

Viral diversity of bat communities in human-dominated landscapes in Mexico

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Abstract

Using integrative epidemiologic techniques, we studied the changing relationships (beta and phylogenetic beta diversity) of multihost systems and virus associations in bat communities in fragmented landscapes from Chiapas, Campeche and Greater Mexico City. We combined computing applications, molecular detection, and nucleotide sequencing of coronaviruses, hantaviruses, paramyxoviruses and pegiviruses with ecological and phylogenetic analyses. A total of 22 viruses were discovered in 1,067 samples from 42 bat species, representing an estimated 78% of all viral richness in the system. Based on 17 virus genotypes discovered with an equal sampling effort, a total viral richness of 23 genotypes was estimated using a Chao2 statistic model. Using a residual model, we categorized host species and habitat types that are prone to harboring higher viral richness. Positive relationships were found between phylogenetic host diversity and both viral diversity ($r = 0.41$, $p < 0.05$) and viral richness ($r = 0.51$, $p < 0.05$). The beta diversity (the rate of change) of viral communities was explained by host beta diversity ($r = 0.86$, $p < 0.05$). To understand the change in viral and host communities, we partitioned beta diversity in nestedness (species loss) and turnover (compositional dissimilarity) components. In Chiapas, the host beta diversity was explained by the nestedness of species composition, while the phylogenetic host diversity was explained by turnover of the host lineages. Campeche showed a high phylogenetic host nestedness and low host turnover. Beta-diversity and beta-phylogenetic diversity indicated that patterns of local species assemblages and regional abiotic features in human-dominated landscapes are significant drivers of viral community composition. Our study represents the first effort in Mexico to study the relationship between viral diversity in bat communities in modified landscapes to understand host-virus relationships.

Keywords: Disease Ecology; Chiroptera; Viral richness; Alpha diversity; Beta diversity; Phylogenetic diversity; Habitat loss.

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Introduction

Land use change appears to be the primary mechanism driving zoonotic diseases (Patz *et al.*, 2004). The expansion of agricultural production and urbanization has simultaneously modified ecosystem structure and function, community structure (species assemblages), patterns of species distribution and biodiversity (Christian *et al.*, 2009; Gibbs *et al.*, 2009). These modified systems have produced suitable environments for multi-species interactions, particularly hosts, vectors, and/or pathogens (McMichael, 2004; Rivard *et al.*, 2007; Jones *et al.*, 2013; Rubio *et al.*, 2014).

Bats, on one hand, have been considered a likely source of highly pathogenic RNA viruses including lyssaviruses (Banyard *et al.*, 2011), Ebola virus (Leroy *et al.*, 2005), Marburg virus (Towner *et al.*, 2009), Nipah virus (Epstein *et al.*, 2006), Hendra virus (Smith *et al.*, 2011), and coronaviruses (CoV) (e.g., SARS-coronavirus, MERS-coronavirus) (Hilgenfeld and Peiris, 2013). On the other hand, bats are recognized as a key group in the maintenance of ecological systems by providing ecological services such as pollination, seed dispersal and agricultural pest control (Medellín, 2009; Kunz *et al.*, 2011). Additionally, due to their response to habitat loss and fragmentation, bats are excellent bioindicators of environmental changes (Medellín *et al.*, 2000).

To properly understand complex interactions in multihost systems, several ecological and phylogenetic tools have been used. In disease ecology, diversity indices have been used to correlate the number of species (*richness*) and the relative abundance of species present in a given community (*alpha diversity*) (Suzán *et al.*, 2009) and in microbiome systems (Anthony *et al.*, 2013a; Olson *et al.*, 2014) with disease prevalence. Diversity indices have also been used to evaluate changes in host-parasite composition in host communities at the local, regional, and biogeographic scales (*alpha, beta gamma diversity*) (Svensson-Coelho and Ricklefs, 2011; Scordato and Kardish, 2014). From an evolutionary perspective, host and pathogen phylogenetic relationships have been studied, and diversity indices have been incorporated to measure changes in host species community assemblages through environmental gradients (Webb *et al.*, 2002; Helmus *et al.*, 2007). These phylogenetic methods offer additional dimensions to explore host-parasite interactions over time, such as host specificity, host-parasite co-evolution, host switching events, and phylogenetic barriers preventing pathogen transmission (Legendre *et al.*, 2002; Streicker *et al.*, 2010; Poulin *et al.*, 2011). The study of ecological and phylogenetic interactions between host-pathogen systems integrates the role of environmental influences on host and pathogen distributions across time and spatial scales and across different levels of biological organization beyond taxonomic levels (Hawley and Altizer, 2011).

In this study, we examined the relationship between host diversity and the diversity of four viral taxa in bats from human-dominated landscapes in Mexico. Two hypotheses were tested related to the effect of host species and host phylogenetic diversities on viral diversity and the influence of habitat type on the composition of host and viral communities. First, we hypothesized that (1) host communities with high species and phylogenetic diversities will support high values of viral diversity; (2) changes in host and viral community composition across a habitat type will be reflected in high values of beta diversity and phylogenetic-beta diversity.

Materials and Methods

Sample collection

Bats were captured at three different sites in Mexico: the Reserva de la Biósfera Montes Azules (RBMA) in Chiapas, the Reserva de la Biósfera Calakmul (RBC) in Campeche, and Greater Mexico City (GMC) including the Distrito Federal and Metropolitan Area. The first two sites, located in southeastern Mexico, represent regions of high species diversity and are characterized by large tracts of continuous primary vegetation, while the GMC site is highly urbanized with vegetation patches. A high evergreen forest characterizes RBMA, while RBC is dominated by tropical semi-deciduous forest; both regions have high anthropogenic pressure. In RBMA and RBC, bats were collected from three different habitat types: 'Forested' (Fd), where signs of human impact are largely absent and the original vegetation persists; 'Fragmented' (F), where areas of primary vegetation are interspersed with agricultural/rangeland; and 'Disturbed' (D), the transition zone between areas of secondary vegetation and agricultural/rangeland or urban areas. In the GMC sites, bats were captured in 'Urban' (U), human-dominated areas and 'Fragmented' habitats. We used 5 mist-nets (each 9 x 3 m wide) that were opened at dusk and remained open for four consecutive hours. Each habitat was sampled once in six months. Bats were identified using a field guide (Medellín *et al.*, 2008). The minimal distance in RBMA was 2 km, while in RBC it was 10 km. A mantel test was performed to ensure site independence due to the geographic distance (RBMA; $r = 0.55$, $p = 0.01$; RBC $r = 0.57$ $p = 0.006$). Oral and rectal swabs and, when possible, blood samples were collected from each animal. Samples were collected in lysis buffer and preserved at -80°C until transfer to the Center for Infection and Immunity, Columbia University, New York for viral screening.

Virus discovery

A total of 1,067 samples from 608 individuals representing 42 bat species were tested for the five viral families/genera (Table S1). Total nucleic acid was extracted from all samples using the EasyMag® (bioMérieux, Inc Darham, NC, USA.) platform, and cDNA synthesis performed using SuperScript® III first strand synthesis supermix (Invitrogen), all according to the manufacturer's instructions. Viral discovery was performed using broadly reactive consensus PCR primers, targeting the L-Segment for hantavirus (HTV) detection (Klempa *et al.*, 2006) and the polymerase (pol) gene for paramyxovirus (PMV) detection (Tong *et al.*, 2008). PCR products of the expected size were cloned into the StrataClone™ PCR cloning vector and sequenced using standard M13R primers. CoV, hepatitis virus (HPV) and pegivirus (PGV) detections have been previously reported, and these viral sequences were detected in the same 1,067 samples (Anthony *et al.*, 2013b; Quan *et al.*, 2013).

Estimates and Completeness of Viral Richness

We evaluated our sampling effort (the number of samples tested for a given virus), using two methods: by producing rarefaction and extrapolation curves and by calculating the values of the residuals of the linear regression between viral richness within a host and sampling effort by host. Rarefaction and extrapolation curves are statistical techniques to estimate the number of species for a given number of

samples (Magurran, 2004; Chao and Jost, 2012), allowing the evaluation of the sampling effort and estimation of the number of host samples required to obtain a viral richness value with 95% confidence (Chao *et al.*, 2014). We evaluated the viral richness, defined as unique viruses discovered in the 1,067 samples, by constructing sample size-based rarefaction and extrapolation curves using a three-fold original sample effort (3,201 samples) (Chao *et al.*, 2014) with the R iNEXT library (Hsieh *et al.*, 2013). The same methodology was used to explore viral completeness by habitat type. For this purpose, we only considered samples with the same number of PCR screenings (CoVs, PMVs and HTVs). To identify host species associated with higher viral richness, we used a methodology proposed by Herbreteau, 2012. We calculated residual values from the linear regression of the logarithm of viral richness and the logarithm of sampling effort for each species and at each disturbance level. These data were logarithmically transformed to stabilize the variance. Host species or disturbance levels with positive or negative residual values were identified as host species with more or less viral richness than expected by the regression model (Herbreteau *et al.*, 2012).

