

Inside

Page 9

Avian influenza A virus infection in nomadic duck flocks in northeastern Bangladesh

Page 15

HIV Counselling and Testing at icddr,b's Dhaka Hospital

Page 21

Surveillance updates

Randomized controlled trial evaluating health impact of treating and safely storing shallow tubewell drinking water in rural Bangladesh

We conducted a randomized controlled trial to assess whether improved microbial quality of tubewell drinking water by chlorination and safe storage would reduce diarrhoea among children aged <2 years in rural Bangladesh. We randomly assigned 1800 households into one of three arms: chlorine plus safe storage, safe storage only and control (use of neither safe storage nor chlorine). In the chlorine plus safe storage arm, 9% of stored water samples had *Escherichia coli* concentrations exceeding 10 colony forming units/100 mL, compared to 21% in the safe storage arm and 61% in the control arm ($p < 0.05$). Compared to controls, diarrhoea prevalence in children aged <2 years was 36% lower in the chlorine plus safe storage arm and 31% lower in the safe storage only arm ($p < 0.05$). This study suggests that safe storage of drinking water can substantially reduce illness among children in rural Bangladesh.

Recent studies in Bangladesh have demonstrated that up to 65% of tubewell water is contaminated by fecal pathogens; however, the level of contamination is low (1-5). Possible



mechanisms for tubewell contamination with fecal pathogens include infiltration into the groundwater aquifers from nearby latrines, septic tanks and ponds, short-circuiting of contaminated surface water into the wells through leaky seals of various tubewell components or harboring of bacteria in contaminated hand pumps (3,6,7). Tubewell water also becomes contaminated during collection, handling and storage in households. The most commonly used storage container in Bangladesh is the *kolshi*, a lidless vessel with a wide brim, which leaves water vulnerable to contamination by contact with hands (8). There is evidence from various settings on contamination of drinking water during household storage, and the extent of point-of-use contamination is typically more pronounced in settings where the source water quality is relatively good, such as in the case of tubewell water in Bangladesh (9,10). We conducted a randomized controlled trial to evaluate the individual and combined impacts of chlorinating and safely storing tubewell water on household water quality and diarrhoea prevalence among children aged <2 years in rural Bangladesh.

We screened 87 randomly selected villages in the Fulbaria sub-district of Mymensingh to identify households that consistently relied on shallow tubewells (tubewells <250 ft) as their primary source of drinking water, did not complain of iron taste in water from their tubewells, and had a child between the ages of six and 18 months living in the household. The lower age limit of six months was chosen because of the national Bangladeshi policy recommending that infants under six months be exclusively breastfed -- infants in households adhering to these guidelines would not consume water. The upper age limit of 18 months was chosen to ensure that the majority of the children would be aged <2 years during the follow-up period, representing the age group that is most vulnerable to waterborne illness and its long-term sequelae (11).

We selected a random subset of 1,800 households from those that met our eligibility criteria. Households in Bangladesh are typically clustered into *baris*, compounds consisting of extended families. If there were multiple eligible households in a compound, only one household was randomly selected from the *bari*. Field staff approached selected households to obtain informed written consent from primary caregivers of children and administer a baseline questionnaire that assessed the following: the pre-intervention health status of children, the water and sanitation practices, the demographics and socioeconomic status of selected households.

We generated the randomization sequence using the random allocation function of STATA software (version 10.1, STATA Corp., College Station, TX). The study area was divided into 15 distinct geographical regions. In each region, eligible households were listed in the order they were identified during screening and block randomization with a block size of three was applied to assign 1800 households to one of three study arms:

i) chlorine plus safe storage: sodium dichloroisocyanurate (NaDCC) tablets and a narrow-mouth vessel with a lid and tap (Figure 1); ii) safe storage only; and iii) control: no intervention. Field teams charged with delivering the interventions and collecting follow-up data were informed about the randomization assignment after completion of participant enrollment and baseline data collection.

Figure 1: Intervention products: NaDCC tablets (left) and safe storage container (right)



Field staff distributed the intervention products to study households following the baseline interview and demonstrated their use, including how to clean the safe storage containers with a brush and detergent that were provided. They left an illustrated instruction sheet at a visible location in the household to serve as a reminder. They instructed participants in both intervention arms to discard any remaining water after 24 hours and collect a fresh 10-liter batch of water and to exclusively give the treated and/or safely stored water to all children aged <5 years living in the households. The promotion team continued to visit households approximately once a month for one year to promote correct and consistent use of the products and replenish the supply of NaDCC tablets. In order to detect a possible Hawthorne effect in which subjects perceive or report spurious health benefits as a consequence of “being watched and unusual attention being paid” (12,13), the control group was visited with the same frequency as the intervention groups. However, field staff did not provide the control group with information on water treatment or safe storage but focused on general information on diarrhoea and oral rehydration therapy, which was

not expected to affect diarrhoea prevalence in this group.

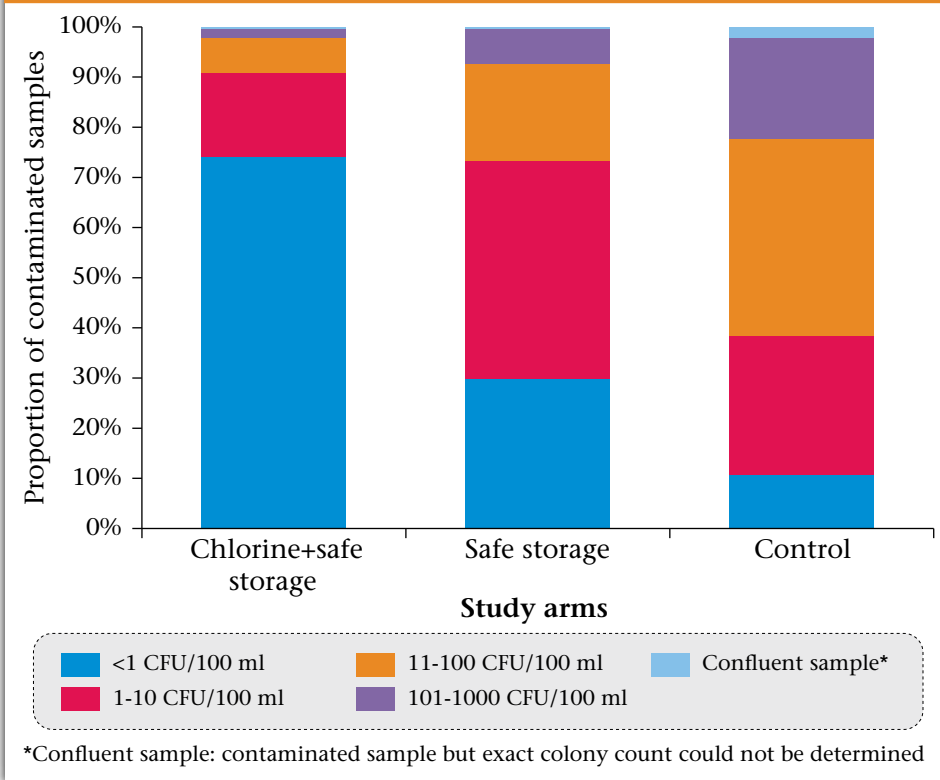
A separate data collection team conducted unannounced monthly follow-up visits between November 2011 and October 2012 to record caregiver-reported prevalence of diarrhoea (defined as three or more loose stools within a 24-hour period) in the previous seven days in children aged <2 years. We specified use of seven-day prevalence *a priori* for our analysis unless we detected evidence of differential recall bias (i.e., difference in the magnitude of effect estimates obtained using two vs. seven-day recall period) (14). In addition to diarrhoea, the field team recorded caregiver-reported prevalence of other health conditions not expected to be related to drinking water quality to detect differential reporting between intervention groups, namely, rashes and ear infections. During each of the 10 follow-up visits, the team monitored intervention uptake by recording self-reported use, conducting spot checks on the presence and status of the intervention products and collecting stored water samples in all households in the chlorine plus safe storage arm to test for free chlorine residual. In a rotating systematic subsample of 10% of households in all study arms, the field team also collected tubewell and stored water samples for microbiological testing. Samples were transported on ice to the field laboratory. Laboratory staff measured free chlorine residual with the *n,n*-diethyl-*p*-phenylenediamine colorimetric method using a digital colorimeter (Hach, Loveland, CO, USA; lower estimated detection limit: 0.02 mg/L; precision ± 0.05 mg/L). *Escherichia coli* was enumerated with membrane filtration using U.S. EPA Method 1604 within eight hours of sample collection (15). Quality control measures, including 10% blanks and 10% duplicates, were followed. *E. coli* concentrations were measured in colony forming units (CFU) per 100 mL, and samples were classified according to the World Health Organization thresholds of no risk (0 CFU/100 mL), low risk (1-10 CFU/100 mL), moderate risk (11-100 CFU/100 mL), and high risk (101-1000 CFU/100 mL) (16). We calculated prevalence ratios (PR), the ratios of the seven-day prevalence of diarrhoea between each intervention group and the control group.

