HHS Public Access
Author manuscript
Ecohealth. Author manuscript; available in PMC 2017 December 01.

Published in final edited form as:
Ecohealth. 2016 December ; 13(4): 720–728. doi:10.1007/s10393-016-1166-0.

Investigating Rare Risk Factors for Nipah Virus in Bangladesh: 2001–2012

Sonia T. Hegde[1,2], **Hossain M. S. Sazzad**[3], **M. Jahangir Hossain**[3,4], **Mahbub-Ul Alam**[3], **Eben Kenah**[5], **Peter Daszak**[6], **Pierre Rollin**[1], **Mahmudur Rahman**[7], **Stephen P. Luby**[1,8], and **Emily S. Gurley**[3]

[1]Centers for Disease Control and Prevention, Atlanta, Georgia

[3]icddr,b (formerly, International Center for Diarrheal Disease Research, Bangladesh), Centre for Communicable Diseases icddr,b, 68, Shaheed Tajuddin Ahmed Sharani, Mohakhali, Dhaka 1212, Bangladesh

[4]Medical Research Council Unit, Banjul, The Gambia

[5]University of Florida, Florida

[6]EcoHealth Alliance, New York

[7]Institute of Epidemiology, Disease Control and Research, Dhaka, Bangladesh

Abstract

Human Nipah encephalitis outbreaks have been identified almost yearly in Bangladesh since 2001. Though raw date palm sap consumption and person-to-person contact are recognized as major transmission pathways, alternative pathways of transmission are plausible and may not have been identified due to limited statistical power in each outbreak. We conducted a risk factor analysis using all 157 cases and 632 controls surveyed in previous investigations during 2004–2012 to identify exposures independently associated with Nipah, since date palm sap was first asked about as an exposure in 2004. To further explore possible rare exposures, we also conducted in-depth interviews with all cases, or proxies, since 2001 that reported no exposure to date palm sap or contact with another case. Cases were 4.9 (95% 3.2–7.7) times more likely to consume raw date palm sap and 7.3 (95% 4.0–13.4) times more likely to have contact with a Nipah case than controls. In-depth interviews revealed that 39/182 (21%) of Nipah cases reporting neither date palm sap consumption nor contact with another case were misclassified. Prevention efforts should be focused on interventions to interrupt transmission through date palm sap consumption and person-to-person contact. Furthermore, pooling outbreak investigation data is a good method for assessing rare exposures.

Correspondence to: Emily S. Gurley, egurley@icddrb.org.
[2]Present Address: University of Michigan, Ann Arbor, Michigan
[8]Present Address: Stanford University, Palo Alto, CA

Electronic supplementary material: The online version of this article (doi:10.1007/s10393-016-1166-0) contains supplementary material, which is available to authorized users.

DISCLAIMER
The findings and conclusions in this report are those of the authors and do not necessarily represent the official position of the CDC Bangladesh or the U.S. Centers for Disease Control and Prevention.

Keywords

Nipah virus; Bangladesh; risk factors; zoonoses; rare exposures; prevention

INTRODUCTION

Nipah virus is a paramyxovirus, first discovered during an outbreak involving pigs and people in Malaysia and Singapore in 1998–1999 (Chua et al. 2000). Pig consumption of dropped, bat-bitten fruit was the most likely transmission pathway from bats to pigs, and contact with ill pigs was the source of infection for most human cases (Chua et al. 2000; Chua 2010a; Chua 2010b; Luby and Gurley 2012). The pigs in the outbreak were culled to prevent further transmission and no human cases have been reported from Malaysia or Singapore since (Chua 2010b; Shapshak et al. 2015).

In Bangladesh, however, recurrent outbreaks of Nipah virus have been identified almost yearly since 2001 (ICDDRB 2003; ICDDRB 2005; Luby et al. 2009b; Luby and Gurley 2012; Sazzad et al. 2013; Luby 2013). Case fatality among patients in Bangladesh has been >70% (Luby et al. 2009b; Luby and Gurley 2012) with one-third of survivors suffering permanent neurological deficits (Sejvar et al. 2007). The most common risk factors for human disease in Bangladesh have been consumption of raw date palm sap, a national delicacy (Luby et al. 2006; Montgomery et al. 2008; Luby et al. 2009b; Rahman et al. 2012), and contact with another human Nipah case, primarily through caregiving (Hsu et al. 2004; Gurley et al. 2007b; Gurley et al. 2007a; Luby et al. 2009a; Homaira et al. 2010). Some cases have neither reported exposure to date palm sap nor contact with another Nipah case during epidemiologic investigations, however, suggesting other possible unidentified pathways of transmission (Luby et al. 2006; Luby et al. 2009b; Luby et al. 2009a).

Identifying alternative routes of Nipah transmission would provide public health officials additional opportunities for prevention and further our understanding of drivers of cross-species transmission of this highly fatal emerging pathogen. During each individual outbreak the number of cases is small, typically less than 20, which limits the statistical power to identify associations with less common exposures (Luby et al. 2009b). Therefore, this study aimed to identify rare risk factors for Nipah virus disease by combining all outbreak data from 2004 to 2012. We also aimed to identify other possible exposures to *Pteropus* fruit bats, the reservoir hosts for Nipah virus, or sick animals that were not adequately addressed in epidemiologic investigations but may have contributed to transmission (Fig. 1) (Halpin et al. 2011).

