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References

1. Dixon LK, Escribano JM, Martins C, Rock DL, Salas ML, Wilkinson PJ. 2005: Asfarviridae. In: Fauquet CM, Mayo MA, Maniloff J, Desselberger U, Ball LA, editors. Virus taxonomy, VIIIth Report of the International Committee on Taxonomy of Viruses. London: Elsevier Academic Press. p. 135–43.
2. Bastos AD, Penrith ML, Crucière C, Edrich JL, Hutchings G, Roger F, et al. Genotyping field strains of African swine fever virus by p72 gene characterization. Arch Virol. 2003;148:693–706. http://dx.doi.org/10.1007/s00705-002-0946-8
3. Boshoff CI, Bastos AD, Gerber LJ, Vosloo W. Genetic characterization of African swine fever viruses from outbreaks in southern Africa (1973–1999). Vet Microbiol. 2007;121:45–55. http://dx.doi.org/10.1016/j.vetmic.2006.11.007
4. Rowlands RJ, Michaud V, Heath L, Hutchings G, Oura C, Vosloo W, et al. African swine fever virus isolate, Georgia, 2007. Emerg Infect Dis. 2008;14:1870–4. http://dx.doi.org/10.3201/eid1412.080591
5. Chapman DA, Darby AC, Da Silva M, Upton C, Radford AD, Dixon LK. Genomic analysis of highly virulent Georgia 2007/1 isolate of African swine fever virus. Emerg Infect Dis. 2011;17:599–605. http://dx.doi.org/10.3201/eid1704.101283
6. Gallardo C, Anchuelo R, Pelayo V, Poudevigne F, Leon T, Nzoussi J, et al. African swine fever virus p72 genotype IX in domestic pigs, Congo, 2009. Emerg Infect Dis. 2011;17:1556–8.
7. Lubisi BA, Bastos ADS, Dwarka RM, Vosloo W. Molecular epidemiology of African swine fever in east Africa. Arch Virol. 2005;150:2439–52. http://dx.doi.org/10.1007/s00705-005-0602-1
8. Misinzo G, Magambo J, Masambu J, Yongolo MG, Van Doorsselaere J, Nauwynck HJ. Genetic characterization of African swine fever viruses from a 2008 outbreak in Tanzania. Transbound Emerg Dis. 2011;58:86–92. http://dx.doi.org/10.1111/j.1865-1682.2010.01177.x
9. Misinzo G, Jumapili F, Ludosha M, Mafie E, Silialis J, Mushi R, et al. Genotyping of African swine fever virus from a 2009 outbreak in Tanzania. Research Opinions in Animal and Veterinary Science. 2012;2:334–8.
10. Wambura PN, Masambu J, Msami H. Molecular diagnosis and epidemiology of African swine fever outbreaks in Tanzania. Vet Res Commun. 2006;30:667–72. http://dx.doi.org/10.1007/s11259-006-3280-x

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Controlling Highly Pathogenic Avian Influenza, Bangladesh

To the Editor: Highly pathogenic avian influenza (HPAI) A(H5N1) virus is a deadly zoonotic pathogen. Since 2003, HPAI infections have been reported in millions of poultry and wild birds from 63 countries (*1*) and in 598 humans, among whom there have been 352 reported deaths in 15 countries (*2*). HPAI (H5N1) virus is endemic in Bangladesh, and the first outbreak occurred in March 2007. Since then, the virus has spread to 49 of 64 districts in Bangladesh, and samples from 536 farms have tested positive for the virus. Bangladesh now ranks among countries worldwide with the highest reported number of HPAI outbreaks (*1*). Intermittent outbreaks in Bangladesh and clusters of disease across the border in northeastern India are dramatic reminders that the emergence of new, mutant viruses in developing countries could lead to a pandemic among humans. Six cases of nonfatal HPAI (H5N1) infection have been reported in Bangladesh (*2*). Live bird markets that are in poor physical condition and that lack or have poor biosecurity are probable sources of HPAI transmission to humans and for bird-to-bird transmission (*3–5*).