The interventions achieved high uptake during the study period. The delivered storage containers were observed to contain water in 87% of observations in the chlorine plus safe storage arm and 91% of spot check observations in the safe storage arm over all follow-up visits. Of the chlorine plus safe storage households that had water in the safe storage containers at the time of the visit, 83% had free chlorine residual above the minimum US Centers for Disease Control and Prevention-recommended value of 0.2 mg/L.

Among tubewell water samples, 41% were positive for *E. coli*. In 14% of samples, *E. coli* counts were above the low-risk limit of 10 CFU/100 mL. There were no differences in tubewell water contamination between any pairs of study arms ($p > 0.05$). However, stored water quality showed marked

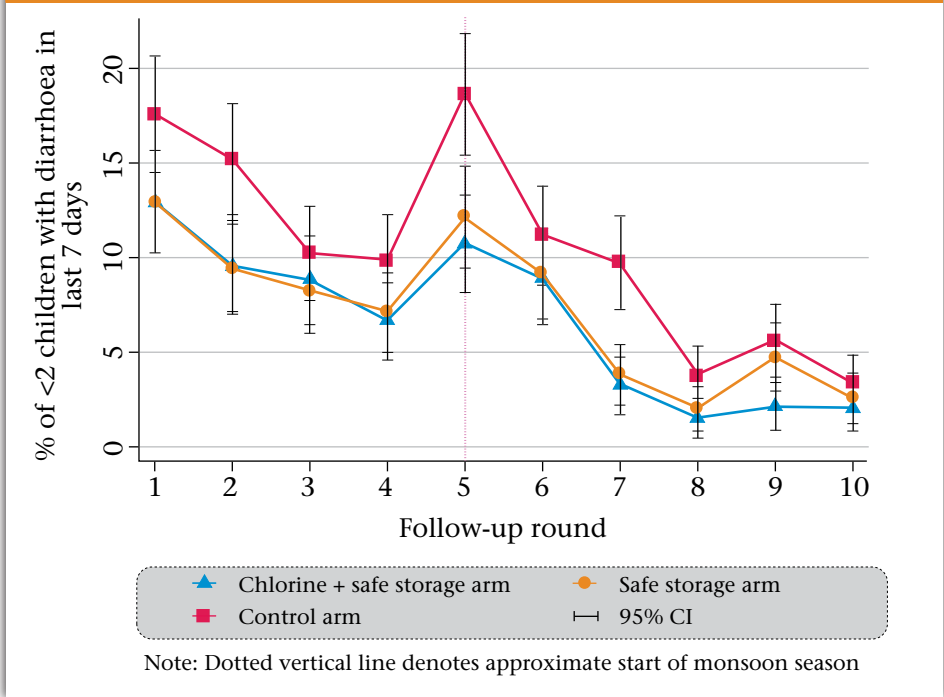
differences between the three arms. In the control arm, 89% of samples were positive for *E. coli* compared to 70% in the safe storage arm and 26% in the chlorine plus safe storage arm (Figure 2). The percentages of samples with *E. coli* >10 CFU/100 mL were 61% in the control arm, 27% in the safe storage arm and 9% in the chlorine plus safe storage arm (Figure 2).

Figure 2: Categories of *E. coli* counts in stored water across three study arms



Diarrhoea prevalence among children aged <2 years in the control arm was 11% over the study period. In all three arms, diarrhoea prevalence peaked at the onset of the monsoon season but decreased with increasing study duration (Figure 3). Compared to the control arm, caregiver-reported diarrhoea in children aged <2 years was significantly reduced in both the chlorine plus safe storage arm (PR=0.64, 95% confidence interval (CI)=0.55-0.73) and the safe storage arm (PR=0.69, 95% CI=0.60-0.80); there was no difference in the chlorine plus safe storage versus safe storage arms (PR=0.92, 95% CI=0.79-1.08). In addition, there were no significant differences in the seven-day prevalence of the negative control outcomes (i.e., rashes and ear infections) across the three study arms.

Figure 3: Temporal trends in diarrhoea prevalence in children <2 years between November 2011 and October 2012



Reported by: Water, Sanitation and Hygiene Research Group, Centre for Communicable Diseases, icddr,b

Supported by: United States Agency for International Development

Comments

Our findings indicate that safe storage, alone or combined with chlorination, was effective in reducing child diarrhoea in rural Bangladesh compared to standard practices, and, given safe storage, there was no additional health benefit from chlorination. One interpretation of these results is that if drinking water is safely stored and handled, there truly is no added benefit from adding chlorine in this particular setting, where the levels of contamination are low. Our water quality testing results support this explanation; the source water quality was relatively good in the study area and contamination of water stored in households was common, as evidenced by the high percentage of stored water samples containing *E. coli* in the control group. It is also possible that chlorination does confer some additional protection but our study had insufficient statistical power to detect a difference between the two intervention groups.