Host and Viral Diversity

To study regional host and viral alpha diversities, abundance matrixes (host and virus) were constructed, where the rows were disturbance level and the columns were (i) host species and (ii) viruses discovered in each disturbance level. Using the R vegan library (Oksanen *et al.*, 2013), a Shannon-Wiener diversity index (Shannon, 1948) was calculated for each matrix. Values ranged from 0, when there is only one species present, to 1, when all species are equally represented in the sample (Magurran, 2004).

Phylogenetic Diversity and Host Specificity

The mammalian super tree (Bininda-Emonds *et al.*, 2007) was used to calculate the phylogenetic diversity (PD) of host communities using the R Picante library (Kembel *et al.*, 2010). The PD was measured by calculating the sum of the total branch length of the host species phylogeny sampled in each habitat type (Faith, 1992). Because the data on host taxonomic diversity were not normally distributed, only phylogenetic analyses were performed. The relationships of viral richness and viral diversity with host PD was explored using a linear model. To quantify host-viral taxonomic associations, we used a modified index of host specificity proposed by Poulin and Mouillot, 2003, that measures the PD of host communities associated with each virus. Viruses with high values of host specificity exhibit plasticity to infect a wide range of hosts, while viruses with lower values are restricted to a few closely related host species (Poulin and Mouillot, 2003; Poulin *et al.*, 2011).

Beta Diversity and Phylogenetic Beta Diversity

A Pearson correlation test was performed to explore the relationship between host BD and PBD with changing compositions of viral communities by habitat type calculated by the Sorensen index. To evaluate the change in composition of viral and host communities (BD) within regions, we used measures of beta-diversity: spa-

tial turnover (β_{SIM}) and nestedness (β_{SNE}) components (Baselga, 2010). Spatial turnover (β_{SIM}) measures the replacement of species by other species due to environmental factors or spatial isolation, such as by habitat fragmentation (Calderón-Patrón *et al.*, 2012). The nestedness component (β_{SNE}) measures whether sites with smaller number of species are subsets of richer sites (Ulrich *et al.*, 2009). These components were calculated for taxonomic and phylogenetic beta-diversity analyses. Phylogenetic beta-diversity (PBD) measures how phylogenetic relatedness changes across space in the same manner that BD measures how species composition changes across space (Graham and Fine, 2008). The PBD between disturbance levels was obtained using the inverse of the PhyloSor index (Bryant *et al.*, 2008). This index represents shared branches between communities from two sites. Values range from 0 when no species are shared to 1 when all species in the two locations are the same. The methodology of Leprieux (2012) was used to calculate the phylogenetic turnover ($P\beta_{SIM}$) and to measure the phylogenetic dissimilarity, nested patterns of species assemblages ($P\beta_{SNE}$). The functions *beta.multi* for BD and *phylo.beta.multi* for PBD from the R betapart library were also applied to calculate the influence of each component on host and viral community composition by habitat type in each region (Baselga and Orme, 2013).

Results

Viral Community Data

A total of 4,139 consensus PCR assays were performed for viral detection, including CoVs ($n = 1,067$), PMVs ($n = 1,067$), HTVs ($n = 1,067$), PGVs ($n = 469$) and HPVs ($n = 469$). A viral richness (S) of 22 virus genotypes in 46 positive samples from a total of 1,067 samples, 13 for CoVs, 2 for PMVs, 2 for HTVs, and 5 for PGVs, was obtained (Table S2). No HPVs and no co-infections were detected. This viral richness is associated with 17 bat species from 12 genera and 4 families (Fig. 1A). In forested habitats, a total of 11 viruses were detected, followed by fragmented habitats (10), disturbed habitats (8) and urban habitats (3) (Fig. 1B). The bat species harboring viral richness greater than one were all phyllostomid bats: *Carollia sowelli* ($S = 5$), *Artibeus lituratus* ($S = 4$), *Artibeus jamaicensis* ($S = 3$), *Artibeus phaeotis* ($S = 3$) and *Trachops cirrhosus* ($S = 2$) (Fig. S1).

Estimates and Completeness of Viral Richness

Based on the 17 virus genotypes discovered, we estimated a maximum richness of 23 genotypes using a Chao2 statistic model (Chao and Jost, 2012). The sampling effort of 1,067 samples represents a completeness of 81% in relation to the estimated viral richness. The rarefaction sample coverage function estimates 97% completeness with a sample size of 3,201 (three-fold sample size) (Fig. 2). The comparison between habitats showed the highest value of completeness (53%) in forested habitat, followed by disturbed (43%) and fragmented (15%) habitats. The estimates of viral richness with a three-fold original sample effort by habitat were 24 from fragmented habitats, 19 from forested habitats and 10 from disturbed habitats (Table 1).

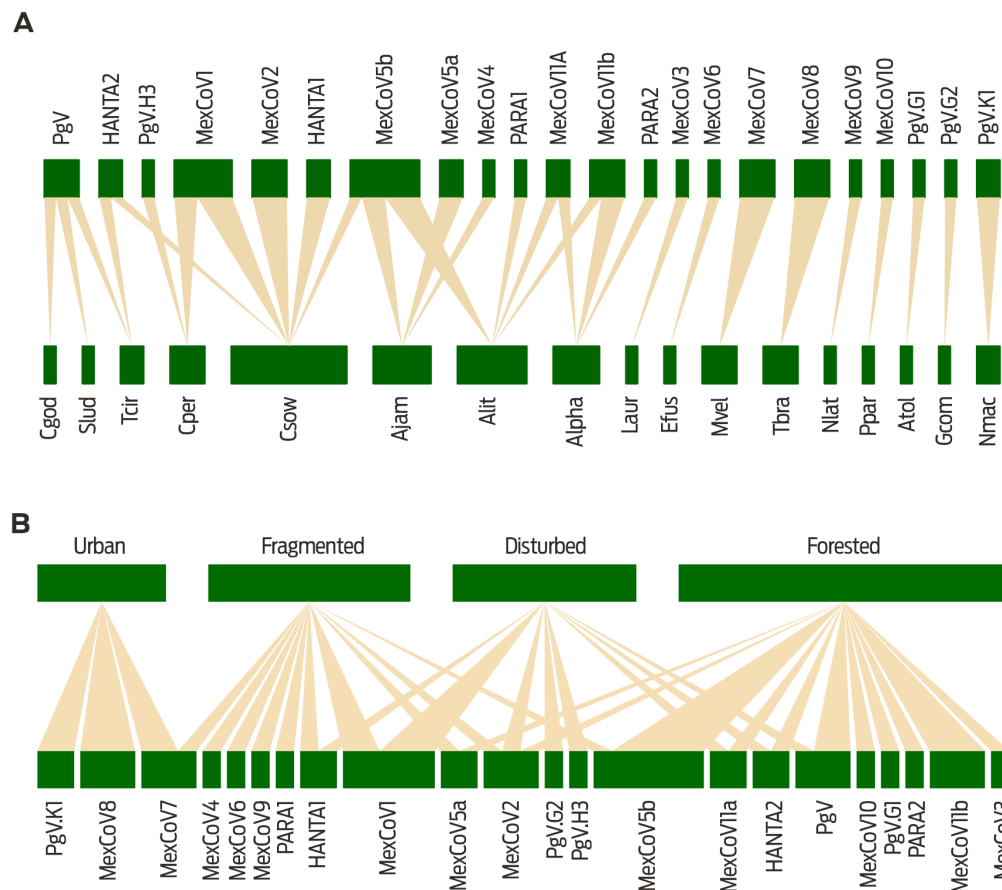


Figure 1. Bipartite graph of 22 virus genotypes discovered in 17 bat species. A. Viral richness associated with bats. The width of the green boxes represents viral and positive hosts abundances. B. Viral richness associated with habitat type.

