingredients and dilution rates. Surface and aerial disinfection were practiced in the four investigated hatcheries, using a range of disinfectants and application methods such as thermal fogging and fumigation. Primary and secondary egg disinfections were uniformly applied across all breeder farms and hatcheries, respectively, with consistent contact times and different disinfectant types and methods. Primary egg disinfection was conducted in all breeder farms utilizing disinfectant D (Hydrogen peroxide + silver ions) or E (Paraformaldehyde) through spraying or fumigation, respectively. Hatching egg secondary disinfection was applied in all hatcheries using Disinfectant D (Hydrogen peroxide + silver ions), F (Hydrogen peroxide + Peracetic acid), or G (NaDCC) via

spraying. Chick processing rooms and transport boxes were also disinfected in the four investigated hatcheries, though disinfectant types and combinations varied, using disinfectant B (QACs + glutaraldehyde) through spraying.

2. Sustainable waste management and disposal practices in surveyed poultry hatcheries

Table 3 provides an overview of the methods used for waste management and disposal in the four surveyed poultry hatcheries. The assessed hatcheries had varying capacities, ranging from twelve million to fifty million eggs annually. Three of the four hatcheries (75%) used multi-stage trolleys for incubation, and the majority (75%) received eggs daily. Waste management strategies varied among the four investigated hatcheries. Separate waste collection units were found in 75% of the hatcheries, primarily in the form of waste rooms located outside the hatchery buildings, while one hatchery used a tank under air pressure. The quantity of waste generated daily ranged from 400 kg to five tons, with three hatcheries limiting waste storage to a maximum of one day, and one storing waste for up to a week. Waste separation before disposal was reported in 75% of hatcheries.

Methods of waste disposal showed substantial variation. Two hatcheries (50%) had formal arrangements for waste disposal, either through contracts with municipalities or external incineration services. One hatchery burned waste on-site, while another disposed of it as regular trash. Notably, no hatchery reported pre-treatment of waste before disposal. All hatcheries (100%) sold egg cartons for reuse, particularly for transporting culled, spoiled, or discarded eggs. Similarly, all rejected eggs, including large, small, broken, over-calcified, infertile, or double-yolk eggs, were sold at all facilities. Floor eggs were managed by selling (25%) or incubating after disinfection (75%).

Regarding non-hatched eggs (e.g., those with early or late embryonic deaths or dead-in-shell embryos), half of the investigated hatcheries sold them as hatching waste, while others used incineration or disposed of them with general waste. All hatcheries condemned deformed chicks, and methods for disposing of condemned or dead chicks included burial, incineration, and use of waste tanks, though one hatchery discarded them in regular trash. Fluff was primarily discarded in garbage or wastewater, with 75% of hatcheries using one of these routes. Additionally, 50% of the Hatcheries (1 and 4) reported selling a portion of their waste to duck or turkey farms. Hatchery 2 did not engage in this practice, and Hatchery 3's policies were unspecified.

3. Microbial load of hatcheries' compartments

Table 4 presents the microbial load of air, floor, and wall surfaces across various compartments of the four hatcheries, measured before and

after cleaning and disinfection. The microbial parameters assessed included Total Bacterial Count (TBC), Total Coliform Count (TCC), and Total Fungal Count (TFC), with values expressed as $\log_{10}$ CFU ± SEM.

3.1. Air:

The microbial load in air samples varied between compartments, with the sorting area recording the highest TBC (3.60 $\log_{10}$ CFU), while reception had the lowest (3.05 $\log_{10}$ CFU). Similarly, the TCC level was high in the hatcher room and sorting areas. The TFC was high in the reception and sorting compartments. However, no significant differences were observed in air microbial loads between compartments ($P > 0.05$), and the effect of disinfection is continuous.

3.2. Floors

Disinfection led to a notable reduction in microbial loads on floor surfaces across all compartments. The highest reductions were observed in TFC levels, with complete fungal elimination (100% log reduction) in the setter, hatcher, and sorting areas. A statistically significant reduction in fungal count was noted in the hatcher ($P = 0.018$), indicating effective disinfection. TBC levels showed moderate reductions, ranging from 4.9% to 20.4%, though none were statistically significant ($P > 0.05$). TCC also decreased post-disinfection in most areas, with reductions approaching 50% in the reception and sorting compartments, yet these were not significant ($P > 0.05$).

3.3. Walls

Wall microbial loads followed a similar trend to floor surfaces, with noticeable decreases in TBC, TCC, and TFC following disinfection in most compartments. Notably, TFC in the hatcher walls dropped to zero, representing a 100% log reduction. While the reductions in TBC and TCC on wall surfaces were substantial, they did not reach statistical significance ($P > 0.05$). An exception was seen in the sorting area, where TBC slightly increased post-disinfection, though not significantly.

4. Effectiveness of the disinfection of hatchery surfaces and equipment

Table 5 summarizes the effectiveness of disinfection practices in reducing microbial contamination across various hatchery surfaces and equipment.

4.1. Eggshell

For floor eggs, disinfection failed to reduce bacterial contamination, as TBC slightly increased post-disinfection (from 4.26 to 4.42 $\log_{10}$ CFU), indicating a 3.76% increase ($P = 0.893$). However, TCC and TFC were fully eliminated, each showing a 100% reduction, although without statistical significance ($P = 0.423$ and 0.150, respectively). In contrast, clean eggs showed modest improvement post-disinfection, with a slight reduction in TBC (3.27 to 3.15 $\log_{10}$ CFU), a 47.52% reduction in TCC, and complete elimination of

fungal contaminants. While the reduction in TFC approached significance (P = 0.079), overall changes in microbial loads on egg surfaces were not statistically significant.

4.2. Hatchery Equipment:

Among equipment surfaces, trays demonstrated the most significant microbial reductions. TBC decreased by 45.06% (P = 0.021), and TCC dropped by 87.60% (P = 0.019), both of which were statistically significant. Fungal contamination also declined by 84.38%, although not significantly (P = 0.210).

4.3 Hatchery Baskets and Chick Transport Boxes:

Disinfection of hatchery baskets led to moderate reductions in microbial loads, with TBC decreasing by 8.27%, TCC by 38.73%, and complete elimination of fungal colonies. However, none of these reductions were statistically significant (P > 0.05). For chick transport boxes, TBC and TCC were reduced by 30.56% and 71.33%, respectively, and TFC by 40%. Although the microbial load reductions were substantial, they did not reach statistical significance (P > 0.05).

5. Spatial contamination differences between corners and centers of floors

Figure 1 illustrates the spatial distribution of microbial loads between center and corner locations on hatchery walls (**Fig. 1A**) and floors (**Fig. 1B**). The results obtained revealed notable differences in microbial contamination patterns, particularly in less accessible areas. On walls (**Fig. 1A**), the mean TBC was higher in the corners than in the center (3.62 and 2.81 $\log_{10}$ CFU, respectively). This suggests that bacterial build-up may be more common in corner areas, potentially due to reduced cleaning efficacy. Similarly, TCC levels were elevated in corners compared to centers (1.15 and 0.69 $\log_{10}$ CFU, respectively), indicating possible localised hygiene challenges. Fungal contamination on walls remained low overall, with no significant difference between center (0.15 $\log_{10}$ CFU) and corner (0.69 $\log_{10}$ CFU) locations, suggesting effective fungal control.

On floors (**Fig. 1B**), the bacterial counts were again higher in corner areas than in center areas (4.12 and 3.58 $\log_{10}$ CFU, respectively). However, coliform counts on floors showed a statistically significant increase in corners (1.46 $\log_{10}$ CFU) compared to the complete absence in center areas (P < 0.01), highlighting floor corners as hotspots for coliform contamination. This may be attributed to moisture accumulation and suboptimal cleaning access. Fungal counts on the floor were modest and showed no significant spatial variation (center: 0.49, corner: 0.89 $\log_{10}$ CFU), reflecting consistent fungal control across locations.

