A systematic review and meta-analysis of the potential non-human animal reservoirs and arthropod vectors of the Mayaro virus

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Abstract

Improving our understanding of Mayaro virus (MAYV) ecology is critical to guide surveillance and risk assessment. We conducted a PRISMA-adherent systematic review of the published and grey literature to identify potential arthropod vectors and non-human animal reservoirs of MAYV. We searched PubMed, Embase, Web of Science, SciELO and grey-literature sources including PAHO databases and dissertation repositories. Studies were included if they assessed MAYV virological/immunological measured occurrence in field-caught, domestic, or sentinel animals or in field-caught arthropods. We conducted an animal seroprevalence meta-analysis using a random effects model. We compiled granular georeferenced maps of non-human MAYV occurrence and graded the quality of the studies using a customized framework. Overall, 57 studies were eligible out of 1523 screened, published between the years 1961 and 2020. Seventeen studies reported MAYV positivity in wild mammals, birds, or reptiles and five studies reported MAYV positivity in domestic animals. MAYV positivity was reported in 12 orders of wild-caught vertebrates, most frequently in the orders Charadriiformes and Primate. Sixteen studies detected MAYV in wild-caught mosquito genera including *Haemagogus, Aedes, Culex, Psorophora, Coquillettidia,* and *Sabethes*. Vertebrate animals or arthropods with MAYV were detected in Brazil, Panama, Peru, French Guiana, Colombia, Trinidad, Venezuela, Argentina, and Paraguay. Among non-human vertebrates, the Primate order had the highest pooled prevalence (PP) at 13.1% (95% CI: 4.3-25.1%). From the three most studied primate genera we found the highest prevalence was in *Alouatta* (PP: 32.2%, 95% CI: 0.0-79.2%), followed by *Callithrix* (PP: 17.8%, 95% CI: 8.6-28.5%), and *Cebus/Sapajus* (PP: 3.7%, 95% CI: 0.0-11.1%). We further found that MAYV occurs in a wide range of vectors beyond *Haemagogus* spp. The quality of evidence behind these findings was variable and prompts calls for standardization of reporting of arbovirus occurrence. These findings support further risk emergence prediction, guide field surveillance efforts, and prompt further *in-vivo* studies to better define the ecological drivers of MAYV maintenance and potential for emergence.

Author Summary

Mayaro virus (MAYV) is an emerging tropical public health threat in the Americas. We conducted a georeferenced, quality-graded systematic review to evaluate the current evidence regarding MAYV occurrence in non-human vertebrates and arthropods. Overall, 57 studies were eligible out of 1523 screened, published between the years 1961 and 2020. Seventeen studies reported MAYV positivity in wild mammals, birds, or reptiles and five studies reported MAYV positivity in domestic animals. MAYV positivity was reported in 12 orders of wild-caught vertebrates, most frequently in the orders Charadriiformes and Primate. Our systematic review identified 12 orders of wild-caught vertebrates and seven mosquito genera with evidence of MAYV occurrence. Primates had the highest pooled MAYV prevalence according to a seroprevalence meta-analysis. The graded quality of evidence behind these findings was variable and prompts calls for standardization of reporting of MAYV and perhaps other emerging arbovirus occurrence in animals and vectors. This study provides important information for public health authorities and disease ecologists concerned with the growing threat of MAYV in Latin America. Our analysis provides a foundation for future laboratory and field studies focused on the MAYV transmission cycle.

Introduction

First detected in Trinidad in 1954 [1], Mayaro virus (MAYV) is a zoonotic *Alphavirus* that is endemic in several Latin American countries. Like Chikungunya virus (CHIKV), MAYV may cause complications such as debilitating arthralgia but often presents with a non-specific constellation of symptoms and signs that may be clinically indistinguishable from other vector borne diseases such as dengue or Zika [2]. There is no current licensed vaccine or antiviral treatment for MAYV infections, and the current standard of clinical treatment is supportive care only [2, 3].

MAYV has caused periodic outbreaks in humans in Brazil [4, 5], Bolivia [6], and Venezuela [7], while surveillance studies and serological surveys have detected MAYV in humans in several countries throughout the Americas including Peru [8], Suriname [9], Mexico [10], Colombia [11], French Guiana [12], and Haiti [13]. These findings demonstrate widespread circulation of the virus throughout the region. A recent 2019 epidemiological alert by the Pan American Health Association (PAHO) has emphasized the need for increased awareness of and extended surveillance for this emerging virus in the Americas [3]. However, the precise areas of risk from MAYV throughout the Americas remain unclear. Understanding the ecology and distribution of MAYV remains a major obstacle in predicting areas that are at high risk of transmission to humans and domestic animals.

Current evidence suggests that MAYV is maintained in nature through a sylvatic transmission cycle involving mosquito vectors and non-human animal reservoirs. Therefore, human MAYV cases reported to date likely represent direct sylvatic spillovers. Residing near forested areas [12] and hunting in the rainforest [14] have been identified as risk factors for

MAYV infection in humans, highlighting the importance of the sylvatic transmission cycle and the potential for spillover events.

Identification of the non-human vertebrate animals (i.e., reservoirs) involved in MAYV transmission is an important step in delineating the human populations at greatest risk. The spillover of MAYV into humans represents a complex interaction of processes involving the density and distribution of reservoirs and vectors, as well as the prevalence and intensity of infection among reservoirs [15].

Identifying the non-human vertebrates that may serve as MAYV reservoirs is a difficult task due to a myriad of issues including, but not limited to, the challenges associated with establishing evidence of infection in wild animal populations [16, 17]. High seroprevalence of a pathogen in an animal population does not necessarily implicate a given host as an efficient reservoir; conversely, low seroprevalence at a single point in time cannot definitively rule out an animal as a reservoir [17]. Due to the relatively short viremia of MAYV (approximately 3-10 days) molecular assays may be unsuccessful in detecting virus [18], necessitating the use of serological assays such as hemagglutination-inhibition (HI) assays, enzyme-linked immunosorbent assays (ELISA), or plaque-reduction neutralization tests (NT).

Several studies have been conducted to clarify the precise vertebrate hosts that may serve as MAYV reservoirs. High seroprevalence among non-human primates (NHPs) in Brazil [19], French Guiana [12], and Panama [20] provides evidence that NHPs may play an important role in the MAYV transmission cycle. MAYV antibodies have also been detected in mammals including rodents and marsupials [21] as well as several avian species [19]. Unfortunately, there is significant heterogeneity in the study methods used to identify potential MAYV reservoirs and

there remains a high level of uncertainty surrounding the role of various non-human vertebrate species in the MAYV transmission cycle.

Studies have also been conducted in wild-caught mosquito populations as well as in controlled laboratory conditions in order to identify potential arthropod vectors of MAYV. One study in Brazil [19] suggested that the canopy-dwelling *Haemagogus janthinomys* mosquito is an important vector of MAYV. Additional mosquito species including *Aedes aegypti, Ae. albopictus,* and several anopheline species have been shown to be competent vectors in laboratory settings [22-24], posing a potential but as yet theoretical risk of urban MAYV cycles. The occurrence of MAYV in the city of Manaus has also led to concerns about the involvement of *Aedes* mosquitoes in a MAYV urban transmission cycle [25].

Although many non-human vertebrate animals and arthropod species have been proposed as capable MAYV reservoirs or vectors, our understanding of the MAYV transmission cycle and ecology remains limited. Collating and evaluating the current evidence regarding the potential MAYV reservoirs and vectors are important steps in characterizing MAYV transmission ecology and identifying the communities at greatest risk for MAYV outbreaks. Therefore, the goal of this systematic review is to evaluate the current evidence regarding MAYV occurrence in non-human vertebrates and arthropods. We present here the first structured evaluation of the potential vector and non-human reservoir range of MAYV, including the development of custom criteria for grading the quality of evidence of arbovirus occurrence in invertebrate and vertebrate non-human hosts**.**

Methods

This systematic review and meta-analysis were conducted according to the PRISMA 2020 Checklist [26] (see **S1 Table**). A protocol was developed but was not uploaded to PROSPERO.

Information Sources

We conducted a systematic review of original research articles, reports, and dissertations that attempted to identify potential non-human animal reservoirs or arthropod vectors of MAYV. We first searched Embase, Web of Science, PubMed, and SciELO databases for English, Spanish, and Portuguese language articles published between 1954 (the year MAYV was first isolated) and March 21, 2020. We searched all databases using the highly sensitive search term "Mayaro". A PubMed alert using the search term "Mayaro" was also set to capture any additional studies that were published between the initial search and May 2021. This database search was extended using bioRxiv (https://www.biorxiv.org/) and medRxiv (https://www.medrxiv.org/) pre-print databases. We complemented these database search results with 'grey literature,' including hand-searched bibliographies of MAYV review articles (including systematic reviews), dissertations from several Brazilian university repositories, the Pan American Health Organization (PAHO) Institutional Repository for Information Sharing database (iris.paho.org), the GIDEON database (https://www.gideononline.com/), and GenBank [27] (https://www.ncbi.nlm.nih.gov/genbank/). In addition, we searched conference handbooks that are available online (2004-2019) from the American Society of Tropical Medicine and Hygiene (https://www.astmh.org/annual-meeting/past-meetings).

Eligibility Criteria

We included studies that evaluated past or current MAYV infection in non-human vertebrates using methods including virus isolation, molecular detection, and serosurveys. We also included studies that screened arthropods for MAYV using virus isolation and molecular detection. Original research studies were considered for eligibility if they assessed MAYV positivity in field-caught, captive, or sentinel non-human vertebrates or field-caught arthropods. Studies that met any of the following exclusion criteria were not included: studies involving only humans; studies not reporting original data (e.g., review articles, perspective pieces, editorials, recommendations, and guidelines); duplicate studies; *in vitro* studies such as vector cell-line or mammal cell line experiments; laboratory-based vector competence studies that did not explicitly demonstrate the detection of MAYV in a wild-caught vector; i*n-vivo* lab-reared animal studies or any laboratory-based study that experimentally inoculated an animal to test theoretical reservoir status.

Selection process

All articles were organized using EndNote software version X9 (Clarivate, Philadelphia, Pennsylvania, USA), and data were abstracted into a Microsoft Excel table. Two reviewers independently screened all titles and abstracts to determine articles that could immediately be excluded and articles that should be included in the second stage of review. Results were compared to reconcile any differences between the two reviewers. The first and second reviewers then independently read the full text of potentially eligible articles identified through screening and selected the articles that were candidates for inclusion in the study. Results were compared to reconcile any differences between the two reviewers. A third-party reviewer adjudicated when consensus was not reached between the two reviewers during the first or second stage review.

From those studies deemed eligible, data were extracted from articles by one reviewer using the data abstraction tool in Microsoft Excel.

Data abstraction

Relevant information was abstracted by one reviewer in an Excel sheet. Information for each article was abstracted across several domains including publication details (author and affiliation, study title, study funding), study methods (date and location of study, study design, laboratory methods to assess MAYV positivity), and study results (sample size, taxonomic classification, proportion of animals testing positive for MAYV, location of vertebrates/arthropods testing positive for MAYV). A second reviewer randomly selected and reviewed five articles for review to validate the data abstraction process.

Grading quality of evidence

We developed a customized grading system to assess the quality of each study included in our review. Several published studies have employed a similar grading system to assess evidence quality of included articles [28-30]. We assigned each study in our systematic review a grade for each of four quality items: clarity of research question/objective (*Was the research question/objective clearly described and stated?*); description of study methods (*Were the study methods presented in a reproducible way?*); description of sampling methods (*Was the sampling method described in detail?*); and validity of diagnostic tests (*Was MAYV positivity measured in a valid way?*). For each quality item, eligible studies were assigned a score of 3 (strong evidence), 2 (moderate evidence), 1 (weak evidence), or unable to judge. Studies were deemed unable to judge if the information provided was insufficient to assign quality scores (e.g., a single GenBank entry or conference abstract).

A score of 3 was assigned for the *description of sampling methods* item if authors thoroughly described the type of trap used, the habitats in which traps were set, how often traps were checked, and the results of trapping (i.e., were animals reported to the species level). For studies that assessed MAYV in vertebrate animals, a score of 3 was assigned for the *validity of diagnostic tests* item if MAYV positivity was assessed using RT-PCR, viral culture, or high-specificity serological method (i.e., plaque reduction NT); a score of 2 was assigned if MAYV positivity was assessed using non-specific serological assay (i.e., HI and ELISA); and a score of 1 was assigned if MAYV positivity was based on presumptive exposure only with no confirmatory assay. For studies that assessed MAYV in arthropods, a score of 3 was assigned for this item if MAYV positivity was assessed using viral culture; a score of 2 was assigned if MAYV positivity was assessed using RT-PCR or metagenomics; and a score of 1 was assigned if MAYV positivity was based on presumptive exposure only with no confirmatory assay. A score of "NA" was assigned for the *validity of diagnostic tests* item if studies did not detect MAYV positivity in any animal or arthropod samples.

Quality review scores were recorded in two different Excel documents for animal reservoir studies and arthropod vector studies, respectively. Two reviewers independently graded the evidence quality for each study and results were compared to reconcile any differences between the two reviewers. A third-party reviewer adjudicated if consensus was not reached between the two reviewers.

Data analysis

Descriptive Analysis

Descriptive statistics were presented by species for potential animal reservoirs showing the total sample size, proportion infected, and locations of infected animals. Descriptive statistics were presented by species for potential arthropod vectors showing the total sample size and total pools tested for virus (if applicable), the number of MAYV isolates or PCR-positive pools, and locations of infected arthropods. Maps were developed using ArcGIS software [31] to display the geographic distribution of MAYV-positive animals and vectors.

Pooled Analysis

Due to the heterogeneity of study designs and outcome measurements, a quantitative meta-analysis across all eligible studies was not possible. Instead, we conducted a seroprevalence meta-analysis using the studies that reported MAYV seroprevalence (i.e., using serological methods including HI, ELISA, or NT) in non-human vertebrate animals. Pooled prevalence estimates were stratified by taxonomic order and an additional analysis was conducted among the various Primate genera. Orders were excluded from the analysis if the total sample size was less than 10 or if no MAYV-positive samples were reported within that order. Pooled seroprevalence was first calculated based on all available data, regardless of test method. This included the samples that tested MAYV-positive based on HI alone (when no confirmatory assay was performed) as well as the samples that were confirmed positive by an NT. Only monotypic reactions to MAYV were included in the meta-analysis in the absence of confirmatory NT. A sensitivity analysis was then conducted using only the MAYV-positive samples that were confirmed using NT. Positive samples that were based on HI alone (without confirmatory NT)

were excluded from this analysis, although all MAYV-negative samples were retained. This sensitivity analysis was conducted to account for the low specificity of HI compared to NT [32] and provided a more conservative estimate of seroprevalence.

Due to the substantial differences across studies including sample size, study design, species sampling methods, and geographical location, a random effects model was used for analysis [33, 34]. The Freeman-Tukey double-arcsine transformation was implemented to calculate a proportion, based on the recommendation of Barendregt et al. [35]. A sensitivity analysis was conducted using a generalized linear mixed model (GLMM) with a logit transformation, due to the potential for misleading results with the double-arccosine transformation [36, 37]. Measures of variance (τ^2), heterogeneity (I^2), and statistical significance are presented for each random effects model. An additional sensitivity analysis was conducted using a fixed effects model. Results of sensitivity analyses are presented in the Supplementary materials.

The I^2 statistic measures inconsistency across study results and is calculated as I^2 = 100% x (Q - df) / Q [38]. The I^2 statistic ranges between 0% and 100%, where a value of 0% represents no heterogeneity and larger values represent increased heterogeneity. Animal seroprevalence estimates with 95% confidence intervals (CIs) weighted by sample size are presented as forest plots. All analyses were conducted using the '*meta*' package in R statistical software version 4.0.2 (R Project for Statistical Computing, Vienna, Austria) [39, 40].

Estimation of bias

An assessment of publication bias was carried out for meta-analyses that included five studies or more. Bias was assessed using funnel plots and tests for funnel plot asymmetry based

on methods proposed by Egger [41]. If the Egger's test revealed bias, the Trim and Fill technique was used to estimate the effect of missing studies on the outcomes of the meta-analysis [42].

Georeferencing of MAYV occurrence

All available location information from each confirmed MAYV infection (animal and mosquito) was extracted from each article and georeferenced based on methods that have been described previously [43, 44]. Each occurrence of MAYV was designated as either a point or polygon location according to the spatial resolution provided in the study. When specific latitude and longitude coordinates were provided, they were verified in GoogleMaps and designated as a point location. If a neighborhood, town, village, or small city was explicitly mentioned in the article and fell within a 5x5 km grid cell, it was designated as a point location and its centroid coordinates were recorded. For studies that report a less precise spatial resolution such as states or counties, first level (ADM1) or second level (ADM2) administrative divisions were recorded as polygons. If the size of a specific named location was greater than a 5x5 km grid cell the occurrence was assigned to a custom polygon created in ArcGIS that encompassed the extent of that location. If place names were duplicated (i.e., the ADM1 and ADM2 units had the same name), the coarsest spatial resolution was used. Country shapefiles were accessed through the geoBoundaries Global Administrative Database [45].