METHODS

Exposure Data

Two kinds of Nipah cases were included in our analysis: laboratory-confirmed and probable cases. Laboratory-confirmed cases tested IgM positive against Nipah virus in serum via an enzyme-linked immunosorbent assay (ELISA) (Kashiwazaki et al. 2004; Gurley et al. 2007a). Probable cases presented with febrile illness and altered mental status and had an epidemiologic link to a laboratory-confirmed case, but died before a blood specimen could be collected or before antibodies may have developed (Ramasundram et al. 2000). Trained interviewers used structured questionnaires to collect basic demographics, household characteristics, travel histories, exposures related to animal, fruit, and contact with a Nipah case. Questionnaires were administered to cases who survived without neurological impairment or, for most cases, two to three proxy respondents. Proxy respondents were selected from within the case household or from others who had daily interactions with the case. Three, sometimes four, geographically matched controls per case were also interviewed. Consumption of raw date palm sap was added to the questionnaire in 2004 and fermented date palm sap, locally known as *tari*, was added in 2012 (Islam 2011). Prior to 2004, questionnaires varied and controls were matched for age, sex, and geography. As a result, we restricted our analysis to data collected since 2004.

Case–Control Analysis

From 2004 to 2012, epidemiologists identified 157 human Nipah cases, including 80 confirmed and 77 probable cases, and enrolled 632 controls. We combined these data and calculated summary statistics of frequencies and means for all possible exposures. We recoded “don’t know” responses as “no” to not reduce the sample size and delete observations with any “don’t know” entry; the proportion of such responses did not exceed 10% for any exposure. This recoding would result in an underestimate in the effect of any one exposure. We used conditional logistic regression to build univariable models to investigate associations between each exposure and being a Nipah case, after adjusting for age and sex. We adjusted *P* values for multiple comparisons using the Bonferroni correction method (Holm 1979). We capped adjusted *P* values at 1, so that if an adjusted *P* value was greater than 1, we only reported 1. We included contact with another Nipah case and history of consuming raw date palm sap in all possible models as we were interested in knowing if other exposures were independently associated with Nipah infection. A multivariable model of best fit was then built using the Akaike Information Criterion and biological plausibility, while still adjusting for age and sex. We calculated odds ratios (ORs) and 95% confidence intervals (CIs), qualifying an association as significant based on a P value < 0.05. Since our probable case definition did not include laboratory confirmation, we conducted a sensitivity analysis using only lab-confirmed Nipah cases.

For significant factors in the model of best fit, we also calculated the attributable risk using the multivariable odds ratio from the model and the percentage of exposed among cases, using the Bruzzi et al. method (Bruzzi et al. 1985; Rockhill et al. 1998). Though calculating the attributable risk from an odds ratio is problematic because odds ratios are not collapsible (Greenland et al. 1999b), because the disease of interest is rare, the odds ratio is a good approximation of the relative risk of Nipah virus. Though the risk of Nipah virus is geographically variable and stronger in the areas where cases and controls are selected, we assume the disease of interest is rare as few Nipah cases occur in the country of Bangladesh (Cummings 2009).

In-Depth Interviews

To further explore possible rare exposures to Nipah, in 2013 we conducted in-depth interviews with 34 Nipah cases identified since 2004 who themselves or their proxies reported neither exposure to date palm sap nor a Nipah case on the case–control study structured questionnaire. Date palm sap consumption was not investigated as an exposure during 2001–2003, so for the 11 cases during that time period who did not report person-to-person exposure, we sought to retrospectively investigate both date palm sap and other possible bat exposures. Of the total 45 cases investigated, trained anthropologists interviewed 11 cases directly and at least three proxy respondents per case for the 34 cases who did not survive. The interviewers collected detailed field notes on general practices of date palm sap and *tari* consumption, including whether cases engaged in these exposures prior to illness, as these exposures were not investigated in the first questionnaire. Dropped fruit consumption, typical animal contact, contact with a Nipah case, occupation, and travel were additionally asked about, both as general practices and exposures prior to illness. We reviewed the field notes and calculated the proportion of cases with plausible exposures to bats, bat secretions or excretions based on interview responses.

Ethics

The Bangladesh Ministry of Health and Family Welfare authorized and actively participated in all reviewed and approved all protocols. Participation in these studies was strictly voluntary. Informed consent was obtained from all individual participants included in the study. For those <18 years of age, individual and parental consent were obtained. icddr,b's Institutional Review Board reviewed and approved the protocol for the in-depth interviews.

RESULTS

Case–Control Analysis

From 2004 to 2012, the median age of Nipah cases was 25 years with a range from 6 months to 75 years; 39% of cases were female. The primary occupation was student or homemaker; the mortality rate was 77%. In the univariable case–control analysis, Nipah cases were significantly more likely than controls to live in a tin-shed house than in a thatched house, to visit an area outside their district or a hospital in the one month prior to becoming ill, and to see bats in trees around the house at night (Table 1). Contact with domestic animals, tree climbing, or other hypothesized risk factors were not significantly associated with being a Nipah case. However, cases were significantly more likely to consume raw date palm sap and have contact with another Nipah case. Controls were significantly more likely to eat washed fruit, either picked from the ground or not, before consuming it.