6. Efficacy of disinfection protocols across the investigated hatcheries

6.1. Total Bacterial Count (TBC)

Table 6 presents the total bacterial counts (TBC) on various surfaces across the four investigated hatcheries, measured before and after the implementation of their respective disinfection programs. Surprisingly, floor surfaces in 75% of hatcheries showed increased bacterial load following disinfection. The most pronounced increase was observed in Hatchery 1, where TBC rose from 3.68 to 4.51 $\log_{10}$ CFU (22.55% increase). However, none of these increases were statistically significant ($P > 0.05$), indicating potential inconsistencies or ineffectiveness in floor disinfection protocols. Hatchery 4 demonstrated the most effective floor disinfection protocol, achieving a 29.27% log reduction in total bacterial count, decreasing from 4.10 to 2.90 $\log_{10}$ CFU ($P = 0.313$). This notable reduction suggests that the disinfection measures implemented in that hatchery were comparatively more effective in controlling surface bacterial contamination than in the other facilities.

Notably, corners in Hatchery 2 showed a significant reduction in total bacterial count (TBC) following disinfection, decreasing from 4.50 to 3.28 $\log_{10}$ CFU (27.11% reduction; $P < 0.0001$). Hatcheries 3 and 4 also exhibited reductions in TBC (17.74% and 10.53%, respectively), although these changes were not statistically significant ($P = 0.274$ and $P = 0.549$, respectively). In contrast, Hatchery 1 showed an unexpected increase in TBC post-disinfection, rising from 3.67 to 5.12 $\log_{10}$ CFU (39.51% increase; $P = 0.208$), suggesting possible defects in the sanitation process. A highly significant inter-hatchery difference was observed following disinfection ($P < 0.0001$), with the highest TBC recorded in Hatchery 1. This variability underscores potential disparities in disinfection efficacy, likely influenced by factors such as the choice of disinfectant, application method, environmental conditions, or personnel compliance.

Wall surfaces generally exhibited increases in bacterial loads following disinfection, particularly in Hatchery 3 (80.93% increase) and Hatchery 1 (69.90% increase). Although these rises were notable, none reached statistical significance ($P > 0.05$), suggesting suboptimal application techniques or the limited effectiveness of disinfectants on vertical surfaces, where coverage and contact time may be inconsistent. In contrast, Hatchery 4 demonstrated a meaningful reduction in TBC on walls, achieving a 40.44% decrease ($P = 0.091$), which may reflect better implementation of disinfection protocols or more effective product use in that facility.

Egg trays exhibited the most effective disinfection efficacy, with

Hatchery 1 achieving complete bacterial elimination (100% reduction; $P = 0.010$), highlighting the efficacy of its tray sanitation protocol. In comparison, other hatcheries showed only modest reductions in total bacterial count, ranging from 23.88% in Hatchery 2 to 7.65% in Hatchery 3, none of which reached statistical significance. The inter-hatchery difference after disinfection was significant ($P = 0.001$), emphasizing variability in sanitation outcomes and underlining Hatchery 1's superior performance in tray disinfection relative to the others.

Chicks' navels and workers' hands emerged as critical contamination points in several hatcheries. Hatchery 4 recorded significantly higher bacterial loads on chicks' navels (7.31 $\log_{10}$ CFU) compared to the other hatcheries (3.22–3.72 $\log_{10}$ CFU; $P = 0.020$), indicating possible hygiene lapses during the hatching or handling process. Similarly, hand hygiene among personnel varied notably across facilities. Workers in Hatchery 3 and Hatchery 4 exhibited alarmingly high bacterial counts on their hands (7.00 and 5.21 $\log_{10}$ CFU, respectively), significantly surpassing those in Hatchery 1 (1.21 $\log_{10}$ CFU; $P = 0.019$). These findings underscore the urgent need for stricter biosecurity training and enforcement of hand hygiene protocols to mitigate cross-contamination risks.

6.2. Total coliform counts (TCC)

Table 7 presents the total coliform count (TCC) on different surfaces across the four hatcheries studied before and after cleaning and disinfection. The effectiveness of these protocols showed noted variation depending on the hatchery and surface type. Notably, floor samples from hatcheries 1 and 2 demonstrated an unexpected increase in TCC post-disinfection, with coliform bacterial loads rising about 66.67 and 34.18% logs, respectively. Although these increases did not reach statistical significance ($P = 0.500$ and 0.183), they suggested potential post-cleaning contamination or ineffective disinfection procedures.

The TCC of corners varied significantly across hatcheries. Hatchery 2, which initially recorded the highest coliform load among hatcheries (3.61 $\log_{10}$ CFU; $P < 0.0001$), demonstrated a significant reduction post-disinfection to 1.01 $\log_{10}$ CFU (a 72.02% decrease; $P < 0.0001$), indicating effective sanitation practices. In contrast, Hatcheries 3 and 4 began with lower TCC values (0.64 and 0.69 $\log_{10}$ CFU, respectively), followed by moderate but statistically nonsignificant reductions of 32.81% and 62.32%. Hatchery one showed a 67.18% increase in TCC on corner surfaces after disinfection, suggesting contamination, ineffective disinfectant use, or protocol flaws.

Walls across hatcheries showed inconsistent disinfection results. In

Hatchery 1, coliform counts increased post-disinfection, from 0.43 to 1.34 $\log_{10}$ CFU, representing a 67.91% rise and pointing to potential contamination during or after cleaning. Hatchery 2 maintained higher TCC both before and after disinfection (2.96 to 3.01 log10 CFU), suggesting the poor efficacy of its disinfection protocol. Conversely, Hatchery 3 eliminated coliform bacteria, indicating successful sanitation. Statistically significant inter-hatchery differences were observed both before (P = 0.046) and after (P = 0.007) disinfection. Notably, Hatchery 2 had the highest residual contamination, whereas Hatchery 4 maintained the lowest TCC levels, emphasizing the variability in hygiene practices and effectiveness among facilities.

Egg tray appeared as the most successfully sanitized of all sampled areas. Hatchery 1 demonstrated complete coliform elimination following disinfection, with TCC dropping from 4.64 to 0.00 $\log_{10}$ CFU (P = 0.014), indicating a highly effective protocol. Hatchery 2 also showed a substantial but not significant reduction of 63.58%. Significant inter-hatchery variation was observed before disinfection (P = 0.007), with Hatcheries 1 and 2 recording the highest initial TCC levels (4.64 and 3.35 $\log_{10}$ CFU, respectively). Conversely, Hatcheries 3 and 4 maintained low or undetectable coliform levels throughout, indicating superior initial hygiene or effective routine sanitation.

Coliform contamination of chicks' navels was low across all hatcheries, with no statistically significant differences observed (P = 0.639). Despite this, the workers' hands showed considerable variation. Hatchery 3 had the highest TCC (5.18 $\log_{10}$ CFU), followed by Hatchery 4 (4.47 $\log_{10}$ CFU), while Hatchery 1 maintained zero detection (P < 0.001). These findings emphasize the critical role of hand hygiene in contamination control protocols.

6.3. Total fungal count (TCC)

Table 8 illustrates the total fungal counts (TFC) on various hatchery surfaces before and after the implementation of disinfection protocols. Overall, the efficacy of fungal control varied notably by both surface type and hatchery, underscoring inconsistencies in sanitation performance. Floor surfaces in Hatcheries 1 and 2 initially harbored detectable fungal contamination (1.35 and 0.50 $\log_{10}$ CFU, respectively), both of which were eliminated post-disinfection, reflecting a 100% reduction. Meanwhile, Hatcheries 3 and 4 maintained fungus-free floors both before and after disinfection. Despite these reductions, the changes were not statistically significant (P > 0.05).

