

### SUPPLEMENTARY FILES

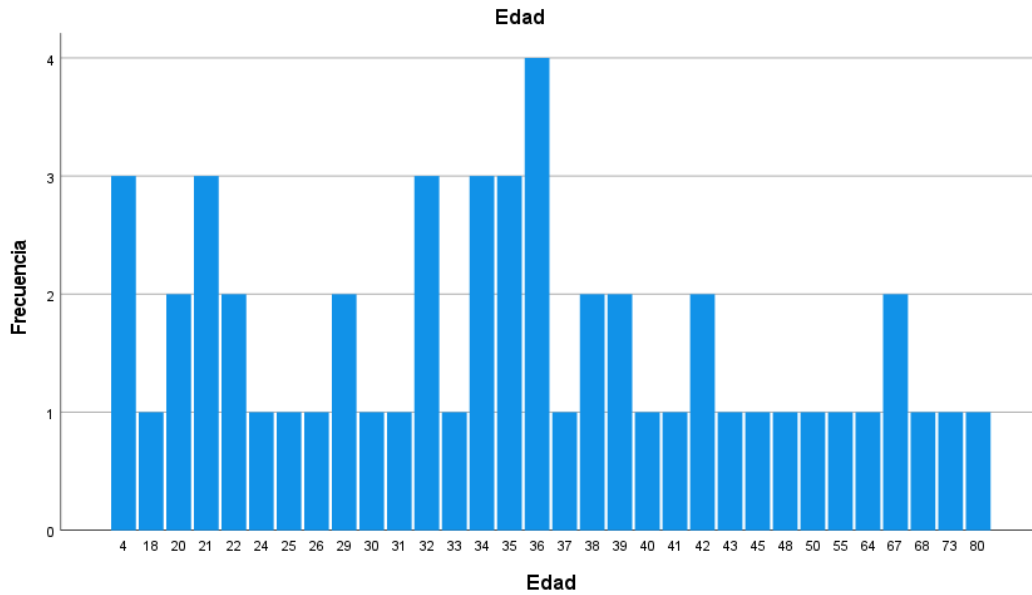


Figure S1. Frequency of monotypic samples of WNV according to patient age