In the multivariable model that best explained the occurrence of Nipah infection, after controlling for age, sex, person-to-person contact, and date palm sap consumption, those infected with Nipah were significantly more likely to report seeing bats in trees around the house at night (OR 3.3, 95% CI 1.2–8.8) and to have visited an area outside one's own sub-district (OR 3.5, 95% CI 1.5–8.2) in the month prior to becoming ill than healthy controls (Table 2). The population attributable fraction for raw date palm sap consumption was 50%; that is, half of the Nipah risk in the population could be eliminated if exposure to raw date palm sap were prevented. Similarly, 30% of Nipah cases could be prevented if exposure to another Nipah case were prevented. The population attributable fraction of Nipah infection was 40% for reporting bats feeding in trees around the house at night and 22% for traveling to another sub-district.

In-Depth Interviews

During the in-depth interviews, 29 (85%) of the 34 cases or their proxies since 2004 with no reported exposure to date palm sap or a Nipah case on the structured questionnaire reported being exposed to date palm sap or a Nipah case. Of the 34 cases, 12 cases or proxies reported having consumed raw date palm sap while four consumed *tari*, which was not asked about in questionnaires as an exposure until 2012. No cases or proxies reported consuming both *tari* and raw data palm sap. Thirteen cases reported exposure to a Nipah case within 5–11 days before their own illness onset. One example of a misclassified case exposure was a patient from the 2004 outbreak in Rajbari, Bangladesh. This patient was breastfed by his mother while the mother was ill with Nipah virus. Although the epidemiologic questionnaire stated that the child had no contact with a febrile patient, upon in-depth interview family members reported that the mother had contact with her 24-month-old child at least during the early stage of her illness. In the 2011 Rangpur outbreak, for a case who was a professional driver, there was no reported contact with a Nipah patient on the questionnaire. The case-patient's colleagues and wife did not report any exposure to date palm sap or *tari* either; however, did report that the case transported a sick person in his taxi, possibly a Nipah case, during the 15 days before his death.

Five cases from 2004 to 2012 reported not having exposures to date palm sap or a Nipah case during the in-depth interview. One case, a 13-year-old girl, was reported by proxies to regularly eat bat-dropped fruit from the ground as well as climb trees to fetch the fruit. Relatives of another case reported that he played with bat-dropped fruit, such as boroi, olive, or banana. Neighbors of a 65-year-old woman reported that she may have consumed raw date palm sap available at the local market and that she regularly ate dropped star fruit from the grounds of her neighbor's house. The fourth case, an electrician, would climb electric pillars and trees to repair cable lines and may have had direct contact with bats who rested on cable lines. However, family members also reported he consumed bat-dropped fruit 6–7 days prior to his onset of illness. The last case, a 5-year-old boy, was fed bat meat as a traditional remedy for his breathing problem.

The in-depth interviews conducted with the 2001–2003 cases that initially reported no exposure to a Nipah case suggested that exposure to another Nipah case was misclassified on the survey tool. Exposure to date palm sap was also identified as an important exposure, but retrospectively as it was not asked about as an exposure prior to 2004 because of the evolution of our understanding of Nipah epidemiology. In the 2001 Meherpur outbreak, one case infected through drinking raw date palm sap likely infected 12 others through person-to-person transmission. Among the three cases that were reinvestigated for exposures, one reported consuming date palm sap and two reported having had contact with the initial case. In the 2003 Naogaon outbreak, six cases were re-classified as date palm sap drinkers and one case as having had contact with a Nipah case. One case from the 2003 Naogaon outbreak, however, reported neither date palm sap consumption nor contact with a Nipah case, but rather direct contact with bat excreta; the case would go under a bat roost to defecate. Overall, based on the qualitative data collected, among the 25 cases from 2001 to 2003 and the 157 cases from 2004 to 2012, only 3% of cases reported neither exposure to date palm sap nor a Nipah case (Table 3). Previously, we believed 25% of all cases reported neither exposure to date palm sap nor a Nipah case (44% of cases from 2001 to 2003 and 22% of cases since 2004).

DISCUSSION

The primary objective of our analysis was to identify rare exposures associated with Nipah virus infection by combining data from multiple outbreaks to increase our power to find associations. What we found, however, was that the exposures repeatedly identified in individual outbreaks—consumption of raw sap and contact with another case—remained the best explanation for infection, even with greater power to find new associations. This is an important finding for the prevention of Nipah virus in Bangladesh because by ruling out other major contributors of infection, it allows the public health community to focus its efforts on interrupting these two transmission pathways.