Corner displayed the greatest variation. Hatchery 2 recorded the highest pre-disinfection fungal count (2.36 $\log_{10}$ CFU), which was

eliminated after disinfection, yielding a statistically significant reduction (LR% = 100%; $P < 0.0001$). Conversely, Hatchery 1 exhibited an unexpected post-disinfection increase from 0.55 to 0.83 $\log_{10}$ CFU (+50.91%), suggesting potential recontamination or ineffective sanitation. Hatcheries 3 and 4 began with minimal fungal loads (0.00 and 0.17 $\log_{10}$ CFU, respectively), which were effectively reduced to undetectable levels. Significant inter-hatchery differences in corner TFC were noted both before and after disinfection ($P < 0.0001$), with Hatchery 2 exhibiting the highest initial count and Hatchery 1 showing the highest residual contamination post-disinfection. These findings highlight the uneven performance of disinfection protocols, particularly in hard-to-clean areas like corners.

Walls were almost clean from fungal contamination across the hatcheries, except for Hatchery 2, which exhibited a notable pre-disinfection fungal load of 2.00 $\log_{10}$ CFU. Following disinfection, this was eliminated, indicating a 100% reduction in fungal counts. In contrast, Hatchery 1 showed a slight and unexpected increase in fungal load, rising from 0.85 to 1 $\log_{10}$ CFU, although this change did not reach statistical significance ($P > 0.05$). These findings reinforce the sporadic nature of fungal contamination on wall surfaces and highlight the importance of implementing more targeted and consistent disinfection strategies for vertical structures, which may be prone to oversight during routine cleaning.

Egg tray disinfection efficacy varied among hatcheries. Hatchery 2 exhibited the most effective outcome, achieving complete fungal elimination from an initially high load of 2.97 $\log_{10}$ CFU, a statistically significant reduction ($P = 0.018$). Hatcheries 1, 3, and 4 maintained undetectable fungal levels before and after disinfection, reflecting either effective ongoing hygiene practices or a lower fungal burden. Importantly, inter-hatchery differences in pre-disinfection fungal counts were statistically significant ($P < 0.0001$), with Hatchery 2 recording the highest initial contamination, underscoring the inconsistencies in baseline sanitation standards across facilities.

No fungal growth was detected in chick navel samples across any hatchery, reflecting effective control of fungal contamination during hatching. However, worker hand swabs revealed variable fungal contamination, with Hatcheries 2 and 3 showing relatively high levels (2.85 and 2.48 $\log_{10}$ CFU, respectively), compared to Hatcheries 1 and 4. Although this variation was not statistically significant ($P = 0.165$), it suggests inconsistent adherence to personal hygiene protocols that may warrant further training and supervision.

7. Hatchability performance

Hatchability performance varied noticeably across the four hatcheries investigated, reflecting differences in fertility rates, embryonic mortality, and operational efficiency **(Table 9)**. Hatchery 1 exhibited the highest average fertility rate (93%) and total hatchability (88.5%), indicating well-managed incubation processes. Hatchery 2 performed comparably in fertility (92%) but achieved a slightly lower hatchability rate of 84% due to increased embryonic mortality. Hatchery 3 recorded a moderate fertility rate (87%) and a corresponding hatchability of 80%. In contrast, Hatchery 4 exhibited the lowest fertility (85%) and hatchability (78.5%), suggesting underlying issues in flock productivity, egg handling practices, or environmental conditions within the incubators that warrant targeted interventions.

Embryonic mortality patterns differed among hatcheries. Hatchery 1 had the lowest early embryonic death rate (0.75%), while Hatcheries 2 and 4 reported higher early mortality (3.5 and 2.5%). Middle-stage mortality remained consistently low across all hatcheries, particularly Hatchery 3 (0.2%). However, late embryonic deaths were most pronounced in Hatchery 1 (3.25%), followed closely by Hatcheries 3 and 4 (3.0%). These late losses may reflect suboptimal humidity or ventilation control during the final incubation phase.

Total embryonic mortality exhibited notable variation among the hatcheries, ranging from 5.1% in Hatchery 1 to a maximum of 7.75% in Hatchery 4. This pattern aligns with overall hatchability outcomes and may reflect differences in environmental control, egg storage conditions, or parental flock health. Despite these discrepancies, contamination rates remained consistently low at 0.5% across all facilities, indicating that biosecurity and egg sanitation measures were effective in preventing microbial penetration into the eggshell. In contrast, cull rates varied, with Hatchery 3 exhibiting the highest proportion of non-viable or poor-quality chicks (0.8%), which may reflect suboptimal incubation conditions or underlying genetic or nutritional issues within the breeder's flock.

In terms of daily productivity, Hatchery 2 stood out with the highest egg output (115,200 eggs/day) and chick production (95,000 chicks/day), indicating its large-scale operation capacity. Hatchery 1, despite its small scale (38,400 eggs/day), maintained powerful performance and efficiency, producing 24,500 chicks/day. Hatcheries 3 and 4 had moderate capacities, each processing 57,600 eggs/day, with corresponding chick outputs of 46,000 and 47,500, respectively.

Table 3. *Waste management and disposal practices in surveyed poultry hatcheries (N = 4).*

Items	Hatcheries				Frequency %
	1	2	3	4	
Hatchery capacity (Million/year)	12	50	18	16	
Hatchery trolleys	Multi-stage with trolley load	Single stage with trolley load	Multi-stage with trolley load	Multi-stage with trolley load	Multi-stage: 75%
Egg receiving rate	Daily	Daily	Daily	Different days	Daily: 75%
Separate unit for waste collection	Waste room outside hatchery	Waste room outside hatchery	A tank under air pressure	Waste room outside hatchery	Waste room: 75%
Amounts of wastes produced daily	400 Kg	2.5 tons	5 tons	600 Kg	
Maximum period for waste storage	Day	Day	Week	Day	Day: 75%
Waste separation before disposal	Yes	Yes	Maybe	Yes	Yes: 75%
Method of waste disposal	Burn pit	Cooperate with any services that burn (incinerator)	Contract with the municipality	Throw it in the regular trash	Contract: 50%
Pre-treatment of wastes	No treatment	No treatment	No treatment	No treatment	100%
Egg carton disposal	Sell and reuse in transporting culls, spoiled, and discarded eggs	Sold	Sell and reuse in transporting culls, spoiled and discarded eggs	Sold	Sell: 100%
Floor eggs	Incubation after disinfection	Sold	Incubation after disinfection	Incubation after disinfection	Incubation: 75%
Rejected eggs; large/ small eggs. Broken eggs; Over calcified egg. Infertile eggs; Double yolk	Sold	Sold	Sold	Sold	100%
Non-hatched eggs Early and late embryonic deaths Dead in shell	Sell as hatching waste	Cooperate with any services that burn (incinerator)	Sell as hatching waste	Garbage	Sell: 50%
Deformed chicks	Condemnation	Condemnation	Condemnation	Condemnation	100%
Condemned and Dead chicks	By burying	Incineration	In a waste tank under pressure	Disposed of in regular waste	
Fluff	Go with the wastewater	Garbage	In a waste tank	Garbage	Garbage & wastewater: 75%
Any waste sold for duck or turkey farms	Yes	No	No information	Yes	Sell: 50%

Table 4. *Microbial load (Mean log_{10} CFU ± SEM) of air, floor, and wall surfaces across different compartments of the investigated hatcheries before and after disinfection*

