Strategies to Prevent (STOP) Spillover

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Assessing the Effectiveness of an Intervention to Reduce Risk of Avian Influenza Virus Transmission in a Live Poultry Shop in Dhaka City, Bangladesh

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Background

- Live bird markets (LBMs) act as a hub for the zoonotic transmission of highly pathogenic avian influenza viruses
- LBMs in Bangladesh are characterized by inadequate infrastructure and biosafety conditions
- We developed and implemented an intervention in a live poultry shop to improve biosecurity and hygiene practices

Objective

To assess the effectiveness of the intervention in reducing influenza A in the shop's environment

Methods

- **Study period:** September 2023 to March 2024
- **Study site:** One intervention and two control shops in Dhaka city
- **The intervention:**
  - ✓ Infrastructural improvements (9-28 Nov, 2023)
    - ❑ Washable tiled floor and wall
    - ❑ Washable cage, tray and slaughtering equipment
    - ❑ Adequate water supply
    - ❑ Ventilation system to maintain unidirectional flow

Intervention shop before renovation

Intervention shop after renovation

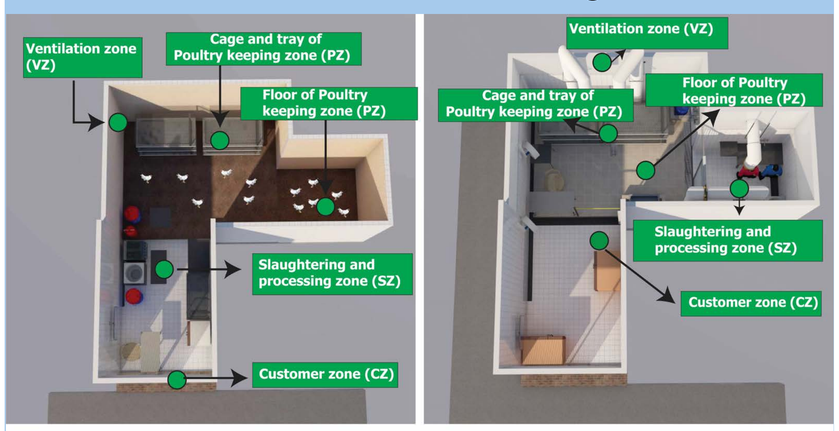
  - ✓ Training of workers (in two rounds in 29 Nov, 2023 and 28 Jan, 2024) (Figure 1)
    - ❑ Biosecurity practices
      - ❖ Cleaning and disinfection of cage, equipment and shop surfaces, waste disposal and rodent control
    - ❑ Biosafety/hygiene practices
      - ❖ Handwashing, use of apron and mask, changing cloths and shoes

Figure 1. Biosecurity and biosafety training to workers in the intervention shop

- **Sample collection**
  - ✓ **Duration**
    - ❑ Before intervention – four weeks (4 x 2 days)
    - ❑ After intervention- eight weeks (8 x 2 days)
  - ✓ **Types of sample collected:**
    - ❑ **Cloacal swab**
      - ❖ Samples form live chicken (Figure 3)
      - ❖ Collection time: before starting the business

Methods (cont.)

- ❑ **Air sample**
  - ❖ Particle size captured: < 1µm, 1-4 µm, and > 4 µm size particles
  - ❖ Machine used: National Institute of Occupational Safety and Hazards (NIOSH) bioaerosol cyclone sampler (BC251)
  - ❖ Collection site: CZ, PZ, SZ and VZ (Figure 2)
  - ❖ Collection time: Busiest hour of selling



Shop area was divided into four zones such as customer zone (CZ), poultry keeping zone (PZ), slaughtering-processing zone (SZ) and ventilation zone (VZ) (if any) . For environmental swab sampling, PZ was further divided into two areas such as cage and tray, and floor

Figure 2: Sample collection points (air and environment) in intervention shop

- ❑ **Environmental swab**
  - ❖ Collection site: the CZ, cage and PZ (cage and tray), floor of PZ and SZ (Figure 2)
  - ❖ Collection time: before and after disinfection protocols
- ❑ **Waste effluent**
  - ❖ Collection site: Drain of the shop
  - ❖ Collection time: At the end of the business hour

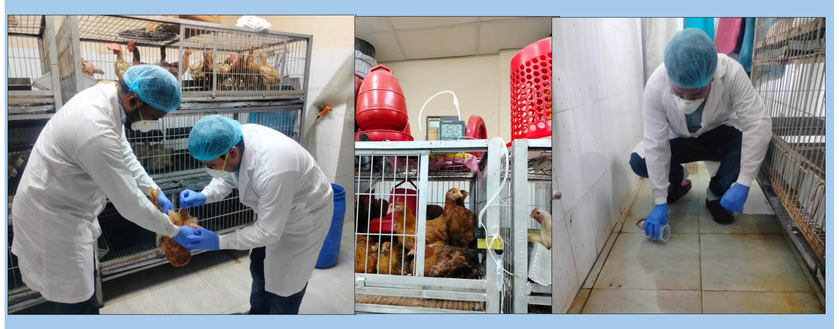


Figure 3: Collecting samples from intervention shop

Data analysis

- ✓ Compared the test results before and after intervention in intervention and control shop
- ✓ Compared the test results between the intervention and control shop before and after intervention
- ✓ Used T-test and Z-test according to the needs and in both cases T and Z signified at 0.05 level

Results

- ❑ **Cloacal swab samples from live chickens**
- ✓ No significant difference in AIV positivity and average AIV Ct values in chickens between intervention and control shops before (P=0.72) and after intervention (P=0.84)
- ❑ **Air samples**
- ✓ PZ and SZ had significantly lower viral load (higher Ct value) in ≥ 1 µm sized particles after intervention (Figure 4)