In 2008, a global project of the United States Agency for International Development, Stamping Out Pandemic and Avian Influenza (STOP AI), was initiated in Bangladesh. The project began with biosecurity training for veterinarians and livestock science graduates on some large-scale commercial farms. The local STOP AI office was established in Dhaka, the capital of Bangladesh, in February 2009, and the organization managed the project through its completion in September 2010 (online Technical Appendix Figure 1, wwwnc.cdc.gov/EID/pdfs/12-0635-Techapp.pdf). STOP AI initially organized 7 highly successful live bird market biosecurity training programs in 5 geographic divisions of Bangladesh; later, STOP AI piloted cleaning and disinfection activities in 2 live bird markets, Mohammadpur and Kaptan Bazaar, in Dhaka by working closely with the United Nations' Food and Agriculture Organization. The Food and Agriculture Organization subsequently conducted cleaning and disinfection activities in 24 other markets within Dhaka and other districts in Bangladesh.

We focused on understanding the inter-relationships among household poultry producers, commercial farmers, suppliers, transporters, processors, and consumers that facilitate the process of producing and moving poultry, i.e., the entire poultry value chain (PVC). We describe how improved biosecurity on poultry farms and hygienic standards in live bird markets can reduce HPAI outbreaks. In resource-limited countries, like Bangladesh, these improvements can be made through training, technical support, financial assistance for infrastructure renovations, and incentive-driven trust-building between service

providers and key PVC stakeholders. To determine whether interventions would reduce the number of HPAI infections, we implemented changes during 2009–2010 in 2 districts in Bangladesh, Gazipur and Dinajpur, that had a high number of cases (online Technical Appendix Figure 2).

Using field-tested questionnaires, we conducted a baseline survey during in-person interviews with 1,372 poultry stakeholders (Table). Stakeholder workshops were held in each district to share survey findings and design biosecurity improvement programs. STOP AI implemented biosecurity training for 1,319 people in 53 subsector-specific 1-day sessions in Gazipur and Dinajpur (online Technical Appendix Table). We created biosecurity improvement models (e.g., farm boundary, footbath) in 12 commercial farms in Gazipur and selected 2 live bird markets in each district for infrastructure improvements, including biogas and compost plants, that were needed for the cleaning and disinfection activities (online Technical Appendix Figure 3). We provided technical support and ≤25% ($750) of the cost for each farm and ≤50% ($10,000) for each market on a cost-sharing basis.

After completion of all interventions, we conducted a final survey of 514 poultry stakeholders, including 70% of the original trainees from both districts (Table). We analyzed pre- and post-intervention survey data by using GraphPad Software (www.graphpad.com/quickcalcs/index.cfm). The results indicated that awareness of the proper disposal of birds that were culled or died because of HPAI had increased in both districts ($p<0.0001$); awareness of human HPAI cases rose substantially ($p<0.0001$); an understanding of how HPAI is spread (e.g., through sick or wild birds) changed ($p<0.001$); use of personal protective equipment (masks, gloves) and other precautionary measures (washing hands) increased ($p<0.0001$); awareness of protecting birds from HPAI (e.g., separately housing chickens and ducks) increased ($p<0.05$); and a preference for purchasing slaughtered birds instead of live birds at the markets increased ($p<0.0001$).

Substantially fewer HPAI outbreaks were reported and no clusters of infection were found during our intervention, 2009–2010 (online Technical Appendix Figure 1), probably indicating that control measures were effective. The challenge now is to sustain the progress that has been made. Several months after completion of the STOP AI interventions, their effect on the incidence of disease in Bangladesh was limited. However, STOP AI could not be expected in the short