Results

General Findings

We identified a total of 57 research items that met our eligibility criteria out of 1523 research items screened, including 46 research articles, seven dissertations, two GenBank entries, one laboratory report, and one abstract (see **Table 1** for a full list of eligible items and citations).

298 Thirty-nine (68%) of the included items assessed MAYV infection in non-human vertebrates
299 while 29 (51%) items assessed MAYV infection in arthropods. Of the 57 eligible items, 24
300 (42%) were included in the vertebrate seroprevalence meta-analysis, and the remaining items
301 were only included in the qualitative analysis. A flow chart describing the article search and
302 selection process is presented in **Fig 1**. Five articles were identified that met the inclusion criteria
303 but were deemed to be reporting the same data as other included articles. These include de
304 Thoisy *et al*., (2001) [46] and Talarmin *et al.*, (1998) [12] (both reporting the same data as de
305 Thoisy *et al.*, (2003) [21]), Aitken *et al.*, (1960) [47] (reporting the same data as Aitken *et al.*,
306 (1969) [48]), Batista *et al*., 2013 [49] (reporting the same data as Paulo *et al*., (2015) [50]), and
307 Woodall (1967) [51] (reporting the same data as Taylor, (1967) [52]). These articles were
308 excluded from this systematic review.

309 **Table 1. Eligible Study Characteristics**

Reference	Study Period	Country	Arthropods Tested (n)	Vertebrate non-human animals tested (n)[a]	MAYV infection reported
Aitken, 1969 [48]	1953-1963	Trinidad	1,568,439	---	Yes
Araujo, 2003 [53]	2002	Brazil	---	555	Yes
Araujo, 2004 [54]	2003	Brazil	---	202	No
Araujo, 2004b [55]	2003	Brazil	---	495	Yes
Araujo, 2012 [56]	2007-2008	Brazil	---	95	Yes
Araujo, 2012b [57]	2009	Brazil	---	102	Yes
Azevedo, 2009 [58]	2008	Brazil	832	---	Yes
Batista, 2012 [59]	2010	Brazil	122	65	Yes
Calisher, 1974 [60]	1967	USA[b]	---	1,300	Yes
Carrera, 2020 [61]	2017	Panama	113	---	No
Casseb, 2010 [62]	2009	Brazil	---	2191	Yes
Casseb, 2016 [63]	2009	Brazil	---	753	Yes
Catenacci, 2017 [64]	2006-2014	Brazil	239	142	Yes
Cruz, 2009 [65]	2006-2008	Brazil	---	85	No
Degallier, 1992 [66]	1974-1988	Brazil	2,005,069	6,248	Yes
De Thoisy, 2003 [21]	1994-1995	French Guiana	---	579	Yes
Diaz, 2007 [67]	1994	Argentina, Paraguay	---	90	No
Esposito, 2015 [68]	1960	Brazil	NA[d]	---	Yes
Ferreira, 2020 [69]	2017-2018	Brazil	10,569	---	Yes
Galindo, 1966 [70]	1959-1962	Panama	377,492	2,444	Yes
Galindo, 1967 [71]	1966	Panama	11,829	---	Yes

Galindo, 1983 [72]	1972-1979	Panama	NA[c]	NA[c]	Yes
GenBank KY618129	1991	Brazil	NA[d]	---	Yes
GenBank KY618130	2011	Brazil	NA[d]	---	Yes
Gibrail, 2015 [73]	2011-2014	Brazil	---	50	No
Gomes, 2019 [74]	2018	Brazil	---	213	Yes
Groot, 1961 [75]	1958-1960	Colombia	41,564	---	Yes
Groot, 1964 [11]	1956-1961	Colombia	---	34	Yes
Henriques, 2008 [76]	2002-2005	Brazil	37,519	---	No
Hoch, 1981 [19]	1978-1979	Brazil	10,667	1785	Yes
Kubiszeski, 2017 [77]	2014-2015	Brazil	778	---	Yes
Laroque, 2014 [78]	2008-2010	Brazil	---	131	Yes
Maia, 2019 [79]	2017	Brazil	4786	---	Yes
Martinez, 2020 [80]	2018-2019	Colombia	169	---	No
Medlin, 2016 [81]	2005-2007	Costa Rica	---	94	No
Medina, 2015 [82]	1999	Venezuela	---	NA[d]	Yes
Moreira-Soto, 2018 [83]	2012-2017	Brazil	---	103	Yes
Nunes, 2009 [84]	2005	Brazil	---	181	No
Paulo, 2015 [50]	2012-2014	Brazil	---	43	Yes
Pauvolid-Correa, 2010 [85]	2007	Brazil	---	135	No
Pauvolid-Correa, 2015 [86]	2009-2011	Brazil	---	748	Yes
Pauvolid-Correa, 2008 [87]	2007	Brazil	1,759	NA[e]	No
Perez, 2019 [88]	2007-2008	Peru	---	90	Yes
Pinheiro, 1974 [89]	1971-1974	Brazil	NA[c]	NA[c]	Yes
Pinheiro, 2019 [90]	2017	Brazil	867	---	No
Powers, 2006 [91]	N/A	N/A	NA[d]	NA[d]	Yes
Price, 1978 [92]	1972-1974	Trinidad	---	997	No
Ragan, 2019 [93]	N/A	N/A	---	NA[c]	No
Sanmartin, 1973 [94]	1967	Colombia	27,437	480	No
Scherer, 1975 [95]	1970-1971	Peru	1,500	NA[c]	No
Serra, 2016 [96]	2013	Brazil	4,556	---	Yes
Seymour, 1983 [20]	1974-1976	Panama	---	304	Yes
Silva, 2017 [97]	2016	Brazil	3,750	---	No
Srihongse, 1974 [98]	1967	Panama/Colombia	---	2026	Yes
Tauro, 2019 [99]	2017	Brazil	125	---	No
Taylor, 1967 [52]	N/A	Brazil/Trinidad	NA[c]	NA[c]	Yes
Turell, 2019 [100]	2001-2002	Peru	---	20	No

[a] Includes wild-caught, sentinel, and domestic animals.
[b] Migratory birds captured in Louisiana.
[c] Unable to determine the total number of animals or arthropods tested for MAYV.
[d] Genomic sequence only. No additional information provided.
[e] Horse seroprevalence data collected but recorded in another study.

Fig 1. Flow diagram for search and selection of articles

Studies were conducted in the following countries: Brazil (n=34), Panama (n=5), Colombia (n=4), Peru (n=3), Trinidad and Tobago (n=2), French Guiana (n=1), Venezuela

(n=1), Costa Rica (n=1), and the United States of America (n=1). Several studies reported data from multiple countries including Argentina/Paraguay (n=1), Panama/Colombia (n=1), and Brazil/Trinidad and Tobago (n=1). The majority of studies were conducted after the year 2000 (n=33), although some studies were conducted between 1950-1969 (n=9), 1970-1989 (n=8), or 1990-1999 (n=4). Quality scores for all included studies are reported in Table 2.

Table 2. Quality Review Scores

	Vertebrate animals				Arthropods			
	Research question	Study methods	Sampling method	MAYV+ test method[a]	Research question	Study methods	Sampling method	MAYV+ test method[a]
Aitken, 1969 [48]	---	---	---	---	3	2	2	3
Araujo, 2003 [53]	3	3	2	2	---	---	---	---
Araujo, 2004 [54]	3	3	3	NA	---	---	---	---
Araujo, 2004b [55]	3	3	2	2	---	---	---	---
Araujo, 2012 [56]	3	3	3	2	---	---	---	---
Araujo, 2012b [57]	3	3	2[b]	2	---	---	---	---
Azevedo, 2009 [58]	---	---	---	---	2	2	2	3
Batista, 2012 [59]	2	3	2	2	2	3	2	NA
Calisher, 1974 [60]	3	3	2	3	---	---	---	---
Carrera, 2020 [61]	---	---	---	---	3	3	3	N/A
Casseb, 2010 [62]	3	3	2[b]	2	---	---	---	---
Casseb, 2016 [63]	3	3	3[b]	2	---	---	---	---
Catenacci, 2017 [64]	3	3	3	N/A	3	3	2	2
Cruz, 2009 [65]	2	3	2	N/A	---	---	---	---
Degallier, 1992 [66]	3	2	2	2	3	2	3	N/A
De Thoisy, 2003 [21]	3	3	2	3	---	---	---	---
Diaz, 2007 [67]	3	2	2	3	---	---	---	---
Esposito, 2015 [68]	---	---	---	---	Unable to judge	Unable to judge	Unable to judge	3
Ferreira, 2020 [69]	---	---	---	---	3	3	3	3
Galindo, 1966 [70]	3	3	2	N/A	3	3	3	3
Galindo, 1967 [71]	---	---	---	---	3	3	2	2
Galindo, 1983 [72]	3	3	3	N/A	3	2	2	3
GenBank KY618129	---	---	---	---	Unable to judge	Unable to judge	Unable to judge	3
GenBank	---	---	---	---	Unable to	Unable to	Unable to	3

KY618130					judge	judge	judge	
Gibrail, 2015 [73]	3	3	2	2	---	---	---	---
Gomes, 2019 [74]	3	3	3[b]	3	---	---	---	---
Groot, 1961 [75]	---	---	---	---	3	3	3	3
Groot, 1964 [11]	3	3	3	2	---	---	---	---
Henriques, 2008 [76]	---	---	---	---	3	3	3	N/A
Hoch, 1981 [19]	3	3	3	3	3	3	3	3
Kubiszeski, 2017 [77]	---	---	---	---	3	3	3	2
Laroque, 2014 [78]	3	3	2	2	---	---	---	---
Maia, 2019 [79]	---	---	---	---	3	3	3	3
Martinez, 2020 [80]	---	---	---	---	3	3	2	N/A
Medlin, 2016 [81]	3	3	3	N/A	---	---	---	---
Medina, 2015 [82]	3	2	2[c]	3	---	---	---	---
Moreira-Soto, 2018 [83]	3	3	3	3	---	---	---	---
Nunes, 2009 [84]	2	3	2	N/A	---	---	---	---
Paulo, 2015 [50]	3	3	3	2	---	---	---	---
Pauvolid-Correa, 2010 [85]	3	2	2[b]	N/A	---	---	---	---
Pauvolid-Correa, 2015 [86]	3	3	3	3	---	---	---	---
Pauvolid-Correa, 2008 [87]	---	---	---	---	3	3	2	N/A
Perez, 2019 [88]	3	2	2	3	---	---	---	---
Pinheiro, 1974 [89]	3	2	2	2	3	2	2	N/A
Pinheiro, 2019 [90]	---	---	---	---	3	3	3	N/A
Powers, 2006 [91]	3	2	Unable to judge	3	3	2	Unable to judge	3
Price, 1978 [92]	3	2	2	N/A	---	---	---	---
Ragan, 2019 [93]	Unable to judge	Unable to judge	Unable to judge	Unable to judge	---	---	---	---
Sanmartin, 1973 [94]	3	3	3	N/A	2	3	2	N/A
Scherer, 1975 [95]	2	3	3[c]	N/A	2	2	2	N/A
Serra, 2016 [96]	---	---	---	---	3	3	3	3
Seymour, 1983 [20]	2	3	2	3	---	---	---	---
Silva, 2017 [97]	---	---	---	---	3	3	3	N/A
Srihongse, 1974 [98]	3	2	2	2	---	---	---	---
Tauro, 2019 [99]	---	---	---	---	3	2	2	N/A
Taylor, 1967 [52]	Unable to judge	Unable to judge	Unable to judge	3	Unable to judge	Unable to judge	Unable to judge	3
Turell, 2019 [100]	3	2	3[c]	N/A	---	---	---	---

[a] Studies were assigned a score of NA for this criterion if no MAYV-positive samples were reported.
[b] Domestic animals only.
[c] Sentinel animals only.

MAYV in wild-caught non-human vertebrate animals

Thirty-nine (68%) studies in our systematic review assessed MAYV infection in wild-caught non-human vertebrate animals (including birds, mammals, and reptiles). Seventeen (44%) of these studies identified at least one non-human vertebrate that was positive for MAYV infection. Of the 27 taxonomic orders studied, 12 (44.4%) had evidence of MAYV infection: Artiodactyla (even-toed ungulates), Caprimulgiformes (nightbirds), Carnivora, Charadriiformes (shorebirds), Cingulata (armadillos), Columbiformes (pigeons and doves), Didelphimorphia (opossums), Passeriformes (passerine birds), Pilosa (sloths and anteaters), Primate, Rodentia, and Squamata (scaled reptiles). The greatest number of MAYV-positive animal species were found in the order Charadriiformes (n=16 positive species) and the order Primate (n=15 positive species). (See **S2 Table** for complete mammal data and **S3 Table** for complete avian data).

Table 3 reports NHP species that were detected with MAYV antibodies. Only studies with positive results are shown on Table 3; other negative studies are listed in the **S2 Table**. High MAYV seroprevalence was confirmed by NT among *Alouatta seniculus* monkeys in individual studies in French Guiana [21] (n=51/98) and among *Callithrix argentata* monkeys in Brazil [19] (n=32/119). In addition, 29 *Cebus libidinosus* monkeys from wildlife screening centers were detected with MAYV antibodies according to HI, although only six were reported as monotypic reactions [78]. Diagnosis in these monkeys was not confirmed by NT. An additional *Cebus libidinosus* monkey presented a heterotypic reaction to MAYV (titer of 1:20) and four additional viruses according to HI (including a titer of 1:640 for Oropouche virus) [73].

351 However, based on the study's protocol, confirmatory NT was only performed for viruses with

352 titers ≥ 1:40.

353 **Table 3. Evidence of MAYV infection in non-human primates**

Species	Positive (n)	Total tested (n)[a]	% Pos	Test method	Notes	Citation
Alouatta seniculus	51	98	**52.0**	HI with confirmatory NT[d]	NA	[21]
	1	1	**100.0**	ELISA with confirmatory plaque-reduction NT	NA	[88]
Callithrix argentata	32	119	**26.9**	HI with confirmatory NT	One isolation also reported but not included in this table.	[19]
Cebus libidinosus[b]	6	100	**6.0**	HI	Six reactions were monotypic, and 23 were heterotypic, with titers of 1:20 (n=1), 1:80 (n=6), 1:160 (n=2), 1:320 (n=6), 1:640 (n=6), and 1:1280 (n=8). Only 6 of the 29 reactions were monotypic.	[78]
Tamarin, Pithecia, Cebus (species not specified)	7	21	**33.3**	HI	Results presented as a table from the Belem Virus Laboratory, but no further information is provided regarding the study methods or primate species.	[52]
Cebus apella	10	62	**16.1**	HI	Titer results for monotypic reactions were 1:80 (n=2), 1:160 (n=7) and 1:640 (n=1). Three additional samples showed positive results for MAYV and another virus.	[59]
Saguinas midas	8	42	**19.1**	HI with confirmatory NT[d]	NA	[21]
Alouatta sp.[c]	7	11	**63.6**	HI	NA	[11]
Lagothrix poeppigii	6	11	**54.5**	ELISA with confirmatory plaque-reduction NT	NA	[88]
Saimiri sciureus	4	6	**66.7**	HI with confirmatory NT[d]	NA	[21]
Pithecia pithecia	4	5	**80.0**	HI with confirmatory NT[d]	NA	[21]
Cebus sp.[c]	4	13	**30.8**	HI	NA	[11]
Alouatta villosa	3	5	**60.0**	Plaque-reduction NT	Samples considered positive if 90% plaque reduction by plasma 1:16 or weaker. The median positive titer was 1:128 (range	[20]

					1:32-1:512).	
Sapajus sp.	3	43	**7.0**	HI and RT-PCR	Positive samples had a monotypic reaction to MAYV with titers of 1:80 (n=1) and 1:160 (n=2). All samples negative by RT-PCR.	[50]
Sapajus xanthosternos	1	2	**50.0**	Plaque-reduction NT	Plaque reduction NTs were performed against MAYV for all CHIKV-positive samples. The sample neutralized both MAYV and CHIKV at titers of 1:40.	[83]
Ateles marginatus	1	1	**100.0**	Plaque-reduction NT	Plaque reduction NTs were performed against MAYV for all CHIKV-positive samples. The sample neutralized both MAYV and CHIKV at titers of 1:40.	[83]
Alouatta belzebul	1	1	**100.0**	HI with confirmatory NT	NA	[19]
Sapajus macrocephalus	1	6	**16.7**	ELISA with confirmatory plaque-reduction NT	NA	[88]
Cacajao calvus	1	3	**33.3**	ELISA with confirmatory plaque-reduction NT	NA	[88]
Callicebus brunneus[e]	1	N/A	**NA**	HI	Sera reacted against MAYV and Tacaiuma virus. No additional information provided.	[66]
Aotus sp.[c]	1	4	**25.0**	HI	NA	[11]
Saimiri sp.[c]	1	1	**100.0**	HI	NA	[11]

MAYV: Mayaro virus; HI: hemagglutination inhibition; ELISA: enzyme-linked immunosorbent assay; RT-PCR: reverse transcription polymerase chain reaction; NT: neutralization test; CHIKV: Chikungunya virus

[a] Denominators presented in this table reflect only studies that reported MAYV positivity. Complete data (including MAYV-negative samples) are included in the seroprevalence meta-analysis and the Supplementary Tables.
[b] Captive primates from a wildlife rescue facility.
[c] Sera analyzed for MAYV may have had cross reactivity with Una virus because the authors used a Colombian isolate that was initially characterized as MAYV but was later identified as Una virus. A differential test was not performed for MAYV. However, the authors identified human sera that was reactive to MAYV alone in the same study region.
[d] Serum samples with titers >1:20 confirmed by seroneutralization. Positive reaction was considered with the total inhibition of the cytopathic effect in the cell monolayer.
[e] Authors also reported that seven monkey sera among the 14 examined were positive for yellow fever and MAYV, of which five were positive for the two agents. The species of these positive samples were: *Pithecia pithecia* (n=1), *Alouatta seniculus* (n=2), *Saimiri sciureus* (n=1), *Saguinus midas* (n=1), and *Ateles paniscus* (n=2). However, they did not note the specific primate species that were positive for MAYV.