Positive relationships were observed between sampling effort and host viral richness ($R^2 = 0.44$, $p < 0.01$) and between sampling effort and habitat type viral richness ($R^2 = 0.37$, $p < 0.05$). *Sturnira lilium*, *Pteronotus parnelli* and *Artibeus jamaicensis* were associated with greater viral richness than expected by the linear model between host viral richness and sampling effort, whereas *Trachops cirrhosus*, *Lonchorhina aurita* and *Eptesicus fuscus* (Fig. 3.A), were identified as host species associated with lower viral richness than expected. The RBMA fragmented habitat type harbored the highest number of viruses compared to the expected value, while the RBC fragmented habitat type was identified as the site with the smallest viral richness compared to the value expected by the model (Fig. 3.B).

Host and Viral Diversity

RBMA was the region with the most virus genotypes discovered (12), followed by RBC (11) and GMC (3) (Fig. 4). As we expected, RBMA was the most diverse region in terms of host species and host phylogeny, with Fd being the most diverse area in both diversity scales ($H = 2.79$, $PD = 484.8$). Interestingly, D presented high values of both species and phylogenetic diversities compared to F (Table 2). A

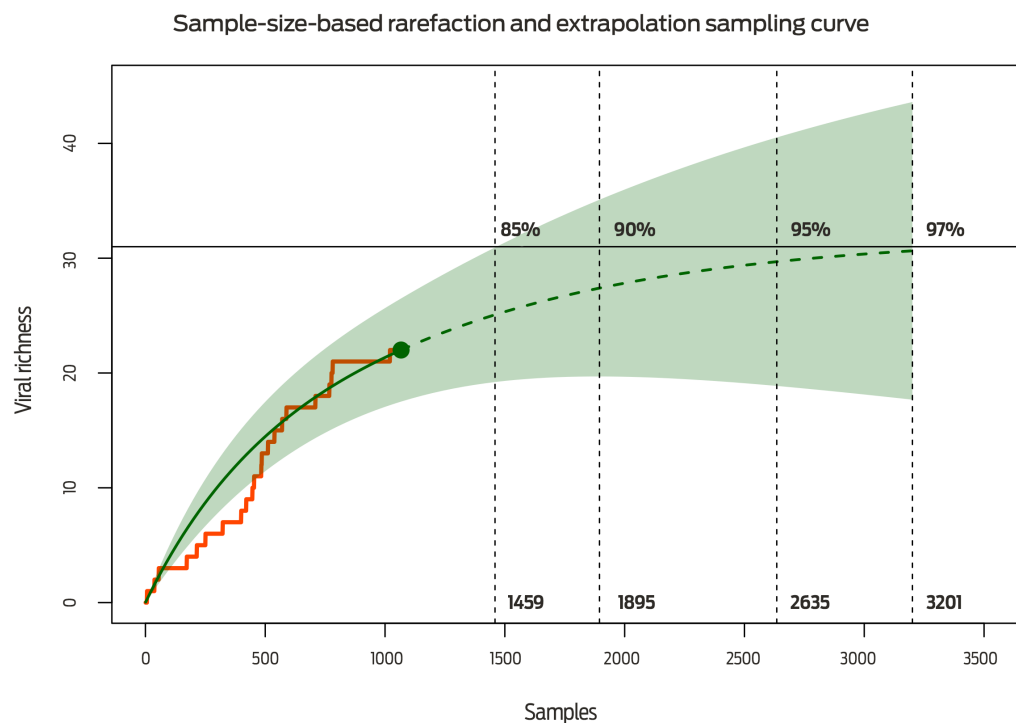


Figure 2. Rarefaction and extrapolation sampling curve based on 1,067 samples. Orange line represents accumulation curve of virus genotypes over samples tested. Green point, viral richness = 22 in 1,067 samples analyzed for virus tested. Solid green line: rarefaction curve, green dashed line: extrapolation-sampling curve. The numbers of samples needed to obtain the completeness percentages of 85, 90, 95 and 97% are presented.

difference in host species diversity ($F = 13.63$, $df = 2$, $p < 0.01$) and host phylogenetic diversity was observed ($F = 16.71$, $df = 2$, $p < 0.01$) at the regional scale, in contrast to viral diversity comparison ($F = 3.73$, $df = 2$, $p > 0.05$). A significant relationship of both viral richness and viral diversity with host phylogenetic diversity was observed. A total of 41% ($p < 0.05$) and 51% ($p < 0.05$) of viral richness and viral diversity variance was explained by phylogenetic diversity, respectively. This result suggests that host community composition at each habitat type determines both viral richness and viral diversity.

Phylogenetic Host Specificity

Of 22 viruses discovered, only six were associated with more than two host species, and 16 viruses were detected in one host species (Table S3). The flavivirus PgV was the virus with the lowest value of phylogenetic host specificity (114.5) and was detected in three species from three different Phyllostomidae subfamilies; Stenodermatinae and Phyllostominae, both from forested sites, and Glossophaginae, from disturbed habitats. Hanta 2 (87.8) was detected in two bat species from different habitats; *Carollia sowelli* (disturbed) and *Trachops cirrhosus* (forested), while the CoV MexCoV 5b was found in three bat species, two of which belong to the same genus; *Artibeus jamaicensis*, *A. lituratus* (forested) and *C. sowelli* (fragmented). MexCoV 11a (68.3), MexCoV 11b (68.3) and MexCoV 1 (65.6) were detected in two bat species from the same genus (Figure 1A).

Table 1. Estimates of viral richness and sample completeness by habitat type.

Habitat	Sample Size	Richness	Sampled Completeness
Total	1067	17	81%
	1677	18	95%
	3201	23	97%
Forested	405	9	53%
	1215	19	76%
	3201	27	95%
Fragmented	217	10	15%
	651	24	40%
	3201	48	94%
Disturbed	395	5	43%
	1185	10	97%
	3201	11	99%
Urban	49	2	100%

Beta Diversity

We found that 86% ($p < 0.05$) of the beta diversity of viral communities was explained by the turnover of host species between habitats. The same trend was observed with host PBD and virus beta diversity but was not statistically significant ($r = 0.79$, $p > 0.05$). For RBMA, the community dissimilarity in host species composition through the habitat gradient was relatively high ($\beta_{SOR} = 0.49$) and is explained by the nestedness component ($\beta_{SNE} = 0.57$), as most of the bat species sampled in low-richness sites are also contained in the richest site ($F_d = 26$ species). PBD showed different behavior; although the value of overall PBD is similar ($P\beta_{SOR} = 0.42$), the phylogenetic composition change was partially explained by the turnover component ($P\beta_{SIM} = 0.25$) due to the low phylogenetic relationship contained in the species subset replaced by nestedness. The overall beta diversity in RBC was relatively low ($\beta_{SOR} = 0.37$), suggesting that the habitat type has a low influence on the species assemblage. The observed pattern in RBC was different from RBMA, while species composition dissimilarity was moderately driven by species replacement ($P\beta_{SIM} = 0.22$). In contrast to RBMA, the PBD analysis indicated a replacement dominated by highly phylogenetically related species ($P\beta_{SNE} = 0.79$). The interpretation of the pattern observed in GMC is limited, as only two communities were sampled and the nestedness component is impossible to calculate. In RBMA and RBC, we observed high overall values of beta diversity in viral communities (Table S3), and both were explained by the turnover component ($\beta_{SIM} = 0.66$), suggesting that changes in habitat quality drive a high replacement of virus species regardless of host composition.