One limitation of our study is that it employed a non-blinded design

with self-reported outcomes. An alternative explanation for the similar health impacts in the two intervention groups may therefore be that the reported reductions in diarrhoea in both groups may be a result of courtesy bias (i.e., giving answers to questions that study participants believe that investigators would like to hear) due to provision of intervention products to participants. However, this is unlikely in our case given the several measures we implemented to minimize biased reporting. The interventions were distributed and promoted by different field staff than those who collected the health data to minimize courtesy bias. We also found no impact of either intervention on rashes or ear infections suggesting no evidence of courtesy bias. Finally, our stored water quality measurements present an objective intermediate outcome on the causal pathway between the interventions and child health, and provide support for a reduction in diarrhoea of similar magnitude in both intervention arms.

These findings indicate that similar to other low-income settings, unsafe handling of water during storage in households was the primary mechanism for contamination of tubewell drinking water in the study area (17,18) and that safe storage alone could substantially reduce waterborne illness among young children in rural Bangladesh.

References

1. Ferguson AS, Layton AC, Mailloux BJ, Culligan PJ, Williams DE, Smartt AE *et al.* Comparison of fecal indicators with pathogenic bacteria and rotavirus in groundwater. *Sci Total Environ* 2012;431:314-22.
2. Islam MS, Siddika A, Khan MN, Goldar MM, Sadique MA, Kabir AN *et al.* Microbiological analysis of tube-well water in a rural area of Bangladesh. *Appl Environ Microbiol* 2001;67:3328-30.
3. Knappett PS, McKay LD, Layton A, Williams DE, Alam MJ, Huq MR *et al.* Implications of fecal bacteria input from latrine-polluted ponds for wells in sandy aquifers. *Environ Sci Technol* 2012;46:1361-70.
4. Luby S, Islam MS, Johnston R. Chlorine spot treatment of flooded tube wells, an efficacy trial. *J Appl Microbiol* 2006;100:1154-8.
5. van Geen A, Ahmed KM, Akita Y, Alam MJ, Culligan PJ, Emch M *et al.* Fecal contamination of shallow tubewells in Bangladesh inversely related to arsenic. *Environ Sci Technol* 2011;45:1199-1205.
6. Ferguson AS, Mailloux BJ, Ahmed KM, van Geen A, McKay LD, Culligan PJ. Hand-pumps as reservoirs for microbial contamination of well water. *J Water Health* 2011;9:708-17.
7. Knappett PS, Escamilla V, Layton A, McKay LD, Emch M, Williams DE *et al.* Impact of population and latrines on fecal contamination of ponds in rural Bangladesh. *Sci Total Environ* 2011;409:3174-82.

8. Clasen T, Menon S. Microbiological performance of common water treatment devices for household use in India. *Int J Environ Health Res* 2007;17:83-93.
9. Trevett A, Carter R, Tyrrel S. The importance of domestic water quality management in the context of faecal-oral disease transmission. *J Water Health* 2005;3:259-70.
10. Wright J, Gundry S, Conroy R. Household drinking water in developing countries: a systematic review of microbiological contamination between source and point-of-use. *Trop Med Int Health* 2004;9:106-17.
11. Walker CLF, Perin J, Aryee MJ, Boschi-Pinto C, Black RE. Diarrhea incidence in low-and middle-income countries in 1990 and 2010: a systematic review. *BMC Public Health* 2012;12:220.
12. Adair JG. The Hawthorne effect: a reconsideration of the methodological artifact. *J Appl Psychol* 1984;69(2):334.
13. Loevinsohn BP. Health education interventions in developing countries: a methodological review of published articles. *Int J Epidemiol* 1990;19:788-94.
14. Arnold BF, Galiani S, Ram PK, Hubbard AE, Briceño B, Gertler PJ *et al.* Optimal recall period for caregiver-reported illness in risk factor and intervention studies: a multicountry study. *Am J Epidemiol* 2013;177:361-70.
15. United States Environmental Protection Agency. Method 1604: Total Coliforms and *Escherichia coli* in Water by Membrane Filtration Using a Simultaneous Detection Technique (MI Medium). Washington, DC: Office of Water, United States Environmental Protection Agency, 2002. 14 p. (<http://www.epa.gov/microbes/documents/1604sp02.pdf>; accessed on 2 October 2013).
16. World Health Organization. Guidelines for Drinking-water Quality: 4th Edition. Geneva: World Health Organization, 2011.
17. Mintz ED, Reiff FM, Tauxe RV. Safe water treatment and storage in the home. A practical new strategy to prevent waterborne disease. *JAMA*. 1995;273:948-53.
18. Roberts L, Chartier Y, Chartier O, Malenga G, Toole M, Rodka H. Keeping clean water clean in a Malawi refugee camp: a randomized intervention trial. *Bull World Health Organ* 2001;79:280-7.

Avian influenza A virus infection in nomadic duck flocks in northeastern Bangladesh

In Bangladesh avian influenza A (H5N1) has circulated in poultry since 2007. To identify the management practices for nomadic ducks and the proportion of ducks positive for avian influenza A RNA and anti-H5 antibodies, we tested duck flocks in northeastern Bangladesh from December 2011 to February 2012. Sixty-two nomadic duck flock owners were interviewed and egg yolk and faecal samples were collected. Of 1,860 egg yolk samples, 886 (48%) were antibody positive for H5 and 60 flocks (97%) had at least one egg with antibodies directed toward H5. Of the 496 faecal samples, 131 (26%) had detectable RNA for avian influenza A by rRT-PCR, but no detectable RNA for H5. Thirty-three flocks (53%) had at least one faecal sample testing positive for avian influenza A RNA. Thus, a substantial proportion of nomadic duck flocks had detectable avian influenza A RNA and had evidence of H5 exposure, suggesting a need for targeted surveillance of nomadic duck flocks.

Wild waterfowl are reservoirs of influenza A viruses and these viruses are generally nonpathogenic to ducks (1). Nomadic ducks may play a role in the maintenance and spread of avian influenza viruses to domestic bird populations in Southeast Asia (2,3). In Thailand, free-grazing ducks were suspected to be the source of H5N1 outbreaks in chickens, contributing to human infections (4). In Vietnam, duck flock management practices were identified as playing a role in transmission of H5N1 among poultry (5). Since March 2007, there have been >500 reported H5N1 outbreaks in chickens in Bangladesh (6).

Bangladesh has the third largest domestic duck population worldwide, with 38.1 million ducks (7). Domestic duck flocks are concentrated around large water bodies (8) and adjacent harvested rice fields which provide feed and serve as points of intersection between domestic ducks and wild waterfowl. Scavenging duck flocks move frequently, facilitating distant spread of viruses (9,10). Small-scale farmers use this scavenging system because no feed supplementation is needed (11). To determine evidence of exposure and shedding of avian influenza, we conducted a cross sectional survey of 62 nomadic duck flocks in Netrokona, Bangladesh.

Nomadic duck flocks were defined as groups of ducks reared for egg production that access feeding sites beyond village boundaries and housed overnight in temporary enclosures. We selected one flock with at least 100 egg-laying ducks from each of 62 owners in the Mohanganj sub-district.