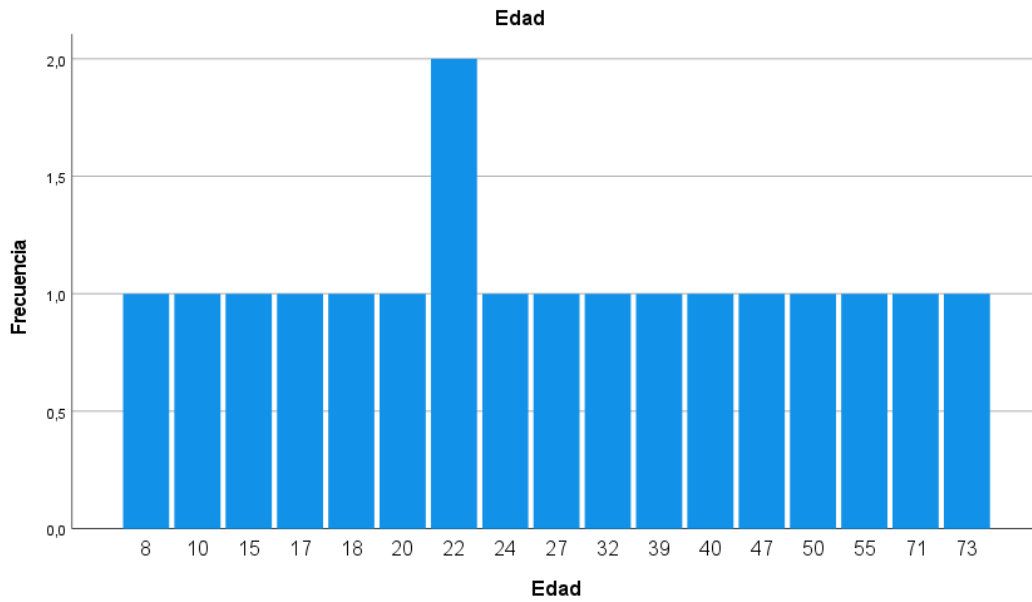
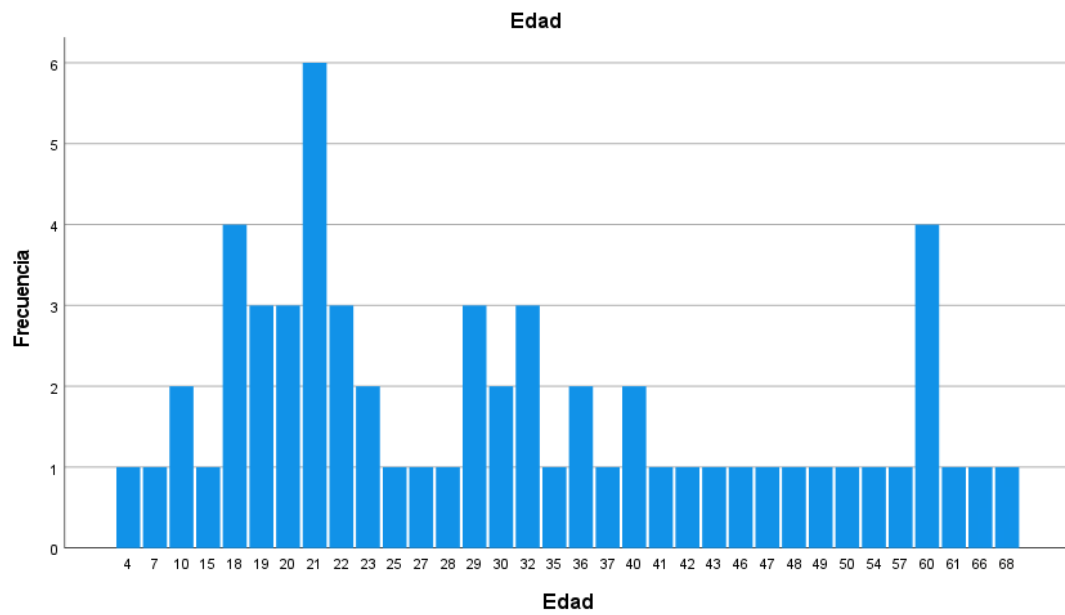
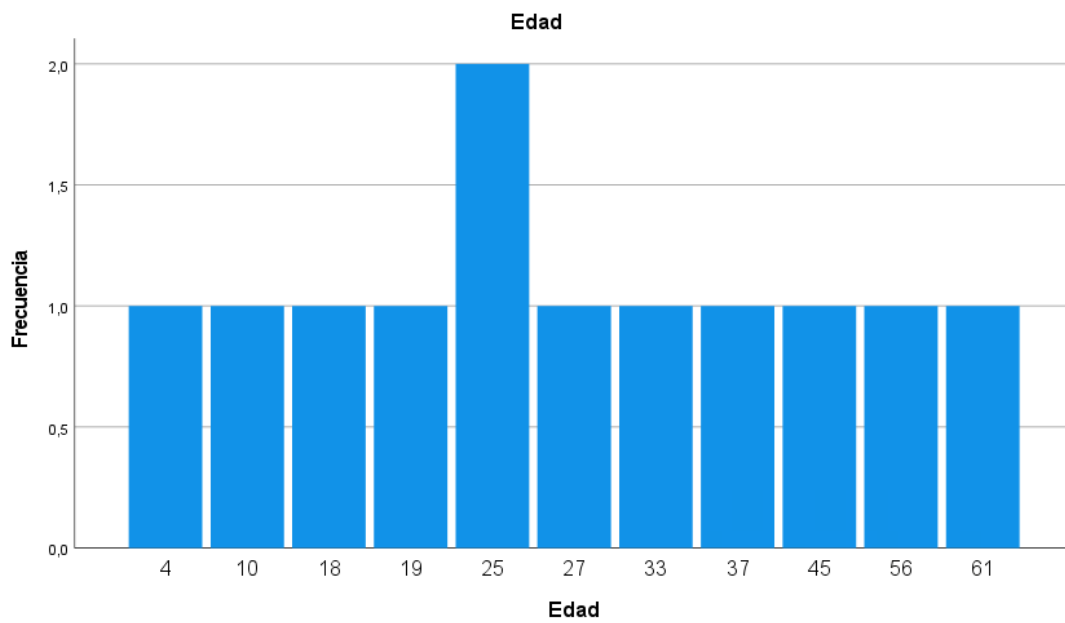