Discussion

In this study, we evaluated the relationship between bat diversity and the diversity of 4 medically important viral families within an environmental gradient in

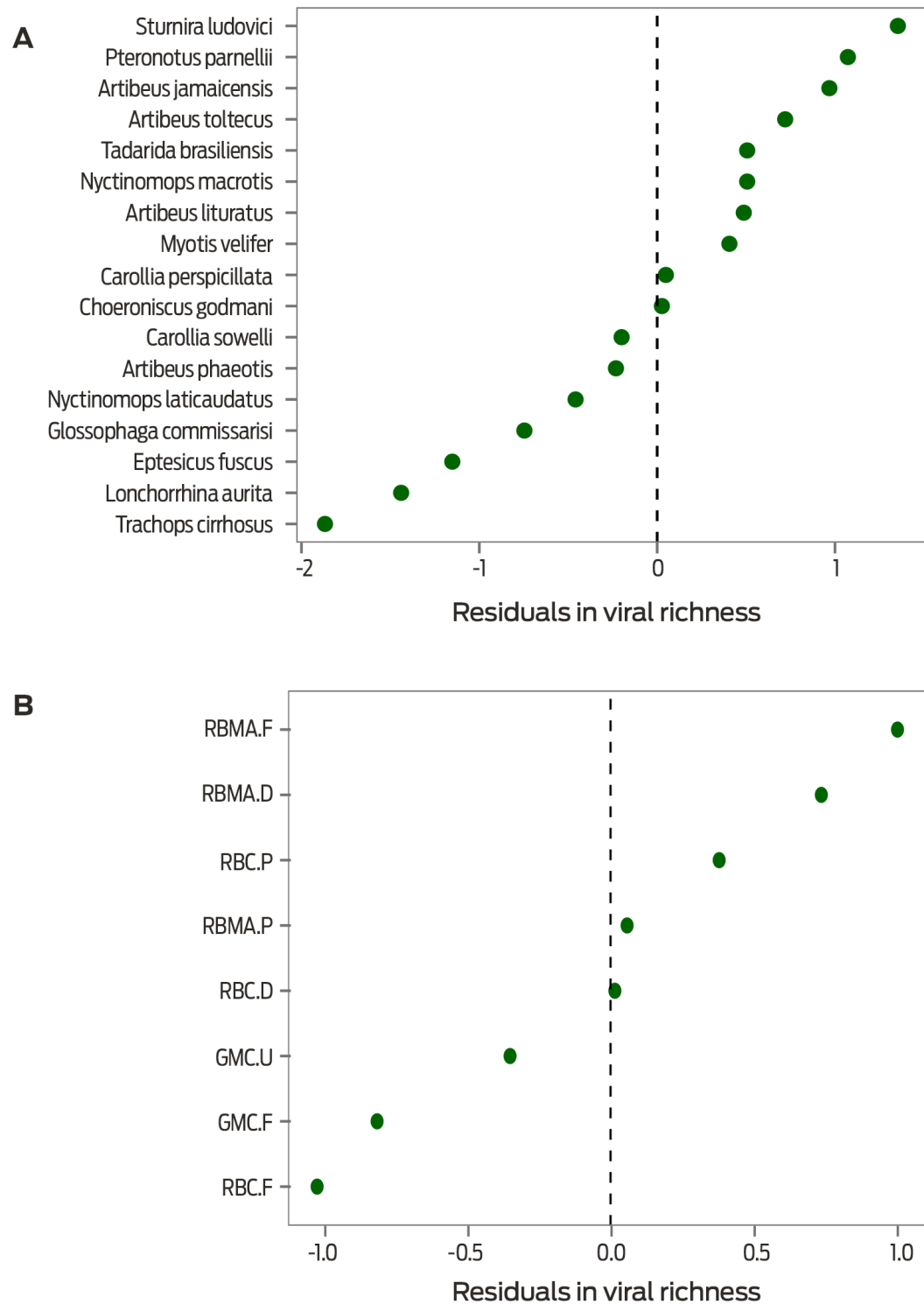


Figure 3. Distribution of residual values from the linear relations (A) between host viral richness and sampling effort and (B) between habitat type viral richness and sampling effort. Host species and habitat type were reordered by residuals of the sampling effort regression.

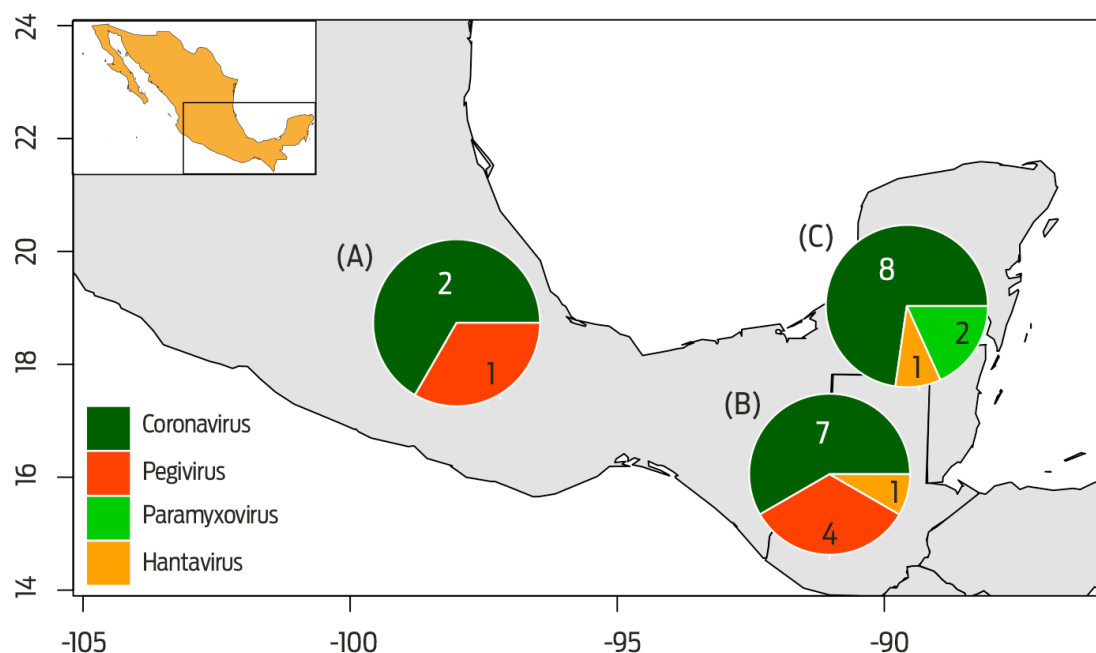


Figure 4. Viral richness in the three regions of study: (A) Greater Mexico City, (B) Montes Azules and (C) Calakmul. Number of virus genotypes discovered per region is shown.

human-dominated landscapes in southern Mexico. Combining computing applications and leading-edge molecular techniques for viral identification with ecological and phylogenetic analyses, we measured the viral community turnover in bat communities. A strong relationship of viral richness and viral diversity with host phylogenetic diversity was detected. Generalist species were associated with more viruses than expected, and a positive relationship of the beta diversity of both viral and bat communities with the habitat gradient were detected.

As hypothesized, significant positive correlation of phylogenetic diversity with viral richness and viral diversity was detected, supporting both the habitat heterogeneity hypothesis (Lawton, 1983), which proposes a strong relationship between environmental diversity (in this case the phylogenetic host diversity) and biological diversity (viral diversity), and the keystone structure hypothesis (Tews, 2004), which refers to “keystone structures” as host species that provide distinct resources, which may be linked to different viral species. Considering bat hosts as habitats, the keystone structures can be categorized in characteristics that relate to reproduction and abundance (reproductive potential, longevity, trophic guild, abundance and adult mass), transmission potential (home range, diet breadth and roost size) and phylogenetic relationships (phylogenetic distance and phylogenetic distinctiveness). Our findings suggest that factors such as fragmentation and habitat loss drive species assemblages, resulting in areas of greater risk for zoonotic disease emergence, as proposed by (Gay *et al.*, 2014; Kamiya *et al.*, 2014; Rubio *et al.*, 2014). Future studies are required to identify which host traits determine the viral community assemblages, but evidently greater viral diversity does not imply greater health risks; in fact, the correlation between viral and host diversity suggests that only under

Table 2. Host and viral diversities per habitat type for the 3 regions. Richness (S), Shannon – Weiner diversity index (H), phylogenetic diversity (PD).

Region	Habitat	Host			Virus	
		S	H	PD	S	H
Montes Azules	Forested	26	2.79	484.8	8	2.043
	Fragmented	14	2.207	332.8	2	0.693
	Disturbed	20	2.622	418.2	5	1.56
Calakmul	Forested	12	1.906	260.1	4	1.906
	Fragmented	8	1.853	208.8	7	1.853
	Disturbed	13	1.986	289.1	4	1.986
Greater Mexico City	Fragmented	1	0	60	1	0
	Urban	4	1.155	137.6	3	1.079

disturbed, deforested conditions can a pathogen have greater chances of becoming a health risk.

The phylogenetic host specificity calculated in this study not only reflects the number of bat species infected by a single virus but also helps to explore the phylogenetic relationships among these species. Our findings show that few viral species possess high host plasticity, such as PgV and HTV 2, while most viruses detected showed high phylogenetic specificity. Due to the quality of our data, we cannot conclude that viruses found in only one bat species are strictly exclusive to them, and further studies are needed. However, the coronavirus family showed high host phylogenetic specificity at the genus level, as shown in [Anthony *et al.*, 2013b](#)).