After obtaining informed consent from flock owners, field workers collected 30 eggs that had been laid that morning in duck shelters and also collected eight pooled faecal swabs. Pooled faecal samples were placed in a single tube containing 5 ml of viral transport medium, were stored at 2-8°C and sent to icddr,b for testing by rRT-PCR specific for influenza A virus and sub-typing for H5. Egg yolk specimens were tested by antibody-capture enzyme-linked immunosorbent assay (cELISA) specific for anti-H5 antibodies.

Characteristics and management practices of nomadic duck flocks are shown in the Table. The median size of nomadic duck flocks was 300 and the median age of ducks was 24 months. Most duck flocks (98%) stayed within scavenging areas in temporary enclosures. The median length of time before moving to new areas was 30 days. All duck flock owners reported that flocks were moved because of scarcity of feed. The median distance for moving during a year was seven kilometers. Most owners (58%) reported moving flocks outside their villages; 16% moved them outside their sub-districts and 28% moved them outside their districts. Flock owners reported that ducks co-fed in water bodies with wild waterfowl. Duck eggs were sold in village markets in flock owners' sub-districts and the majority of owners (92%) reported selling ducks to vendors and/or retail customers. The main reason for selling flocks was decreased egg production. Forty-eight percent of flock owners disposed of dead ducks in adjacent water bodies and the majority of the owners (77%) reported washing their hands only with water (vs. with soap and water or ash) after collecting eggs.

Of the 496 pooled faecal samples collected, 131 (26%) had detectable RNA for avian influenza A but none had detectable RNA for H5. Thirty-three flocks (53%, 95% confidence interval [CI]: 40-60) had at least one faecal sample that tested positive for avian influenza A.

Of 1,860 egg yolk samples collected, 886 (48%, 95% CI: 42-53) had detectable antibodies to H5. Sixty flocks (97%, 95% CI: 89-100) had at least one egg containing anti-H5 antibodies (Figure 1).

Reported by: Department of Livestock Services, Ministry of Fisheries & Livestock, Government of Bangladesh, Centre for Communicable Diseases, icddr,b

Supported by: Centers for Disease Control and Prevention, Atlanta, USA

Comments

Exposure to avian influenza A (H5) is common in nomadic ducks in northeastern Bangladesh, and they frequently shed influenza A virus into the environment.

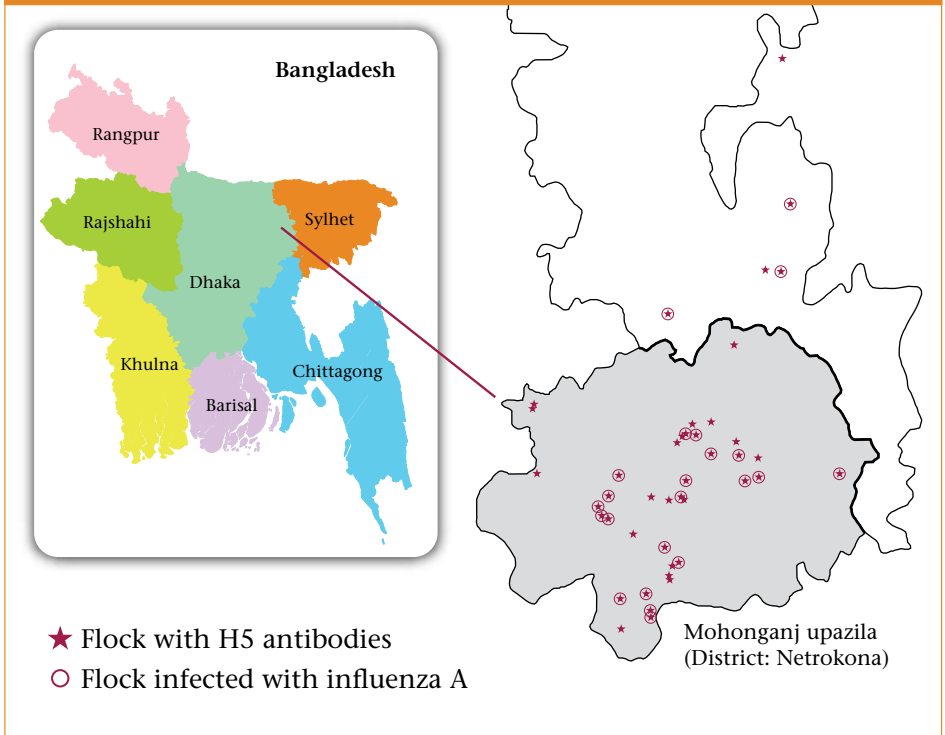
More than one-quarter of nomadic ducks in this study shed avian influenza A. A previous report from Bangladesh that analyzed individual cloacal swabs of domestic waterfowl, most of which were raised on backyard farms, found that 3.4% were positive for avian influenza A (12). Differences in findings

Table: Characteristics and management practices of nomadic duck flocks in northeastern Bangladesh, 2011

Demographic characteristics	N=62 n (%)
Age of ducks in the duck flocks in months, median (range)	24 (8-36)
Flock size, median (range)	300 (105-1100)
Nomadic duck flock management practices	
Movement practices	
Longest distance in kilometres of movement of duck flocks in the past year, median (range)	7 (1-150)
Places of movement of duck flocks in the past year	
Within own village	8 (13)
Into another village	36 (58)
Into another sub-district	10 (16)
Into another district	8 (13)
Marketing practices	
Reasons for selling duck flocks	
Scarcity of feed	5 (8)
Decreased egg production	56 (90)
Disease outbreak(s)	1 (2)
Methods of selling duck flocks	
Brought to market	5 (8)
Vendors came to herding places to purchase	57 (92)
Method of selling duck eggs	
Brought the eggs to village markets	62 (100)
Bio-security/bio-safety practices	
Kept duck flocks away from chickens	7 (11)
Used disinfectant in night shelters	10 (16)
Took measures to prevent duck flocks from mixing with wild waterfowl in common feeding grounds	5 (8)
Methods of disposal of dead ducks	
Burying	29 (47)
Throwing into a water body	31 (50)
Burning	2 (3)
Reported handwashing techniques after collecting eggs	
With water alone	48 (77)
With soap and water	13 (21)
With ash	1 (2)
Vaccinated flock for either duck plague or cholera	36 (58)

from our study and those of the previous study may be partially explained by differences in where the waterfowl were raised and how samples were collected.

Figure 1: Nomadic duck flocks infected with influenza A and with H5 positive antibodies in northeastern Bangladesh, 2011



This study also demonstrated that the proportion of nomadic duck eggs with antibodies to H5 was higher (48%) than proportions reported in studies from Indonesia (3%) and Vietnam (18%) (3,14). In the Indonesia and Vietnam studies, serum samples were tested by hemagglutination inhibition assay. Previous cELISA testing has demonstrated 100% sensitivity and 90.9% specificity using yolk samples (15). We utilized egg yolk samples as an alternative to collecting serum because of animal welfare concerns about the impact of blood collection on egg production. Differences in findings in our study from those of previous studies might be explained in part by overestimation of anti-H5 antibody production by cELISA due to cross-reactivity with other viral antigens.