While raw date palm sap consumption and contact with an infected person remained the predominant transmission pathways of human Nipah virus infection in Bangladesh, case-patients were more likely than controls to report that *Pteropus* bats visit trees near their homes at night. This association could represent a proxy for bats contaminating raw date palm sap at night and a bias of case-patients having prior knowledge that the disease is spread by bats. As the cumulative effects of multiple factors are not necessarily additive, the attributable risk of Nipah infection for bats being around the house at night (40%) is likely largely accounted for in the attributable risk for drinking date palm sap (50%) (Benichou 2001). Additionally, while case-patients were more likely to travel outside their sub-district, this association could represent our inability to capture exposures to date palm sap or a Nipah case that occurred during travels, as village proxies likely did not travel with case-patients and, therefore, could not report on exposures during travel. In support of this, other studies suggest that date palm sap is not available in every village and that availability of the sap is a primary determinant of whether individuals consume it or not (Nahar et al. 2015). Although we do not know of outbreaks occurring in areas where these people traveled, they may have encountered cases that went unobserved by our surveillance system. Therefore, the attributable risk of Nipah infection for traveling to another sub-district (22%) could largely be accounted for in the attributable risk of having contact with a Nipah case (30%) or drinking date palm sap (50%).

Zoonotic transmission of Nipah from bat-dropped fruit to pigs, and subsequently from pigs to humans, was the most likely transmission route in the 1999 Malaysian outbreak. Although there is anecdotal evidence of one goat to human transmission event in Bangladesh (Luby et al. 2009a), we found no evidence of an association between exposure to sick animals and being a human case (Chua 2010a; b; Luby and Gurley 2012). Although the in-depth interviews identified some potential exposures to bat secretions, like exposures to dropped fruit, it did not identify any common exposures that we have not evaluated in previous studies. Though we cannot rule out contact with sick livestock or bat-dropped fruit as occasional modes of Nipah transmission, this study suggests these exposures have not been important contributors to spillover infections in Bangladesh.

In the univariable analysis, eating washed fruit was significantly protective; however, less so after adjusting for multiple comparisons. This could be due to chance variation, as based on a 95% confidence interval, 1 in 20 significant findings will be spurious (Rothman et al. 2012). The more likely explanation though may be that due to the high case fatality of Nipah, controls were able to report exposures more completely than cases. While it is possible that sour fruits with high vitamin C content may have a protective effect against viral infection through ascorbic acid, there was no observed association between eating fruit and Nipah infection in the multivariable model of best fit (Atherton et al. 1978).

In the case–control studies conducted during outbreaks, there was evidence of misclassification of exposures. Since most Nipah case-patients die, proxy respondents were asked about case-patient exposures. As proxies may have had limited knowledge of case-patients' activities prior to illness, this likely leads to inaccurate reporting and an underestimate of relevant exposures. This differential misclassification of exposures would lead to a bias towards the null, however, because the reported exposures are likely an underestimate. Therefore, the reported associations are likely an underestimate of true associations. Furthermore, we could not control for having a proxy respondent in the analysis since, as is inherent in case-control studies, selection of study participants is related to the outcome. Because the exposures of consuming date palm sap and having contact with a Nipah case are strongly associated with the outcome, which has a high case fatality, these exposures are spuriously linked to having a proxy respondent (Greenland et al. 1999a; Cole et al. 2010; Rothman et al. 2012). This limitation is inherent to investigations of diseases with high case fatality and should be considered in data analysis and interpretation.

While reporting bias likely contributed to the misclassification of exposures, there are numerous examples of discrepancies in quantitative and qualitative findings from outbreak investigations that used both methods. In the 2005 Tangail outbreak of Nipah, qualitative in-depth investigations reported much higher date palm sap consumption than the quantitative survey (Luby et al. 2006). In the Bangladesh 2007 outbreak of illness from consumption of *Xanthium stumarium*, a plant also known as *ghagra shak*, qualitative investigators also found higher levels of reported exposure during in-depth interviews than in quantitative surveys (Gurley et al. 2010).

Qualitative in-depth interviews are open-ended discussions between study participants and the investigators that aim to build rapport. By spending more time in case villages and building rapport, participants may be more willing to mention certain exposures that are socially stigmatizing or that participants suspect may be a cause of the outbreak. Interviews that use structured questionnaires are less likely to elicit honest responses of sensitive information, as they are relatively shorter by comparison and less likely to build rapport (Steckler et al. 1992). Qualitative interviews are also imperfect, however; they can result in confirmation bias as a result of prodding by the interviewer, as discussed in anthropologic research literature (Bernard 1984). Furthermore, when multiple proxy respondents are interviewed, it is important to note how discrepancies between answers will be interpreted. Thus, outbreak investigations could benefit from the use of both approaches, particularly when the hypotheses about exposures causing disease are still being formed.

A limitation of studying Nipah virus and other rare, emerging infections is that our knowledge of relevant exposures changes over time and subsequent changes in questionnaires compromise our ability to compare data across years. This is true for our analysis where date palm sap was not included in the questionnaire until 2004 and *tari* consumption not until 2012.

Furthermore, as some case-patients die before a blood sample can be collected, cases are classified as either probable, having an epidemiologic link with another case, fever, but no lab-confirmation, as well as lab-confirmed. An epidemiologic link can signify contact, or living in the same village, or sharing date palm sap. A concern with the use of probable cases in our analysis is that probable cases are not laboratory confirmed. In a sensitivity analysis conducted with only lab-confirmed Nipah cases, we find similar results and that our model is still significant with slightly reduced effect estimates for drinking raw date palm sap and larger effect estimates for having contact with another case (Supplement Table 1).