Changes in viral community composition through the evaluated anthropogenic landscapes showed a strong dependence on host species turnover; however, this relationship was not statistically significant when host PBD was considered. In general, different patterns between regions and between diversity levels were reported. For instance, RBMA is characterized by a nestedness process in host community composition, and changes reflect of a high PD host pool, while in RBC, the beta diversity values were relatively low due to low PD host diversity, and the PBD was explained by the nestedness component. As hypothesized, we found high values of beta diversity in viral communities, supporting the hypothesis of perturbation, where land use change modifies parasite dynamics in multihost systems by cross-species shifting in parasite transmission ([Murray and Daszak, 2013](#)). It has been widely proposed that habitat transformation drives the exposure of novel hosts to a rich pool of parasites, especially in high-diversity regions, influencing the cross-species transmission rate ([Lloyd-Smith *et al.*, 2009](#); [Brearley *et al.*, 2013](#); [Murray and Daszak, 2013](#)).

The use of beta analyses at both the taxonomic and phylogenetic scales has provided a useful tool to understand whether host species or environmental filters can determine parasite composition ([Svensson-Coelho and Ricklefs, 2011](#); [Scordato and Kardish, 2014](#)). We could not demonstrate that host phylogeny determines the composition of viral communities due to spatial scale limitations in our study. Further studies are necessary to test the correlation between host phylogeographi-

cal structure and the beta diversity of viral communities within larger spatial scales or in communities separated by geographic barriers (vicariance processes).

Conclusions

Our study showed that habitat quality and abiotic characteristics are important factors driving changes in viral diversity in human-dominated landscapes. This viral richness and viral diversity are both explained by host phylogenetic diversity. The host turnover component represents an unexplored dimension with a high potential for estimating viral diversity, especially in transformed landscapes. We integrated viral and host diversity analyses within an environmental gradient in human-dominated landscapes in vertebrates. This effort represents the first study in Mexico measuring viral community turnover in bats to monitor potential viral richness in wildlife and its associations with biodiversity, a necessary subject to understand how changes in ecosystem function and anthropogenic changes can promote disease emergence.

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Conflicts of interest

The authors declare that they have no conflicts of interest.

Author contributions

Oscar Rico Chávez and Rafeal Ojeda Flores: Contributed equally to this work as joint first authors.

Oscar Rico Chávez: Performed the fieldwork and ecological and phylogenetic analyses, and wrote the manuscript.

Rafael Ojeda Flores: Performed laboratory work and wrote the manuscript.

Jesús Sotomayor Bonilla: Performed fieldwork and reviewed the manuscript.

Carlos Zambrana Torrelío and Alonso Aguirre: Coordinated the project and reviewed the manuscript.

Elizabeth Loza Rubio: Reviewed the manuscript.

Gerardo Suzán: Conducted the research and wrote and reviewed the manuscript.

References

- 1) Anthony SJ, Epstein JH, Murray KA, Navarrete-Macias, I, Zambrana-Torrelío CM, Solovyov A, Ojeda-Flores R, Arrigo NC, Islam A, Ali Khan S, Hosseini P, Bogich TL, Olival KJ, Sanchez-Leon MD, Karesh WB, Goldstein T, Luby SP, Morse SS, Mazet JAK, Daszak P, Lipkin WI. 2013a. A Strategy to estimate unknown viral diversity in mammals. *mBio* 4(5):e00598-13. DOI:10.1128/mBio.00598-13. <http://mbio.asm.org/content/4/5/e00598-13.full>
- 2) Anthony SJ, Ojeda-Flores R, Rico-Chávez O, Navarrete-Macias I, Zambrana-Torrelío CM, Rostal MK, Epstein JH, Tipps T, Liang E, Sanchez-Leon M, Sotomayor-Bonilla J, Aguirre AA, Ávila-Flores R, Medellín RA, Goldstein T, Suzán G, Daszak P, Lipkin WI. 2013b. Coronaviruses in bats from Mexico. *The Journal of General Virology* 94: 1028–1038. DOI:10.1099/vir.0.049759-0. <http://www.ncbi.nlm.nih.gov/pmc/articles/PMC3709589/>
- 3) Banyard AC, Hayman D, Johnson N, McElhinney L, Fooks AR. 2011. Bats and lyssaviruses. *Advances in Virus Research* 79, 239–289. DOI:10.1016/B978-0-12-387040-7.00012-3. <http://www.sciencedirect.com/science/article/pii/B9780123870407000123>
- 4) Baselga A. 2010. Partitioning the turnover and nestedness components of beta diversity. *Global Ecology and Biogeography* 19(Issue 1):134–143. DOI: 10.1111/j.1466-8238.2009.00490.x. <http://onlinelibrary.wiley.com/doi/10.1111/j.1466-8238.2009.00490.x/full>
- 5) Baselga A, Orme C. 2013. Betapart: an R package for the study of beta diversity. *Methods in Ecology and Evolution* 3(Issue 5):808–812. DOI:10.1111/j.2041-210X.2012.00224.x. <http://onlinelibrary.wiley.com/doi/10.1111/j.2041-210X.2012.00224.x/full>
- 6) Bininda-Emonds ORP, Cardillo M, Jones KE, MacPhee RDE, Beck RMD, Grenyer R, Price SA, Vos RA, Gittleman JL, Purvis A. 2007. The delayed rise of present-day mammals. *Nature* 446:507–512. DOI:10.1038/nature07347. <http://www.nature.com/nature/journal/v446/n7135/full/nature05634.html>
- 7) Brearley G, Rhodes J, Bradley A, Baxter G, Seabrook L, Lunney D, Liu Y, McAlpine C. 2013. Wildlife disease prevalence in human-modified landscapes. *Biological Reviews of the Cambridge Philosophical Society* 88(Issue 2):427–442. DOI:10.1111/brv.12009. <http://onlinelibrary.wiley.com/doi/10.1111/brv.12009/full>
- 8) Bryant JA, Lamanna C, Morlon H, Kerkhoff AJ, Enquist BJ, Green JL. 2008. Microbes on mountainsides: contrasting elevational patterns of bacterial and plant diversity. *Proceedings of the National Academy of Sciences* 105(Suppl 1):11505–11511. DOI:10.1073/pnas.0801920105. <http://www.pnas.org/content/111/20/7166.full.pdf+html>
- 9) Calderón-Patrón JM, Moreno CE, Zuria I. 2012. La diversidad beta: medio siglo de avances. *Revista Mexicana de Biodiversidad* 83:879–891. DOI:10.7550/rmb.25510. http://www.scielo.org.mx/scielo.php?script=sci_arttext&pid=S1870-34532012000300034

- 10) Chao A, Gotelli NJ, Hsieh TC, Sander EL, Ma KH, Colwell RK, Ellison AM. 2014. Rarefaction and extrapolation with Hill numbers : a framework for sampling and estimation in species diversity studies. *Ecological Monographs* 84(Issue 1):45–67. DOI:10.1890/13-0133.1. <http://www.esajournals.org/doi/abs/10.1890/13-0133.1>
- 11) Chao A, Jost L. 2012. Coverage-based rarefaction and extrapolation: standardizing samples by completeness rather than size. *Ecology* 93 (Issue 12):2533–2547. DOI:10.1890/11-1952.1. <http://www.esajournals.org/doi/abs/10.1890/11-1952.1>
- 12) Christian K, Isabelle LV, Frédéric J, Vincent D. 2009. More species, fewer specialists: 100 years of changes in community composition in an island biogeographical study. *Diversity and Distributions* 15 (Issue 4):641–648. DOI:10.1111/j.1472-4642.2009.00569.x. <http://onlinelibrary.wiley.com/doi/10.1111/j.1472-4642.2009.00569.x/full>
- 13) Epstein JH, Field HE, Luby S, Pulliam JRC, Daszak P. 2006. Nipah virus: impact, origins, and causes of emergence. *Current Infectious Disease Reports* 8:59–65. DOI:10.1007/s11908-006-0036-2. <http://link.springer.com/article/10.1007%2Fs11908-006-0036-2#page-1>
- 14) Faith DP. 1992. Conservation evaluation and phylogenetic diversity. *Biological Conservation* 61(1):1–10. DOI:10.1016/0006-3207(92)91201-3. http://www.planta.cn/forum/files_planta/1_119.pdf
- 15) Gay N, Olival KJ, Bumrungsri S, Siriaronrat B, Bourgarel M, Morand S. 2014. Parasite and viral species richness of Southeast Asian bats: fragmentation of area distribution matters. *International Journal for Parasitology. Parasites and Wildlife* 3(2):161–170. DOI:10.1016/j.ijppaw.2014.06.003. <http://www.ncbi.nlm.nih.gov/pmc/articles/PMC4142259/>
- 16) Gibbs KE, MacKey RL, Currie DJ. 2009. Human land use, agriculture, pesticides and losses of imperiled species. *Diversity and Distributions* 15 (Issue 2):242–253. DOI:10.1111/j.1472-4642.2008.00543.x. <http://onlinelibrary.wiley.com/doi/10.1111/j.1472-4642.2008.00543.x/full>
- 17) Graham CH, Fine PVA. 2008. Phylogenetic beta diversity: linking ecological and evolutionary processes across space in time. *Ecology letters* 11(12):1265–1277. DOI:10.1111/j.1461-0248.2008.01256.x. <http://onlinelibrary.wiley.com/doi/10.1111/j.1461-0248.2008.01256.x/pdf>
- 18) Hawley DM, Altizer SM. 2011. Disease ecology meets ecological immunology: understanding the links between organismal immunity and infection dynamics in natural populations. *Functional Ecology* 25(Issue 1):48–60. DOI:10.1111/j.1365-2435.2010.01753.x. <http://onlinelibrary.wiley.com/doi/10.1111/j.1365-2435.2010.01753.x/full>
- 19) Helmus MR, Savage K, Diebel MW, Maxted JT, Ives AR. 2007. Separating the determinants of phylogenetic community structure. *Ecology letters* 10(Issue 10):917–925. DOI:10.1111/j.1461-0248.2007.01083.x. <http://onlinelibrary.wiley.com/doi/10.1111/j.1461-0248.2007.01083.x/full>
- 20) Herbreteau V, Bordes F, Jittapalpong S, Supputamongkol Y, Morand S. 2012. Rodent-borne diseases in Thailand: targeting rodent carriers and risky habitats. *Infection Ecology and Epidemiology* 2:18637. DOI:10.3402/iee.v2i0.18637. <http://dx.doi.org/10.3402/iee.v2i0.18637>