Item	Compartments	TBC					TCC					TFC			
		Before	After	LR%	P		Before	After	LR%	P		Before	After	LR%	P
Air *	Reception	3.05					0.80					2.23			
	Setter	3.16					1.03					1.24			
	Hatcher	3.13					2.11					1.40			
	Sorting	3.60					1.52					2.21			
	SEM	0.13					0.32					0.32			
	P-value	*0.542*					*0.52*					*0.625*			
Floor	Reception	3.94	3.16	*19.80*	*0.161*		1.31	0.69	*47.33*	*0.478*		0.73	0.17	*76.71*	*0.242*
	Setter	3.06	2.91	*4.90*	*0.831*		0.00	0.00	-	*1.000*		0.51	0.00	*100*	*0.186*
	Hatcher	4.22	3.36	*20.38*	*0.204*		1.19	1.46	*↑22.69*	*0.783*		**1.03[a]**	**0.00[b]**	*100*	***0.018***
	Sorting	4.23	3.61	*14.66*	*0.373*		1.95	0.94	*51.79*	*0.430*		0.74	0.00	*100*	*0.088*
	SEM	0.26	0.22				0.31	0.26				0.19	0.04		
	P-value	*0.300*	*0.731*				*0.134*	*0.184*				*0.809*	*0.444*		
Walls	Reception	3.04	2.60	*14.47*	*0.620*		1.18	0.42	*64.41*	*0.169*		0.46	0.18	*60.87*	*0.489*
	Setter	3.63	3.56	*1.93*	*0.840*		0.72	0.29	*59.72*	*0.361*		0.31	0.36	*↑16.13*	*0.905*
	Hatcher	3.85	3.43	*10.91*	*0.565*		1.33	0.45	*66.17*	*0.308*		0.93	0.00	*100*	*0.112*
	Sorting	2.83	3.66	*↑29.33*	*0.149*		1.14	1.23	*↑7.89*	*0.703*		0.42	0.24	*42.86*	*0.660*
	SEM	0.24	0.22				0.28	0.22				0.18	0.10		
	P-value	*0.401*	*0.334*				*0.874*	*0.482*				*0.666*	*0.613*		

[a,b] Different superscripts in the same row indicate significant differences before and after disinfection (Paired sample *t*-test, $P \leq 0.05$).
Differences between compartments within the same column were analyzed using one-way ANOVA ($P \leq 0.05$).
*Air was subjected to continuous disinfection; therefore, before-and-after comparisons were not conducted.
SEM: Pooled standard error of means.
LR%: Log reduction percentage; TBC: Total Bacterial Count; TCC: Total Coliform Count; TFC: Total Fungal Count.
↑: Indicates an increase rather than a reduction in microbial load after disinfection.

Table 5. *Effectiveness of disinfection on microbial loads of hatchery surfaces and equipment (Mean log_{10} CFU ± SEM).*

Surfaces	TBC					TCC					TFC			
	Before	After	LR%	P		Before	After	LR%	P		Before	After	LR%	P
Floor Eggs	4.26	**4.42***	*↑3.76*	*0.893*		1.43	0.00	*100*	*0.423*		1.30	0.00	*100*	*0.150*
Clean eggs	3.27	**3.15**	*3.67*	*0.861*		1.01	0.53	*47.52*	*0.724*		1.15	0.00	*100*	*0.079*
SEM	0.41	0.30				0.46	0.35				0.35	0.00		
***P*-value**	*0.308*	***0.037***				*0.952*	*0.516*				*0.850*	*1.000*		
Trays	**2.33[a]**	**1.28[b]**	*45.06*	***0.021***		**1.29[a]**	**0.16[b]**	*87.60*	***0.019***		0.32	0.05	*84.38*	*0.210*
Hatch cages	2.66	2.44	*8.27*	*0.690*		1.42	0.87	*38.73*	*0.371*		0.31	0.00	*100*	*0.139*
Transport boxes	3.01	2.09	*30.56*	*0.290*		1.43	0.41	*71.33*	*0.319*		0.10	0.06	*40.00*	*0.374*
SEM	0.20	0.24				0.26	0.18				0.10	0.03		
***P*-value**	*0.446*	*0.089*				*0.871*	*0.207*				*0.727*	*0.624*		

[a,b] Different superscripts in the same row indicate significant differences before and after disinfection (Paired sample *t*-test, $P \leq 0.05$).
* Asterisk indicates a significance between surfaces in the same column (Tukey, $P \leq 0.05$).
SEM: Pooled standard error of means
LR%: Log reduction percentage; TBC: Total Bacterial Count; TCC: Total Coliform Count; TFC: Total Fungal Count.
↑: Indicates an increase rather than a reduction in microbial load after disinfection.

Table 6. *Comparison of total bacterial counts (TBC) on the investigated hatcheries' surfaces before and after implementation of the disinfection programs (Mean log_{10} CFU ± SEM).*

Surfaces	Disinfection	Hatchery 1	Hatchery 2	Hatchery 3	Hatchery 4	SEM	*P*- value
Floors	**Before**	3.68	3.31	2.85	4.10	0.36	*0.695*
	After	4.51	3.52	3.14	2.90	0.46	*0.672*
	LR %	*↑22.55*	*↑6.34*	*↑10.18*	*29.27*		
	***P*- value**	*0.616*	*0.730*	*0.764*	*0.313*		
Corners	**Before**	3.67	**4.50***	3.72	3.42	0.21	*0.283*
	After	**5.12[a]**	**3.28[b]**	**3.06[b]**	**3.06[b]**	0.18	***<0.0001***
	LR %	*↑39.51*	*27.11*	*17.74*	*10.53*		
	***P*- value**	*0.208*	***<0.0001***	*0.274*	*0.549*		
Walls	**Before**	2.06	3.54	2.15	3.66	0.43	*0.415*
	After	3.50	3.67	3.89	2.18	0.43	*0.506*
	LR %	*↑69.90*	*↑3.67*	*↑80.93*	*40.44*		
	***P*- value**	*0.377*	*0.834*	*0.280*	*0.091*		
Trays	**Before**	**4.25***	4.48	3.27	4.11	0.27	*0.481*
	After	**0.00[b]**	**3.41[a]**	**3.02[a]**	**3.78[a]**	0.49	***0.001***
	LR %	*100*	*23.88*	*7.65*	*8.03*		
	***P*- value**	***0.010***	*0.125*	*0.802*	*0.701*		
Chicks' navel		**3.22[b]**	**3.36[b]**	**3.72[b]**	**7.31[a]**	0.42	***0.020***
Workers' hands		**1.21[c]**	**4.20[bc]**	**7.00[a]**	**5.21[b]**	0.79	***0.019***

[a, b, c] Different superscripts within the same row indicate statistically significant differences between hatcheries (Tukey, $P \leq 0.05$).
*An asterisk indicates a significant difference between values before and after disinfection within the same column (Paired sample *t*-test, $P \leq 0.05$).
SEM: Pooled standard error of means
LR%: Log reduction percentage.
↑: Indicates an increase rather than a reduction in microbial load after disinfection.