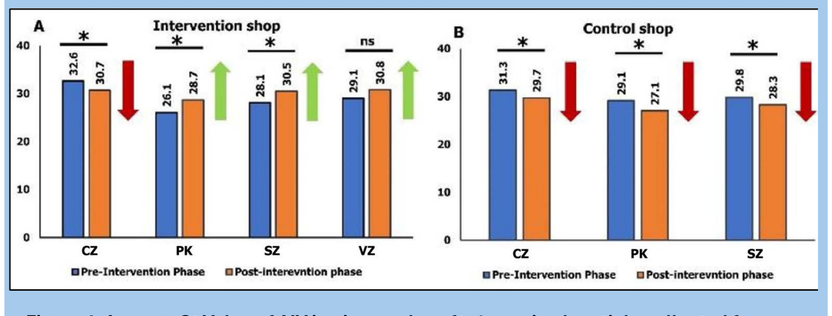


Figure 4. Average Ct Value of AIV in air samples of ≥ 1 µm sized particle collected from A) Intervention shop B) Control shop; * signifies P<0.05; ns signifies non-significant; indicates decreased Ct value, indicates increased Ct value

Results (cont.)

- ❑ **Environmental samples**
  - In intervention shop, reduction in AIV positivity (%) were observed in all sampling zones after detergent cleaning (Table 1)

Table 1. AIV A positivity (%) in environmental swab samples of control and intervention shops during post-intervention phase

Zone of shop	Control shop		P value	Intervention shop		P value
	Before n/N (%)	After n/N (%)		Before n/N (%)	After n/N (%)	
CZ	23/32 (72)	28/32 (88)	0.12	13/16 (81)	9/16 (56)	0.06
PZ (cage and tray)	31/32 (97)	32/32 (100)	0.31	30/32 (94)	24/32 (75)	0.04
PZ (floor)	29/32 (91)	30/32 (94)	0.64	15/16 (94)	5/16 (31)	0.00
SZ	30/32 (94)	31/32 (97)	0.55	14/16 (88)	11/16 (69)	0.19

- Among all zones of intervention shop, most significant changes were noticed in PZ (floor) (30.5 vs 33.8; P=0.02) and in SZ (27.9 vs 32.5; P=0.01) (Figure 5)

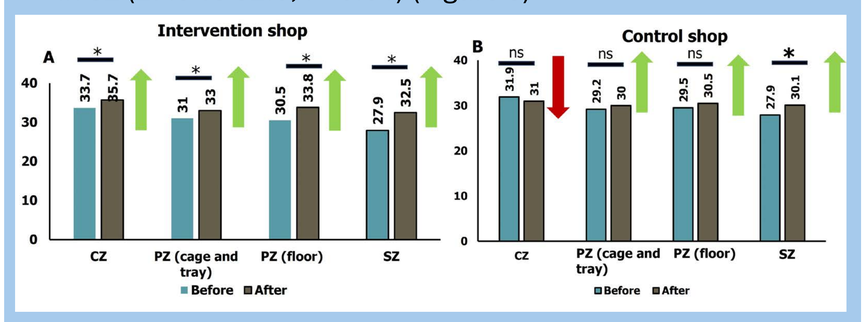


Figure 5. (A) Average Ct Value of AIV in environmental samples of collected from intervention shop and (B) control shop; * signifies P<0.05; ns signifies non-significant; indicates decreased Ct value, indicates increased Ct value

- After intervention, a notable decrease in AIV positivity in waste effluent samples were found in intervention shop (31%) compared to control shop (100%) (Figure 6.A)
- In addition, viral load was also reduced significantly (higher Ct value) in intervention shop's waste effluent samples compared to control shop's samples (26.8 vs 32.3, P=0.001) (Figure 6.B)

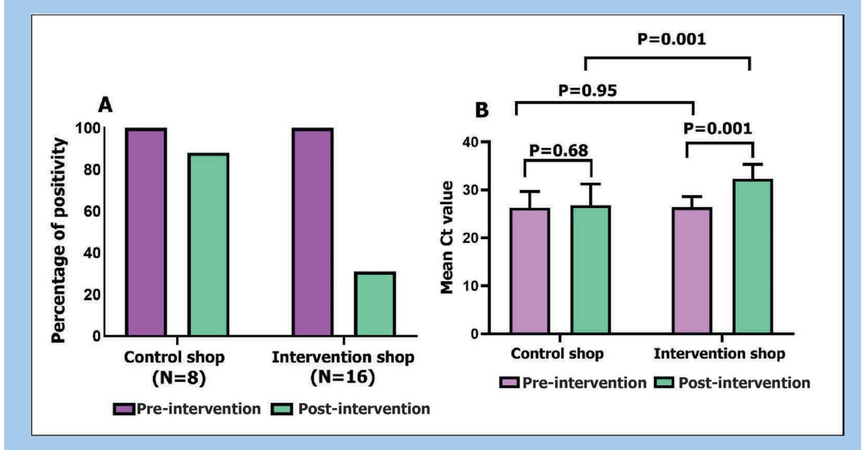


Figure 6. (A) AIV positivity (%) and (B) average Ct value of AIV in WES collected from control and intervention shop; * signifies P<0.05; ns signifies non-significant

Conclusions

- The intervention reduced the viral load of AIV in air and environmental samples in the intervention shop, though it did not completely eliminate pathogen presence
- This study demonstrates that the approaches considering infrastructural, logistic and behavioural requirements to maintain biosecurity and hygiene can be effective at reducing pathogen presence in LBMs, reducing transmission risk to humans

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