Although we did not detect H5 in faecal samples of the nomadic duck flocks, previous studies in live bird markets in Bangladesh have demonstrated H5

RNA in 0.6% of ducks, 88% of which had been raised on backyard poultry farms (12). All sampled ducks that were positive for avian influenza A or H5 antibody appeared healthy when tested, indicating that shedding influenza A virus without signs of infection may play a role in the spread of avian influenza A viruses in Bangladesh. Epidemiological studies suggest that asymptomatic ducks are important in maintaining, perpetuating and transmitting these viruses to other susceptible hosts (16).

This study was subject to several limitations. First, the study only examined duck flocks in one sub-district and may not be representative of nomadic duck flocks elsewhere in the country. Second, the study was conducted during the winter and exposure to and shedding of avian influenza may be subject to seasonal variations. Finally, egg yolk testing by cELISA may have overestimated anti-H5 antibody production due to cross-reactivity with other viral antigens.

Nomadic duck flocks may act as sentinels for the identification of re-assortment of influenza viruses, some with pandemic potential. Our results indicate a need for active surveillance of nomadic duck flocks at the interface with wild waterfowl to detect the emergence of novel avian influenza viruses.

References

1. Sturm-Ramirez KM, Hulse-Post DJ, Govorkova EA, Humberd J, Seiler P, Puthavathana P *et al.* Are ducks contributing to the endemicity of highly pathogenic H5N1 influenza virus in Asia? *J Virol* 2005;79:11269-79.
2. Gilbert M, Chaitaweesub P, Parakamawongsa T, Premashthira S, Tiensin T, Kalpravidh W *et al.* Free-grazing ducks and highly pathogenic avian influenza, Thailand. *Emerg Infect Dis* 2006;12:227-34.
3. Henning J, Henning KA, Morton JM, Long NT, Ha NT, Vu le T *et al.* Highly pathogenic avian influenza (H5N1) in ducks and in-contact chickens in backyard and smallholder commercial duck farms in Viet Nam. *Prev Vet Med* 2011;101:229-40.
4. Songserm T, Jam-on R, Sae-Heng N, Meemak N, Hulse-Post DJ, Sturm-Ramirez KM *et al.* Domestic ducks and H5N1 influenza epidemic, Thailand. *Emerg Infect Dis* 2006;12:575-81.
5. Minh PQ, Morris RS, Schauer B, Stevenson M, Benschop J, Nam HV *et al.* Spatio-temporal epidemiology of highly pathogenic avian influenza outbreaks in the two deltas of Vietnam during 2003-2007. *Prev Vet Med* 2009;89:16-24.
6. Ministry of Fisheries and Livestock, Bangladesh. Daily bird flu Situation. Dhaka: Ministry of Fisheries and Livestock, Government of the People's Republic of Bangladesh, 2011. (<http://www.mofl.gov.bd/>; accessed on 1 August 2013).

7. Dolberg, F. Poultry sector country review: Bangladesh. Rome: FAO Animal Production and Health Division, Food and Agriculture Organization of the United Nations, 2008. 57 p. (<ftp://ftp.fao.org/docrep/fao/011/ai319e/ai319e00.pdf>; accessed on 19 July 2012).
8. Government of Bangladesh. 2nd National Avian and Pandemic Influenza Preparedness and Response Plan, Bangladesh 2009-2011. Dhaka: Ministry of Health and Family Welfare. 2009. 118 p. (http://www.who.int/influenza/preparedness/plans/influenza_preparedness_plan_bangladesh/en/index.html; accessed on 19 July 2012).
9. Gilbert M, Xiao X, Chaitaweesub P, Kalpravidh W, Premasathira S, Boles S *et al.* Avian influenza, domestic ducks and rice agriculture in Thailand. *Agric Ecosyst Environ* 2007;119:409-15.
10. Minh PQ, Stevenson MA, Schauer B, Morris RS, Quy TD. A description of the management of itinerant grazing ducks in the Mekong River Delta of Vietnam. *Prev Vet Med* 2010;94:101-7.
11. Rahman MM, Khan MJ, Chowdhury SD, Akbar MA. Duck rearing system in southern coastal districts of Bangladesh. *Bang J Anim Sci* 2009;38:132-41.
12. Khan MSU, Gurley ES, Rahman M, Hossain MJ, Nasreen S, Azim T *et al.* Live bird market surveillance for avian influenza in Bangladesh, 2007-2009. (Abstract) 2010 International Conference on Emerging Infectious Diseases, Atlanta, July 11-14, 2010.
13. Runstadler JA, Happ GM, Slemmons RD, Sheng ZM, Gundlach N, Petrula M *et al.* Using RRT-PCR analysis and virus isolation to determine the prevalence of avian influenza virus infections in ducks at Minto Flats State Game Refuge, Alaska, during August 2005. *Arch Virol* 2007;152:1901-10.
14. Henning J, Wibawa H, Morton J, Usman BT, Junaidi A, Meers J. Scavenging ducks and transmission of highly pathogenic avian influenza, Java, Indonesia. *Emerg Infect Dis* 2010;16:1244-50.
15. Jeong OM, Kim MC, Kang HM, Ha GW, Oh JS, Yoo JE *et al.* Validation of egg yolk antibody based C-ELISA for avian influenza surveillance in breeder duck. *Vet Microbiol* 2010;144:287-92.
16. Hulse-Post DJ, Sturm-Ramirez KM, Humberd J, Seiler P, Govorkova EA, Krauss S *et al.* Role of domestic ducks in the propagation and biological evolution of highly pathogenic H5N1 influenza viruses in Asia. *Proc Natl Acad Sci U S A* 2005;102:10682-7.
17. Nazir J, Haumacher R, Ike AC, Marschang RE. Persistence of avian influenza viruses in lake sediment, duck feces, and duck meat. *Appl Environ Microbiol* 2011;77:4981-5.
18. Keawcharoen J, van Riel D, van Amerongen G, Bestebroer T, Beyer WE, van Lavieren R *et al.* Wild ducks as long-distance vectors of highly

pathogenic avian influenza virus (H5N1). *Emerg Infect Dis* 2008;14:600.

19. Terregino C, De Nardi R, Guberti V, Scremin M, Raffini E, Martin AM *et al.* Active surveillance for avian influenza viruses in wild birds and backyard flocks in Northern Italy during 2004 to 2006. *Avian Pathol* 2007;36:337-44.

HIV counselling and testing at icddr,b's Dhaka Hospital

HIV prevalence in Bangladesh is low and diagnosis of HIV-infected individuals is often difficult. HIV counselling and testing is the entry point for prevention and treatment. Voluntary Counselling and Testing (VCT) was introduced at icddr,b's Dhaka Hospital in 2002 and in 2008, the hospital established Provider-initiated HIV Testing and Counselling (PITC) services. The objective of this study was to describe the characteristics of HIV-infected patients diagnosed at Dhaka Hospital using different counselling and testing approaches. We collected demographic and clinical data from persons who were tested by Dhaka Hospital's VCT and PITC services during May 2008 to May 2012. During this time, 4,236 persons were tested for HIV, 85% by VCT and 15% by PITC. A total of 7% of those tested were found to be HIV-positive. Although a greater number of cases were identified by VCT than by PITC (151 vs. 126), a substantially greater proportion of those tested by PITC were found to be HIV-infected (19% vs. 4%). Both VCT and PITC were important for HIV diagnosis at Dhaka Hospital and both should be expanded at Dhaka Hospital and in other hospitals in Bangladesh.