Table 7. *Comparison of total coliform counts (TCC) on the investigated hatcheries' surfaces before and after implementation of the disinfection programs (Mean log_{10} CFU ± SEM).*

Surfaces	Disinfection	Hatchery 1	Hatchery 2	Hatchery 3	Hatchery 4	SEM	*P*-value
Floors	**Before**	0.85	1.58	0.00	0.00	0.34	*0.237*
	After	2.55	2.12	0.00	0.00	0.55	*0.251*
	LR %	*↑66.67*	*↑34.18*				
	***P*- value**	*0.500*	*0.183*	*1.000*	*1.000*		
Corners	**Before**	**0.43^{b}**	**3.61^{a*}**	**0.64^{b}**	**0.69^{b}**	0.28	***<0.0001***
	After	1.31	**1.01**	0.43	0.26	0.20	*0.303*
	LR %	*↑67.18*	*72.02*	*32.81*	*62.32*		
	***P*- value**	*0.172*	***<0.0001***	*0.754*	*0.407*		
Walls	**Before**	**0.43^{b}**	**2.96^{ab}**	**0.75^{b}**	**0.00^{b}**	0.36	***0.046***
	After	**1.34^{ab}**	**3.01^{a}**	**0.00^{b}**	**0.00^{b}**	0.42	***0.007***
	LR %	*↑67.91*	*↑1.69*	*100*			
	***P*- value**	*0.423*	*0.535*	*0.391*	*1.000*		
Trays	**Before**	**4.64^{a*}**	**3.35^{ab}**	**0.00^{c}**	**1.16^{bc}**	0.63	***0.007***
	After	**0.00**	1.22	0.00	0.00	0.30	*0.441*
	LR %	*100*	*63.58*		*100*		
	***P*- value**	***0.014***	*0.236*	*1.000*	*0.423*		
Chicks' navel		0.00	0.40	0.77	0.00	0.24	0.639
Workers' hands		**0.00^{c}**	**3.42^{b}**	**5.18^{a}**	**4.47^{ab}**	0.74	**<0.001**

[a, b, c] Different superscripts within the same row indicate statistically significant differences between hatcheries (Tukey, $P \leq 0.05$).
*An asterisk indicates a significant difference between values before and after disinfection within the same column (Paired sample *t*-test, $P \leq 0.05$).
SEM: Pooled standard error of means
LR%: Log reduction percentage.
↑: Indicates an increase rather than a reduction in microbial load after disinfection.

Table 8. *Comparison of total fungal counts (TFC) on the investigated hatcheries' surfaces before and after implementation of the disinfection programs (Mean log_{10} CFU ± SEM).*

Surfaces	Disinfection	Hatchery 1	Hatchery 2	Hatchery 3	Hatchery 4	SEM	*P*- value
Floor	**Before**	1.35	0.50	0.00	0.00	0.27	*0.245*
	After	0.00	0.00	0.00	0.00	0.00	*1.000*
	LR %	*100*	*100*	*0*	*0*		
	***P*- value**	*0.189*	*0.391*	*1.000*	*1.000*		
Corners	**Before**	**0.55^{b}**	**2.36^{a*}**	**0.00^{b}**	**0.17^{b}**	0.13	***<0.0001***
	After	**0.83^{a}**	**0.00^{b}**	**0.00^{b}**	**0.00^{b}**	0.02	***<0.0001***
	LR %	*↑50.91*	*100*	*0*	*100*		
	***P*- value**	*0.885*	***<0.0001***	*1.000*	*0.333*		
Walls	**Before**	0.85	2.00	0.00	0.00	0.23	*0.058*
	After	1.00	0.00	0.00	0.00	0.18	*0.217*
	LR %	*↑15.00*	*100*	*0*	*0*		
	***P*- value**	*0.500*	*1.000*	*1.000*	*1.000*		
Trays	**Before**	**0.00^{b}**	**2.97^{a*}**	**0.00^{b}**	**0.00^{b}**	0.40	***<0.0001***
	After	0.00	**0.00**	0.00	0.00	0.00	*1.000*
	LR %	*0*	*100*	*0*	*0*		
	***P*- value**	*1.000*	***0.018***	*1.000*	*1.000*		
Chicks' navel		0.00	0.00	0.00	0.00	0.00	*1.000*
Workers' hands		0.00	2.85	2.48	0.00	0.65	*0.165*

[a, b] Different superscripts within the same row indicate statistically significant differences between hatcheries (Tukey, $P \leq 0.05$).

*An asterisk indicates a significant difference between values before and after disinfection within the same column (Paired sample *t*-test, $P \leq 0.05$).

SEM: Pooled standard error of means

LR%: Log reduction percentage.

↑: Indicates an increase rather than a reduction in microbial load after disinfection.

Table 9. *Hatchability performance parameters across the investigated commercial hatcheries.*

Parameters	Hatchery 1	Hatchery 2	Hatchery 3	Hatchery 4
Average fertility %	93	92	87	85
Early dead %	0.75	3.5	2.5	4
Middle dead %	1.25	1	0.2	0.75
Late dead %	3.25	2.5	3	3
Total embryonic deaths%	5.1	7	5.7	7.75
Contaminated %	0.5	0.5	0.5	0.5
Culls %	0.2	0.5	0.8	0.5
Average total hatchability%	88.5	84	80	78.5
Eggs production/ day	38,400	115,200	57,600	57,600
Average chick production/ day	24,500	95,000	46,000	47,500

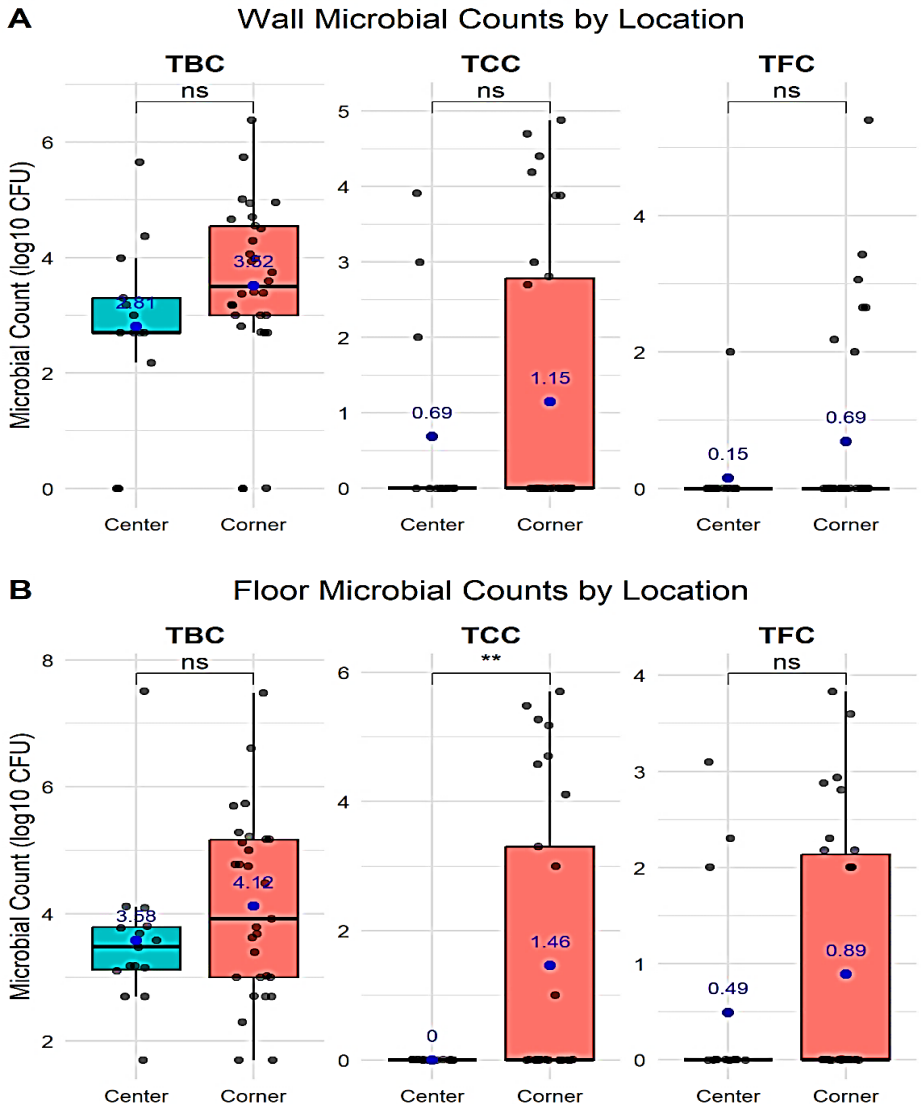

Fig. 1. Comparison of microbial counts (log_{10} CFU) between center and corner locations on walls (Panel A) and floors (Panel B) in hatcheries. TBC: Total bacterial count; TCC: Total coliform count; TFC: Total fungal count; CFU: Colony-forming units. Blue points indicate mean values. Statistical significance was determined using the independent sample *t*-test. Asterisks represent significance levels: $P < 0.05$ (*), $P < 0.01$ (**), ns: not significant.