- 21) Hilgenfeld R, Peiris M. 2013. From SARS to MERS: 10 years of research on highly pathogenic human coronaviruses. *Antiviral Research* 100(Issue 1):286–295. DOI:10.1016/j.antiviral.2013.08.015. <http://www.sciencedirect.com/science/article/pii/S0166354213002234>
- 22) Hsieh TC, Ma KH, Chao A. 2013. iNEXT: An R package for interpolation and extrapolation in measuring species diversity [unpublished manuscript].
- 23) Jones BA, Grace D, Kock R, Alonso S, Rushton J, Said MY, McKeever D, Mutua F, Young J, McDermott J, Pfeiffer DU. 2013. Zoonosis emergence linked to agricultural intensification and environmental change. *Proceedings of the National Academy of Sciences of the United States of America* 110(21):8399–8404. <http://www.pnas.org/content/110/21/8399.full>
- 24) Kamiya T, O'Dwyer K, Nakagawa S, Poulin R. 2014. Host diversity drives parasite diversity: meta-analytical insights into patterns and causal mechanisms. *Ecography* 37(Issue 7):689–697. DOI:10.1111/j.1600-0587.2013.00571.x. <http://onlinelibrary.wiley.com/doi/10.1111/j.1600-0587.2013.00571.x/full>
- 25) Kembel SW, Cowan PD, Helmus MR, Cornwell WK, Morlon H, Ackerly DD, Blomberg SP, Webb CO. 2010. Picante: R tools for integrating phylogenies and ecology. *Bioinformatics* 26(11):1463–1464. DOI:10.1093/bioinformatics/btq166. <http://bioinformatics.oxfordjournals.org/content/26/11/1463.long>
- 26) Klempa B, Fichet-Calvet E, Lecompte E, Auste B, Aniskin V, Meisel H, Denys C, Koivogui L, ter Meulen J, Krüger DH. 2006. Hantavirus in African wood mouse, Guinea. *Emerging Infectious Diseases* 12(5):838–840. DOI:10.3201/eid1205.051487. <http://www.ncbi.nlm.nih.gov/pmc/articles/PMC3374458/>
- 27) Kunz TH, Braun de Torrez E, Bauer DM, Lobova TA, Fleming TH. 2011. Ecosystem services provided by bats. *Annals of the New York Academy of Sciences*, 1223:1-38. Issue: The Year in Ecology and Conservation. DOI: 10.1111/j.1749-6632.2011.06004.x. <http://onlinelibrary.wiley.com/doi/10.1111/j.1749-6632.2011.06004.x/pdf>
- 28) Lawton JH. 1983. Plant architecture and the diversity of phytophagous insects. *Annual Review of Entomology* 28:23–39. DOI:10.1146/annurev.en.28.010183.000323.
- 29) Legendre P, Desdevises Y, Bazin E. 2002. A statistical test for host-parasite coevolution. *Systematic Biology* 51(2):217–234. DOI:10.1080/10635150252899734. <http://sysbio.oxfordjournals.org/content/51/2/217.full.pdf+html>
- 30) Leprieur F, Albouy C, De Bortoli J, Cowman PF, Bellwood DR, Mouillot D. 2012. Quantifying phylogenetic beta diversity: distinguishing between "true" turnover of lineages and phylogenetic diversity gradients. *Plos One* 7(8):e42760. DOI: 10.1371/journal.pone.0042760. <http://journals.plos.org/plosone/article?id=10.1371/journal.pone.0042760>
- 31) Leroy EM, Kumulungui B, Pourrut X, Rouquet P, Hassanin A, Yaba P, Délicat A, Paweska JT, Gonzalez JP, Swanepoel R. 2005. Fruit bats as reservoirs of Ebola virus. *Nature* 438:575–576. DOI:10.1038/438575a. <http://www.nature.com/nature/journal/v438/n7068/full/438575a.html>
- 32) Lloyd-Smith JO, George D, Pepin KM, Pitzer VE, Pulliam JRC, Dobson AP, Hudson PJ, Grenfell BT. 2009. Epidemic dynamics at the human-animal interface. *Science* 326(5958):1362–1367. DOI:10.1126/science.1177345. <http://www.ncbi.nlm.nih.gov/pmc/articles/PMC3891603/>

- 33) Magurran AE. 2004. *Measuring Biological Diversity*. Oxford, UK: Blackwell Science Ltd.
- 34) McMichael AJ. 2004. Environmental and social influences on emerging infectious diseases: past, present and future. *Philosophical Transactions of the Royal Society of London. Series B, Biological Sciences* 359:1049–1058. DOI:10.1098/rstb.2004.1480. <http://rstb.royalsocietypublishing.org/content/royptb/359/1447/1049.full.pdf>
- 35) Medellín, RA. 2009. Sustaining transboundary ecosystem services provided by bats. In: López-Hoffman L, McGovern E, Varady R, Flessa K. (eds.) *Conservation of Shared Environments: Learning from the United States and Mexico*. Tucson, USA: University of Arizona Press. pp. 170–187.
- 36) Medellín R, Arita HT, Sánchez HO. 2008. *Identificación de los murciélagos de México. Clave de campo*. DF, México: Instituto de Ecología, UNAM-CONABIO.
- 37) Medellín R, Equihua M, Amin M. 2000. Bat diversity and abundance as indicators of disturbance in neotropical rainforests. *Conservation Biology* 14(Issue 6):1666–1675. DOI:10.1046/j.1523-1739.2000.99068.x. <http://onlinelibrary.wiley.com/doi/10.1111/j.1523-1739.2000.99068.x/full>
- 38) Murray KA, Daszak P. 2013. Human ecology in pathogenic landscapes: two hypotheses on how land use change drives viral emergence. *Current Opinion in Virology* 3(1):79–83. DOI:10.1016/j.coviro.2013.01.006. <http://www.ncbi.nlm.nih.gov/pmc/articles/PMC3713401/>
- 39) Oksanen J, Blanchet GF, Kindt R, Legendre P, Minchin PR, O'Hara RB, Simpson GL, Solymos P, Stevens MHH, Wagner H. 2013. *Vegan: Community Ecology Package. R package version 2.0-10* <http://cran.r-project.org/web/packages/vegan/index.html> [access: November 4th, 2014].
- 40) Olson SH, Parmley J, Soos C, Gilbert M, Latorre-Margalef N, Hall JS, Hansbro PM, Leighton F, Munster V, Joly D. 2014. Sampling strategies and biodiversity of influenza A subtypes in wild birds. *PLoS ONE* 9(3):e90826. DOI:10.1371/journal.pone.0090826. <http://journals.plos.org/plosone/article?id=10.1371/journal.pone.0090826>
- 41) Patz JA, Daszak P, Tabor GM, Aguirre AA, Pearl M, Epstein J, Wolfe ND, Kilpatrick AM, Foufopoulos J, Molyneux D, Bradley DJ, Amerasinghe FP, Ashford RW, Barthelemy D, Bos R, Bradley DJ, Buck A, Butler C, Chivian ES, Chua KB, Clark G, Colwell R, Confalonieri UE, Corvalan C, Cunningham AA, Dein J, Dobson AP, Else JG, Epstein J, Field H, Furu P, Gascon C, Graham D, Haines A, Hyatt AD, Jamaluddin A, Kleinau EF, Koontz F, Koren HS, LeBlancq S, Lele S, Lindsay S, Maynard N, McLean RG, McMichael T, Molyneux D, Morse SS, Norris DE, Ostfeld RS, Pearl MC, Pimentel D, Rakototiana L, Randriamanajara O, Riach J, Rosenthal JP, Salazar-Sanchez E, Silbergeld E, Thomson M, Vittor AY, Yameogo L, Zakarov V. 2004. Unhealthy landscapes: policy recommendations on land use change and infectious disease emergence. *Environmental Health Perspectives* 112(10):1092–1098. DOI:10.1289/ehp.6877. <http://www.ncbi.nlm.nih.gov/pmc/articles/PMC1247383/pdf/ehp0112-001092.pdf>
- 42) Poulin R, Krasnov BR, Mouillot D. 2011. Host specificity in phylogenetic and geographic space. *Trends in Parasitology* 27(Issue 8):355–361. DOI:10.1016/j.pt.2011.05.003. <http://www.sciencedirect.com/science/article/pii/S147149221100095X>