In 2012, an estimated 35.3 million people were living with HIV globally and among them, 2.3 million were newly infected (1). By the end of 2013, Bangladesh's Ministry of Health and Family Welfare (MOHFW) had confirmed HIV in 3,241 persons, of whom 1,299 (40%) had developed the acquired immunodeficiency syndrome (AIDS) and 472 (15%) had died (2). UNAIDS and the World Health Organization estimated that in 2012, 8,000 (range: 3,100-82,000) people in Bangladesh were living with HIV (1).

The discrepancy between case detection and estimated prevalence may be explained by limited HIV testing in Bangladesh due to low capacity for HIV testing and the unwillingness of many people to be tested because of the stigma of HIV(2) as well as a lack of awareness of risks for infection.

HIV counselling and testing is the critical entry point for engagement into care and prevention (4). Until recently, the primary model for providing HIV counselling and testing has been client-initiated, Voluntary Counselling and Testing (VCT) whereby individuals seek an HIV test at health care or community-based facilities (5).

Globally, and particularly in low-resource settings, many HIV-infected persons are unaware of their HIV status, despite the availability of VCT (4,6). There is evidence that many opportunities to diagnose HIV and provide HIV prevention counselling at health care facilities are missed. Provider-initiated HIV Testing and Counselling (PITC) (7) refers to HIV testing and counselling recommended by health care providers to persons at health care facilities. PITC is typically performed on the basis of clinical manifestations and/or perceived risks for HIV infection with the objective of determining the HIV status of patients in order to guide patient management. Currently, most HIV testing in Bangladesh is conducted through VCT services and PITC is performed infrequently.

The Jagori Outpatient Department, the outpatient HIV unit at icddr,b's Dhaka Hospital, introduced VCT in 2002. From January 2002 to May 2013, approximately 12,000 people received VCT services there. In 2008, Dhaka Hospital established the Jagori Ward, an inpatient HIV unit. The Jagori Ward provides free clinical care to HIV-infected patients and it provides PITC services to patients referred by providers from Dhaka Hospital and from other health care facilities. The objective of this study was to describe the characteristics of HIV-infected patients diagnosed at Dhaka Hospital using different counselling and testing approaches.

In early 2008, Dhaka Hospital providers were given an orientation and sensitization training on HIV that included training about PITC. They were encouraged to refer patients for PITC if they had clinical manifestations suggestive of HIV or risk factors for HIV. All persons presenting to the Jagori Outpatient Department for VCT or referred for PITC at Dhaka Hospital during May 2008 to May 2013 were included in this retrospective observational study. Written informed consent was obtained from all persons tested by VCT or PITC, which included consent for the use of data collected for research purposes. Counselling and testing staff and clinical fellows entered demographic, clinical and laboratory data into an inpatient electronic patient record system at Dhaka Hospital. Medical record data relevant to this study, including age, sex, residence, marital status, history of migrant work, self-reported high-risk behaviour (including having multiple sex partners,

sex with a commercial sex worker, or male-male sex), self reported probable mode of HIV transmission, provider referral, and clinical information (clinical information was obtained only from persons who had been both referred for PITC by Dhaka Hospital providers and also admitted to the Jagori Ward) were entered into a database that did not contain personally identifiable information. Statistical analyses were performed using SPSS 17.0 (SPSS Inc., Chicago, IL, USA). This study was approved by icddr,b's Research Review Committee and Ethical Review Committee.

A total of 4,236 persons were tested for HIV at Dhaka Hospital during the study period. Of these, 3,582 (85%) were tested by VCT and 654 (15%) were tested by PITC. Among those tested by PITC, 114 (17.4%) were referred by providers at Dhaka Hospital and 540 (82.6%) were referred by providers from other hospitals and clinics. A total of 277 (6.5%) persons tested positive for HIV (Table 1); 195 (70.4%) were male and the average age was 32.8 (standard deviation: ± 10.3) years. Among HIV-infected persons, 151 (54.5%) were identified through VCT and 126 (45.5%) were identified through PITC; 4.2% of those tested by VCT were HIV-positive and 19.3% of those tested by PITC were HIV-positive. Among those found to be HIV-infected, 59 (39%) of those tested by VCT and 97 (76%) of those tested by PITC had a history of migrant work.

Persons referred for PITC by Dhaka Hospital providers

Among patients who were referred for PITC by Dhaka Hospital providers, 80 (70%) were patients in the hospital's Long Stay Unit, 27 (23.7%) were patients in the hospital's Intensive Care Unit, and 7 (6.3%) were patients in the hospital's Short Stay Unit. Table 2 shows characteristics of patients, by HIV status, who were referred for PITC by Dhaka Hospital providers. Overall, 14 (12.3%) persons were found to be HIV-positive, and all of them were transferred to the Jagori Ward for care and treatment for HIV following diagnosis. The most common mode of HIV transmission (64%) was high risk sexual behaviour. Among the HIV-infected patients, reasons for referral for PITC included diarrhoea (86%), pulmonary tuberculosis (82%), and fever, oral thrush and/or weight loss (55%). The median CD4 count was 103 (range 3-240) cells/mm³; 11 (78.6%) had a CD4 count <200 cells/mm³ and 5 (35.7%) had a CD4 count <50 cells/mm³. Three patients died within one week of their HIV diagnosis and the rest of the patients were discharged from the Jagori Ward.

Reported by: Jagori Unit, Dhaka Hospital, icddr,b

Supported by: icddr,b

Comments

In low-income countries that have low HIV prevalence, the identification and diagnosis of HIV-infected individuals is often difficult. In this study of two different HIV counselling and testing methods at a hospital in Dhaka,

Bangladesh, we found that while more HIV cases were identified by VCT than PITC, the proportion who tested positive was substantially higher among those tested by PITC than by VCT.

Table 1: Characteristics of persons testing positive for HIV at Dhaka Hospital by type of HIV counselling and testing method, May 2008-May 2013

Characteristic	Counselling and testing method	
	Voluntary counselling and testing (N=151) n (%)	Provider-initiated testing and counselling (N=126) n (%)
Sex		
Male	103 (68)	91 (72)
Female	47 (31)	35 (28)
Transgender	1 (0.7)	0 (0)
Age		
<18 years	6 (4)	6 (5)
≥18 years	145 (96)	120 (95)
History of migrant work		
Yes	59 (39)	97 (76)
Probable Mode of HIV transmission (by self report)		
High-risk sexual behaviour*	107 (71)	91 (72)
Received blood products	4 (3)	5 (4)
Perinatal transmission	4 (3)	5 (4)
Injecting drug use	7 (5)	0 (0)
Multiple risk behaviours	23 (15)	9 (7)
No risk behaviours disclosed	6 (4)	16 (13)

*Includes having multiple sex partners, sex with a commercial sex worker, or male-male sex.