Additionally, cases were matched to controls initially for geographic location, age, and sex. Only geographic location was used as a matching variable in later years to avoid complications of overmatching. To control for this change in methods, we adjusted for age and sex in all models. This likely resulted in a loss of statistical efficiency as some cases were matched for age and sex in addition to controlling for age and sex in the models. Lastly, imperfect recall is also a concern because in-depth interviews in this study were conducted 1–10 years after the exposures occurred, which may have further influenced the misclassification of exposures.

CONCLUSION

While this analysis did not identify new risk factors for Nipah infection, it confirmed the centrality of transmission through contaminated sap and contact with infected patients, allowing public health officials to further focus efforts to interrupt these transmission pathways. Our study also highlights the benefits of using both qualitative and quantitative methods in outbreak investigations; using the strengths of each approach is advantageous particularly when knowledge of the transmission of a disease is still evolving.

Efforts to prevent this highly fatal emerging infection should focus on interrupting transmission from bats to humans through date palm sap and reducing the exposure to Nipah-infected patients in Bangladesh. The use of bamboo skirts around the date palm sap collection area on trees prevents bat contamination of sap and can be practiced more widely, thereby reducing population risk (Khan et al. 2012; Nahar et al. 2014). For Nipah infection spread, handwashing with soap has had a significantly protective effect (Gurley et al. 2007a) and interventions to promote handwashing and increase access to handwashing stations in hospitals are urgently needed. Though some infection control has been ongoing, limited infection control within hospitals and by family caregivers continue to be major contributors to disease transmission (Ram et al. 2011; Islam et al. 2014). The recent West Africa Ebola outbreak made evident the difficulty of infection control in limited resource settings and the need for interventions to prevent person-to-person transmission, especially for similarly fatal, emerging infections (Chertow et al. 2014). Importantly, improving such interventions could cause additional benefits by reducing other bat-borne pathogens, including emerging and endemic pathogens, which could transmit via contaminated fluids or from person-to-person.

Supplementary Material

Refer to Web version on PubMed Central for supplementary material.

Acknowledgments

Funded by icddr,b and CDC cooperative agreement (no. 5U01CI000628-01), NIH Grant No. 07-015-0712-52200 (Bangladesh-NIH/Emerging). icddr,b is grateful to the Governments of Bangladesh, Canada, Sweden and the UK for providing core/unrestricted support.

References

Atherton JG, Kratzing CC, Fisher A. The effect of ascorbic acid on infection chick-embryo ciliated tracheal organ cultures by coronavirus. Arch Virol. 1978; 56:195–199. [PubMed: 205194]

Benichou J. A review of adjusted estimators of attributable risk. Stat Methods Med Res. 2001; 10:195–216. [PubMed: 11446148]

Bernard H. The Problem of Informant Accuracy: The Validity of Retrospective Data. Annu Rev Anthropol. 1984; 13:495–517. DOI: 10.1146/annurev.anthro.13.1.495

Bruzzi P, Green SB, Byar DP, Brinton LA, Schairer C. Estimating the population attributable risk for multiple risk factors using case-control data. Am J Epidemiol. 1985; 122:904–914. [PubMed: 4050778]

Chertow DS, Kleine C, Edwards JK, Scaini R, Giuliani R, Sprecher A. Ebola virus disease in West Africa–clinical manifestations and management. N Engl J Med. 2014; 371:2054–2057. DOI: 10.1056/NEJMp1413084 [PubMed: 25372854]

Chua KB. Epidemiology, surveillance and control of Nipah virus infections in Malaysia. Malays J Pathol. 2010; 32:69–73. [PubMed: 21329176]

Chua KB. Risk factors, prevention and communication strategy during Nipah virus outbreak in Malaysia. Malays J Pathol. 2010; 32:75–80. [PubMed: 21329177]

Chua KB, Bellini WJ, Rota PA, Harcourt BH, Tamin A, Lam SK, Ksiazek TG, Rollin PE, Zaki SR, Shieh W, Goldsmith CS, Gubler DJ, Roehrig JT, Eaton B, Gould AR, Olson J, Field H, Daniels P, Ling AE, Peters CJ, Anderson LJ, Mahy BW. Nipah virus: a recently emergent deadly paramyxovirus. Science. 2000; 288:1432–1435. [PubMed: 10827955]

Cole SR, Platt RW, Schisterman EF, Chu H, Westreich D, Richardson D, Poole C. Illustrating bias due to conditioning on a collider. Int J Epidemiol. 2010; 39:417–420. DOI: 10.1093/ije/dyp334 [PubMed: 19926667]

Cummings P. The relative merits of risk ratios and odds ratios. Arch Pediatr Adolesc Med. 2009; 163:438–445. DOI: 10.1001/archpediatrics.2009.31 [PubMed: 19414690]

Greenland S, Pearl J, Robins JM. Causal diagrams for epidemiologic research. Epidemiology. 1999; 10:37–48. [PubMed: 9888278]

Greenland S, Robins JM, Pearl J. Confounding and Collapsibility in Causal Inference. Stat Sci. 1999; 14:29–46.