- 43) Poulin R, Mouillot D. 2003. Parasite specialization from a phylogenetic perspective: a new index of host specificity. *Parasitology* 126(Issue 5):473–480. DOI:10.1017/S0031182003002993. http://journals.cambridge.org/download.php?file=%2FPAR%2FPAR126_05%2FS0031182003002993a.pdf&code=919662fc57bed7954f7b01a59f9543
- 44) Quan PL, Firth C, Conte JM, Williams SH, Zambrana-Torrel CM, Anthony SJ, Ellison JA, Gilbert AT, Kuzmin IV, Niezgoda M, Osinubi MOV, Recuenco S, Markotter W, Breiman RF, Kalemba L, Malekani J, Lindblade KA, Rostal MK, Ojeda-Flores R, Suzan G, Davis LB, Blau DM, Ogunkoya AB, Alvarez Castillo DA, Moran D, Ngam S, Akaibe D, Agwanda B, Briesse T, Epstein JH, Daszak P, Rupprecht CE, Holmes EC, Lipkin WI. 2013. Bats are a major natural reservoir for hepaciviruses and pegiviruses. *Proceedings of the National Academy of Sciences of the United States of America* 110:8194–8199. DOI:10.1098/rspb.2014.2124. <http://dx.doi.org/10.1098/rspb.2014.2124>
- 45) Rivard DH, Poitevin J, Plasse D, Carleton M, Currie DJ, Ward DH, Curriet DJ. 2000. Changing species richness and composition in canadian national parks 14(issue 4):1099–1109. DOI:10.1046/j.1523-1739.2000.98247.x. <http://onlinelibrary.wiley.com/doi/10.1046/j.1523-1739.2000.98247.x/pdf>
- 46) Rubio AV, Avila-Flores R, Suzán G. 2014. Responses of small mammals to habitat fragmentation: epidemiological considerations for rodent-borne hantaviruses in the Americas. *EcoHealth*. DOI:10.1007/s10393-014-0944-9. <http://dx.doi.org/10.1007/s10393-014-0944-9>
- 47) Scordato ESC, Kardish MR. 2014. Prevalence and beta diversity in avian malaria communities: host species is a better predictor than geography. *The Journal of Animal Ecology* 83(issue 6):1387–1397. DOI: 10.1111/1365-2656.12246. <http://onlinelibrary.wiley.com/doi/10.1111/1365-2656.12246/full>
- 48) Shannon CE. 1948. A mathematical theory of communication. *System Technical Journal*, 379–423.
- 49) Smith I, Broos A, de Jong C, Zeddeman A, Smith C, Smith G, Moore F, Barr J, Crameri G, Marsh G, Tachedjian M, Yu M, Kung YH, Wang LF, Field H. 2011. Identifying hendra virus diversity in pteropid bats. *PLoS ONE*, 6(9):e25275. DOI:10.1371/journal.pone.0025275. <http://journals.plos.org/plosone/article?id=10.1371/journal.pone.0025275>
- 50) Streicker DG, Turmelle AS, Vonhof MJ, Kuzmin IV, McCracken GF, Rupprecht CE. 2010. Host phylogeny constrains cross-species emergence and establishment of rabies virus in bats. *Science* 329(5993):676–679. New York, NY, USA. DOI:10.1126/science.1188836. <http://www.ncbi.nlm.nih.gov/pubmed/20689015>
- 51) Suzán G, Marcé E, Giermakowski JT, Mills JN, Ceballos G, Ostfeld RS, Armién B, Pascale JM, Yates TL. 2009. Experimental evidence for reduced rodent diversity causing increased hantavirus prevalence. *PLoS ONE* 4(5):e5461. DOI:10.1371/journal.pone.0005461. <http://journals.plos.org/plosone/article?id=10.1371/journal.pone.0005461>
- 52) Svensson-Coelho M, Ricklefs RE. 2011. Host phylogeography and beta diversity in avian haemosporidian (*Plasmodiidae*) assemblages of the Lesser Antilles. *Journal of Animal Ecology* 80(issue 5):938–946. DOI:10.1111/j.1365-2656.2011.01837.x. <http://onlinelibrary.wiley.com/doi/10.1111/j.1365-2656.2011.01837.x/full>

- 53) Tews J, Brose U, Grimm V, Tielbörger K, Wichmann MC, Schwager M, Jeltsch F. 2004. Animal species diversity driven by habitat heterogeneity/diversity: the importance of keystone structures. *Journal of Biogeography* 31(1):79-92. DOI: 10.1046/j.0305-0270.2003.00994.X. <http://onlinelibrary.wiley.com/doi/10.1046/j.0305-0270.2003.00994.x/full>
- 54) Tong S, Chern SWW, Li Y, Pallansch MA, Anderson LJ. 2008. Sensitive and broadly reactive reverse transcription-PCR assays to detect novel paramyxoviruses. *Journal of Clinical Microbiology* 46(8):2652–2658. DOI:10.1128/JCM.00192-08. <http://jcm.asm.org/content/46/8/2652.full>
- 55) Towner JS, Amman BR, Sealy TK, Reeder Carroll SA, Comer JA, Kemp A, Swanepoel R, Paddock CD, Balinandi S, Khristova ML, Formenty PBH, Albarino CG, Miller DM, Reed ZD, Kayiwa JT, Mills JN, Cannon DL, Greer PW, Byaruhanga E, Farnon EC, Atimnedi P, Okware S, Katongole-Mbidde E, Downing R, Tappero JW, Zaki SR, Ksiazek TG, Nichol ST, Rollin PE (2009). Isolation of genetically diverse Marburg viruses from Egyptian fruit bats. *PLoS Pathog* 5(7):e1000536. DOI:10.1371/journal.ppat.1000536. <http://journals.plos.org/plospathogens/article?id=10.1371/journal.ppat.1000536>
- 56) Ulrich DH, Almeida-Neto M, Gotelli NJ. 2009. A consumer's guide to nestedness analysis. *Oikos* 118(Issue 1):3–17. DOI:10.1111/j.1600-0706.2008.17053.x. <http://onlinelibrary.wiley.com/doi/10.1111/j.1600-0706.2008.17053.x/full>
- 57) Webb CO, Ackerly DD, McPeck MA, Donoghue MJ. 2002. Phylogenies and community ecology. *Annual Review of Ecology and Systematics* 33:475–505. DOI:10.1146/annurev.ecolsys.33.010802.150448. <http://www.annualreviews.org/doi/full/10.1146/annurev.ecolsys.33.010802.150448>

Table S1. Number of samples and bats per disturbance level by region.

Region	Disturbance level	No. of Samples	Bats captured
Montes Azules	Forested	253	127
	Fragmented	142	79
	Disturbed	293	134
Calakmul	Forested	152	129
	Fragmented	65	38
	Disturbed	102	71
Greater Mexico City	Fragmented	10	5
	Urban	49	25
Total		1067	608

Supplementary material

Table S2. Virus discovered in bat species from three regions.
Virus, name of unique virus, *N*, number of virus per bat species.