Table. Analysis of pre- and postintervention survey data for biosecurity practices for HPAI (H5N1) virus in Gazipur and Dinajpur districts, Bangladesh, 2009–10*

| | No. persons surveyed (% aware of practice; 95% CI) | | | |
| | Gazipur | | Dinajpur | |
Biosecurity practice	Baseline survey, n = 821	Final survey, n = 300†	Baseline survey, n = 525	Final survey, n = 209†
Awareness of bird deaths caused by HPAI	191 (23; 21–26)	219 (73; 68–78)	116 (22; 19–26)	88 (42; 36–49)
Awareness of bird culling resulting from HPAI	163 (20; 17–23)	186 (62; 56–67)	56 (11; 8–14)	47 (22; 17–29)
Awareness of HPAI cases among humans	138 (17; 14–20)	172 (57; 52–63)	21 (4; 3–6)	113 (54; 47–61)
Understand how HPAI is spread				
Do not know how HPAI is spread	209 (25; 23–29)	42 (14; 11–18)	286 (54; 50–59)	21 (10; 7–15)
Perceive that wild birds are the cause	466 (57; 53–60)	207 (69; 64–74)‡	134 (26; 22–29)	154 (74; 67–80)
Recognize sick poultry as a vector	43 (5; 4–7)	140 (47; 41–52)	53 (10; 8–13)	61 (29; 23–36)
Awareness of how to protect people				
Wear masks	190 (23; 20–26)	204 (68; 63–73)	44 (8; 6–11)	94 (45; 38–52)
Wear gloves	122 (15; 13–17)	136 (45; 40–50)	54 (10; 8–13)	75 (36; 30–43)
Wash hands	207(25; 23–29)	166(55; 50–60)	58 (11; 9–14)	139 (67; 60–73)
Kids should not handle birds	3 (0; 0.1–1)	203 (68; 62–73)	1 (0; 0.01–1)	54 (26; 20–32)
No need to protect	243 (30; 27–33)	0 (0)	294 (56; 52–60)	4 (2; 0.6–5)
Awareness of how to protect birds				
Separate chickens and ducks	28 (3; 3–4)	92 (31; 26–36)	18 (3; 2–5)	36 (17; 13–23)
Clean and disinfect poultry cages	288 (35; 32–38)	182 (61; 55–66)	39 (7; 5–10)	99 (47; 41–54)
Restrict entry to farms	226 (28; 25–31)	131(44; 38–49)	56 (11; 8–14)	56 (27; 21–33)
Vaccinate against Newcastle disease	12 (1; 0.8–2)	67 (22; 18–27)	7 (1; 0.6–3)	47 (22; 17–29)
Properly dispose of feces	158 (19; 17–22)	79 (26; 22–32)§	16 (3; 2–5)	117 (56; 49–63)
Wear proper clothing	38 (5; 3–6)	67 (22; 18–27)	29 (6; 4–9)	43 (21; 16–27)
Clean and disinfect transport vehicles	35 (4; 3–6)	105 (35; 30–40)	10 (2; 1–4)	20 (10; 6–14)
Keep dogs and cats away from farms	92 (11; 9–14)	99 (33; 28–39)	22 (4; 3–6)	33 (16; 11–21)
Do not know	170 (21; 18–24)	46 (15; 12–20)¶	267 (51; 47–55)	2 (1; 0.04–4)
Bird purchase preference				
Dead bird (slaughtered at market)	152 (19; 16–21)	136 (45; 40–51)	50 (10; 7–12)	70 (33; 27–40)
Live bird (slaughtered at home)	652 (79; 77–82)	167 (56; 50–61)	484 (92; 90–94)	136 (65; 58–71)

*HPAI, highly pathogenic avian influenza.
†Two-sided χ^2 test of significance compared with baseline data had p value of <0.0001, except as noted.
‡p = 0.0002.
§p = 0.013.
¶p = 0.049.

term to dramatically reduce the high incidence of HPAI in Bangladesh. We have progressively and dramatically increased the scope and benefits of our pilot PVC implementation program, but additional work is needed. To help spread PVC approaches throughout the country, community leaders, imams of local mosques, and school teachers can be trained to implement awareness programs on safe practices for raising poultry and regular cleaning and disinfection of live bird markets. The strengthening of biosecurity measures will help control the spread of HPAI virus and other zoonotic diseases.