Gurley ES, Montgomery JM, Hossain MJ, Bell M, Azad AK, Islam MR, Molla MA, Carroll DS, Ksiazek TG, Rota PA, Lowe L, Comer JA, Rollin P, Czub M, Grolla A, Feldmann H, Luby SP, Woodward JL, Breiman RF. Person-to-person transmission of Nipah virus in a Bangladeshi community. Emerg Infect Dis. 2007; 13:1031–1037. [PubMed: 18214175]

Gurley ES, Montgomery JM, Hossain MJ, Islam MR, Molla MA, Shamsuzzaman SM, Akram K, Zaman K, Asgari N, Comer JA, Azad AK, Rollin PE, Ksiazek TG, Breiman RF. Risk of nosocomial transmission of Nipah virus in a Bangladesh hospital. Infect Control Hosp Epidemiol. 2007; 28:740–742. [PubMed: 17520553]

Gurley ES, Rahman M, Hossain MJ, Nahar N, Faiz MA, Islam N, Sultana R, Khatun S, Uddin MZ, Haider MS, Islam MS, Ahmed BN, Rahman MW, Mondal UK, Luby SP. Fatal outbreak from consuming Xanthium strumarium seedlings during time of food scarcity in northeastern Bangladesh. PLoS One. 2010; 5:e9756.doi: 10.1371/journal.pone.0009756 [PubMed: 20305785]

Halpin K, Hyatt AD, Fogarty R, Middleton D, Bingham J, Epstein JH, Rahman SA, Hughes T, Smith C, Field HE, Daszak P, Group HER. Pteropid bats are confirmed as the reservoir hosts of henipaviruses: a comprehensive experimental study of virus transmission. Am J Trop Med Hyg. 2011; 85:946–951. [PubMed: 22049055]

Holm S. A Simple Sequentially Rejective Multiple Test Procedure. Scand J Stat. 1979; 6:65–70. DOI: 10.2307/4615733

Homaira N, Rahman M, Hossain MJ, Epstein JH, Sultana R, Khan MS, Podder G, Nahar K, Ahmed B, Gurley ES, Daszak P, Lipkin WI, Rollin PE, Comer JA, Ksiazek TG, Luby SP. Nipah virus outbreak with person-to-person transmission in a district of Bangladesh, 2007. Epidemiol Infect. 2010; 138:1630–1636. [PubMed: 20380769]

Hsu VP, Hossain MJ, Parashar UD, Ali MM, Ksiazek TG, Kuzmin I, Niezgoda M, Rupprecht C, Bresee J, Breiman RF. Nipah virus encephalitis reemergence, Bangladesh. Emerg Infect Dis. 2004; 10:2082–2087. DOI: 10.3201/eid1012.040701 [PubMed: 15663842]

ICDDRB. Outbreaks of Encephalitis Due to Nipah/Hendra-like Viruses, Western Bangladesh. Heal Sci Bull icddr,b. 2003; 1:1–6.

ICDDRB. Nipah Virus Outbreak from Date Palm Juice. Heal Sci Bull. 2005; 3:1–5.

Islam MS, Luby SP, Sultana R, Rimi NA, Zaman RU, Uddin M, Nahar N, Rahman M, Hossain MJ, Gurley ES. Family caregivers in public tertiary care hospitals in Bangladesh: risks and opportunities for infection control. Am J Infect Control. 2014; 42:305–310. DOI: 10.1016/j.ajic.2013.09.012 [PubMed: 24406254]

Islam S. Nipah transmission from bats to humans associated with drinking traditional liquor (tari) in northern Bangladesh. Heal Sci Bull. 2011; 10:16–20.

Kashiwazaki Y, Na YN, Tanimura N, Imada T. A solid-phase blocking ELISA for detection of antibodies to Nipah virus. J Virol Methods. 2004; 121:259–261. DOI: 10.1016/j.jviromet.2004.06.015 [PubMed: 15381364]

Khan SU, Gurley ES, Hossain MJ, Nahar N, Sharker MA, Luby SP. A randomized controlled trial of interventions to impede date palm sap contamination by bats to prevent nipah virus transmission in Bangladesh. PLoS One. 2012; 7:e42689. [PubMed: 22905160]

Luby SP. The pandemic potential of Nipah virus. Antivir Res. 2013; 100:38–43. [PubMed: 23911335]

Luby SP, Gurley ES. Epidemiology of henipavirus disease in humans. Curr Top Microbiol Immunol. 2012; 359:25–40. [PubMed: 22752412]

Luby SP, Gurley ES, Hossain MJ. Transmission of human infection with Nipah virus. Clin Infect Dis. 2009; 49:1743–1748. [PubMed: 19886791]

Luby SP, Hossain MJ, Gurley ES, Ahmed BN, Banu S, Khan SU, Homaira N, Rota PA, Rollin PE, Comer JA, Kenah E, Ksiazek TG, Rahman M. Recurrent zoonotic transmission of Nipah virus into humans, Bangladesh, 2001-2007. Emerg Infect Dis. 2009; 15:1229–1235. [PubMed: 19751584]

Luby SP, Rahman M, Hossain MJ, Blum LS, Husain MM, Gurley E, Khan R, Ahmed BN, Rahman S, Nahar N, Kenah E, Comer JA, Ksiazek TG. Foodborne transmission of Nipah virus, Bangladesh. Emerg Infect Dis. 2006; 12:1888–1894. [PubMed: 17326940]