Among the 12 additional NHP species with evidence of past MAYV infection, nine were confirmed by NT and three by HI alone. In addition, MAYV positivity was reported in the following NHP genera, although animals were not reported to species: *Aotus* (n=1/4), *Alouatta*

(n=7/11), *Cebus* (n=4/13), *Sapajus* (n=3/43), and *Saimiri* (n=1/1). The authors reporting MAYV positivity in the *Aotus*, *Alouatta*, *Cebus*, and *Saimiri* genera noted that these results should be interpreted with caution due to potential for cross-reactivity with Una virus (UNAV) [11]. In one study conducted in Brazil, two of 11 Chikungunya virus (CHIKV)-positive serum samples (in the species *Sapajus xanthosternos* and *Ateles marginatus*) neutralized MAYV with titers of 1:40 in plaque reduction NTs [83]. These two samples were considered MAYV-positive and included in our meta-analysis. One additional study [67] detected neutralizing antibodies against both UNAV and MAYV in 21 *Alouatta caraya* monkeys. However, all 21 monkeys were diagnosed with UNAV based on a 4-fold titer difference between the two viruses. Therefore, we considered these monkeys MAYV-negative and did not include them in our meta-analysis. Finally, in 1963 the Belem Virus laboratory reported MAYV infection in seven NHPs based on HI tests alone [52]. These monkeys were described as Tamarin, Pithecia, and Cebus although no further information was provided regarding sampling method, testing protocol, or primate species.

MAYV antibodies were also detected in 21 bird species from the order Charadriiformes (n=16) and Passeriformes (n=5). All MAYV-positive birds were found in Brazil, with the exception of one MAYV isolate from a migrating bird captured in Louisiana USA [60]. A high MAYV-seroprevalence (n=34/122) was reported by the Belem Laboratory in 1963 among *Columbigallina* birds, although no additional information was provided regarding sampling method or bird species. MAYV antibodies were also detected in seven avian families that were not identified to genus or species. Only one study that detected MAYV antibodies in birds performed confirmatory NT [19]. All other diagnoses (with the exception of the virus isolation) were made by HI tests alone. See **Table 4** for additional information regarding avian species that were infected with MAYV.

396 **Table 4. Evidence of MAYV infection in birds**

Order	Species	Positive (n)	Total (n)[a]	% Pos	Test method	Notes	Citation
Columbiformes	*Columbigallina* sp.	34	121	**28.1**	HI	Results presented as a table from the Belem Virus Laboratory, but no further information is provided regarding the methods or species.	[52]
Charadriiformes	*Sterna hirundo*	23	342	**6.7**	HI	NA	[53]
Charadriiformes	*Sterna trudeaui*	12	56	**21.4**	HI	NA	[53]
Charadriiformes	*Arenaria interpres*	8	28	**28.6**	HI	NA	[53]
		1	NA	**NA**	HI	Titers 1:40	[55]
Charadriiformes	*Calidris canutus*	7	51	**13.7**	HI	NA	[53]
Passeriformes	Fringillidae family, unspecified species	6	131	**4.6**	HI with confirmatory NT	NA	[19]
Passeriformes	Formicariidae family, unspecified species	5	444	**1.1**	HI with confirmatory NT	NA	[19]
Charadriiformes	*Limosa haemastica*	5	17	**29.4**	HI	NA	[53]
Charadriiformes	*Tringa flavipes*	4	5	**80.0**	HI	NA	[53]
Charadriiformes	*Calidris pusilla*	3	NA	**NA**	HI	Titers 1:40 for all positive samples	[55]
		1	30	**3.3**	HI	Monotypic reaction with titers ≥ 1:20 to MAYV	[56]
Charadriiformes	*Sterna superciliaris*	2	8	**25.0**	HI	N/A	[53]
Charadriiformes	*Actitis macularius*	2	22	**9.1**	HI	Monotypic reaction with titers ≥ 1:20 to MAYV	[56]
Passeriformes	Dendrocolaptidae family, unspecified species	1	97	**1.0**	HI with confirmatory NT	NA	[19]
Passeriformes	*Icterus spurius*	1	223	**0.45**	Virus isolation by inoculation into suckling mice	NA	[60]
Passeriformes	*Arremon tactiturnus*	1	NA	**NA**	HI (confirmatory NT unclear)	NA	[66]
Passeriformes	Pipridae family, unspecified species	1	229	**0.44**	HI with confirmatory NT	NA	[19]
Passeriformes	*Cercomacra tyrannina*	1	NA	**NA**	HI (confirmatory NT unclear)	NA	[66]
Passeriformes	*Formicivora grisea*	1	NA	**NA**	HI (confirmatory NT unclear)	NA	[66]
Passeriformes	*Tyrannus*	1	NA	**NA**	HI	NA	[66]

	melancholicus				(confirmatory NT unclear)		
Passeriformes	Tyrannidae family, unspecified species	1	102	**0.98**	HI with confirmatory NT	NA	[19]
Charadriiformes	*Pluvialis squatarola*	1	4	**25.0**	HI	Monotypic reaction with titers ≥ 1:20 to MAYV	[56]
Charadriiformes	*Haematopus palliatus*	1	6	**16.7**	HI	NA	[53]
Charadriiformes	*Sterna eurygnatha*	1	7	**14.3**	HI	NA	[53]
Charadriiformes	*Sterna maxima*	1	1	**100**	HI	NA	[53]
Charadriiformes	*Sterna niotica*	1	1	**100**	HI	NA	[53]
Charadriiformes	*Calidris fuscicollis*	1	11	**9.1**	HI	NA	[53]
Charadriiformes	*Calidris minutilla*	1	6	**16.7**	HI	Monotypic reaction with titers ≥ 1:20 to MAYV	[56]
Caprimulgiformes	Caprimulgidae family, unspecified species	1	5	**20.0**	HI with confirmatory NT	NA	[19]
Columbiformes	Columbidae family, unspecified species	1	34	**2.9**	HI with confirmatory NT	NA	[19]
Passeriformes	*Molothrus* sp.	1	NA	**NA**	HI	Titers 1:80	[55]

MAYV: Mayaro virus; HI: hemagglutination inhibition; NT: neutralization test

[a] Denominators presented in this table reflect only studies that reported MAYV positivity. Complete data (including MAYV-negative samples) is reflected in the seroprevalence meta-analysis and the Supplementary Tables.

Additional wild-caught mammals with evidence of MAYV infection are presented in **Table 5**. Six rodent species as well as unidentified rodents in the *Echimys* and *Proechimys* genera were detected with MAYV antibodies in French Guiana [21], Peru [88], and Panama [20]. In addition, four species in the order Didelphimorphia, three species in the order Pilosa, and one species each in the orders Carnivora, Artiodactyla, and Cingulata were detected with MAYV antibodies in French Guiana [21] and Peru [88]. Additional positive samples were detected in the orders Rodentia, Didelphimorphia, and Pilosa although the species were not identified.

Table 5. Evidence of MAYV infection in mammals (excluding non-human primates)

Order	Species	Positive (n)	Total (n)[a]	% Pos	Test method	Notes	Citation
Rodentia	Wild rodents, unspecified	71	960	**7.4**	HI	Results presented as a table from the Belem Virus Laboratory, but no further	[52]

						information is provided regarding the methods or species.	
Didelphimorphia	Opossum, unspecified	9	122	**7.4**	HI	Results presented as a table from the Belem Virus Laboratory, but no further information is provided regarding the methods or species.	[52]
Pilosa	*Choloepus didactylus*	7	26	**26.9**	HI with confirmatory NT[b]	NA	[21]
Didelphimorphia	*Marmosa* sp.	7	46	**15.2**	HI	NA	[52]
Pilosa	*Tamandua tetradactyla*	6	26	**23.1**	HI with confirmatory NT[b]	NA	[21]
Cingulata	*Dasypus novemcinctus*	4	40	**10.0**	HI with confirmatory NT[b]	NA	[21]
		2	4	**50.0**	ELISA with confirmatory plaque reduction NT	NA	[88]
Rodentia	*Dasyprocta leporina*	5	29	**17.2**	HI with confirmatory NT[b]	NA	[21]
Didelphimorphia	*Philander opossum*	5	27	**18.5**	HI with confirmatory NT[b]	NA	[21]
Rodentia	*Coendou prehensilis*	3	26	**11.5**	HI with confirmatory NT[b]	NA	[21]
Rodentia	*Dasyprocta punctata*	3	5	**60.0**	Plaque reduction NT	Samples considered positive if 90% plaque reduction by plasma 1:16 or weaker. The median positive titer was 1:128 (range 1:32-1:512).	[20]
Rodentia	*Dasyprocta fuliginosa*	3	27	**11.1**	ELISA with confirmatory plaque reduction NT	NA	[88]
Rodentia	*Coendou melanurus*	2	15	**13.3**	HI with confirmatory NT[b]	NA	[21]
Didelphimorphia	*Didelphis albiventris*	2	19	**10.5**	HI with confirmatory NT[b]	NA	[21]
Rodentia	*Echimys* sp.	1	21	**4.8**	HI with confirmatory NT[b]	NA	[21]
Rodentia	*Agouti paca*	1	10	**10.0**	ELISA with confirmatory plaque reduction NT	NA	[88]

Rodentia	*Proechimys* sp.	1	18	**5.6**	HI with confirmatory NT[b]	NA	[21]
Didelphimorphia	*Caluromys philander*	1	5	**20.0**	HI with confirmatory NT[b]	NA	[21]
Didelphimorphia	*Didelphis marsupialis*	1	29	**3.5**	HI with confirmatory NT[b]	NA	[21]
Carnivora	*Potos flavus*	1	9	**11.1**	HI with confirmatory NT[b]	NA	[21]
Artiodactyla	*Pecari tajacu*	1	6	**16.7**	ELISA with confirmatory plaque reduction NT	NA	[88]
Pilosa	*Bradypus tridactylus*	1	29	**3.5**	HI with confirmatory NT[b]	NA	[21]
Pilosa	*Bradypus* sp.	1	3	**33.3**	HI	NA	[52]

MAYV: Mayaro virus; HI: hemagglutination inhibition; ELISA: enzyme-linked immunosorbent assay; RT-PCR: reverse transcription polymerase chain reaction; NT: neutralization test

[a] Denominators presented in this table reflect only studies that reported MAYV positivity. Complete data (including MAYV-negative samples) is reflected in the seroprevalence meta-analysis and the Supplementary Tables.
[b] Serum samples with titers >1:20 confirmed by seroneutralization. Positive reaction was considered with the total inhibition of the cytopathic effect in the cell monolayer.

Successful isolation of MAYV was reported from the following viremic animals: a silvery marmoset (*Callithrix argentata*) captured during a MAYV outbreak in Belterra, Brazil [19] and a migrating orchard oriole (*Icterus spurius*) captured in Louisiana [60]. In addition, the Belem Virus Laboratory reported MAYV isolation from two lizard species in 1963 [52] (*Tropidurus torquatus* and *Ameiva ameiva*) although no further information was provided regarding study methods or procedures.

The geographic distribution of animals (wild-caught, domestic, and sentinel) infected with MAYV is presented in **Fig 2**. The infected animals were identified in six countries overall, including Brazil, Peru, French Guiana, Colombia, Venezuela, and Panama, although the majority of infected animals were found in Brazil. Overall, 12 locations were geo-referenced as points,

four locations as ADM1 polygons, 15 locations as ADM2 polygons, and two locations as custom polygons.

Fig 2. Georeferenced locations of MAYV positivity in non-human animals and arthropods. The finest spatial scale is presented where possible. One MAYV isolate detected in a migrating bird in Louisiana is not included in the map.

MAYV in domestic or sentinel animals

Nine studies analyzed MAYV seroprevalence in domestic animals (equids, sheep, poultry, dogs, pigs, cattle, and buffaloes), and five studies analyzed MAYV seroprevalence in sentinel animals (monkeys, mice, and hamsters). Domestic and sentinel animals with evidence of MAYV positivity are reported in **Table 5** and complete results are reported in the **S4 Table**. In domestic animals, evidence of MAYV infection was detected in equids, cattle/buffalo, and dogs. Six studies assessed MAYV seroprevalence in Brazilian equids [54, 57, 63, 74, 85, 86], and antibodies against MAYV were detected in four of these studies. Notably, Gomes et al. [74] reported MAYV neutralizing antibodies in 48 equids out of 213 (23%) based on ELISA. However, only 16 of the 48 equids were considered positive based on the study's diagnostic criterion of 4-fold greater plaque reduction NT_{90} titer than that of the other viruses under study. In addition, Casseb et al. [63] detected MAYV antibodies in 40 horses using HI, although only four of the 40 reactions were monotypic, and confirmatory NTs were not performed. Additional domestic animals with evidence of MAYV infection included cattle/buffalo (n=14/1103 positive reactions by HI; 5/14 monotypic reactions [62]) and dogs (n=2/7 positive reactions by HI [53]). In addition, neutralizing antibodies (plaque reduction NT_{90} titer $\geq$10) against MAYV were detected in three sheep in Brazil [86]. However, these animals did not meet the original study's diagnostic criterion for MAYV diagnosis based on 4-fold greater plaque reduction NT_{90} titer

450 than that of the other viruses under study. Evidence of MAYV infection was also detected by HI
451 in two sentinel monkeys placed in the tree canopy in Panama [98], and one MAYV isolate was
452 obtained from a sentinel hamster in Venezuela [82].

453 **Table 6. Domestic and sentinel animals with evidence of MAYV infection**

Animal Type	Total Positive	Number Tested[a]	% Pos	Test Method	Notes	Citation
Domestic Equids	16	213	**7.5**	ELISA with confirmatory plaque reduction NT	Forty-eight horses had antibodies to MAYV by ELISA. Sixteen of 48 (33%) were considered positive by plaque reduction NT_{90} for MAYV with titers 1:10 (n=12), 1:20 (n=3) and 1:40 (n=1).	[74]
	4	753	**0.5**	HI	Forty reactions overall. Four of 40 reactions were monotypic while 36 of 40 were heterotypic.	[63]
	11	102	**10.8**	HI	Not clear if the 11 reactions are monotypic or heterotypic.	[57]
	10	748	**1.5**	Plaque reduction NT	Forty-four horses had neutralizing antibody (titer $\geq$ 10) against MAYV, but only ten met the diagnostic criteria of 4-fold greater plaque reduction NT_{90} titer than the three other viruses (VEEV, EEEV, WEEV). Positive samples had titers of 1:20 (n=6) and 1:40 (n=4)	[86]
Domestic Cattle/Buffalo	5	1103	**0.5**	HI	Positive reactions were considered any reaction with a titer equal to or greater than 1:20. Fourteen reactions overall, and five of 14 reactions were monotypic.	[62]
Domestic Dog	2	7	**28.6**	HI	N/A	[53]
Sentinel Hamster	1	N/A	**N/A**	RT-PCR		[82]
Sentinel Monkeys	2	13	**15.4**	HI	N/A	[98]

454 MAYV: Mayaro virus; HI: hemagglutination inhibition; ELISA: enzyme-linked immunosorbent assay; RT-PCR:
455 reverse transcription polymerase chain reaction; NT: neutralization test; VEEV: Venezuelan equine encephalitis
456 virus; EEEV: Eastern equine encephalitis virus; WEEV: Western equine encephalitis virus

457 [a] Denominators presented in this table reflect only studies that reported MAYV positivity. Complete data (including
458 MAYV-negative samples) are reflected in the seroprevalence meta-analysis and the Supplementary Tables.
459

Pooled prevalence of MAYV in non-human vertebrate animals

Twenty-four studies overall were included in the pooled prevalence meta-analysis. Eight studies were excluded because they did not clearly state how many animals were tested for MAYV within each order [55, 66, 72, 84, 89, 93] or did not present serologic results [60, 70]. Another study was excluded because authors reported the number of "Group A" positive serum samples, but did not specify individual viruses [98]. Studies were also excluded if they only reported sequence data or only included sentinel animals [82, 91, 95, 100]. Finally, a study that sampled bats exclusively was excluded because no MAYV-positive samples were reported in the order Chiroptera [92].