Region	Habitat	Sample	Species	Virus	N
Montes Azules	Forested	Oral	<i>Carollia sowelli</i>	Mex Cov 2	1
	Forested	Oral	<i>Artibeus phaeotis</i>	Mex Cov 11a	1
	Forested	Oral	<i>Artibeus phaeotis</i>	Mex Cov 11b	1
	Forested	Oral	<i>Trachops cirrhosus</i>	HANTA 2	1
	Forested	Rectal	<i>Lonchorhina aurita</i>	Mex CoV 3	1
	Forested	Rectal	<i>Artibeus jamaicensis</i>	Mex CoV 5a	1
	Forested	Blood	<i>Trachops cirrhosus</i>	PgV 1	1
	Forested	Blood	<i>Sturnira ludovici</i>	PgV 1	1
	Forested	Blood	<i>Artibeus toltecus</i>	PgV.G1	1
	Fragmented	Rectal	<i>Artibeus jamaicensis</i>	Mex CoV 5a	1
	Fragmented	Blood	<i>Eptesicus fuscus</i>	Mex CoV 6	1
	Disturbed	Oral	<i>Carollia sowelli</i>	Mex CoV 1	1
	Disturbed	Oral	<i>Carollia sowelli</i>	HANTA 2	1
	Disturbed	Rectal	<i>Carollia sowelli</i>	Mex CoV 1	1
	Disturbed	Blood	<i>Choeroniscus godmani</i>	PgV	1
	Disturbed	Blood	<i>Glossophaga commissarisi</i>	PgV.G2	1
	Disturbed	Blood	<i>Carollia perspicillata</i>	PgV.H3	1
Calakmul	Forested	Rectal	<i>Artibeus jamaicensis</i>	Mex CoV 5b	2
	Forested	Rectal	<i>Artibeus lituratus</i>	Mex CoV 5b	3
	Forested	Rectal	<i>Artibeus lituratus</i>	Mex CoV 11b	1
	Forested	Rectal	<i>Artibeus phaeotis</i>	Mex CoV 11b	1
	Forested	Rectal	<i>Artibeus phaeotis</i>	PMV 2	1
	Forested	Rectal	<i>Pteronotus parnellii</i>	Mex CoV 10	1
	Fragmented	Rectal	<i>Carollia perspicillata</i>	Mex CoV 1	1
	Fragmented	Rectal	<i>Carollia sowelli</i>	Mex CoV 1	1
	Fragmented	Rectal	<i>Carollia sowelli</i>	Mex CoV 2	1
	Fragmented	Rectal	<i>Carollia sowelli</i>	Mex CoV 5b	1
	Fragmented	Rectal	<i>Carollia sowelli</i>	HANTA 1	1
	Fragmented	Rectal	<i>Artibeus jamaicensis</i>	Mex CoV 4	1
	Fragmented	Rectal	<i>Artibeus lituratus</i>	PMV 1	1
	Fragmented	Rectal	<i>Nyctinomops laticaudatus</i>	Mex CoV 9	1
	Disturbed	Rectal	<i>Carollia perspicillata</i>	Mex CoV 1	1
	Disturbed	Rectal	<i>Carollia sowelli</i>	Mex CoV 2	1
	Disturbed	Rectal	<i>Artibeus lituratus</i>	Mex CoV 11a	1
	Disturbed	Blood	<i>Carollia sowelli</i>	HANTA 1	1
Greater Mexico City	Fragmented	Rectal	<i>Myotis velifer</i>	Mex CoV 7	1
	Urban	Rectal	<i>Myotis velifer</i>	Mex Cov 7	2
	Urban	Rectal	<i>Tadarida brasiliensis</i>	Mex CoV 8	3
	Urban	Rectal	<i>Nyctinomops macrotis</i>	PgV.K1	1
	Urban	Blood	<i>Nyctinomops macrotis</i>	PgV.K1	1

Supplementary material

Table S3. Host specificity index (HSI) for each virus measured as the phylogenetic diversity of host community associated to each virus. (S) = Host richness.

Virus	HSI	S
PgV	114.5	3
HANTA2	87.8	2
MexCoV5b	85.8	3
MexCoV11a	68.3	2
MexCoV11b	68.3	2
MexCoV1	65.6	2
MexCoV2	60	1
MexCoV3	60	1
MexCoV4	60	1
MexCoV5a	60	1
MexCoV6	60	1
MexCoV7	60	1
MexCoV8	60	1
MexCoV9	60	1
MexCoV10	60	1
PgV.G1	60	1
PgV.G2	60	1
PgV.H3	60	1
PgV.K1	60	1
HANTA1	60	1
PARA1	60	1
PARA2	60	1

Table S4. Multiple site dissimilarities accounting for the spatial turnover and the nestedness components of beta diversity and phylogenetic beta diversity. β_{SNE} , nestedness component, β_{SIM} , turnover component, β_{SOR} , value of overall beta diversity. $P\beta_{SNE}$, phylogenetic nestedness component, $P\beta_{SIM}$, phylogenetic turnover, $P\beta_{SOR}$, dissimilarity value accounting for phylogenetic beta diversity.

Region	Host						Virus		
	β_{SNE}	β_{SIM}	β_{SOR}	$P\beta_{SNE}$	$P\beta_{SIM}$	$P\beta_{SOR}$	β_{SNE}	β_{SIM}	β_{SOR}
Total	0.08	0.70	0.79	0.06	0.65	0.72	0.08	0.70	0.78
Montes Azules	0.57	0.33	0.49	0.17	0.25	0.42	0.13	0.66	0.80
Calakmul	0.14	0.22	0.37	0.79	0.36	0.44	0.06	0.66	0.73
Greater Mexico City	0.60	0	0.60	0.45	0	0.45	0.50	0	0.50

Supplementary material

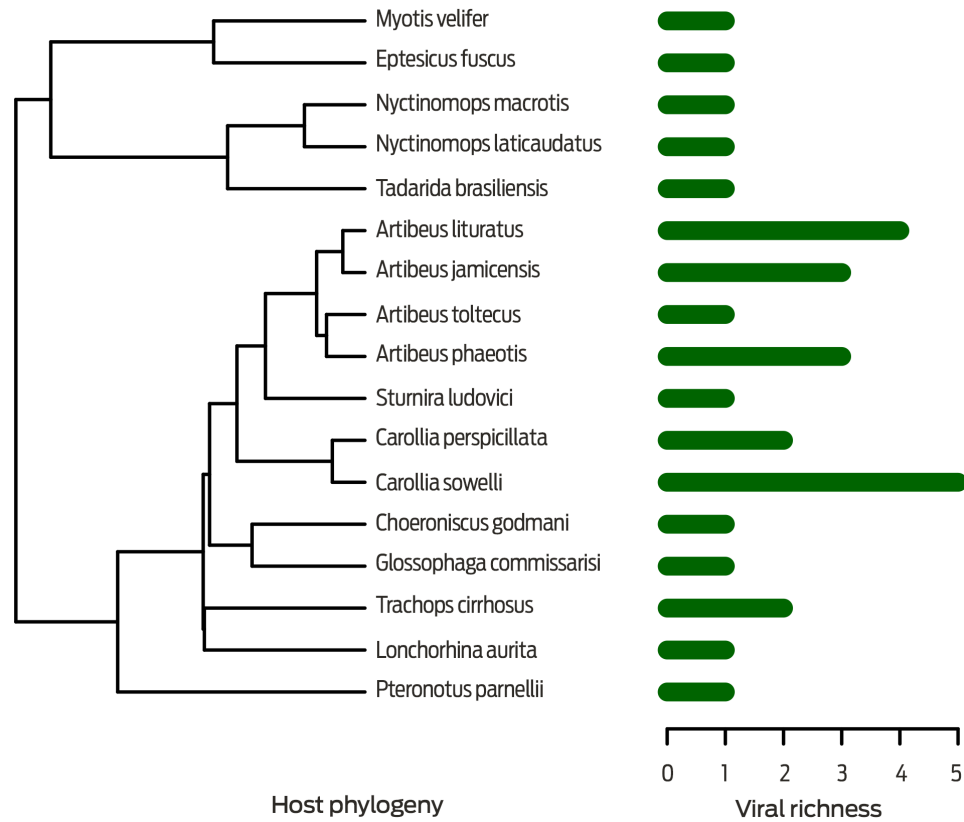


Figure S1. Host phylogeny and viral richness.