PITC has proven to be an important intervention that increases HIV testing, particularly in settings where it complements existing VCT services (8). In this study, we demonstrated that both VCT and PITC contributed towards the diagnosis of HIV infection at an urban hospital in a country with low HIV prevalence. Persons presenting for VCT make a deliberate decision to undergo testing, but may or may not be at risk for HIV. On the other hand, patients are typically referred for PITC because they have clinical manifestations suggestive of HIV infection and so it would be expected that many of those identified with HIV are diagnosed late in the course of their infection. We

found that almost 80% of the patients who were referred for PITC by Dhaka Hospital providers and who both tested positive and were admitted to the Jagori Ward had CD4 counts <200 cells/mm³, which is consistent with findings from other studies (9,10). While risk behaviors of persons found to be HIV-positive by VCT were fairly comparable to those identified with HIV infection by PITC, the number and proportion of persons with a history of migrant work among those diagnosed with HIV were substantially higher for those tested by PITC than for those tested by VCT. One possible explanation for this finding is that many HIV-infected persons with a history of migrant work may not belong to populations targeted for HIV interventions (e.g., men who have sex with men, sex workers and clients, people who inject drugs) and so may not seek VCT services and therefore be diagnosed only after developing signs or symptoms of HIV-associated infections.

Table 2: Characteristics of patients referred for Provider-initiated Testing and Counselling by Dhaka Hospital providers, May 2008-May 2013

Characteristic	HIV-positive (N=14) n (%)	HIV-negative (N=100) n (%)
Sex		
Male	6 (43)	58 (58)
Age		
<18 years	3 (21)	10 (10)
≥18 years	11 (79)	90 (90)
Residence		
Urban	11 (79)	75 (75)
Marital status		
Single	1 (7)	17 (17)
Married	8 (57)	63 (63)
Divorced	0 (0)	10 (10)
Widowed	2 (14)	0 (0)
Not applicable (children)	3 (21)	10 (10)

This study has several limitations. First, this study was only conducted at one hospital and the findings may not be representative of all persons tested for HIV or of all persons testing positive for HIV by VCT or PITC in Bangladesh. Second, there was a lack of detailed risk information for all participants which made it difficult to determine how the risk profiles of persons who were tested and those who tested positive differed by counselling and testing approach. In addition, the lack of detailed clinical information on persons

testing positive, particularly those seeking VCT and those referred for PITC by providers from other health care facilities, prevented adequate assessment of differences in stage of infection at diagnosis by counselling and testing approach. Lastly, the lack of clinical information about patients referred for PITC by providers outside Dhaka Hospital resulted in the availability of only a small number of PITC patients in whom to compare factors by HIV status.

Our findings underscore the importance of offering both VCT and PITC services at Dhaka Hospital. To identify cases earlier in the course of HIV infection, it will be very important to maintain and extend VCT. However, PITC is extremely important because this approach can result in a high diagnostic yield and identify HIV-infected persons who do not present for VCT. PITC should be extended in health care settings where it is currently implemented and initiated more widely in health care settings throughout Bangladesh, particularly in areas where HIV prevalence is high. The Government of Bangladesh should consider incorporating PITC into the National HIV Testing and Counselling Guidelines. All health care providers should receive training on the clinical manifestations of and risk behaviours associated with HIV infection, the importance of HIV counselling and testing, and the strengths and limitations of VCT and of PITC in the diagnosis of HIV infection.

References

1. UNAIDS. Global report: UNAIDS report on the global AIDS epidemic 2013. Geneva: United Nations Programme on HIV/AIDS, 2013. (http://www.unaids.org/en/media/unaids/contentassets/documents/epidemiology/2013/gr2013/UNAIDS_Global_Report_2013_en.pdf; accessed on 15 December 2013).
2. Dhaka Tribune. World AIDS day observed: 370 new HIV cases detected in the country. Dhaka Tribune 2013 Dec 2. (<http://www.dhakatribune.com/health/2013/dec/02/world-aids-day-observed>; accessed on 15 December 2013).
3. Azim T, Khan SI, Nahar Q, Reza M, Alam N, Saifi R *et al*. 20 Years of HIV in Bangladesh: experiences and way forward. Washington DC: World Bank; 2009.
4. Bunnell R, Opio A, Musinguzi J, Kirungi W, Ekwaru P, Mishra V *et al*. HIV transmission risk behavior among HIV-infected adults in Uganda: results of a nationally representative survey. *AIDS* 2008;22:617-24.
5. Kiene SM, Bateganya M, Wanyenze R, Lule H, Mayer K, Stein M. Provider-initiated HIV testing in health care settings: should it include client-centered counselling? *SAHARA J* 2009;6:115-9.
6. Cherutich P, Kaiser R, Galbraith J, Williamson J, Shiraishi RW, Ngare C *et*

- al.* Lack of knowledge of HIV status a major barrier to HIV prevention, care and treatment efforts in Kenya: results from a nationally representative study. *PLoS One* 2012;7:e36797.
7. World Health Organization. Guidance on provider-initiated HIV testing and counselling in health facilities. Geneva: World Health Organization 2007.
 8. Kennedy CE, Fonner VA, Sweat MD, Okero FA, Baggaley R, O'Reilly KR. Provider-initiated HIV testing and counseling in low- and middle-income countries: a systematic review. *AIDS Behav* 2013;17:1571-90.
 9. Bassett IV, Giddy J, Nkera J, Wang B, Losina E, Lu Z *et al.* Routine voluntary HIV testing in Durban, South Africa: the experience from an outpatient department. *J Acquir Immune Defic Syndr* 2007;46:181-6.
 10. Silvestri DM, Modjarrad K, Blevins ML, Halale E, Vermund SH, McKinzie JP. A comparison of HIV detection rates using routine opt-out provider-initiated HIV testing and counseling versus a standard of care approach in a rural African setting. *J Acquir Immune Defic Syndr* 2011;56:e9-32.

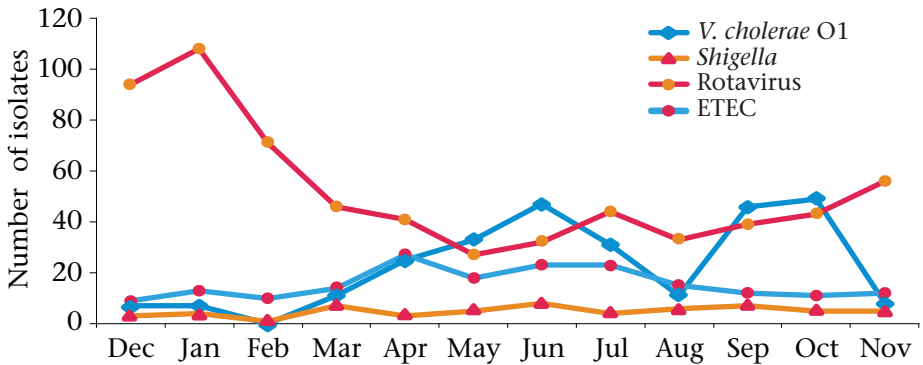
Surveillance updates

With each issue of HSB, updates of surveillance data described in earlier issues are provided. These updated tables and figures represent the most recent observation period available at the time of publication. We hope these updates will be helpful to health professionals who are interested in current patterns of disease and drug resistance in Bangladesh.