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References

1. World Health Organization for Animal Health. Update on highly pathogenic avian influenza in animals (type H5 and H7) [cited 2012 Mar 19]. http://www.oie.int/animal-health-in-the-world/update-on-avian-influenza/2012/
2. World Health Organization. Cumulative number of confirmed human cases of avian influenza A(H5N1) reported to WHO [cited 2012 Mar 19]. http://www.who.int/influenza/human_animal_interface/H5N1_cumulative_table_archives/en/index.html
3. Biswas PK, Christensen JP, Ahmed SS, Barua H, Das A, Rahman MH. at al. Avian influenza outbreaks in chickens, Bangladesh. Emerg Infect Dis. 2008;14:1909–12. http://dx.doi.org/10.3201/eid1412.071567
4. Biswas PK, Christensen JP, Ahmed SS, Das A, Rahman MH, Barua H, et al. Risk for infection with highly pathogenic avian influenza virus (H5N1) in backyard chickens, Bangladesh. Emerg Infect Dis. 2009;15:1931–6. http://dx.doi.org/10.3201/eid1512.090643
5. Biswas PK, Rahman MH, Das A, Ahmed SS, Giasuddin M, Christensen JP. Risk for highly pathogenic avian influenza H5N1 virus infection in chickens in small-scale commercial farms, in a high-risk area, Bangladesh, 2008. Transbound Emerg Dis. 2011;58:519–25. http://dx.doi.org/10.1111/j.1865-1682.2011.01235.x

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Hepatitis E Virus Genotype 3 in Shellfish, United Kingdom

To the Editor: Bivalve mollusks (shellfish), such as mussels and oysters, are filter feeders; they concentrate microorganisms of human and animal origin (up to 100×) from the surrounding environment. Several recent reports have linked the incidence of human infection with hepatitis E virus (HEV) to consumption of undercooked pork, game products, and shellfish (*1,2*). Infectious HEV has been found in swine manure and wastewater (*3*); therefore, application of manure to land and subsequent runoff could contaminate coastal water, leading to contamination of shellfish and, subsequently, possible human infection. Because they are filter feeders, bivalve mollusks are biologically relevant sentinels and can indicate potential pathogens that are contaminating the environment. It is essential to ensure that this sustainable resource of coastal areas, where mussels and oysters are farmed or collected wild, is not subjected to environmental contamination that could lead to public health risks.

Risk management for bivalve mollusks, aimed at control of fecal pollution, relies heavily on the use of *Escherichia coli* as an indicator of fecal (sewage) contamination and is enacted under European food regulations (Regulation 854/2004, www.cefas.co.uk/media/455777/extract_reg_no_854_2004.pdf). However, although these regulations probably reduce the number of infections, especially bacterial infections, they are not viewed as adequately controlling the risk for viral infections. Specific risks are posed by the robustness of viruses in the environment and the different behavior of viruses within bivalve mollusks compared with behavior within bacterial fecal indicators.

HEV is deemed to be inactivated during processing procedures used to prepare mussels for consumption; however, HEV is only 50% inactivated at 56°C and 96% at 60°C for 1 hour, it is stable when exposed to trifluorotrichloroethane, and it is resistant to inactivation by acidic and alkaline conditions (*4*). Most shellfish are usually eaten raw, but viable virus can also pose a risk to public health in shellfish that are lightly steamed or preserved by smoking and/or in acetic acid. Indeed, a recent study by the Food Standards Agency, in which >800 oyster samples from 39 growing beds in the United Kingdom were collected and screened during 2009–2011, found norovirus at low levels in at least 76% of oysters (*5*). Other studies identified hepatitis A virus and norovirus in shellfish production areas and in ready-to-eat products in the United Kingdom (*1,6*). In fact,

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Technical Appendix

Table. Poultry subsector–specific biosecurity training in Gazipur and Dinajpur districts, Bangladesh, 2010*

Category	Gazipur		Dinajpur		Total	
	Sessions	People	Sessions	People	Sessions	People
Commercial farmers	8	200	6	150	14	350
Hatchery owners	1	21	1	25	2	46
Poultry transporters	5	125	6	150	11	275
Live bird market vendors	4	98	6	150	10	248
Backyard producers	4	100	12	300	16	400
Total	**22**	**544**	**31**	**775**	**53**	**1319**