Eleven orders of nonhuman vertebrate animals (including domestic equids) were included in the meta-analysis. Orders were excluded from the analysis due to insufficient sample size (N<10) or if no MAYV-positive samples were reported. These include the orders Apodiformes (MAYV prevalence: 0/3), Caprimulgiformes (MAYV prevalence: 1/6), Chiroptera (MAYV prevalence: 0/1546), Crocodilia (MAYV prevalence: 0/87), Cuculiformes (MAYV prevalence: 0/5), Galliformes (MAYV prevalence: 0/1), Gruiformes (MAYV prevalence: 0/2), Psittaciformes (MAYV prevalence: 0/3), Tinamiformes (MAYV prevalence: 0/2), Pelecaniformes (MAYV prevalence: 0/2), and Podicipediformes (MAYV prevalence: 0/2).

The primate order appeared in 14 studies that were included in the meta-analysis. When all positive samples were included, the pooled MAYV seroprevalence among primates was 13.1% (95% CI: 4.3-25.1%) according to the random effects model, with statistically significant heterogeneity across studies (I^2 = 95%, p < 0.01). After excluding positive samples that were not confirmed by NT, the pooled MAYV seroprevalence among primates decreased to 4.9 (95% CI: 0.0-15.2; I^2 = 96%; p < 0.01) according to the random effects model. When the analyses were

repeated using the GLMM with logit transformation, seroprevalence estimates for primates decreased to 8.7% (95% CI: 3.1-22.0%) overall and to 0.7% (95% CI: 0.0-9.1%) when only NT-positive samples were included. Additional meta-analysis results for the various primate genera are presented in **S6 and S7 Tables**. The seroprevalence for the most frequently sampled primate genera was 32.2% (95% CI: 0.0-79.2%) for the *Alouatta* genus, 17.8% (95% CI: 8.6-28.5%) for the *Callithrix* genus, and 3.7% (95% CI: 0.0-11.1%) for the *Cebus/Sapajus* genus.

Meta-analysis results for additional non-human vertebrate orders are presented in **Table 7** and forest plots for mammal orders and avian orders are presented in **Figs 3 and 4,** respectively. When all positive samples were included in the analysis, the highest seroprevalence was observed in the orders Charadriiformes (prevalence: 7.1%; 95% CI: 2.2-13.8%) and Cingulata (prevalence: 3.0%; 95% CI: 0.0-24.5%). When the analysis was repeated using GLMM with logit transformation, the seroprevalence increased to 10.0% (95% CI: 2.7-30.8%) for the order Cingulata and 9.2% (95% CI: 4.4-18.2%) for the order Charadriiformes. All results of the sensitivity analysis using GLMM with logit transformation are reported in the **S5 Table**. An additional sensitivity analysis using fixed effects models is presented in the **S8 and S9 Tables**.

Table 7. Pooled Prevalence Table (Random effects with Freeman-Tukey double arcsine transformation)

Order	Positives Included[a]	Studies (n)	Total (n)	Positive (n)	Pooled Prevalence (%)	95% CI	I^2 (%)	τ^2	p-value
Mammals									
Primate	HI and NT	13	897	153	13.1	4.3; 25.1	95	0.0692	<0.01
	NT only	13	858	114	4.9	0.0; 15.2	96	0.0851	<0.01
Pilosa	HI and NT	7	297	15	0.0	0.0; 6.6	84	0.0338	<0.01
	NT only	7	296	14	0.0	0.0; 3.9	82	0.0305	<0.01
Rodentia	HI and NT	7	1557	90	1.3	0.0; 6.5	91	0.0160	<0.01
	NT only	7	1486	19	0.1	0.0; 3.7	90	0.0153	<0.01
Domestic Equids	HI and NT	6	1955	41	1.1	0.0; 4.5	90	0.0085	<0.01
	NT only	6	1940	26	0.0	0.0; 1.9	90	0.0087	<0.01
Didelphimorphia	HI and NT	6	369	25	2.0	0.0; 7.2	68	0.0101	<0.01
	NT only	6	353	9	0.1	0.0; 4.2	74	0.0141	<0.01

Carnivora Order	HI and NT	5	40	2	0.1	0.0; 8.1	0	0	0.71
	NT only	5	40	2	0.1	0.0; 8.1	0	0	0.71
Cingulata Order	HI and NT	4	70	6	3.0	0.0; 24.5	35	0.0198	0.20
	NT only	4	70	6	3.0	0.0; 24.5	35	0.0198	0.20
Artiodactyla	HI and NT	2	26	1	2.3	0.0; 20.7	46	0.0172	0.17
	NT only	2	26	1	2.3	0.0; 20.7	46	0.0172	0.17
Birds[b]									
Charadriiformes	HI and NT	3	641	71	7.1	2.2; 13.8	61	0.0045	0.08
Passeriformes	HI and NT	4	1166	14	0.0	0.0; 0.0	27	0.0010	0.25
Columbiformes	HI and NT	4	171	35	2.2	0.0; 27.1	87	0.0591	<0.01

MAYV: Mayaro virus; HI: hemagglutination inhibition; NT: neutralization test; CI: confidence interval

[a] The first analysis (HI and NT) included all positive samples, regardless of test method. A sensitivity analysis was conducted that included only positive samples that were confirmed with NT.

[b] Only one study reporting MAYV positivity in birds used confirmatory NT. Therefore, a sensitivity analysis was not conducted.

Fig 3. Forest plots of mammal orders from meta-analysis of pooled MAYV seroprevalence. Estimates are based on random effects model with Freeman-Tukey double arcsine transformation. All samples that tested MAYV-positive are included, regardless of test method.

Fig 4. Forest plots of avian orders from meta-analysis of pooled MAYV seroprevalence. Estimates are based on random effects model with Freeman-Tukey double arcsine transformation. All samples that tested MAYV-positive are included, regardless of test method.

MAYV in wild-caught arthropods

Twenty-eight of the studies in our systematic review analyzed MAYV infection in wild-caught arthropods. Seventeen (61%) of the 28 studies reported at least one arthropod that was positive for MAYV infection. Of the mosquito genera studied, seven were found to be infected with MAYV: *Aedes, Culex, Haemagogus, Psorophora, Sabethes, Wyeomyia,* and *Mansonia*. For detailed information on all infected mosquito species, see **Table 8**. The majority of infected vectors were identified using viral isolation techniques, although three studies reported MAYV positivity using RT-PCR alone. In addition, one study reported isolation of MAYV from an *Ixodes* tick [91] while another study reported isolation from a *Gigantolaelaps* mite [52].

Complete results, including studies that did not detect MAYV in arthropods, are reported in the **S10 Table**.

The geographic distribution of vectors infected with MAYV is presented in **Fig 2**. MAYV-positive arthropods were identified in four countries overall, including Brazil, Colombia, Panama, and Trinidad. Overall, 15 locations were geo-referenced as points, two locations as ADM1 polygons, two locations as ADM2 polygons, two locations as ADM3 polygons, and two as custom polygons.

Table 8. Evidence of MAYV infection in arthropods

Genus	Species	Notes	Year	Citation
Haemagogus	*Hg. janthinomys*	Pools of *Hg. janthinomys* yielded nine isolates by injection into suckling mice	1978	[19]
		A pool of two *Hg. janthinomys* yielded one strain by inoculation into newborn mice and C6/36 cells and confirmed by complement fixation and immunofluorescent assays	2008	[58]
		Mayaro virus isolate BeAr505578, complete genome. GenBank accession no. KY618129	1991	GenBank: KY618129
		Mayaro virus isolate BeAr505411. Genbank accession no. DQ487382	1991	[91]
	Hg. equinus	One MAYV isolate detected by viral culture using Vero cells with confirmation in microplates.	1973-76	[72]
	Hg. lucifer	Two MAYV isolates detected by viral culture using Vero cells with confirmation in microplates.	1973-76	[72]
	NA	Twenty-five isolates reported. No further information provided.	NA	[52]
		Mayaro virus isolate BeAr350396. GenBank accession no. DQ487388	1978	[91]
		Complete Genome Sequence of Mayaro Virus Strain BeAr 20290. GenBank accession no. KT754168.	1960	[68]
Aedes	*Ae. aegypti*	Two out of 57 (3.5%) pools positive by PCR and isolated in C6/36 cells.	2017	[79]
		Four out of 171 (2.3%) pools positive by RT-PCR. One pool yielded an isolate after inoculation in Vero cells.	2013	[96]
	Ae. serratus	Addendum to the article states that one additional MAYV strain was isolated from *Ae. serratus* pools. No further	1960	[75]

		information provided.		
Mansonia	*M. venezuelensis*[a]	MAYV was isolated in baby mice from a pool of 49 wild-caught *M. venezuelensis* mosquitoes.	1957	[48]
		One isolation. No further information provided. GenBank accession no. DQ487384.	1957	[52, 91]
Culex	*C. nigripalpus*	One pool out of 152 (0.7%) positive by RT-PCR.	2014-15	[77]
	C. quinqefasciatus	Twelve out of 403 (3%) pools positive by RT-PCR. One pool was isolated after inoculation in Vero cells.	2013	[96]
		Twelve out of 179 (6.7%) pools positive by RT-PCR and isolation in Vero cells.	2017-18	[69]
	C. vomerifer	Wild-caught mosquitoes were allowed to feed on caged hamsters. The sera of one hamster produced MAYV antibodies by HI.	1966	[71]
	NA	Mayaro virus strain BeAr757954, complete genome. GenBank accession no. KY618130.	2011	GenBank: KY618130
		One isolation. No further information provided.	NA	[52]
Psorophora	*P. ferox*	A pool of *P. ferox* yielded one isolate by inoculation into Swiss mice.	1959-62	[70]
		Addendum to the article states that five additional MAYV strains were isolated from *P. ferox* pools. No further information provided.	1960	[75]
	NA	Four out of 748 (0.5%) pools yielded strains isolated by inoculation into Swiss mice from. Pools of 50 mosquitoes each were composed of *P. albipes*, *P. ferox*, or a combination of the two.	1958	[75]
Wyeomyia	NA	One pool out of 304 (0.3%) positive by RT-PCR.	2006-14	[64]
Sabethes	NA	Two isolations. No further information provided	NA	[52]
Gigantolaelaps	NA	One isolation. No further information provided	NA	[52]
Ixodes	NA	Genbank accession no. DQ487378	1961	[91]

[a] The mosquito *Mansonia venezuelensis* is now referred to as *Coquillettidia venezuelensis.*

Analysis of publication bias

Publication bias was assessed among six animal orders (including domestic equids) and two primate genera. The results of Egger's test did not reveal evidence of publication bias for the included studies. Therefore, the Trim fill technique was not carried out. Funnel plots are presented in **S1 and S2 Figures**, and results of Egger's test are reported in the **S11 Table**.

Discussion

To our knowledge, this study is the first attempt to systematically review the existing evidence of non-human animal reservoirs and arthropod vectors of MAYV, and the first study to quantitatively analyze the pooled seroprevalence of potential reservoirs. We identified 57 studies that assessed MAYV infection in non-human vertebrate animals and arthropods. Overall, the studies found evidence of MAYV infection in 12 wild-caught animal orders and seven arthropod genera across seven Latin American countries and the USA.

The majority of animal species that were found to be infected with MAYV belonged to the orders Primate and Charadriiformes (shorebirds). Several MAYV-positive species were also detected in the orders Rodentia, Didelphimorphia, and Pilosa. Overall, the highest MAYV pooled prevalence occurred in the Primate order. This finding points to the potential role of NHPs as an important reservoir in the MAYV transmission cycle.

The role of NHPs in sylvatic transmission cycles of arboviruses has been demonstrated with varying degrees of evidence [101]. Several arboviruses have been successfully isolated from wild NHPs, including dengue [102], CHIKV [103], and Zika [104] viruses. While isolation of a virus from NHPs is important for establishing the existence of a sylvatic cycle, it is difficult to achieve due to the short duration of viremia [101]. In our review, we identified only one study that successfully isolated MAYV from a NHP [19]. In the absence of viral detection, antibody seroprevalence has been used as evidence of the role of NHPs in sylvatic transmission cycles [105, 106]. Therefore, the high seroprevalence of MAYV among NHPs, including 52% seropositivity among *A. seniculus* monkeys in a 1994-95 survey in French Guiana [21], points to the potential importance of NHPs as MAYV reservoirs. Furthermore, Hoch et al. [19] reported

558 substantial viremia in *C. argentata* marmosets that were experimentally infected with MAYV
559 and noted that viremia titer was likely sufficient to infect vectors. Due to the high MAYV
560 seroprevalence among marmosets during the Belterra outbreak, the isolation of MAYV from a
561 single *C. argentata* marmoset, and the results of experimental infection studies, the authors
562 concluded that marmosets were likely the amplifying hosts of MAYV.

563 The importance of birds in the MAYV transmission cycle was hypothesized following
564 viral isolation from a migrating oriole (*Icterus spurius*) in Louisiana [60]. Avian species have
565 been implicated as definitive or potential reservoirs of several Alphaviruses, including Sindbis
566 virus [107], Ross River virus [108], and Eastern/Western equine encephalitis virus [109].
567 However, their role in MAYV transmission remains poorly understood. Our systematic review
568 identified seven studies that found MAYV positivity in birds in the orders Passeriformes,
569 Caprimulgiformes, Columbiformes, and Charadriiformes with relatively high seroprevalence
570 reported in several bird species in the latter two orders [52, 53]. While some have theorized that
571 MAYV has been introduced into certain areas by migratory birds [59], this hypothesis requires
572 further study in order to elucidate the role of birds in MAYV transmission.

573 Although evidence of MAYV infection was detected in several vertebrate species,
574 identifying the primary non-human animal reservoirs remains a difficult task. The precise
575 definition of a disease "reservoir" has been a source of disagreement [17, 110]. One definition
576 proposed by Haydon et al., (2002) defined a reservoir as "one or more epidemiologically
577 connected populations or environments in which the pathogen can be permanently maintained
578 and from which infection is transmitted to the defined target population" [17] . In addition, in
579 2005 Kuno and Chang outlined three basic criteria for the identification of reservoirs including
580 isolation of the virus from the suspected reservoir population, high antibody prevalence in field-

581 caught animals, and evidence of viremia in laboratory settings, although they posited that
582 definitive identification of a reservoir requires evidence of long-term infection [111]. The role of
583 various non-human vertebrates in the MAYV transmission cycle should be explored further in
584 longitudinal seroprevalence surveys and experimental transmission studies in laboratory settings.

585 The sylvatic *Hg. janthinomys* mosquito has long been considered as the primary vector of
586 MAYV. This is in part based on the isolation of MAYV from several pools of *Hg. janthinomys*
587 mosquitoes in the context of a major MAYV outbreak in Belterra, Brazil in 1978 [19]. Our
588 systematic review also identified several additional mosquito species including *Ae. aegypti* and
589 *Cx. quinqefasciatus* with evidence of MAYV infection. A caveat, however, is that the isolation
590 of a virus or detection of viral RNA through PCR is not sufficient to establish that arthropod as a
591 biological vector [112], i.e. involved in the biological transmission of pathogens [111]. The
592 World Health Organization (WHO) established three criteria to define a confirmed vector: (1)
593 viral isolation in the absence of vertebrate blood; (2) biological transmission of the virus in
594 experimental conditions; and (3) presence of certain temporal, geographic and other
595 epidemiological or ecological parameters that allow transmission to occur [112]. Thus, certain
596 arthropods that are capable of ingesting and transmitting a virus may not be established as
597 confirmed vectors if the other parameters are not in place.