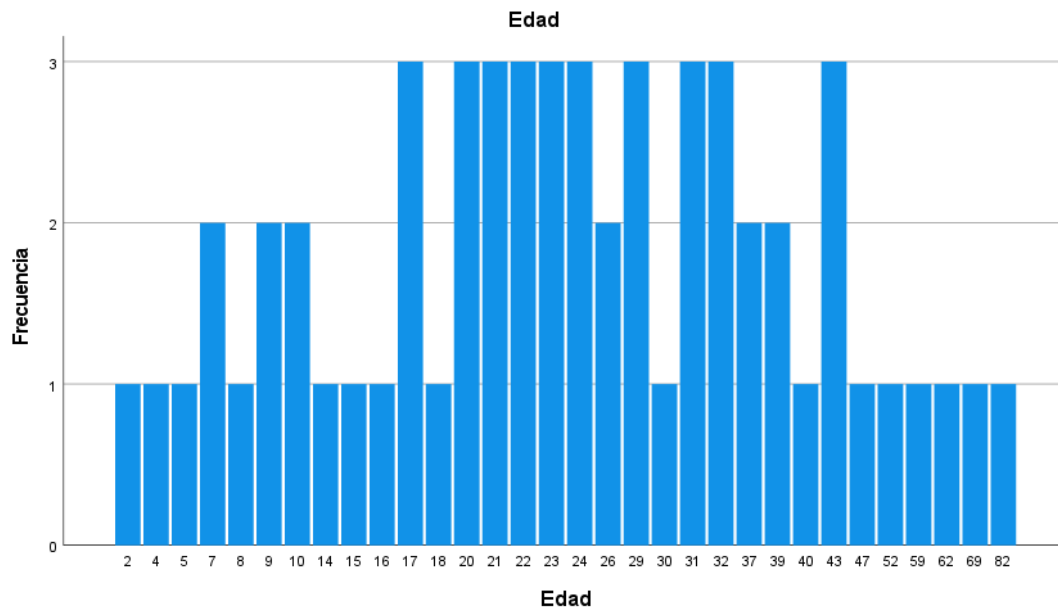
Figure S2. Frequency of monotypic samples of SLEV according to patient age



**Figure S3.** Frequency of monotypic samples of VEEV according to patient age



**Figure S4.** Frequency of monotypic samples of WEEV according to according to patient age.



**Figure S5.** Frequency of monotypic samples of EEEV according to according to patient age

**Table S1.** Number of samples of humans, sentinel hamsters, equines, and mosquitoes collected by location and date

Location*	Collection date	Samples			
		Humans	Sentinel Hamsters	Equines	Mosquitoes
Vinces	9-13/3/09	115	5	0	278
Manglaralto	25-27/3/09	54	5	15	266
Vinces	25-30/5/09	120	5	20	722
Puyo	7-11/9/09	244	7	0	34
Manglaralto	21-25/9/09	129	7	21	243
Puyo	26-30/10/09	149	7	10	8
Vinces	1-5/02/2010	39	6	0	65,318
Manglaralto	22-26/03/2010	46	5	17	281
Puyo	3-7/05/2010	94	5	0	84
Vinces	19-23/07/2010	57	5	11	1,301
Manglaralto	20-24/09/2010	105	6	14	308
Puyo	18-22/10/2010	177	6	5	30
Puyo	14-18/2/2011	103	5	2	15
Vinces	21-25/3/2011	43	5	9	3,212
Manglaralto	16-20/5/2011	77	5	13	82
Puyo	22-26/8/2011	105	6	0	65
Manglaralto	26-30/9/2011	85	5	14	709
Vinces	21-25/11/2011	100	5	37	549
Total		1,842	100	186	73,505

\* The samples were collected from the following sites: Vinces: Hospital Nicolás Cotto Infante, Subcentro de Salud San Lorenzo de Vinces, Isla de Bejuical, Subcentro de Salud Antonio Sotomayor, Abras de Mantequilla, El Recuerdo, Puerto Nuevo, Campo Alegre, Playones La Luz, Palenque, San Miguel de Palenque, Haciendas La María y El Rocío. Manglaralto: Hospital de Manglaralto, —Dos Mangas—, Santa María del Fiat, Haciendas San Francisco, El Edén y La Carmela, Fincas Lauricel Rodríguez, Manuel Baque y Benito Chiquito, —Olón—, Finca El Retiro, —La entrada—, Finca La Española. Puyo: Subcentro de Salud El Dorado, Fuerte Militar Amazonas-Brigada de Selva 17 Pastaza-Shell, Hospital Provincial de Puyo, Quesería 10 de Agosto, Recinto Fátima, Reserva de monos, Depósito de agua potable

**Table S2.** Number of febrile patients (n) per sampling location analyzed for IgM Abs against WNV and DENV and percentage of reactive samples

Location	n (female/ male)	WNV IgM* % of positives (number of positives/n)	DENV IgM % of positives (number of positives/n)
Vinces	69 (36/33)	1.66 (1/60)	0 (0/60)
Manglaralto	20 (16/4)	5.55 (1/18)	0 (0/18)
Puyo	13 (3/10)	0 (0/13)	30.76 (4/13)
Total	102 (55/47)	1.96 (2/102)	4.65 (4/86)