**Proportion of diarrhoeal pathogens susceptible to antimicrobial drugs:
December 2012-November 2013**

Antimicrobial agents	<i>Shigella</i> N=58	<i>V. cholerae</i> O1 N=275
Mecillinam	83.0	Not tested
Ampicillin	58.5	Not tested
TMP-SMX	26.4	0.7
Ciprofloxacin	49.0	100.0
Tetracycline	Not tested	1.5
Azithromycin	83.0	100.0
Ceftriaxone	100.0	Not tested

**Monthly isolation of *V. cholerae* O1, *Shigella*, *Rotavirus* and *ETEC*:
December 2012-November 2013**



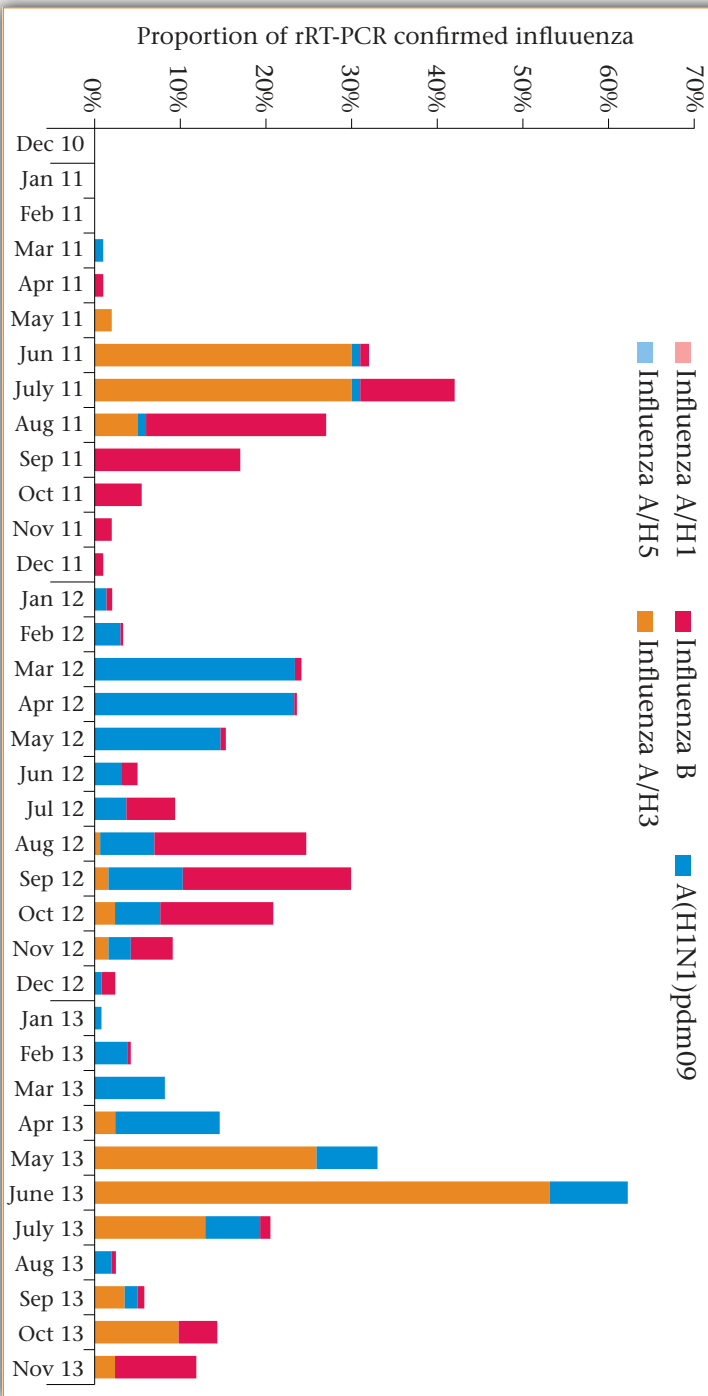
Source: Hospital Surveillance, Dhaka Hospital, icddr,b

Antimicrobial susceptibility pattern of *S. typhi* among children <5 years during October-December 2013

Antimicrobial agent	Total tested (N)	Susceptible n (%)	Reduced susceptibility n (%)	Resistant n (%)
Ampicillin	18	13 (72.0)	0 (0.0)	5 (28.0)
Cotrimoxazole	18	14 (78.0)	0 (0.0)	4 (22.0)
Chloramphenicol	18	13 (72.0)	0 (0.0)	5 (28.0)
Ceftriaxone	18	18 (100.0)	0 (0.0)	0 (0.0)
Ciprofloxacin	18	0 (0.0)	18 (100.0)	0 (0.0)
Nalidixic Acid	18	0 (0.0)	0 (0.0)	18 (100.0)

Source: icddr,b's urban surveillance in Kamalapur (Dhaka)

Proportion of laboratory-confirmed influenza among hospitalized severe acute respiratory illness (SARI) and outpatient influenza like illness (ILI) cases between December 2010 and November 2013



Source: Patients participating in hospital-based influenza surveillance in Dhaka National Medical College Hospital, Community-based Medical College Hospital (Myrmentsingh), Jahurul Islam Medical College Hospital (Kishoregoni), Rajshahi Medical College Hospital, Shaheed Ziaur Rahman Medical College Hospital (Bogra), LAMB Hospital (Dinajpur), Bangabandhu Memorial Hospital (Chittagong), Comilla Medical College Hospital, Khulna Medical College Hospital, Jessore General Hospital, Jalalabad Ragib-Rabeya Medical College Hospital (Sylhet) and Sher-e-Bangla Medical College Hospital (Barisal)



Nomadic duck flock in Mohanganj sub-district, Netrokona, Bangladesh

This publication of HSB is funded by icddr,b and its donors who provide unrestricted support for its operations and research. Currently donors providing unrestricted support include: Government of the People's Republic of Bangladesh, Australian Agency for International Development (AusAID), Canadian International Development Agency (CIDA), Swedish International Development Cooperation Agency (Sida) and Department for International Development (UK aid). We gratefully acknowledge these donors for their support and commitment to icddr,b's research efforts.

icddr,b

GPO Box 128
Dhaka 1000, Bangladesh

www.icddr.org/hsb

Editors

James D. Heffelfinger
M Sirajul Islam Molla
Emily S. Gurley
Diana DiazGranados

Guest editors

Audrie Lin

Contributing authors

1st article:

Abu Mohd. Naser Titu

2nd article:

Shamim Sarkar

3rd article:

Mohammed Moshtaq Pervez

Translation and editing

M Sirajul Islam Molla

Mahbub-ul-Alam

Design and pre-press

Mahbub-ul-Alam

Printed by

Dina Offset Printing Press