598 Experimental transmission studies support the role of *Ae. aegypti* as a possible MAYV
599 vector with high MAYV infection rates and transmission potential [22-24, 113]. For example,
600 Long et al., revealed *Ae. aegypti* to be a capable MAYV vector with a relatively short extrinsic
601 incubation period [22]. Furthermore, MAYV titers in the saliva of *Ae. aegypti* were similar to
602 other Alphavirus-vector systems including EEEV in *Culiseta melanura* and VEEV in *Ae.*
603 *albopictus* and *Ae. taeniorhynchus*. In contrast, *Cx. quinquefasciatus* mosquitoes exhibited low

MAYV infection rates and inability to transmit MAYV in laboratory settings [113]. It is also important to note that the competence of a given vector species to transmit MAYV may be impacted by the MAYV genotype that is present in a given area. In laboratory conditions, genotype L infection rates were significantly higher than genotype D infection rates among *Ae. aegypti* mosquitoes [113].

The spillover of MAYV into urban populations has been a source of concern for Latin American health authorities [114]. The implication is that anthropophilic, urban-dwelling mosquitoes like *Ae. aegypti* as effective vectors of MAYV would increase the potential for urban MAYV outbreaks [115]. Concerns of urban MAYV transmission were amplified after antibodies to MAYV were discovered in 33 of 631 sera (5.2%) in the city of Manaus, Brazil in 2007-08 [25] although it is unclear if humans can serve as amplification hosts. For example, Long et al. noted that the short duration of MAYV viremia and the relatively low viremic titers in humans reduces the probability of urban spread [22]. Our systematic review identified two recent studies conducted in the city of Cuiaba in which MAYV was isolated from pools of wild-caught *Ae. aegypti* mosquitoes [79, 96]. One of these studies also reported vertical transmission of MAYV [79]. This represents another mechanism that may lead to maintenance of the virus in urban mosquito populations. Although *Ae. aegypti* mosquitoes have not been conclusively implicated as MAYV vectors, the isolation of MAYV from wild-caught *Ae. aegypti* mosquitoes combined with the evidence of vector competence in laboratory settings [22-24, 113] suggests that MAYV could spill over into an urban cycle. This hypothesis requires further study to explore natural MAYV infection in city-dwelling mosquitoes and additional controlled vector competence studies.

Our systematic review revealed substantial heterogeneity across included studies, even within animal orders. Heterogeneity may complicate the interpretation of pooled seroprevalence estimates [38]. An additional limitation involves the validity of serological assays used to detect MAYV infection in animals. While plaque reduction NT is considered the "gold standard test" for detecting neutralizing antibodies to MAYV, some of the studies in the review instead relied on the less-specific HI test for antibody detection [101]. Furthermore, antibodies to other alphaviruses in the Semliki Forest serocomplex (e.g., CHIKV) may cross-react in serological tests [116]. Therefore, interpretation of seroprevalence estimates should be done with caution especially in the absence of confirmatory NT. Finally, unpublished data and articles with low quality scores were included in this review due to the paucity of eligible studies. Therefore, readers should consider the heterogeneity of study quality when interpreting the results of pooled seroprevalence estimates

.

Conclusions

MAYV is an emerging arbovirus that poses a major threat to human populations in Latin America. In order for public health authorities to effectively design MAYV surveillance and control programs, an understanding of the disease ecology is essential. This systematic review adds to existing knowledge regarding the potential animal reservoirs and arthropod vectors that are involved in the MAYV transmission cycle. These baseline data and maps of MAYV occurrence can direct risk emergence modeling and prediction efforts. Future studies involving experimental infection of primates and other non-human vertebrates are necessary to determine the animal species that may serve as amplifying hosts. Furthermore, additional experimental

transmission studies may provide critical information regarding the potential for *Ae. aegypti* to facilitate urban spread of MAYV.

Acknowledgments

We would like to thank Dr. Mauro Ramos for his assistance with reviewing Portuguese language articles. ELE is a Scientific researcher of the Consejo de Investigaciones Científicas y Tecnológicas (CONICET) from Argentina.

FUNDING STATEMENT: This work was in part conducted by the Infectious Disease Clinical Research Program (IDCRP), a Department of Defense (DoD) program executed by the Uniformed Services University of the Health Sciences (USU) through a cooperative agreement with The Henry M. Jackson Foundation for the Advancement of Military Medicine, Inc. (HJF). This project has been supported with federal funds from the National Institute of Allergy and Infectious Diseases, National Institutes of Health (NIH), under Inter□Agency Agreement Y1-Al-5072 and from the Defense Health Program, U.S. Department of Defense, under award HU0001190002.

A.A.'s participation in this work was supported by funding from Armed Forces Health Surveillance Branch - Global Emerging Infections Surveillance (GEIS) Project #P0044_20_NS and NASA Applied Sciences Program – Health and Air Quality, Grant #17-HAQ17-0065.

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Navy, or Air Force. Mention of trade names, commercial products, or organizations does not imply endorsement by the U.S. Government.

CONFLICT OF INTEREST: The authors declare no conflicts of interest.

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676 References
677
678 1. Anderson CR, Downs WG, Wattley GH, Ahin NW, Reese AA. Mayaro virus: a new
679 human disease agent. II. Isolation from blood of patients in Trinidad, B.W.I. Am J Trop Med
680 Hyg. 1957;6(6):1012-6. Epub 1957/11/01. doi: 10.4269/ajtmh.1957.6.1012. PubMed PMID:
681 13487973.

682 2. Suhrbier A, Jaffar-Bandjee MC, Gasque P. Arthritogenic alphaviruses--an overview. Nat
683 Rev Rheumatol. 2012;8(7):420-9. Epub 2012/05/09. doi: 10.1038/nrrheum.2012.64. PubMed
684 PMID: 22565316.

685 3. Pan American Health Organization / World Health Organization. Epidemiological Alert:
686 Mayaro Fever. Washington, D.C.: PAHO/WHO: 2019 May 1, 2019. Report No.

687 4. Causey OR, Maroja OM. Mayaro virus: a new human disease agent. III. Investigation of
688 an epidemic of acute febrile illness on the river Guama in Para, Brazil, and isolation of Mayaro
689 virus as causative agent. Am J Trop Med Hyg. 1957;6(6):1017-23. Epub 1957/11/01. PubMed
690 PMID: 13487974.

691 5. LeDuc JW, Pinheiro FP, Travassos da Rosa AP. An outbreak of Mayaro virus disease in
692 Belterra, Brazil. II. Epidemiology. Am J Trop Med Hyg. 1981;30(3):682-8. Epub 1981/05/01.
693 doi: 10.4269/ajtmh.1981.30.682. PubMed PMID: 6266264.

694 6. Schaeffer M, Gajdusek DC, Lema AB, Eichenwald H. Epidemic jungle fevers among
695 Okinawan colonists in the Bolivian rain forest. I. Epidemiology. Am J Trop Med Hyg.
696 1959;8(3):372-96. doi: 10.4269/ajtmh.1959.8.372.

697 7. Auguste AJ, Liria J, Forrester NL, Giambalvo D, Moncada M, Long KC, et al.
698 Evolutionary and Ecological Characterization of Mayaro Virus Strains Isolated during an
699 Outbreak, Venezuela, 2010. Emerg Infect Dis. 2015;21(10):1742-50. Epub 2015/09/25. doi:
700 10.3201/eid2110.141660. PubMed PMID: 26401714; PubMed Central PMCID:
701 PMCPMC4593426.

8. Forshey BM, Guevara C, Laguna-Torres VA, Cespedes M, Vargas J, Gianella A, et al. Arboviral etiologies of acute febrile illnesses in Western South America, 2000-2007. PLoS Negl Trop Dis. 2010;4(8):e787. Epub 2010/08/14. doi: 10.1371/journal.pntd.0000787. PubMed PMID: 20706628; PubMed Central PMCID: PMCPMC2919378.

9. Jonkers AH, Spence L, Karbaat J. Arbovirus infections in Dutch military personnel stationed in Surinam. Further studies. Trop Geogr Med. 1968;20(3):251-6. Epub 1968/09/01. PubMed PMID: 5683357.

10. Navarrete-Espinosa J, Gomez-Dantes H. Arbovirus causales de fiebre hemorrágica en pacientes del Instituto Mexicano del Seguro Social. Rev Med Inst Mex Seguro Soc. 2006;44(4):347-53. Epub 2006/08/15. PubMed PMID: 16904038.

11. Groot H. Estudios sobre virus transmitidos por artropodos en Colombia. Rev Acad Colomb Cienc. 1964;12(46):191-217. doi: 10.18257/raccefyn.565.

12. Talarmin A, Chandler LJ, Kazanji M, de Thoisy B, Debon P, Lelarge J, et al. Mayaro virus fever in French Guiana: isolation, identification, and seroprevalence. Am J Trop Med Hyg. 1998;59(3):452-6. Epub 1998/09/28. doi: 10.4269/ajtmh.1998.59.452. PubMed PMID: 9749643.

13. Blohm G, Elbadry MA, Mavian C, Stephenson C, Loeb J, White S, et al. Mayaro as a Caribbean traveler: Evidence for multiple introductions and transmission of the virus into Haiti. Int J Infect Dis. 2019;87:151-3. Epub 2019/08/06. doi: 10.1016/j.ijid.2019.07.031. PubMed PMID: 31382049.

14. Izurieta RO, Macaluso M, Watts DM, Tesh RB, Guerra B, Cruz LM, et al. Hunting in the Rainforest and Mayaro Virus Infection: An emerging Alphavirus in Ecuador. J Glob Infect Dis. 2011;3(4):317-23. Epub 2012/01/10. doi: 10.4103/0974-777x.91049. PubMed PMID: 22223990; PubMed Central PMCID: PMCPMC3249982.

15. Plowright RK, Parrish CR, McCallum H, Hudson PJ, Ko AI, Graham AL, et al. Pathways to zoonotic spillover. Nat Rev Microbiol. 2017;15(8):502-10. Epub 2017/05/31. doi: 10.1038/nrmicro.2017.45. PubMed PMID: 28555073; PubMed Central PMCID: PMCPMC5791534.

16. Viana M, Mancy R, Biek R, Cleaveland S, Cross PC, Lloyd-Smith JO, et al. Assembling evidence for identifying reservoirs of infection. Trends Ecol Evol. 2014;29(5):270-9. Epub 2014/04/15. doi: 10.1016/j.tree.2014.03.002. PubMed PMID: 24726345; PubMed Central PMCID: PMCPMC4007595.

17. Haydon DT, Cleaveland S, Taylor LH, Laurenson MK. Identifying reservoirs of infection: a conceptual and practical challenge. Emerg Infect Dis. 2002;8(12):1468-73. Epub 2002/12/25. doi: 10.3201/eid0812.010317. PubMed PMID: 12498665; PubMed Central PMCID: PMCPMC2738515.

18. Pezzi L, Reusken CB, Weaver SC, Drexler JF, Busch M, LaBeaud AD, et al. GloPID-R report on Chikungunya, O'nyong-nyong and Mayaro virus, part I: Biological diagnostics. Antiviral Res. 2019;166:66-81. Epub 2019/03/25. doi: 10.1016/j.antiviral.2019.03.009. PubMed PMID: 30905821.

19. Hoch AL, Peterson NE, LeDuc JW, Pinheiro FP. An outbreak of Mayaro virus disease in Belterra, Brazil. III. Entomological and ecological studies. Am J Trop Med Hyg. 1981;30(3):689-98. Epub 1981/05/01. doi: 10.4269/ajtmh.1981.30.689. PubMed PMID: 6266265.

20. Seymour C, Peralta PH, Montgomery GG. Serologic evidence of natural togavirus infections in Panamanian sloths and other vertebrates. Am J Trop Med Hyg. 1983;32(4):854-61. Epub 1983/07/01. doi: 10.4269/ajtmh.1983.32.854. PubMed PMID: 6309027.

21. de Thoisy B, Gardon J, Salas RA, Morvan J, Kazanji M. Mayaro virus in wild mammals, French Guiana. Emerg Infect Dis. 2003;9(10):1326-9. Epub 2003/11/12. doi: 10.3201/eid0910.030161. PubMed PMID: 14609474; PubMed Central PMCID: PMCPMC3033094.

22. Long KC, Ziegler SA, Thangamani S, Hausser NL, Kochel TJ, Higgs S, et al. Experimental transmission of Mayaro virus by Aedes aegypti. Am J Trop Med Hyg. 2011;85(4):750-7. Epub 2011/10/07. doi: 10.4269/ajtmh.2011.11-0359. PubMed PMID: 21976583; PubMed Central PMCID: PMCPMC3183788.

23. Wiggins K, Eastmond B, Alto BW. Transmission potential of Mayaro virus in Florida Aedes aegypti and Aedes albopictus mosquitoes. Med Vet Entomol. 2018;32(4):436-42. Epub 2018/07/15. doi: 10.1111/mve.12322. PubMed PMID: 30006976.

24. Brustolin M, Pujhari S, Henderson C, Rasgon J. Emergent viruses and their interactions in Aedes aegypti: Mayaro and zika virus coinfected mosquitoes can successfully transmit both pathogens. Am J Trop Med Hyg. 2019;101(5):50. doi: 10.4269/ajtmh.abstract2019.

25. Mourao MP, Bastos Mde S, de Figueiredo RP, Gimaque JB, Galusso Edos S, Kramer VM, et al. Mayaro fever in the city of Manaus, Brazil, 2007-2008. Vector Borne Zoonotic Dis. 2012;12(1):42-6. Epub 2011/09/20. doi: 10.1089/vbz.2011.0669. PubMed PMID: 21923266; PubMed Central PMCID: PMCPMC3249893.

26. Page MJ, McKenzie JE, Bossuyt PM, Boutron I, Hoffmann TC, Mulrow CD, et al. The PRISMA 2020 statement: An updated guideline for reporting systematic reviews. Int J Surg. 2021;88:105906. Epub 2021/04/02. doi: 10.1016/j.ijsu.2021.105906. PubMed PMID: 33789826.

27. Clark K, Karsch-Mizrachi I, Lipman DJ, Ostell J, Sayers EW. GenBank. Nucleic Acids Res. 2016;44(D1):D67-D72. Epub 2015/11/20. doi: 10.1093/nar/gkv1276. PubMed PMID: 26590407.

28. Ding H, Gao YM, Deng Y, Lamberton PH, Lu DB. A systematic review and meta-analysis of the seroprevalence of Toxoplasma gondii in cats in mainland China. Parasit Vectors. 2017;10(1):27. Epub 2017/01/15. doi: 10.1186/s13071-017-1970-6. PubMed PMID: 28086987; PubMed Central PMCID: PMCPMC5237326.

29. Rodríguez-Monguí E, Cantillo-Barraza O, Prieto-Alvarado FE, Cucunubá ZM. Heterogeneity of Trypanosoma cruzi infection rates in vectors and animal reservoirs in Colombia: a systematic review and meta-analysis. Parasit Vectors. 2019;12(1):308. Epub 2019/06/22. doi: 10.1186/s13071-019-3541-5. PubMed PMID: 31221188; PubMed Central PMCID: PMCPMC6585012.

30. Guernier V, Goarant C, Benschop J, Lau CL. A systematic review of human and animal leptospirosis in the Pacific Islands reveals pathogen and reservoir diversity. PLoS Negl Trop Dis.

783 2018;12(5):e0006503. Epub 2018/05/15. doi: 10.1371/journal.pntd.0006503. PubMed PMID:
784 29758037; PubMed Central PMCID: PMCPMC5967813.

785 31. ESRI. ArcGIS Desktop: Release 10. Redlands, CA: Environmental Systems Research
786 Institute.; 2011.

787 32. Acosta-Ampudia Y, Monsalve DM, Rodriguez Y, Pacheco Y, Anaya JM, Ramirez-
788 Santana C. Mayaro: an emerging viral threat? Emerg Microbes Infect. 2018;7(1):163. Epub
789 2018/09/27. doi: 10.1038/s41426-018-0163-5. PubMed PMID: 30254258; PubMed Central
790 PMCID: PMCPMC6156602.

791 33. Haidich AB. Meta-analysis in medical research. Hippokratia. 2010;14(Suppl 1):29-37.
792 Epub 2011/04/14. PubMed PMID: 21487488; PubMed Central PMCID: PMCPMC3049418.

793 34. Higgins JPT, Thomas J, Chandler J, Cumpston M, Li T, Page MJ, et al. Cochrane
794 Handbook for Systematic Reviews of Interventions version 6.0 Cochrane; 2019. Available from:
795 www.training.cochrane.org/handbook.

796 35. Barendregt JJ, Doi SA, Lee YY, Norman RE, Vos T. Meta-analysis of prevalence. J
797 Epidemiol Community Health. 2013;67(11):974-8. Epub 2013/08/22. doi: 10.1136/jech-2013-
798 203104. PubMed PMID: 23963506.