*Each training session covered the following topics: an introduction to avian influenza; basic principles of biosecurity; biosecurity measures for specific subgroup; cleaning and disinfection principles; and biosecurity planning for specific subsector (replaced by "public health importance" for backyard subsector). All materials were translated into Bengali. Each of the training sessions had a capacity of 25 participants and each backyard session contained $\geq$40% woman participants. A demonstration for backyard producers included thorough cleaning and disinfection of a backyard poultry house done by the trainer assisted by a female participant. We learned that virtually all respondents who received our training tried to implement biosecurity practices they learned, but backyard farmers tended to adopt more biosecurity practices than other beneficiaries, and women tend to adopt more than men.
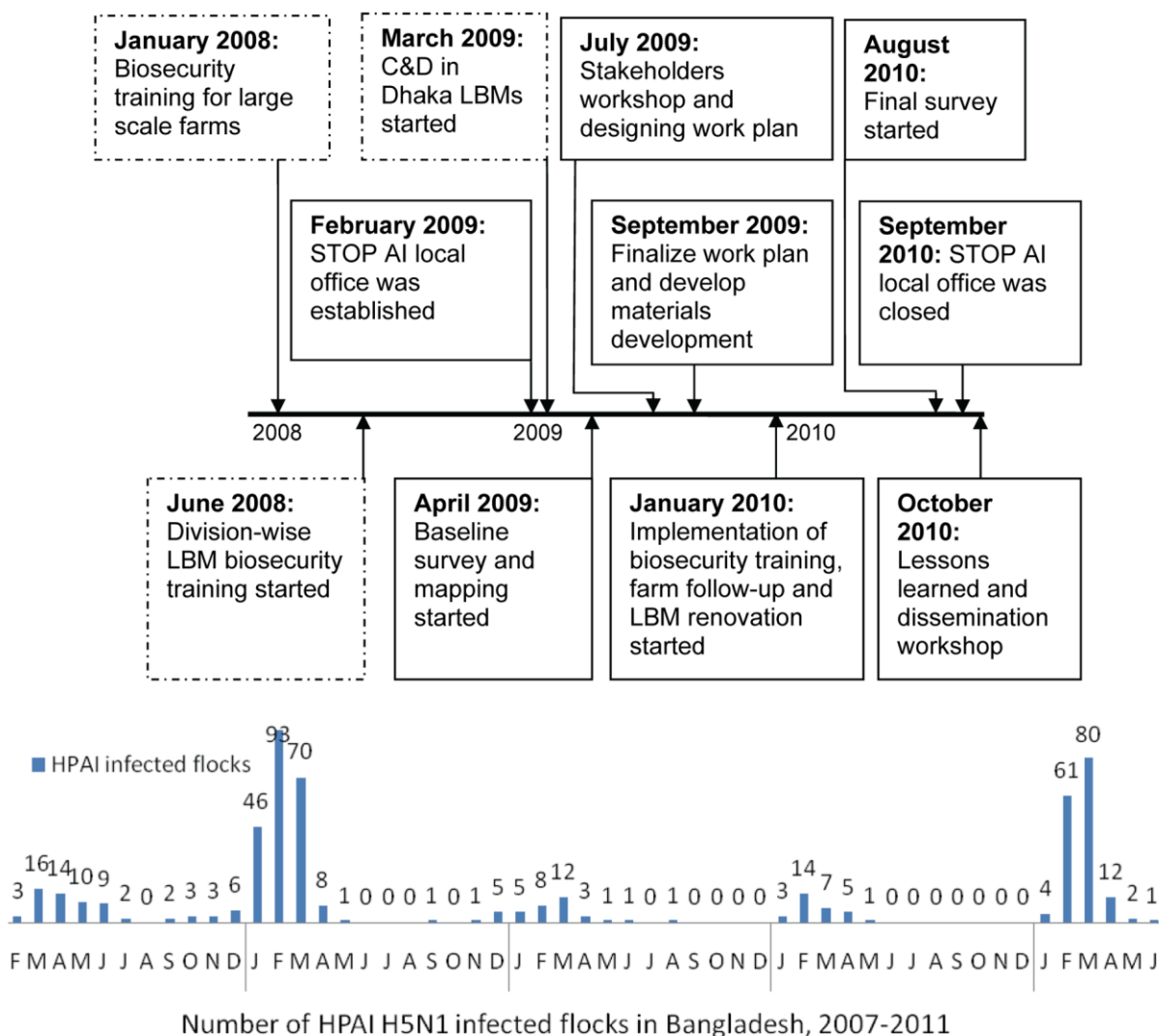
Figure 1. Timeline showing key events of Stamping Out Pandemic and Avian Influenza (STOP AI) project activities, 2008–2010, and weekly number of highly pathogenic avian influenza (HPAI) subtype H5N1 virus outbreaks, Bangladesh, 2007–2011. The activities within dashed-dotted areas are not included in this poultry value chain study. Significantly less numbers of outbreaks were reported during and after the STOP AI intervention. Revised HPAI control strategy (active surveillance and culling) implemented by the government of Bangladesh during that period also may have helped to reduce the number of HPAI outbreaks (1). C&D, cleaning and disinfection; LBM, live bird market.