Montgomery JM, Hossain MJ, Gurley E, Carroll GD, Croisier A, Bertherat E, Asgari N, Formenty P, Keeler N, Comer J, Bell MR, Akram K, Molla AR, Zaman K, Islam MR, Wagoner K, Mills JN, Rollin PE, Ksiazek TG, Breiman RF. Risk factors for Nipah virus encephalitis in Bangladesh. Emerg Infect Dis. 2008; 14:1526–1532. [PubMed: 18826814]

Nahar N, Mondal UK, Hossain MJ, Khan MSU, Sultana R, Gurley ES, Luby SP. Piloting the promotion of bamboo skirt barriers to prevent Nipah virus transmission through date palm sap in Bangladesh. Glob Health Promot. 2014; 21:7–15. DOI: 10.1177/1757975914528249

Nahar, N.; Paul, RC.; Sultana, R.; Gurley, ES.; Abedin, J.; Sumon, SA.; Banik, KC.; Asaduzzaman, M.; Rimi, NA.; Rahman, M.; Luby, SP. Raw sap consumption habits and its association with knowledge of Nipah virus in two endemic districts in Bangladesh. PLOS ONE. 2015. http://dx.doi.org/10.1371/journal.pone.0142292

Shapshak, P.; Sinnott, JT.; Somboonwit, C.; Kuhn, J. Global Virology I-Identifying and Investigating Viral Diseases. New York: Springer Science; 2015. p. 125-146.

Rahman MA, Hossain MJ, Sultana S, Homaira N, Khan SU, Rahman M, Gurley ES, Rollin PE, Lo MK, Comer JA, Lowe L, Rota PA, Ksiazek TG, Kenah E, Sharker Y, Luby SP. Date palm sap linked to Nipah virus outbreak in Bangladesh, 2008. Vector Borne Zoonotic Dis. 2012; 12:65–72. [PubMed: 21923274]

Ram PK, Jahid I, Halder AK, Nygren B, Islam MS, Granger SP, Molyneaux JW, Luby SP. Variability in hand contamination based on serial measurements: implications for assessment of hand-cleansing behavior and disease risk. Am J Trop Med Hyg. 2011; 84:510–516. DOI: 10.4269/ajtmh.2011.10-0299 [PubMed: 21460002]

Ramasundram V, Tan CT, Chua KB, Chong HT, Goh KJ, Chew NK, Tan KS, Thayaparan T, Kunjapan SR, Petharunam V, Loh YL, Ksiazek TG, Lam SK. Kinetics of IgM and IgG seroconversion in Nipah virus infection. Neurol J Southeast Asia. 2000; 5:23–28.

Rockhill B, Newman B, Weinberg C. Use and misuse of population attributable fractions. Am J Public Health. 1998; 88:15–19. DOI: 10.2105/AJPH.88.1.15 [PubMed: 9584027]

Rothman, KJ.; Lash, TL.; Greenland, S. Modern Epidemiology. Philadelphia: Lippincott Williams & Wilkins; 2012.

Sazzad HM, Hossain MJ, Gurley ES, Ameen KM, Parveen S, Islam MS, Faruque LI, Podder G, Banu SS, Lo MK, Rollin PE, Rota PA, Daszak P, Rahman M, Luby SP. Nipah virus infection outbreak with nosocomial and corpse-to-human transmission, Bangladesh. Emerg Infect Dis. 2013; 19:210–217. [PubMed: 23347678]

Sejvar JJ, Hossain J, Saha SK, Gurley ES, Banu S, Hamadani JD, Faiz MA, Siddiqui FM, Mohammad QD, Mollah AH, Uddin R, Alam R, Rahman R, Tan CT, Bellini W, Rota P, Breiman RF, Luby SP. Long-term neurological and functional outcome in Nipah virus infection. Ann Neurol. 2007; 62:235–242. [PubMed: 17696217]

Steckler A, McLeroy KR, Goodman RM, Bird ST, McCormick L. Toward integrating qualitative and quantitative methods: an introduction. Health Educ Q. 1992; 19:1–8. DOI: 10.1177/109019819201900101 [PubMed: 1568869]

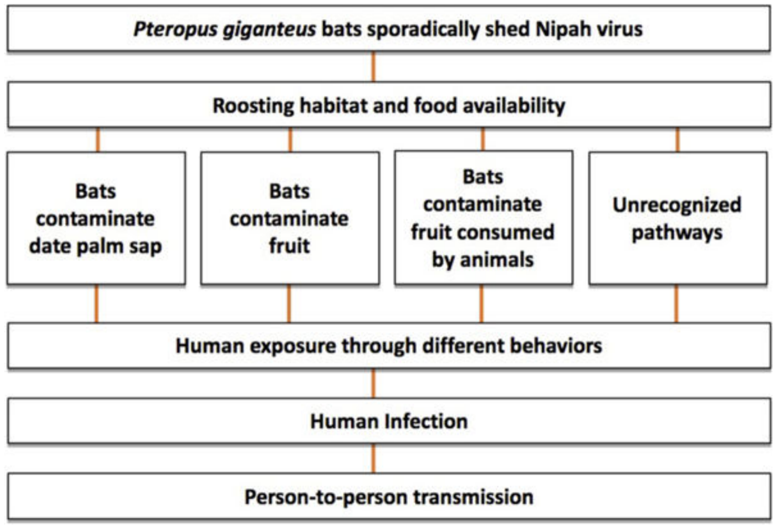

Fig. 1.
Framework for investigating zoonotic transmission of Nipah virus in Bangladesh.