799 36. Schwarzer G, Chemaitelly H, Abu-Raddad LJ, Rücker G. Seriously misleading results
800 using inverse of Freeman-Tukey double arcsine transformation in meta-analysis of single
801 proportions. Res Synth Methods. 2019;10(3):476-83. Epub 2019/04/05. doi: 10.1002/jrsm.1348.
802 PubMed PMID: 30945438; PubMed Central PMCID: PMCPMC6767151.

803 37. Warton DI, Hui FK. The arcsine is asinine: the analysis of proportions in ecology.
804 Ecology. 2011;92(1):3-10. Epub 2011/05/13. doi: 10.1890/10-0340.1. PubMed PMID:
805 21560670.

806 38. Higgins JPT, Thompson SG, Deeks JJ, Altman DG. Measuring inconsistency in meta-
807 analyses BMJ. 2003;327:557-60.

808 39. Quintana DS. From pre-registration to publication: a non-technical primer for conducting
809 a meta-analysis to synthesize correlational data. Front Psychol. 2015;6(1549). doi:
810 10.3389/fpsyg.2015.01549.

811 40. Balduzzi S, Rücker G, Schwarzer G. How to perform a meta-analysis with R: a practical
812 tutorial. Evid Based Ment Health. 2019;22(4):153-60. Epub 2019/09/30. doi: 10.1136/ebmental-
813 2019-300117. PubMed PMID: 31563865.

814 41. Egger M, Davey Smith G, Schneider M, Minder C. Bias in meta-analysis detected by a
815 simple, graphical test. BMJ. 1997;315(7109):629-34. Epub 1997/10/06. doi:
816 10.1136/bmj.315.7109.629. PubMed PMID: 9310563; PubMed Central PMCID:
817 PMCPMC2127453.

818 42. Duval S, Tweedie R. Trim and fill: A simple funnel-plot-based method of testing and
819 adjusting for publication bias in meta-analysis. Biometrics. 2000;56(2):455-63. Epub
820 2000/07/06. doi: 10.1111/j.0006-341x.2000.00455.x. PubMed PMID: 10877304.

821 43. Messina JP, Brady OJ, Pigott DM, Brownstein JS, Hoen AG, Hay SI. A global
822 compendium of human dengue virus occurrence. Sci Data. 2014;1:140004. Epub 2014/01/01.
823 doi: 10.1038/sdata.2014.4. PubMed PMID: 25977762; PubMed Central PMCID:
824 PMCPMC4322574.

825 44. Pigott DM, Golding N, Messina JP, Battle KE, Duda KA, Balard Y, et al. Global
826 database of leishmaniasis occurrence locations, 1960-2012. Sci Data. 2014;1:140036. Epub
827 2014/01/01. doi: 10.1038/sdata.2014.36. PubMed PMID: 25984344; PubMed Central PMCID:
828 PMCPMC4432653.

829 45. Runfola D, Anderson A, Baier H, Crittenden M, Dowker E, Fuhrig S, et al.
830 geoBoundaries: A global database of political administrative boundaries. PloS One.
831 2020;15(4):e0231866. Epub 2020/04/25. doi: 10.1371/journal.pone.0231866. PubMed PMID:
832 32330167; PubMed Central PMCID: PMCPMC7182183 Allen Hamilton, and Deloitte,
833 respectively. This does not alter our adherence to PLOS ONE policies on sharing data and
834 materials.

835 46. de Thoisy B, Vogel I, Reynes JM, Pouliquen JF, Carme B, Kazanji M, et al. Health
836 evaluation of translocated free-ranging primates in French Guiana. Am J Primatol. 2001;54(1):1-
837 16. Epub 2001/05/01. doi: 10.1002/ajp.1008. PubMed PMID: 11329164.

838 47. Aitken TH, Downs WG, Anderson CR, Spence L, Casals J. Mayaro virus isolated from a
839 Trinidadian mosquito, Mansonia venezuelensis. Science (New York, NY). 1960;131(3405):986.
840 Epub 1960/04/01. doi: 10.1126/science.131.3405.986. PubMed PMID: 13792204.

841 48. Aitken TH, Spence L, Jonkers AH, Downs WG. A 10-year survey of Trinidadian
842 arthropods for natural virus infections (1953-1963). J Med Entomol. 1969;6(2):207-15. Epub
843 1969/05/01. doi: 10.1093/jmedent/6.2.207. PubMed PMID: 5807863.

844 49. Batista PM, Andreotti R, Almeida PS, Marques AC, Rodrigues SG, Chiang JO, et al.
845 Detection of arboviruses of public health interest in free-living New World primates (Sapajus
846 spp.; Alouatta caraya) captured in Mato Grosso do Sul, Brazil. Rev Soc Bras Med Trop.
847 2013;46(6):684-90. Epub 2014/01/30. doi: 10.1590/0037-8682-0181-2013. PubMed PMID:
848 24474008.

849 50. Paulo M, Renato A, Da Carneiro Rocha T, Eliane C, Navarro da Silva M. Serosurvey of
850 arbovirus in free-living non-human primates (Sapajus spp.) in Brazil. J Environ Anal Chem.
851 2015;2(155):2380-91.1000155.

852 51. Woodall JP. Virus Research in Amazonia. Atas do Simpósio Sobre a Biota Amazônica;
853 Para, Brazil1967. p. 31-63.

854 52. Taylor RM. Catalogue of arthropod-borne viruses of the world: a collection of data on
855 registered arthropod-borne animal viruses: US Public Health Service; 1967.

856 53. Araujo FAA, Wada MY, da Silva EV, Cavalcante GC, Magalhaes VS, de Andrade Filho
857 GV, et al. Primeiro inquérito sorológico em aves migratórias e nativas do Parque Nacional da
858 Lagoa do Peixe/RS para detecção do vírus do Nilo Ocidental. In: Ministério da Saúde Secretaria
859 de Vigilância em Saúde, editor. Boletim Eletrônico Epidemiologico, 2003.

860 54. Araújo FAA, Vianna RdST, Andrade Filho GVd, Melhado DL, Todeschini B, Cavalcante
861 e Cavalcanti G, et al. Segundo inquérito sorológico em aves migratórias e residentes do parque

862 nacional da Lagoa do Peixe/RS para detecção do vírus da Febre da Febre do Nilo Ocidental e
863 outros vírus. In: Ministério da Saúde Secretaria de Vigilância em Saúde, editor. Boletim
864 Eletrônico Epidemiologico, 2004.

865 55. Araújo FAA, Vianna RdST, Wada MY, Silva ÉVd, Doretto L, Cavalcante GCe, et al.
866 Inquérito sorológico em aves migratórias e residentes de Galinhos/RN para detecção do vírus da
867 Febre do Nilo Ocidental e outros vírus. In: Ministério da Saúde Secretaria de Vigilância em
868 Saúde, editor. Boletim Eletrônico Epidemiológico, 2004.

869 56. Araujo FAA, Lima PC, Andrade MA, de Sá Jayme V, Ramos DG, Da Silveira SL.
870 Soroprevalência de anticorpos “anti-arbovírus” de importância em saúde pública em aves
871 selvagens, Brasil–2007 e 2008. Ciênc Anim Brasil. 2012;13(1):115-23. doi:
872 10.5216/cab.v13i1.16834.

873 57. Araujo FAA, Andrade MA, Jayme VS, Santos AL, Roman APM, Ramos DG, et al.
874 Anticorpos antialfavírus detectados em equinos durante diferentes epizootias de encefalite
875 equina, Paraíba, 2009. Rev Bras Ciênc Vet. 2012;19(1):80-5. doi: 10.4322/rbcv.2014.086.

876 58. Azevedo RS, Silva EV, Carvalho VL, Rodrigues SG, Neto JPN, Monteiro HA, et al.
877 Mayaro fever virus, Brazilian amazon. Emerg Infect Dis. 2009;15(11):1830. doi:
878 10.3201/eid1511.090461.

879 59. Batista PM, Andreotti R, Chiang JO, Ferreira MS, Vasconcelos PF. Seroepidemiological
880 monitoring in sentinel animals and vectors as part of arbovirus surveillance in the state of Mato
881 Grosso do Sul, Brazil. Rev Soc Bras Med Trop. 2012;45(2):168-73. Epub 2012/04/27. doi:
882 10.1590/s0037-86822012000200006. PubMed PMID: 22534986.

883 60. Calisher CH, Gutierrez E, Maness KS, Lord RD. Isolation of Mayaro virus from a
884 migrating bird captured in Louisiana in 1967. Bull Pan Am Health Organ. 1974;8(3):243-8. Epub
885 1974/01/01. PubMed PMID: 4418030.

886 61. Carrera JP, Cucunubá ZM, Neira K, Lambert B, Pittí Y, Liscano J, et al. Endemic and
887 Epidemic Human Alphavirus Infections in Eastern Panama: An Analysis of Population-Based

888 Cross-Sectional Surveys. Am J Trop Med Hyg. 2020. Epub 2020/10/31. doi: 10.4269/ajtmh.20-
889 0408. PubMed PMID: 33124532.

890 62. Casseb AdR. Soroprevalência de anticorpos e padronização do teste ELISA sanduíche
891 indireto para 19 tipos de arbovírus em herbívoros domésticos [Ph.D. Thesis]. Belém:
892 Universidade Federal do Pará; 2010. Available from:
893 http://repositorio.ufpa.br/jspui/handle/2011/4760.

894 63. Casseb AdR, Brito TC, Silva MRMd, Chiang JO, Martins LC, Silva SPd, et al.
895 Prevalence of antibodies to equine alphaviruses in the State of Pará, Brazil. Arq Inst Biol.
896 2016;83. doi: 10.1590/1808-1657000202014.

897 64. Catenacci LS. Abordagem one health para vigilância de arbovirus na Mata Atlântica do
898 sul da Bahia, Brasil. [Ph.D. Thesis]. Ananindeua: Instituto Evandro Chagas; 2017. Available
899 from: https://patua.iec.gov.br/handle/iec/3073.

900 65. Cruz ACR, Prazeres AdSCd, Gama EC, Lima MFd, Azevedo RdSS, Casseb LMN, et al.
901 Vigilância sorológica para arbovírus em Juruti, Pará, Brasil. Cadernos de saude publica.
902 2009;25(11):2517-23.

903 66. Degallier N, Travassos da Rosa AP, Vasconcelos PFC, Hervé JP, Sa Filho GC, Travassos
904 da Rosa JFS, et al. Modifications of arbovirus transmission in relation to construction of dams in
905 Brazilian Amazonia Journal of the Brazilian Association for the Advancement of Science.
906 1992;44.

907 67. Diaz LA, Diaz Mdel P, Almiron WR, Contigiani MS. Infection by UNA virus
908 (Alphavirus; Togaviridae) and risk factor analysis in black howler monkeys (Alouatta caraya)
909 from Paraguay and Argentina. Trans R Soc Trop Med Hyg. 2007;101(10):1039-41. Epub
910 2007/07/31. doi: 10.1016/j.trstmh.2007.04.009. PubMed PMID: 17658571.

911 68. Esposito DL, da Fonseca BA. Complete Genome Sequence of Mayaro Virus
912 (Togaviridae, Alphavirus) Strain BeAr 20290 from Brazil. Genome Announc. 2015;3(6). Epub
913 2015/12/19. doi: 10.1128/genomeA.01372-15. PubMed PMID: 26679574; PubMed Central
914 PMCID: PMCPMC4683219.

915 69. da Silva Ferreira R, de Toni Aquino da Cruz LC, Souza VJ, da Silva Neves NA, de Souza
916 VC, Filho LCF, et al. Insect-specific viruses and arboviruses in adult male culicids from
917 Midwestern Brazil. Infect Genet Evol. 2020:104561. Epub 2020/09/23. doi:
918 10.1016/j.meegid.2020.104561. PubMed PMID: 32961364.

919 70. Galindo P, Srihongse S, De Rodaniche E, Grayson MA. An ecological survey for
920 arboviruses in Almirante, Panama, 1959-1962. Am J Trop Med Hyg. 1966;15(3):385-400. Epub
921 1966/05/01. doi: 10.4269/ajtmh.1966.15.385. PubMed PMID: 4380043.

922 71. Galindo P, Srihongse S. Transmission of arboviruses to hamsters by the bite of naturally
923 infected Culex (Melanoconion) mosquitoes. Am J Trop Med Hyg. 1967;16(4):525-30. Epub
924 1967/07/01. doi: 10.4269/ajtmh.1967.16.525. PubMed PMID: 4952151.

925 72. Galindo P, Adames A, Peralta P, Johnson C, Read R. Impacto de la hidroeléctrica de
926 Bayano en la transmisión de arbovirus. Rev Med Pan. 1983;8:89-134.

927 73. Gibrail MM. Detecção de anticorpos para arbovirus em primatas não humanos no
928 município de Goiânia, Goiás [M.Sc. Thesis]. Goiânia: Universidade Federal de Goiás; 2015.
929 Available from: https://repositorio.bc.ufg.br/tede/handle/tede/5552.

930 74. Gomes FA, Jansen AM, Machado RZ, Jesus Pena HF, Fumagalli MJ, Silva A, et al.
931 Serological evidence of arboviruses and coccidia infecting horses in the Amazonian region of
932 Brazil. PloS One. 2019;14(12):e0225895. Epub 2019/12/13. doi: 10.1371/journal.pone.0225895.
933 PubMed PMID: 31830142.

934 75. Groot H, Morales A, Vidales H. Virus isolations from forest mosquitoes in San Vicente
935 de Chucuri, Colombia. Am J Trop Med Hyg. 1961;10:397-402. Epub 1961/05/01. doi:
936 10.4269/ajtmh.1961.10.397. PubMed PMID: 13708940.

937 76. Henriques DA. Caracterização molecular de arbovírus isolados da fauna diptera
938 nematocera do Estado de Rondônia (Amazônia ocidental brasileira) [Ph.D. Thesis]. São Paulo:
939 Universidade de São Paulo; 2008. Available from:
940 https://teses.usp.br/teses/disponiveis/42/42132/tde-27032009-124003/pt-br.php.

941 77. Kubiszeski JR. Arboviroses emergentes no município de Sinop-MT: pesquisa de vetores
942 [Ph.D. Thesis]. Sinop: Universidade Federal de Mato Grosso; 2016. Available from:
943 https://teses.usp.br/teses/disponiveis/42/42132/tde-27032009-124003/pt-br.php.

944 78. Laroque PO, Valença-Montenegro MM, Ferreira DRA, Chiang JO, Cordeiro MT,
945 Vasconcelos PFC, et al. Levantamento soroepidemiológico para arbovírus em macaco-prego-
946 galego (Cebus flavius) de vida livre no estado da Paraíba e em macaco-prego (Cebus libidinosus)
947 de cativeiro do nordeste do Brasil. Pesq Vet Bras. 2014;34:462-8.

948 79. Maia LMS, Bezerra MCF, Costa MCS, Souza EM, Oliveira MEB, Ribeiro ALM, et al.
949 Natural vertical infection by dengue virus serotype 4, Zika virus and Mayaro virus in Aedes
950 (Stegomyia) aegypti and Aedes (Stegomyia) albopictus. Med Vet Entomol. 2019;33(3):437-42.
951 Epub 2019/02/19. doi: 10.1111/mve.12369. PubMed PMID: 30776139.

952 80. Martinez D, Hernandez C, Munoz M, Armesto Y, Cuervo A, Ramirez JD. Identification
953 of Aedes (Diptera: Culicidae) Species and Arboviruses Circulating in Arauca, Eastern Colombia.
954 Front Ecol Evol. 2020;8. doi: 10.3389/fevo.2020.602190. PubMed PMID:
955 WOS:000596835300001.

956 81. Medlin S, Deardorff ER, Hanley CS, Vergneau-Grosset C, Siudak-Campfield A, Dallwig
957 R, et al. Serosurvey of Selected Arboviral Pathogens in Free-Ranging, Two-Toed Sloths
958 (Choloepus Hoffmanni) and Three-Toed Sloths (Bradypus Variegatus) In Costa Rica, 2005-07. J
959 Wildl Dis. 2016;52(4):883-92. Epub 2016/08/02. doi: 10.7589/2015-02-040. PubMed PMID:
960 27479900; PubMed Central PMCID: PMCPMC5189659.

961 82. Medina G, Garzaro DJ, Barrios M, Auguste AJ, Weaver SC, Pujol FH. Genetic diversity
962 of Venezuelan alphaviruses and circulation of a Venezuelan equine encephalitis virus subtype
963 IAB strain during an interepizootic period. Am J Trop Med Hyg. 2015;93(1):7-10. Epub
964 2015/05/06. doi: 10.4269/ajtmh.14-0543. PubMed PMID: 25940191; PubMed Central PMCID:
965 PMCPMC4497907.