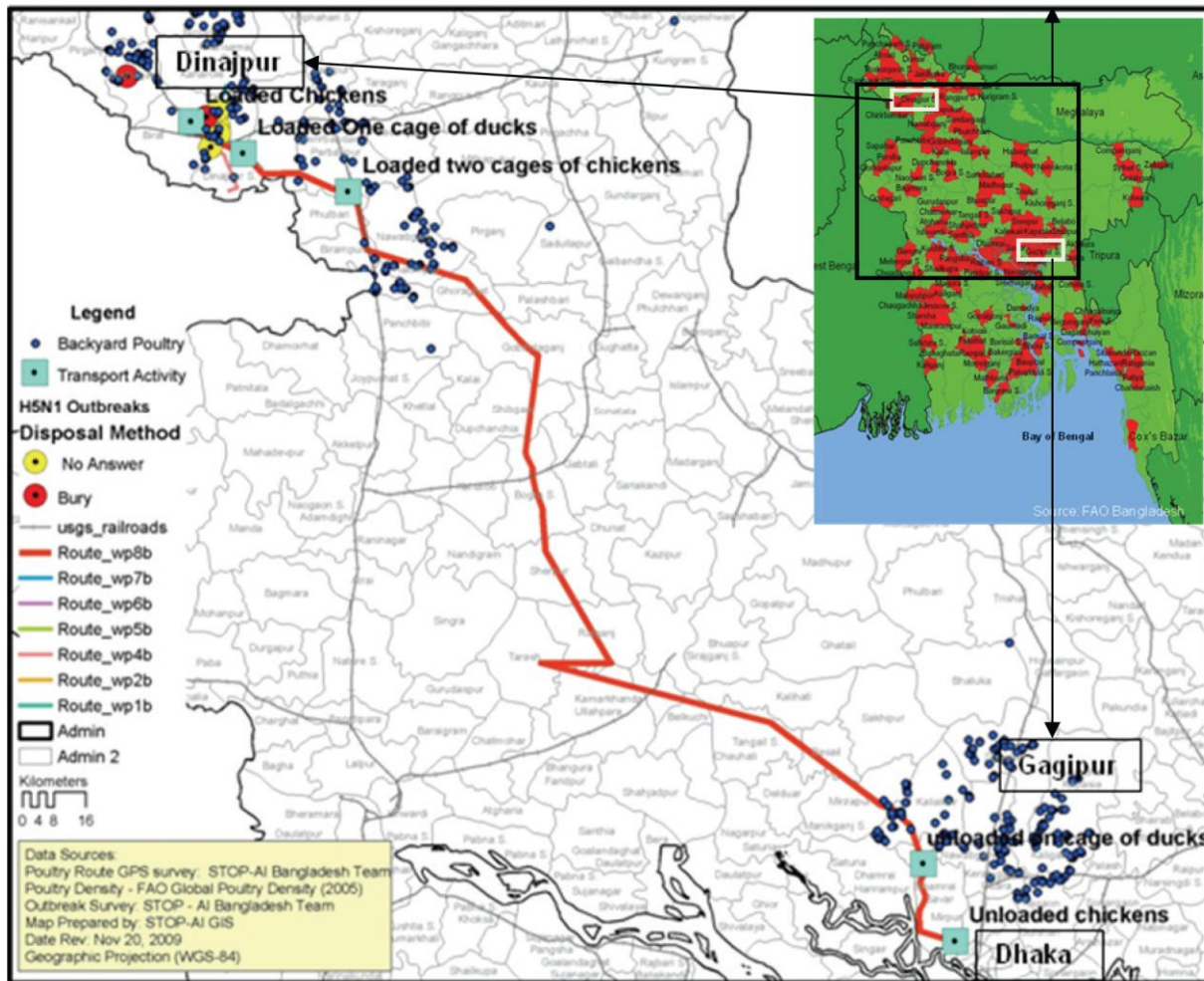
Figure 2. Map of Bangladesh showing locations of intervention districts, Gazipur and Dinajur, 2009–2010, and highly pathogenic avian influenza (HPAI) subtype H5N1 virus outbreaks (red clusters on right top), 2007–2011. Geographic positioning system coordinates were recorded by using personal navigators (eTrex Venture, Garmin, Olathe, KS, USA), and a geographic information system program (Arc View 9.1; Environmental System Research Institute, Redlands, CA, USA) was used to analyze data. Some outbreak farms (large circles) in Dinajpur and backyard poultry (small circles) in Gazipur and Dinajpur illustrate the functional relationship among poultry value chain stakeholders in two districts. Movement of poultry from Dinajpur to Dhaka (red line) by public buses and transport activities (squares on red line) could identify patterns of risk. Gazipur and Dinajpur are located in the northwestern and central parts of Bangladesh, respectively, and both are on an oblique line connecting with southeastern part of the country. The highway connecting this line is the main poultry trading route in the country (*2*). Epidemiologic studies have shown that the highest intensity and magnitude of the subtype H5N1 outbreaks was systematic and persistent in this line (*1,3*). Previous studies have confirmed the role of the highway network in HPAI (H5N1) epidemiology in other countries (*4,5*).