Discussion

1. Hatcheries' biosecurity and hygiene

Biosecurity and hygiene assessments across the four hatcheries revealed inconsistent implementation of standard protocols. While basic infrastructure like fencing and car stations was present in all, only half maintained visitor logs or posted biosecurity signage. Key practices, such as footwear disinfection, visitor attire policies, and poultry ownership restrictions, were inconsistently applied, exposing biosecurity weaknesses. Effective biosecurity depends on isolation, movement control, and sanitation ***(Scott et al., 2018)***. Best practices also include remote hatchery locations at least 1.6 km from other farms, fencing, restricted-access signage, facility maps, and stringent control of personnel and vehicle access ***(World Organisation for Animal Health (WOAH), 2016)***.

Iodophors, composed of iodine and a solubilizing agent, rely on free iodine (I_2) for rapid and broad-spectrum antimicrobial action, effective even at low concentrations. Iodine disrupts microbial function by targeting sulfur-containing amino acids (e.g., cysteine, methionine), nucleotides, and fatty acids, leading to cell death ***(McDonnell & Russell, 1999)***. In contrast, quaternary ammonium compounds (QACs) are cationic surfactants that interact with negatively charged microbial surfaces via their hydrophilic heads, while their hydrophobic tails disrupt cell membranes, compromising microbial integrity ***(Ioannou et al., 2007)***. QACs are effective against gram-positive and gram-negative bacteria and enveloped viruses but are less effective in the presence of organic matter or against spores and non-enveloped viruses ***(Bek et al., 2000)***. Glutaraldehyde, a potent sterilant, retains efficacy even in organic-rich environments and acts by alkylating critical microbial functional groups, thereby disrupting RNA, DNA, and protein synthesis ***(Rutala et al., 2008)***. When combined, QACs and glutaraldehyde form a highly effective disinfectant mix, capable of neutralizing both enveloped and non-enveloped viruses in poultry operations ***(Figueroa et al., 2017)***. Half of the surveyed hatcheries used phenol-based disinfectants in dipping wheels at facility entrances. At high concentrations, phenols function as potent protoplasmic toxins, damaging cell membranes and precipitating intracellular proteins. At lower concentrations, phenol and its derivatives disrupt microbial enzyme systems and cause membrane leakage, leading to bacterial death ***(Rutala et al., 2008)***. The four hatcheries studied had capacities ranging from 12 to 50 million eggs annually, leading to variations in the number of hatchers, and incubation systems used. While 25% used single-stage trolleys, 75% operated multi-stage systems. Proper incubator management is

crucial to prevent overheating and embryo loss ***(Kolańczyk, 2020)***. In this study, all hatcheries maintained similar storage conditions, with temperatures between 15–20°C and relative humidity levels of 65–80%. However, Hatchery 4 stored eggs for a longer period. Optimal storage conditions are around 18 to 19°C for eggs stored for under a week and about 15 to 16°C for longer durations, with 60 to 70% humidity. Prolonged storage negatively impacts hatchability, with each day beyond six days reducing hatchability by 0.5 to 1.5% and delaying hatch time by around 20 minutes ***(Cobb-Vantress, 2018)***.

Half of the surveyed hatcheries employed high-risk procedures. Additionally, the same two hatcheries had rodent control plans in place, a vital element of biosecurity. All employees wear masks, gloves, and use hand sanitizers, which is crucial for effective sanitation in a hatchery. Poor sanitation can lead to low hatch rates, higher mortality during brooding, and ongoing health issues, impacting profitability. A 70% alcohol solution should be available at the entrance for hand sanitization upon entering or leaving the hatchery. Effective hand disinfection products often contain alcohol or mixtures with iodophors or chlorhexidine, targeting significant pathogens through quick action ***(Kampf & Kramer, 2004; Chojecka et al., 2017)***. Alcohol disrupts cell membranes and denatures proteins, affecting metabolism and causing cell lysis ***(McDonnell & Russell, 1999)***.

The hatchery's disinfection process began with dry cleaning to remove organic residues that can reduce disinfectant efficacy, followed by wet cleaning with detergents as recommended by CDC guidelines ***(Rutala et al., 2008)***. Half of the hatcheries primarily used quaternary ammonium compounds (QACs), while the others combined QACs with glutaraldehyde via spraying. Since hatcheries favour microorganism growth, multiple disinfectants with various modes of action, such as alcohols, phenolics, halogens, peroxides, aldehydes, QACs, and chlorhexidine, are employed to disrupt microbial cells ***(Soliman et al., 2009; Van Immerseel et al., 2009)***. The effectiveness of these disinfectants depends on several factors, including organic matter presence, water quality, temperature, pH, contact time, and concentration ***(Stringfellow et al., 2009; Møretrø et al., 2012)***.

Aerial disinfection in 75% of hatcheries was performed using thermal fogging, which produces ultra-fine droplets (1 to 50 μm) that vaporize and form aerosols, creating visible fog. This method ensures the even distribution of disinfectants in hard-to-reach areas without leaving residues and is cost-effective for large spaces like poultry facilities by reducing labor and environmental impact ***(Mitchell, 2015)***. One

hatchery used glycolic acid, a biodegradable disinfectant effective against bacteria, yeast, fungi, and viruses; it is non-staining, non-corrosive to steel and aluminium, provides coverage, prevents *Aspergillus*, and controls *Salmonella* ***(Kersia Group, 2022)***.

Paraformaldehyde was the primary disinfectant for hatching eggs in 75% of breeder farms, regarded as the gold standard for broad-spectrum fumigation due to its strong antimicrobial activity against bacteria, fungi, and viruses ***(Motola et al., 2020)***. Previous research reported that effective *Salmonella* disinfection required fumigation at 25°C for 20 minutes with 600 mg/m³ formaldehyde gas without damaging embryos ***(Cadirci, 2009)***. Most investigated hatcheries used hydrogen peroxide with silver ions or peracetic acid for secondary disinfection of hatching eggs. Hydrogen peroxide is a powerful oxidizer and a safer alternative to formaldehyde, reducing microbial contamination significantly by generating hydroxyl free radicals targeting cell components ***(McDonnell & Russell, 1999)***. Its effectiveness depends on concentration and is negatively affected by organic matter and heat ***(Dvorak, 2005; Møretrø et al., 2012)***. Silver ions' antimicrobial action is through interaction with thiol groups ***(McDonnell & Russell, 1999)***. Peracetic acid is effective even in the presence of organic matter and breaks down into non-toxic products (acetic acid, water, oxygen, and hydrogen peroxide), disrupting proteins and membranes similarly to hydrogen peroxide ***(McDonnell & Russell, 1999; Rutala et al., 2008)***.

One hatchery used sodium dichloroisocyanurate (NaDCC), which releases chlorine upon dissolution and forms hypochlorous acid (HOCl), a potent oxidizer effective against many pathogens ***(Black & Veatch Corporation, 2009)***. Compared to sodium hypochlorite, NaDCC offers higher chlorine content, better stability, resistance to organic matter inactivation, and sustained chlorine release via a “chlorine reservoir” for prolonged disinfection efficacy ***(Bloomfield, 1996; Kuechler, 1999)***.