966 83. Moreira-Soto A, Carneiro ID, Fischer C, Feldmann M, Kummerer BM, Silva NS, et al.
967 Limited Evidence for Infection of Urban and Peri-urban Nonhuman Primates with Zika and

968 Chikungunya Viruses in Brazil. mSphere. 2018;3(1). doi: 10.1128/mSphere.00523-17. PubMed
969 PMID: WOS:000425277500024.

970 84. Nunes MR, Barbosa TF, Casseb LM, Nunes Neto JP, Segura Nde O, Monteiro HA, et al.
971 Eco-epidemiologia dos arbovirus na area de influencia da rodovia Cuiaba-Santarem (BR 163),
972 Estado do Para, Brasil. Cad Saude Publica. 2009;25(12):2583-602. Epub 2010/03/02. doi:
973 10.1590/s0102-311x2009001200006. PubMed PMID: 20191150.

974 85. Pauvolid-Correa A, Tavares FN, Costa EV, Burlandy FM, Murta M, Pellegrin AO, et al.
975 Serologic evidence of the recent circulation of Saint Louis encephalitis virus and high prevalence
976 of equine encephalitis viruses in horses in the Nhecolandia sub-region in South Pantanal,
977 Central-West Brazil. Mem Inst Oswaldo Cruz. 2010;105(6):829-33. Epub 2010/10/15. doi:
978 10.1590/s0074-02762010000600017. PubMed PMID: 20945001.

979 86. Pauvolid-Correa A, Juliano RS, Campos Z, Velez J, Nogueira RM, Komar N.
980 Neutralising antibodies for Mayaro virus in Pantanal, Brazil. Mem Inst Oswaldo Cruz.
981 2015;110(1):125-33. Epub 2015/03/06. doi: 10.1590/0074-02760140383. PubMed PMID:
982 25742272; PubMed Central PMCID: PMCPMC4371226.

983 87. Pauvolid-Correa A. Estudo sobre arbovírus em populações de eqüinos e artrópodes na
984 sub-região da Nhecolândia no Pantanal de Mato Grosso do Sul [M.Sc. Thesis]. Rio de Janeiro:
985 Fundação Oswaldo Cruz; 2008. Available from: https://www.arca.fiocruz.br/handle/icict/21142.

986 88. Perez JG, Carrera JP, Serrano E, Pitti Y, Maguina JL, Mentaberre G, et al. Serologic
987 Evidence of Zoonotic Alphaviruses in Humans from an Indigenous Community in the Peruvian
988 Amazon. Am J Trop Med Hyg. 2019. Epub 2019/10/02. doi: 10.4269/ajtmh.18-0850. PubMed
989 PMID: 31571566.

990 89. Pinheiro FP, Bensabath G, Andrade AH, Lins ZC, Fraihi H, Tang AT, et al. Infectious
991 diseases along Brazil's Trans-Amazon Highway: surveillance and research. Bull Pan Am Health
992 Organ. 1974;8(111).

993 90. Pinheiro GG, Rocha MN, de Oliveira MA, Moreira LA, Andrade JD. Detection of
994 Yellow Fever Virus in Sylvatic Mosquitoes during Disease Outbreaks of 2017-2018 in Minas

995 Gerais State, Brazil. Insects. 2019;10(5). doi: 10.3390/insects10050136. PubMed PMID:
996 WOS:000476846800018.

997 91. Powers AM, Aguilar PV, Chandler LJ, Brault AC, Meakins TA, Watts D, et al. Genetic
998 relationships among Mayaro and Una viruses suggest distinct patterns of transmission. Am J
999 Trop Med Hyg. 2006;75(3):461-9. Epub 2006/09/14. PubMed PMID: 16968922.

1000 92. Price JL. Serological evidence of infection of Tacaribe virus and arboviruses in
1001 Trinidadian bats. Am J Trop Med Hyg. 1978;27(1 Pt 1):162-7. Epub 1978/01/01. doi:
1002 10.4269/ajtmh.1978.27.162. PubMed PMID: 204207.

1003 93. Ragan IK, Hartwig A, Bowen RA. Cold blood: Reptiles and amphibians as reservoir and
1004 over wintering hosts for arboviruses. Am J Trop Med Hyg. 2019;101(5):261. doi:
1005 10.4269/ajtmh.abstract2019.

1006 94. Sanmartín C, Mackenzie RB, Trapido H, Barreto P, Mullenax CH, Gutiérrez E, et al.
1007 Encefalitis equina venezolana en Colombia, 1967. Bol Oficina Sanit Panam. 1973;74(2):108-37.
1008 Epub 1973/02/01. PubMed PMID: 4265714.

1009 95. Scherer WF, Madalengoitia J, Flores W, Acosta M. The first isolations of eastern
1010 encephalitis, group C, and Guama group arboviruses from the Peruvian Amazon region of
1011 western South America. Bull Pan Am Health Organ. 1975;9(1):19-26. Epub 1975/01/01.
1012 PubMed PMID: 238693.

1013 96. Serra OP, Cardoso BF, Ribeiro AL, Santos FA, Slhessarenko RD. Mayaro virus and
1014 dengue virus 1 and 4 natural infection in culicids from Cuiaba, state of Mato Grosso, Brazil.
1015 Mem Inst Oswaldo Cruz. 2016;111(1):20-9. Epub 2016/01/20. doi: 10.1590/0074-02760150270.
1016 PubMed PMID: 26784852; PubMed Central PMCID: PMCPMC4727432.

1017 97. Silva JWP. Aspectos ecológicos de vetores putativos do Vírus Mayaro e Vírus Oropuche
1018 em estratificação vertical e horizontal em ambientes florestais e antropizados em uma
1019 comunidade rural no Amazonas [M.Sc. Thesis]. Manaus, AM: Oswaldo Cruz Foundation,
1020 Instituto Leônidas and Maria Deane; 2017. Available from:
1021 https://www.arca.fiocruz.br/handle/icict/23337.

1022 98. Srihongse S, Galindo P, Eldridge BF. A survey to assess potential human disease hazards
1023 along proposed sea level canal routes in Panama and Colombia. V. Arbovirus infection in non
1024 human vertebrates. Mil Med. 1974;139(6):449-53.

1025 99. Tauro LB, Cardoso CW, Souza RL, Nascimento LC, Santos DRD, Campos GS, et al. A
1026 localized outbreak of Chikungunya virus in Salvador, Bahia, Brazil. Mem Inst Oswaldo Cruz.
1027 2019;114:e180597. Epub 2019/03/08. doi: 10.1590/0074-02760180597. PubMed PMID:
1028 30843962; PubMed Central PMCID: PMCPMC6396974.

1029 100. Turell MJ, Gozalo AS, Guevara C, Schoeler GB, Carbajal F, Lopez-Sifuentes VM, et al.
1030 Lack of Evidence of Sylvatic Transmission of Dengue Viruses in the Amazon Rainforest Near
1031 Iquitos, Peru. Vector Borne Zoonotic Dis. 2019;19(9):685-9. Epub 2019/04/10. doi:
1032 10.1089/vbz.2018.2408. PubMed PMID: 30964397; PubMed Central PMCID:
1033 PMCPMC6716187.

1034 101. Valentine MJ, Murdock CC, Kelly PJ. Sylvatic cycles of arboviruses in non-human
1035 primates. Parasit Vectors. 2019;12(1):463. Epub 2019/10/04. doi: 10.1186/s13071-019-3732-0.
1036 PubMed PMID: 31578140; PubMed Central PMCID: PMCPMC6775655.

1037 102. Cornet M, Saluzzo JF, Hervy JP, Digoutte JP, Germain M, Chauvancy MF. Dengue 2 au
1038 Sénégal oriental: une pousse épizootique en milieu selvatique; isolements du virus à partir de
1039 moustiques et d'un singe et considérations épidémiologiques. Cah Orstom Ser Ent Med Parasitol.
1040 1984;22:313-23.

1041 103. Diallo M, Thonnon J, Traore-Lamizana M, Fontenille D. Vectors of Chikungunya virus
1042 in Senegal: current data and transmission cycles. Am J Trop Med Hyg. 1999;60(2):281-6. Epub
1043 1999/03/11. doi: 10.4269/ajtmh.1999.60.281. PubMed PMID: 10072152.

1044 104. Dick GW, Kitchen SF, Haddow AJ. Zika virus. I. Isolations and serological specificity.
1045 Trans R Soc Trop Med Hyg. 1952;46(5):509-20. Epub 1952/09/01. doi: 10.1016/0035-
1046 9203(52)90042-4. PubMed PMID: 12995440.

1047 105. Althouse BM, Guerbois M, Cummings DAT, Diop OM, Faye O, Faye A, et al. Role of
1048 monkeys in the sylvatic cycle of chikungunya virus in Senegal. Nat Commun. 2018;9(1):1046.

1049 Epub 2018/03/15. doi: 10.1038/s41467-018-03332-7. PubMed PMID: 29535306; PubMed
1050 Central PMCID: PMCPMC5849707.

1051 106. Kading RC, Borland EM, Cranfield M, Powers AM. Prevalence of antibodies to
1052 alphaviruses and flaviviruses in free-ranging game animals and nonhuman primates in the greater
1053 Congo basin. J Wildl Dis. 2013;49(3):587-99. Epub 2013/06/20. doi: 10.7589/2012-08-212.
1054 PubMed PMID: 23778608.

1055 107. Lundström JO, Lindström KM, Olsen B, Dufva R, Krakower DS. Prevalence of sindbis
1056 virus neutralizing antibodies among Swedish passerines indicates that thrushes are the main
1057 amplifying hosts. J Med Entomol. 2001;38(2):289-97. Epub 2001/04/12. doi: 10.1603/0022-
1058 2585-38.2.289. PubMed PMID: 11296837.

1059 108. Stephenson EB, Peel AJ, Reid SA, Jansen CC, McCallum H. The non-human reservoirs
1060 of Ross River virus: a systematic review of the evidence. Parasit Vectors. 2018;11(1):188. Epub
1061 2018/03/21. doi: 10.1186/s13071-018-2733-8. PubMed PMID: 29554936; PubMed Central
1062 PMCID: PMCPMC5859426.

1063 109. Barba M, Fairbanks EL, Daly JM. Equine viral encephalitis: prevalence, impact, and
1064 management strategies. Vet Med (Auckl). 2019;10:99-110. Epub 2019/09/10. doi:
1065 10.2147/vmrr.S168227. PubMed PMID: 31497528; PubMed Central PMCID:
1066 PMCPMC6689664.

1067 110. Kuno G, Mackenzie JS, Junglen S, Hubálek Z, Plyusnin A, Gubler DJ. Vertebrate
1068 reservoirs of arboviruses: myth, synonym of amplifier, or reality? Viruses. 2017;9(7):185.

1069 111. Kuno G, Chang GJ. Biological transmission of arboviruses: reexamination of and new
1070 insights into components, mechanisms, and unique traits as well as their evolutionary trends.
1071 Clin Microbiol Rev. 2005;18(4):608-37. Epub 2005/10/15. doi: 10.1128/cmr.18.4.608-637.2005.
1072 PubMed PMID: 16223950; PubMed Central PMCID: PMCPMC1265912.

1073 112. World Health Organization Scientific Group. Arthropod-borne and rodent-borne viral
1074 diseases. Geneva, Switzerland: World Health Organization, 1985.

113. Pereira TN, Carvalho FD, De Mendonça SF, Rocha MN, Moreira LA. Vector competence of Aedes aegypti, Aedes albopictus, and Culex quinquefasciatus mosquitoes for Mayaro virus. PLoS Negl Trop Dis. 2020;14(4):e0007518. Epub 2020/04/15. doi: 10.1371/journal.pntd.0007518. PubMed PMID: 32287269; PubMed Central PMCID: PMCPMC7182273.

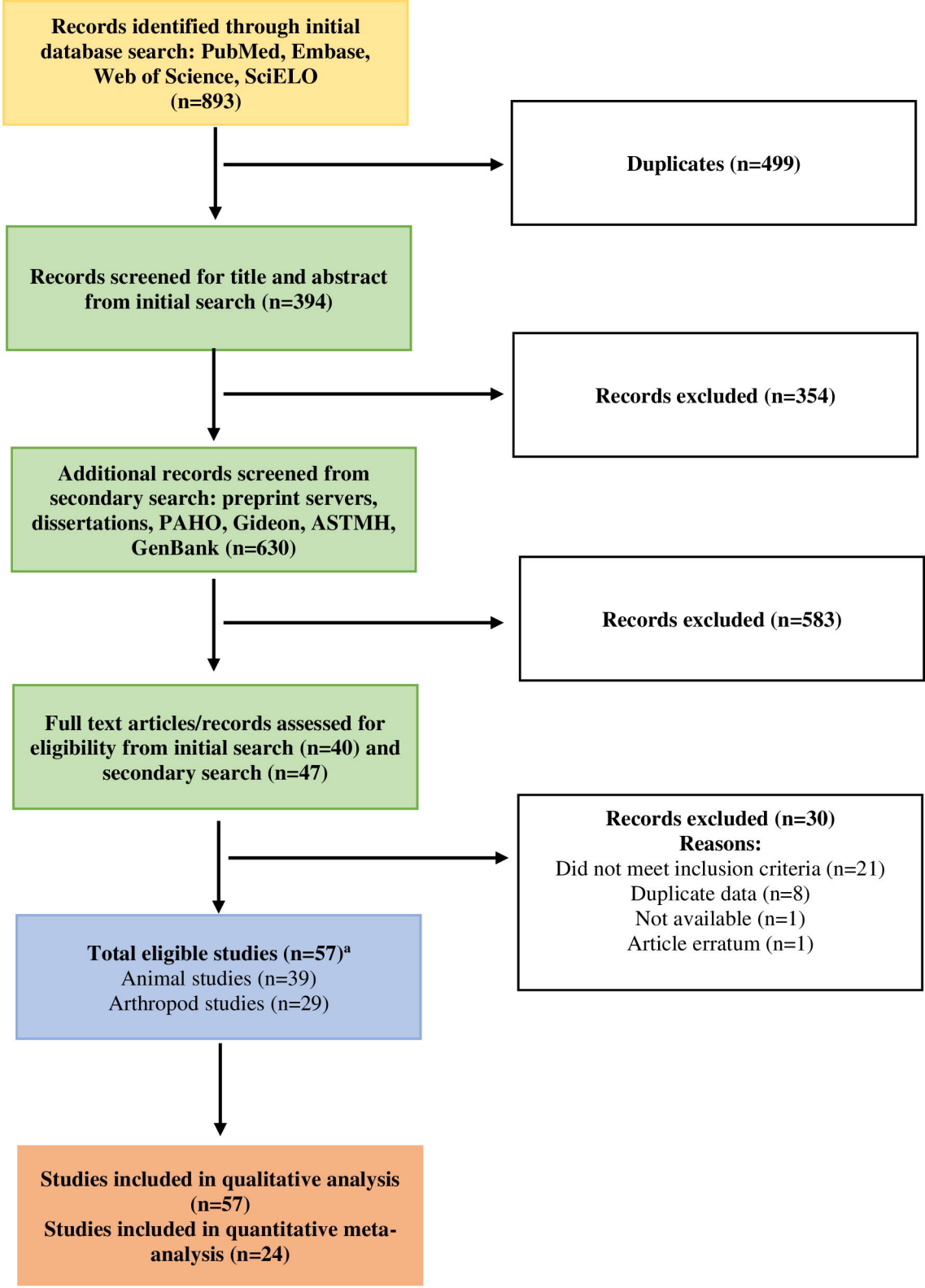
114. Mackay IM, Arden KE. Mayaro virus: a forest virus primed for a trip to the city? Microbes Infect. 2016;18(12):724-34. Epub 2016/12/19. doi: 10.1016/j.micinf.2016.10.007. PubMed PMID: 27989728.

115. Figueiredo MLGd, Figueiredo LTM. Emerging alphaviruses in the Americas: Chikungunya and Mayaro. Revista da Sociedade Brasileira de Medicina Tropical. 2014;47(6):677-83. doi: 10.1590/0037-8682-0246-2014.