Figure 3. Infrastructure improvements in live bird markets (LBMs) in Dinajpur and Gazipur districts, Bangladesh, 2010. After assessing several LBMs in each district, 2 were selected in Dinajpur (A, C) and 2 in Gazipur (E, G) for our intervention. The renovated Hakimpur (B) and Parbatipur (D) LBMs in Dinajpur, and Sreepur (F) and Konabari LBMs (H) in Gazipur included tiling of poultry stalls and floors; supplying metal cages, pressure sprayer for cleaning and disinfection, improved water supply and drainage, modern slaughter facilities for hygienic processing of poultry meat; and establishing compost and biogas for waste disposal. In all 4 markets, biogas plants are being used for proper disposal of LBM waste and large animal slaughter waste, and the connected generators are being used to run lights and fans on a regular basis because frequent electric failure (load shedding) is common in Bangladesh.

References

1. Loth L, Gilbert M, Osmani MG, Kalam AM, Xiao X. Risk factors and clusters of highly pathogenic avian influenza H5N1 outbreaks in Bangladesh. Prev Vet Med. 2010;96:104–13.

2. Dolberg, F. Poultry sector country review: Bangladesh. Rome: Food and Agriculture Organization of the United Nations; 2009.

3. Ahmed SS, Ersbøll AK, Biswas PK, Christensen JP, Toft N. Spatio-temporal magnitude and direction of highly pathogenic avian influenza (H5N1) outbreaks in Bangladesh. PLoS One. 2011;6:e24324.

4. Rivas AL, Chowell G, Schwager SJ, Fasina FO, Hoogesteijn AL, Smith SD, et al. Lessons from Nigeria: the role of roads in the geo-temporal progression of avian influenza (H5N1) virus. Epidemiol Infect. 2010;138:192–8.

5. Ward MP, Maftei D, Apostu C, Suru A. Geostatistical visualisation and spatial statistics for evaluation of the dispersion of epidemic highly pathogenic avian influenza subtype H5N1. Vet Res. 2008;39:22.