2. Hatcheries’ waste management and disposal

The fast growth of the commercial poultry sector has resulted in intensified waste generation, including waste from hatcheries. Improper disposal can create environmental hazards and raise management costs ***(Lipdo et al., 2024)***. Sustainable waste management is a key component in reducing the environmental footprint of hatchery operations. Among the hatcheries visited, 75% stored waste in designated waste rooms outside of production areas, while one hatchery utilized a pressurized tank system. Efficient waste handling and disposal are critical aspects of biosecurity, as

having proper and separate waste storage rooms is essential for preventing contamination, controlling odors, and reducing the risk of pathogen transmission ***(Amu et al., 2005)***. Only one facility used a method like that described by ***(Glatz et al., 2011)*** utilizing sealed tanks or Bio-bins for composting. These containers improve air circulation, reduce odors and microbial load, and meet biosecurity standards, while promoting sustainable practices by converting organic waste into soil fertilizers ***(Glatz et al., 2011)***.

Hatchery waste disposal methods varied by their investment level. Hatcheries that collaborate with municipal or external services for incineration demonstrate a greater commitment to environmentally responsible waste disposal compared to those relying on open burning or regular trash systems. Open burning poses serious environmental and health risks by releasing toxic substances and particulates, contaminating soil, water, and air, and affecting both wildlife and human health ***(Secretariat of the Stockholm Convention, 2008)***. Incineration, while reducing waste volume and potentially generating energy, is costly and emits harmful pollutants ***(Wiliams et al., 1999; Tangri, 2023)***. Raw poultry waste may contain pathogens like *Clostridium*, *Salmonella*, and *Enterobacteria*, making proper treatment essential to prevent disease transmission and protect public health ***(Wiliams et al., 1999)***. Lack of pre-treatment for hatchery waste, observed across all facilities, presents a missed opportunity for safer and more sustainable disposal, particularly for biohazardous materials like dead chicks and embryonic remains.

All the hatcheries demonstrated some degree of material recycling, particularly with egg cartons and non-hatched eggs, which contribute positively toward circular economy principles. Half of the hatcheries sold egg cartons, potentially to layer farms, while the other half reused them. However, reuse poses a risk of *Salmonella Enteritidis* cross-contamination between cartons and eggshells ***(Regmi et al., 2021)***, underscoring the need for strict disinfection protocols or avoidance. Additionally, 75% of hatcheries used floor eggs for incubation after disinfection, despite their higher contamination risk and lower hatchability, as indicated by ***(Fasenko et al., 2000; Khabisi et al., 2012)***. During pre-incubation or candling, eggs are commonly discarded due to infertility, shell defects, cracks, double yolks, or abnormal size. Selection should consider egg uniformity, as it aids in optimizing incubation parameters. Ideal eggs should be clean, consistent in color, and have smooth, unbroken, and structurally sound shells ***(Cobb-Vantress, 2018)***. Eggs unsuitable for hatching are often sold, formally or informally, for use in animal feed, industrial processing,

or occasionally human consumption, offering hatcheries a way to reduce waste and increase revenue, as stated by ***(Glatz et al., 2011)***.

Cracked eggshells pose additional health concerns, as they increase the likelihood of *Salmonella* contamination ***(Patel et al., 1996)***. Selling eggs to other farms poses potential risks of transmitting pathogens like *Salmonella*, *E. coli*, and *Campylobacter* ***(Jones et al., 2004)***. Additionally, using unprocessed infertile or embryonated eggs in animal feed can recycle pathogens into the production cycle ***(Musgrove et al., 2005)***. Proper disposal of dead birds is also essential, as their remains may attract stray dogs, potentially spreading disease. Some countries prohibit using hatchery waste in byproduct meals due to the risk of pathogen transmission ***(EFSA Panels on Animal Health and Welfare (AHAW) & on Biological Hazards (BIOHAZ), 2011; Glatz et al., 2011)***

3. Hatcheries' compartments microbial load

This study found higher airborne microbial loads in hatcher rooms than in setters, which showed the lowest counts. These findings align with earlier reports by ***Magwood (1964)*** and ***Davies & Wray (1994)***, who noted peak microbial air contamination during hatching due to airborne dust, fluff, and dried feces. Airborne bacteria also correlated with surface contamination, likely due to staff activity ***(Lazarov et al., 2018)***. The floors of hatcher and sorting rooms had the highest contamination levels, which is in line with findings by ***Moustafa (2009)*** and ***Kim & Kim (2010)***, who identified floors as critical contamination points due to organic matter buildup, especially in hatchers. The hatcher is confirmed as the most contaminated zone, underscoring the need for buffer zones, restricted movement between compartments, and compartment-specific disinfection strategies, especially in high-load areas like sorting and hatching ***(Lazarov et al., 2018)***.

4. Hatcheries' surface and equipment microbial load

Eggshell disinfection generally resulted in effective microbial reductions, ranging from 3.67% to 100%. However, floor eggs showed a statistically significant difference compared to clean eggs ($P = 0.037$), suggesting that nest-collected eggs are inherently cleaner and more responsive to sanitation. Primary disinfection is essential, as high microbial loads on unsanitized eggs can decrease hatchability and chick viability ***(Scott & Swetnam, 1993; Moustafa, 2009)***.

No significant differences were observed in post-disinfection efficacy across eggshells, hatch cages, and transport boxes, possibly due to improper disinfectant concentration or exposure time. While formaldehyde remains the benchmark disinfectant, its effectiveness depends on precise

application conditions ***(Motola et al., 2023)***. Other disinfectants, like peracetic acid or hydrogen peroxide-silver ion blends, may underperform if used inappropriately or in the presence of organic matter, which can neutralize their activity and hinder biofilm removal. Trays in this study demonstrated significant microbial reductions post-disinfection, likely due to their exposure to the cleaner setter environment. This finding agrees with previous research noted that the setters and trays had minimal contamination and less likely to be *Salmonella*-positive compared to other areas ***(Kim & Kim, 2010; Oastler et al., 2022)***.

5. Hatcheries' corners microbial load

Comparative analysis of microbial counts between center and corner locations on walls and floors in hatcheries highlighted notable spatial differences in contamination patterns. Corners of both walls and floors typically had higher microbial loads, particularly for coliform ($P < 0.01$). These areas are hard-to-reach and tend to accumulate dust, fluff, and moisture, creating ideal conditions for microbial growth, indicating that these zones are critical for contamination control. This finding aligns with a study by ***Kim & Kim (2010)***, who found high bacterial contamination on surfaces in hatchery corridors and non-operating hatchers, even when air contamination was low, highlighting the importance of thorough surface cleaning.

6. Hatcheries' disinfection efficacy

Comparing the biosecurity programs across the four hatcheries revealed that egg trays and corners were the most responsive surfaces to disinfection, showing significant decreases in TBC, TCC, and TFC. Hatchery 1 showed an increase in TBC, TCC, and TFC on the floors, corners, and walls after disinfection. This increase is likely due to a lack of pre-cleaning with detergent. Additionally, Hatchery 1's poor biosecurity measures and the lack of movement restrictions among employees contributed to the spread of microbes throughout the compartments. Hatcheries 2 and 3 showed slight increases in TBC and TCC on floors and walls after disinfection, but with good overall log reductions for both hatcheries. Hatchery 4 recorded reductions in TBC, TCC, and TFC across surfaces despite maintaining inadequate biosecurity practices. This improvement is likely due to its use of QAC and glutaraldehyde in a higher concentration than in Hatchery 3. These findings highlight the importance of investigating the contamination on the surfaces inside hatcheries, as stated by ***Kim & Kim (2010)***.

7. Hatchability performance

Hatchability assessment across the four hatcheries highlighted the influence of biosecurity and management practices on performance outcomes. Hatcheries 1

and 2 exhibited higher fertility and hatchability rates, along with lower embryonic mortality and cull percentages, indicating better overall reproductive efficiency.