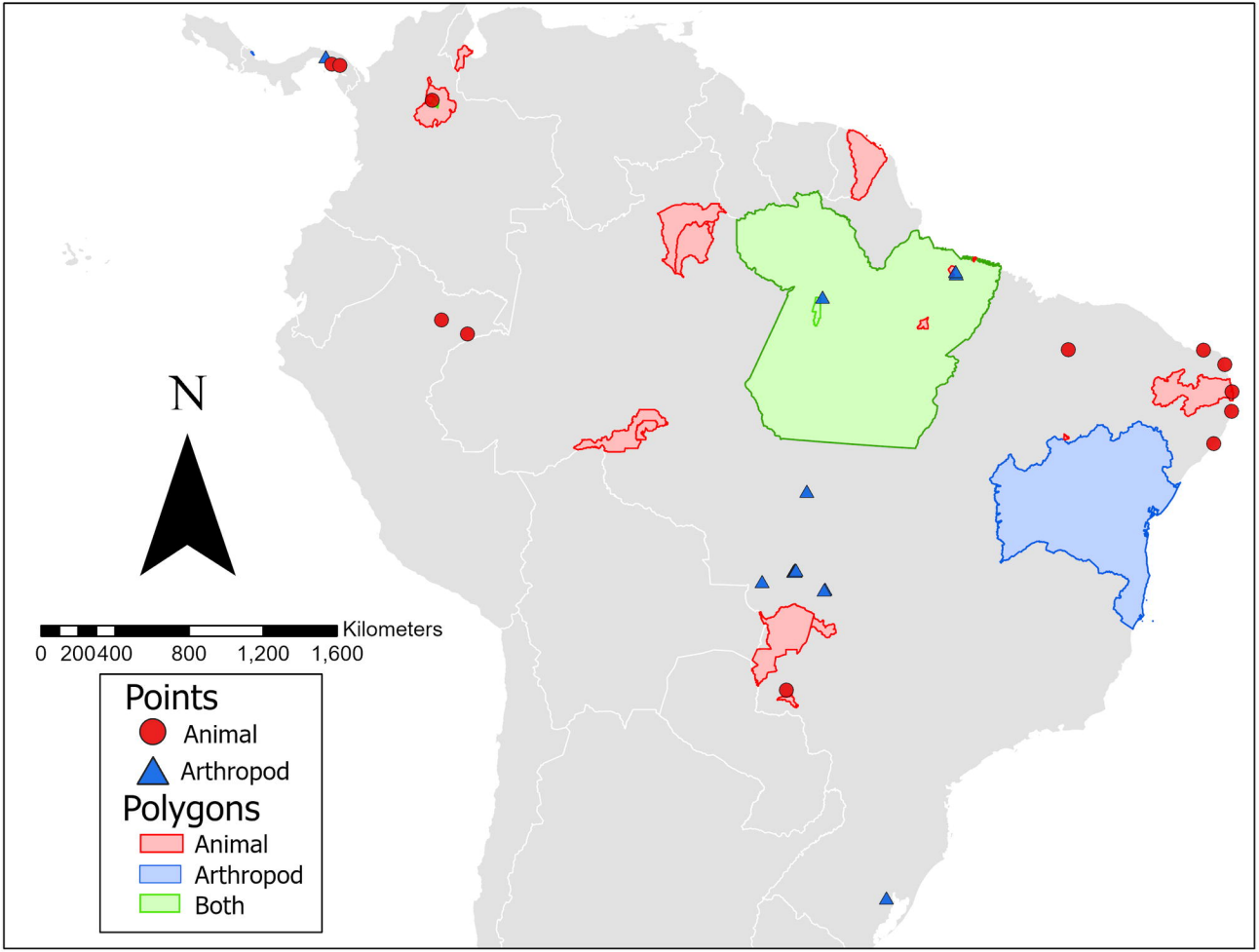
116. Hassing RJ, Leparc-Goffart I, Tolou H, van Doornum G, van Genderen PJ. Cross-reactivity of antibodies to viruses belonging to the Semliki forest serocomplex. Eurosurveillance. 2010;15(23).

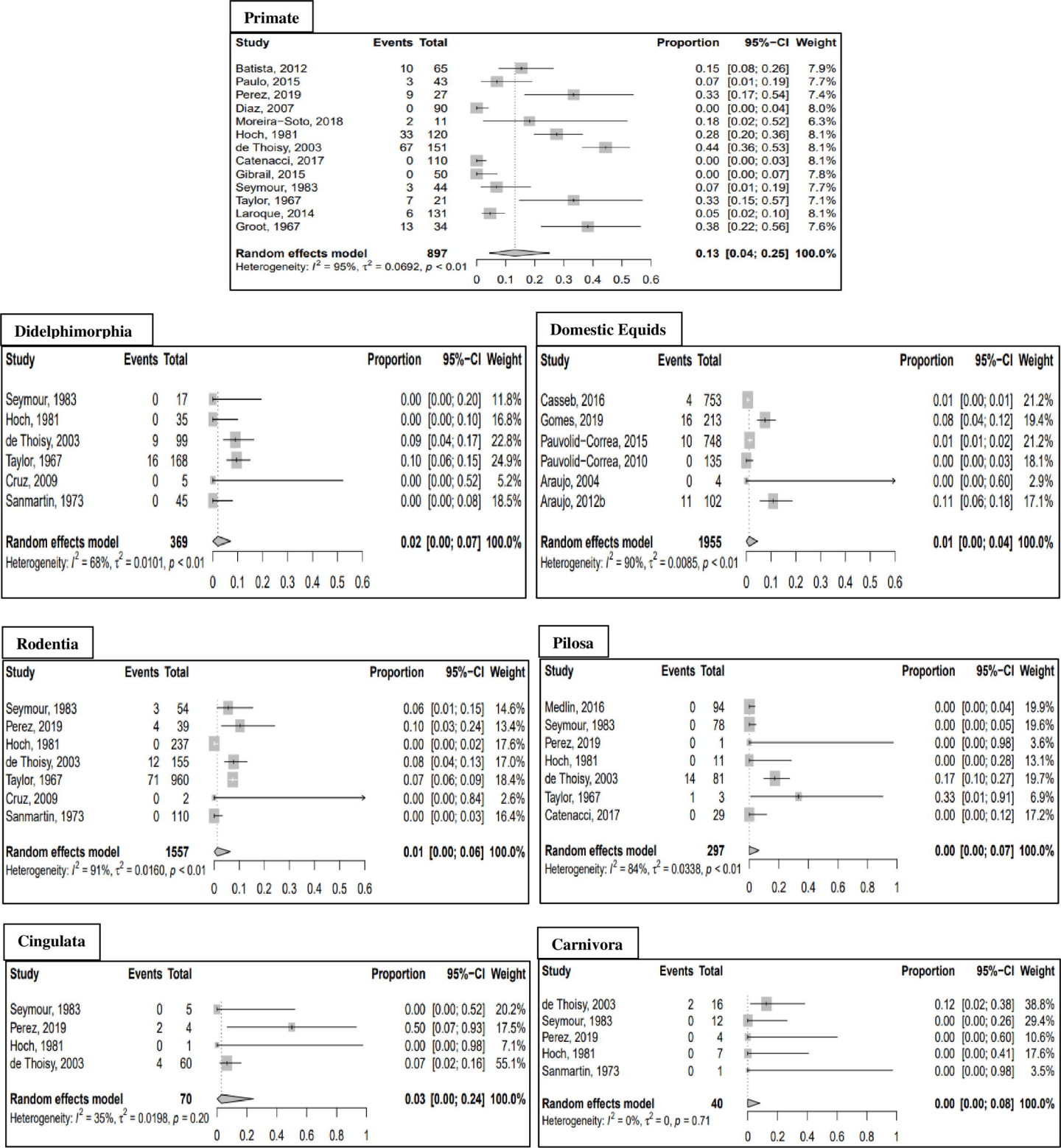
Supplementary Materials

S1 Table. PRISMA Checklist
S2 Table. MAYV positivity by taxa of wild mammals in included studies
S3 Table. MAYV positivity by taxa of wild birds in included studies
S4 Table. MAYV positivity in domestic or sentinel animals studied
S5 Table. Pooled Prevalence Table (Random effects using GLMM with logit transformation)
S6 Table. Primate Genera Pooled Prevalence Table (Random effects with Freeman-Tukey double arcsine transformation)
S7 Table. Primate Genera Pooled Prevalence Table (Random effects using GLMM with logit transformation)
S8 Table. Pooled Prevalence Table (Fixed effects with Freeman-Tukey double arcsine transformation)
S9 Table. Pooled Prevalence Table (Fixed effects using GLMM with logit transformation)
S1 Fig. Funnel plots for estimates of MAYV seroprevalence in non-human animal reservoirs
S2 Fig: Funnel plots for estimates of MAYV seroprevalence in non-human primate genera
S10 Table. Complete arthropod results by genus
S11 Table. Egger's test for publication bias



[a] Eleven articles assessed MAYV in both non-human animals and arthropods

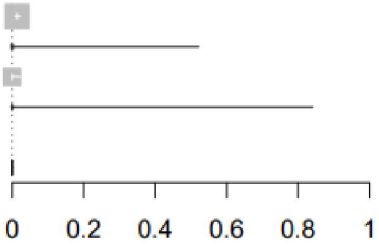

N
0 200400 800 1,200 1,600
Kilometers
Points
Animal
Arthropod
Polygons
Animal
Arthropod
Both


Primate
Study Events Total Proportion 95%−CI Weight
Batista, 2012 10 65 0.15 [0.08; 0.26] 7.9%
Paulo, 2015 3 43 0.07 [0.01; 0.19] 7.7%
Perez, 2019 9 27 0.33 [0.17; 0.54] 7.4%
Diaz, 2007 0 90 0.00 [0.00; 0.04] 8.0%
Moreira−Soto, 2018 2 11 0.18 [0.02; 0.52] 6.3%
Hoch, 1981 33 120 0.28 [0.20; 0.36] 8.1%
de Thoisy, 2003 67 151 0.44 [0.36; 0.53] 8.1%
Catenacci, 2017 0 110 0.00 [0.00; 0.03] 8.1%
Gibrail, 2015 0 50 0.00 [0.00; 0.07] 7.8%
Seymour, 1983 3 44 0.07 [0.01; 0.19] 7.7%
Taylor, 1967 7 21 0.33 [0.15; 0.57] 7.1%
Laroque, 2014 6 131 0.05 [0.02; 0.10] 8.1%
Groot, 1967 13 34 0.38 [0.22; 0.56] 7.6%
Random effects model 897 0.13 [0.04; 0.25] 100.0%
Heterogeneity: I^2 = 95%, τ^2 = 0.0692, p < 0.01
0 0.1 0.2 0.3 0.4 0.5 0.6
Didelphimorphia
Study Events Total Proportion 95%−CI Weight
Seymour, 1983 0 17 0.00 [0.00; 0.20] 11.8%
Hoch, 1981 0 35 0.00 [0.00; 0.10] 16.8%
de Thoisy, 2003 9 99 0.09 [0.04; 0.17] 22.8%
Taylor, 1967 16 168 0.10 [0.06; 0.15] 24.9%
Cruz, 2009 0 5 0.00 [0.00; 0.52] 5.2%
Sanmartin, 1973 0 45 0.00 [0.00; 0.08] 18.5%
Random effects model 369 0.02 [0.00; 0.07] 100.0%
Heterogeneity: I^2 = 68%, τ^2 = 0.0101, p < 0.01
0 0.1 0.2 0.3 0.4 0.5 0.6
Domestic Equids
Study Events Total Proportion 95%−CI Weight
Casseb, 2016 4 753 0.01 [0.00; 0.01] 21.2%
Gomes, 2019 16 213 0.08 [0.04; 0.12] 19.4%
Pauvolid−Correa, 2015 10 748 0.01 [0.01; 0.02] 21.2%
Pauvolid−Correa, 2010 0 135 0.00 [0.00; 0.03] 18.1%
Araujo, 2004 0 4 0.00 [0.00; 0.60] 2.9%
Araujo, 2012b 11 102 0.11 [0.06; 0.18] 17.1%
Random effects model 1955 0.01 [0.00; 0.04] 100.0%
Heterogeneity: I^2 = 90%, τ^2 = 0.0085, p < 0.01
0 0.1 0.2 0.3 0.4 0.5 0.6
Rodentia
Study Events Total Proportion 95%−CI Weight
Seymour, 1983 3 54 0.06 [0.01; 0.15] 14.6%
Perez, 2019 4 39 0.10 [0.03; 0.24] 13.4%
Hoch, 1981 0 237 0.00 [0.00; 0.02] 17.6%
de Thoisy, 2003 12 155 0.08 [0.04; 0.13] 17.0%
Taylor, 1967 71 960 0.07 [0.06; 0.09] 18.4%
Cruz, 2009 0 2 0.00 [0.00; 0.84] 2.6%
Sanmartin, 1973 0 110 0.00 [0.00; 0.03] 16.4%
Random effects model 1557 0.01 [0.00; 0.06] 100.0%
Heterogeneity: I^2 = 91%, τ^2 = 0.0160, p < 0.01
0 0.1 0.2 0.3 0.4 0.5 0.6
Pilosa
Study Events Total Proportion 95%−CI Weight
Medlin, 2016 0 94 0.00 [0.00; 0.04] 19.9%
Seymour, 1983 0 78 0.00 [0.00; 0.05] 19.6%
Perez, 2019 0 1 0.00 [0.00; 0.98] 3.6%
Hoch, 1981 0 11 0.00 [0.00; 0.28] 13.1%
de Thoisy, 2003 14 81 0.17 [0.10; 0.27] 19.7%
Taylor, 1967 1 3 0.33 [0.01; 0.91] 6.9%
Catenacci, 2017 0 29 0.00 [0.00; 0.12] 17.2%
Random effects model 297 0.00 [0.00; 0.07] 100.0%
Heterogeneity: I^2 = 84%, τ^2 = 0.0338, p < 0.01
0 0.2 0.4 0.6 0.8 1
Cingulata
Study Events Total Proportion 95%−CI Weight
Seymour, 1983 0 5 0.00 [0.00; 0.52] 20.2%
Perez, 2019 2 4 0.50 [0.07; 0.93] 17.5%
Hoch, 1981 0 1 0.00 [0.00; 0.98] 7.1%
de Thoisy, 2003 4 60 0.07 [0.02; 0.16] 55.1%
Random effects model 70 0.03 [0.00; 0.24] 100.0%
Heterogeneity: I^2 = 35%, τ^2 = 0.0198, p = 0.20
0 0.2 0.4 0.6 0.8 1
Carnivora
Study Events Total Proportion 95%−CI Weight
de Thoisy, 2003 2 16 0.12 [0.02; 0.38] 38.8%
Seymour, 1983 0 12 0.00 [0.00; 0.26] 29.4%
Perez, 2019 0 4 0.00 [0.00; 0.60] 10.6%
Hoch, 1981 0 7 0.00 [0.00; 0.41] 17.6%
Sanmartin, 1973 0 1 0.00 [0.00; 0.98] 3.5%
Random effects model 40 0.00 [0.00; 0.08] 100.0%
Heterogeneity: I^2 = 0%, τ^2 = 0, p = 0.71
0 0.2 0.4 0.6 0.8 1

Passeriformes

Study	Events	Total	Proportion	95%−CI	Weight
Hoch, 1981	14	1003	0.01	[0.01; 0.02]	65.9%
Cruz, 2009	0	5	0.00	[0.00; 0.52]	1.8%
Sanmartin, 1973	0	156	0.00	[0.00; 0.02]	31.5%
Araujo, 2012	0	2	0.00	[0.00; 0.84]	0.8%
Random effects model		**1166**	**0.00**	**[0.00; 0.00]**	**100.0%**

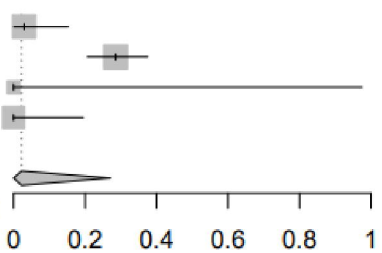
Heterogeneity: $I^2 = 27\%$, $\tau^2 = 0.0010$, $p = 0.25$



Columbiformes

Study	Events	Total	Proportion	95%−CI	Weight
Hoch, 1981	1	34	0.03	[0.00; 0.15]	30.5%
Taylor, 1967	34	119	0.29	[0.21; 0.38]	33.0%
Cruz, 2009	0	1	0.00	[0.00; 0.98]	9.0%
Sanmartin, 1973	0	17	0.00	[0.00; 0.20]	27.5%
Random effects model		**171**	**0.02**	**[0.00; 0.27]**	**100.0%**

Heterogeneity: $I^2 = 87\%$, $\tau^2 = 0.0591$, $p < 0.01$



Charadriiformes

Study	Events	Total	Proportion	95%−CI	Weight
Araujo, 2003	66	541	0.12	[0.10; 0.15]	54.2%
Sanmartin, 1973	0	9	0.00	[0.00; 0.34]	8.7%
Araujo, 2012	5	91	0.05	[0.02; 0.12]	37.1%
Random effects model		**641**	**0.07**	**[0.02; 0.14]**	**100.0%**

Heterogeneity: $I^2 = 61\%$, $\tau^2 = 0.0045$, $p = 0.08$