Table 1

Age- and Sex-Adjusted Univariable Analysis for Risk Factors for Human Nipah Cases, Bangladesh, 2004–2012.

Exposures[‡]	Cases (N = 157) (%)[†]	Controls (N = 632) (%)[†]	aOR[*]	95% CI	*P*	aP[**]
Occupation	100	100	1.03	(0.96–1.10)	0.424	1
House type (relative to living in a thatched house)						
Semi-pucca	8	13	1.53	(0.49–4.76)	0.463	1
Tin-shed	83	65	4.40	(1.83–10.61)	0.001	0.033
Concrete or brick building	0.8	2	1.08	(0.12–9.86)	0.948	1
Other	0.8	0.5	2.21	(0.12–40.31)	0.593	1
Climbed any tree	29	22	1.38	(0.79–2.43)	0.257	1
Animal contact	58	56	0.86	(0.52–1.43)	0.562	1
Ill animal contact	14	16	0.91	(0.49–1.68)	0.767	1
Dead animal contact	9	8	1.26	(0.60–2.64)	0.542	1
Killed a sick animal	4	4	0.66	(0.17–2.60)	0.551	1
Ate a sick animal	10	8	1.23	(0.55–2.73)	0.610	1
Bats in trees around the						
House in the day	14	11	1.29	(0.72–2.30)	0.399	1
House at night	57	41	2.30	(1.51–3.50)	<0.001	0.033
Work/school in the day	6	5	1.11	(0.50–2.45)	0.793	1
Work/school at night	15	12	1.46	(0.83–2.58)	0.186	1
See an increase in						
Rodents	10	10	0.96	(0.52–1.78)	0.908	1
Shrews	4	6	0.59	(0.24–1.46)	0.250	1
Large bats	3	2	1.52	(0.50–4.59)	0.457	1
Small bats	0.8	4	3.81	(1.11–13.04)	0.033	1
Consumed bats or small bats as food or medicine	0.7	0.2	3.77	(0.22–66.03)	0.363	1
Eaten						
Any fruit	92	94	0.85	(0.41–1.79)	0.671	1
Plum/Boroi	46	58	0.57	(0.38–0.87)	0.009	0.297
Star fruit	10	17	0.52	(0.28–0.99)	0.047	1
Guava	21	32	0.45	(0.27–0.73)	0.001	0.033

Exposures‡	Cases (N = 157) (%)†	Controls (N = 632) (%)†	aOR*	95% CI	*P*	aP**
Pick any fruit from the ground	40	35	1.33	(0.81–2.19)	0.259	1
Washed before eating						
Any fruit	48	63	0.47	(0.30–0.73)	0.001	0.033
Boroi	16	27	0.41	(0.21–0.81)	0.010	0.33
Star fruit	5	15	0.13	(0.01–1.12)	0.063	1
Guava	9	19	0.09	(0.01–0.75)	0.026	0.858
Visited any area outside own sub-district	34	18	2.38	(1.51–3.74)	0.000	0
Visited any hospital or clinic	37	23	4.45	(2.53–7.84)	0.000	0
Consumed raw date palm sap	54	25	4.91	(3.16–7.65)	0.000	0
Contact with Nipah case	37	19	7.28	(3.96–13.37)	0.000	0

*Adjusted for sex and categorical age.

**Adjusted Bonferroni *P* value.

†Proportions based on non-missing data.

‡Exposures are within the one month before the cases' onset of illness.

Table 2

Nipah Risk Factor Multivariable Model of Best Fit, Bangladesh, 2004–2012.

Risk factors	N	aOR*	95% CI	*P* value	AR†
Lived in a tin-shed house	367	1.92	(0.59–6.27)	0.279	
Bats in trees around house at night	367	3.25	(1.20–8.83)	0.021	0.40
Visited any area outside own sub-district	367	3.50	(1.49–8.22)	0.004	0.22
Washed any fruit before eating	367	0.54	(0.25–1.19)	0.128	
Date palm sap consumption	367	16.7	(6.50–42.7)	0.000	0.50
Contact with Nipah case	367	8.38	(2.59–27.2)	0.000	0.30

*Adjusted for sex and age category.

†Attributable risk of factors with significant associations.

Table 3

Exposures reported by Nipah cases from epidemiologic questionnaires and updated with responses from in-depth interviews.

Risk factors	All cases[†] n=182 % (n)[*]
Date palm sap consumption	53 (96)
Raw date palm sap consumption	51 (92)
Tari consumption	2 (4)
Contact with Nipah case	44 (80)
Neither date palm sap nor contact with Nipah case	3 (6)

[*]Proportions based on non-missing data.

[†]Data based on initial epidemiologic survey and updated using in-depth interviews done for all cases 2001–2012.