Hatcheries 1 and 2 employ different types of incubation systems. Single-stage (SS) system employed in Hatchery 2, though more costly, offers precise environmental control, especially temperature regulation, which enhances embryo development, nutrient absorption, and organ formation ***(Araújo et al., 2016)***. These systems are also more hygienic, as they can be fully emptied and disinfected, reducing contamination risk and improving biosecurity. Studies consistently show that SS systems achieve better hatchability and chick quality than multi-stage (MS) systems ***(Mauldin, 2006; Mesquita et al., 2021)***. Conversely, MS systems in Hatchery 1 are more energy-efficient by reusing heat from older embryos, but they may suffer from inconsistent temperatures, potentially harming developing embryos ***(Araújo et al., 2016)***.

However, Hatcheries 1 and 3 showed the lowest total embryonic mortality rates (5.1% and 5.7%), but Hatchery 1 exhibited the lowest early (0.75%). This may be linked to the distinct use of hydrogen peroxide+silver ions for primary egg sanitization, unlike the other hatcheries that used formaldehyde gas. The type of egg disinfectant significantly influences hatchability and chick viability. Formaldehyde, though widely used for its broad-spectrum antimicrobial activity ***(Motola et al., 2023)***, can cause harm when improperly applied, especially during early incubation. Excessive exposure has been shown to damage embryonic tracheal tissue and disrupt DNA and RNA function through alkylation ***(Hayretdağ & Kolankaya, 2008)***. Conversely, hydrogen peroxide is a safer option; ***Sheldon (1990)*** found that its use improved hatchability by 2–3%, increasing it from 87.6% with formaldehyde to 90.5%.

Hatchery 1, while demonstrating the highest hatchability among the four hatcheries, also recorded the highest rate of late embryonic death, potentially due to incorrect incubation parameters such as temperature, humidity, and ventilation. These conditions can cause oxygen deficiency, especially in multistage (MS) incubation systems, where heat from older embryos may create thermal imbalances ***(Araújo et al., 2016)***. Additionally, bacterial infections may contribute to late embryonic death. Studies by ***Ibrahim et al. (2024)*** highlight *Pseudomonas aeruginosa* as a significant cause of in-shell embryonic mortality, often introduced through environmental contamination or eggshell penetration, leading to elevated embryo and chick losses.

Hatchery 4, which stored eggs for extended periods (3 to 35 days), recorded the lowest hatching performance with total embryonic

mortality rates of 7.75%. Prolonged egg storage negatively affects hatchability and increases microbial load. Although short-term storage (up to 7 days at 18–20°C and 75% RH) does not significantly impair hatchability, extended storage durations lead to greater embryonic loss ***(Fasenko et al., 2001)***. Additionally, Hatchery 4's low fertility rates (85%) and elevated early embryonic mortality (4%) likely reflect the use of aged breeder flocks. Breeder age influences egg quality; older hens lay eggs with thinner, more porous shells, accelerating gas exchange and moisture loss during storage and incubation. Studies have shown that older breeders exhibit significantly higher early embryonic mortality, increased cull rates, and reduced hatchability ***(Perić et al., 2022)***.

Conclusion:

The results obtained highlight the persistent risk of microbial contamination in key hatchery zones, such as hatchers and egg reception areas, and emphasize the profound impact these contaminants can have on hatching results. This underscores the need for a comprehensive biosecurity strategy that includes systematic monitoring, microbial assessment, and targeted sanitation protocols. Effective disinfection depends not only on the product but also on its application. Alternatives to formaldehyde, such as hydrogen peroxide, have proven safer and more effective, reducing early embryonic mortality and enhancing hatchability. However, even the best products fail without proper application, contact time, and attention to environmental conditions. Ultimately, hatchery success relies on a multi-layered approach: strict hygiene practices, informed disinfectant choices, continuous staff training, and section-specific cleaning programs. When these elements work together, the result is not just cleaner facilities, but healthier chicks, higher hatchability, and a stronger, more resilient poultry industry.

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الأمن الحيوي، والتخلص من المخلفات، وتدابير الاستدامة في بعض معامل تفريخ الدواجن التجارية

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تُعدّ معامل التفريخ جزءًا أساسيًا من سلسلة إنتاج الدواجن، حيث تمثل حلقة وصل حيوية بين مزارع طيور الأمهات ومزارع التسمين التجارية. كما تمثل محورا أساسيًا لضمان إنتاجية قطعان الدواجن، الا أنها قد تكون مصدرا أساسيا وهاما من مصادر التلوث الميكروبي ونقل مسببات الأمراض المختلفة. مما يؤثر بالسلب على نسب الفقس وجودة الكتاكيت الفاقسة. أجريت هذه الدراسة لتقييم برامج الأمن الحيوي وإجراءات التخلص من المخلفات المتبعة في بعض معامل التفريخ التجارية، ومدى تأثيرها على نتائج الفقس وجودة الكتاكيت. ولذلك، تم اجراء الفحوصات البكتيرية والفطرية لعينات من الهواء، وأيدي العاملين، وبيض التفريخ، والأسطح البيئية، وسرة الكتاكيت، التي قد تم جمعها من أربعة من معامل التفريخ التجارية. وقد تم فحص هده العينات قبل وبعد تطبيق إجراءات التطهير، وفقًا لبرامج الأمن الحيوي المتبعة في هده المعامل وقد تم حساب قيم لوغاريتم 10 ومقارنة النتائج إحصائيًا.
أظهرت النتائج عدم التزام برامج الأمن الحيوي المتبعة في معامل التفريخ الأربعة بالبروتوكولات القياسية. حيث سجلت جميع العينات مستويات مختلفة من المحتوي الميكروبي، كما أظهرت درجات متفاوتة من المقاومة للمطهرات الكيميائية المستخدمة. وقد برزت الأرضيات، وكذلك زوايا الجدران والأرضيات، كنقاط تلوث رئيسية. وأيضا برزت سُرّة الكتاكيت وأيدي العاملين كنقاط تلوث حرجة في جميع المعامل التي تمت زيارتها. أما بالنسبة لمخلفات معامل التفريخ فقد خزّن 75% من المعامل النفايات في غرف نفايات مخصصة خارج مناطق الإنتاج، بينما استخدم معمل واحد فقط نظام خزانات مضغوطة. بينما قامت جميع المعامل بإعادة تدوير كراتين البيض وبيع بعض النفايات. أظهر اثنان فقط من المعامل معدلات خصوبة ونسب فقس مقبولة نسبيًا، إلى جانب انخفاض نسب نفوق الأجنة ونسب الكتاكيت الفرزة. وقد أظهرت النتائج مدى تأثير ممارسات الأمن الحيوي واستخدام نظم التحضين المختلفة على معدلات الفقس وجودة الكتاكيت.
وقد خلص البحث الى أن التطبيق الجيد لبرامج الأمن الحيوي الشاملة والتي تتضمن اتباع الطرق الصحية الفعالة لإزالة الملوثات والإدارة المستدامة للمخلفات يؤدى الى انخفاض الحمل الميكروبي كما يعزز نتائج الفقس وجودة الكتاكيت، مما يساهم في توفير بيئة أكثر أمانًا وكفاءة لإنتاج الدواجن كما يعد دمج الصرف الصحي المستهدف مع إدارة المخلفات المنظمة والمستدامة أمرًا ضروريًا لتحسين الحالة الصحية لمعامل التفريخ وتقليل التأثيرات البيئية.

الكلمات المفتاحية: معامل التفريخ، الأمن الحيوي، نسبة الفقس، صحة معامل التفريخ، مخلفات معامل التفريخ، بيض التفقيس، والمُطهرات.