**Table S3.** Equines showing Abs to *Alphavirus* by HI test of N=149. EEEV: Eastern equine encephalitis virus; WEEV: Western equine encephalitis virus; VEEV: Venezuelan equine encephalitis virus. The reciprocal of the highest dilution of serum showing complete inhibition of hemagglutination is shown for each virus. \*= monotypic reactions, \*\* highest dilution showing complete inhibition of hemagglutination in heterotypic reactions

Sample Code	Date	EEEV	WEEV	VEEV	Location
EGV789	24/09/2009	40	0	40	Manglaralto
EGV791	24/09/2009	40	0	40	Manglaralto
EGV 2239	28/09/2011	40	0	40	Manglaralto
EGV 2240	28/09/2011	20	0	20	Manglaralto
EGV 2241	28/09/2011	20	20	40**	Manglaralto
EGV1979	18/05/2011	0	20*	0	Manglaralto
EGV2010	19/05/2011	0	20*	0	Manglaralto
EGV2012	19/05/2011	0	20*	0	Manglaralto
EGV229	26/03/2009	0	0	20*	Manglaralto
EGV1145	25/03/2010	0	0	40*	Manglaralto
EGV 2262	29/09/2011	0	0	40*	Manglaralto
EGV321	27/05/2009	40	80	80	Vinces
EGV322	27/05/2009	40*	0	0	Vinces
EGV323	27/05/2009	80	80	80	Vinces
EGV324	27/05/2009	40	80	80	Vinces
EGV328	27/05/2009	160	80	160	Vinces
EGV329	27/05/2009	160	1,280**	80	Vinces
EGV330	27/05/2009	40	80	80	Vinces
EGV331	27/05/2009	160	320**	40	Vinces
EGV332	27/05/2009	40	320**	40	Vinces
EGV333	27/05/2009	40	320**	0	Vinces
EGV334	27/05/2009	40	160**	40	Vinces
EGV1292	20/07/2010	40*	0	0	Vinces
EGV318	27/05/2009	0	40	40	Vinces
EGV326	27/05/2009	0	20	20	Vinces
EGV335	27/05/2009	0	80	160**	Vinces
EGV1290	20/07/2010	0	0	2 0*	Vinces
EGV1885	23/03/2011	0	0	20*	Vinces
EGV1888	23/03/2011	0	0	40*	Vinces
EGV1711	21/10/2010	0	0	40*	Puyo

**Table S4.** Hamsters showing Abs against Alphavirus by HI test of N=84 after exposure in the field. EEEV: Eastern equine encephalitis virus; WEEV: Western equine encephalitis virus; VEEV: Venezuelan equine encephalitis virus. The reciprocal of the highest dilution of serum showing complete inhibition of hemagglutination is shown for each virus. \*= monotypic reactions, \*\* highest dilution showing complete inhibition of hemagglutination in heterotypic reactions

Sample code	Date	EEEV	WEEV	VEEV	Location
EGV1163	25/03/2010	20*	0	0	Manglaralto
EGV1165	25/03/2010	20*	0	0	Manglaralto
EGV1513	24/09/2010	20	0	20	Manglaralto
EGV1514	24/09/2010	80*	0	0	Manglaralto
EGV1515	24/09/2010	80*	0	0	Manglaralto
EGV1516	24/09/2010	40**	0	20	Manglaralto
EGV1517	24/09/2010	20	0	20	Manglaralto
EGV1518	24/09/2010	40*	0	0	Manglaralto
EGV2018	31/05/2011	40	40	20	Manglaralto
EGV 2271	30/09/2011	20	0	40**	Manglaralto
EGV2019	31/05/2011	0	20	40**	Manglaralto
EGV829	25/09/2009	0	0	20*	Manglaralto
EGV830	25/09/2009	0	0	20*	Manglaralto
EGV832	25/09/2009	0	0	20*	Manglaralto
EGV1164	25/03/2010	0	0	20*	Manglaralto
EGV2015	31/05/2011	0	0	20*	Manglaralto
EGV2016	31/05/2011	0	0	40*	Manglaralto
EGV2017	31/05/2011	0	0	20*	Manglaralto
EGV 2273	30/09/2011	0	0	20*	Manglaralto
EGV1075	05/02/2010	20*	0	0	Vinces
EGV1077	05/02/2010	20*	0	0	Vinces
EGV1373	23/07/2010	20*	0	0	Vinces
EGV1374	23/07/2010	20	0	20	Vinces
EGV1376	23/07/2010	40*	0	0	Vinces
EGV1918	25/03/2011	20*	0	0	Vinces
EGV1078	05/02/2010	0	0	40*	Vinces
EGV1374	23/07/2010	20	0	20	Vinces
EGV1377	23/07/2010	0	0	20*	Vinces
EGV657	11/09/2009	1,280**	0	20	Puyo
EGV1280	07/05/2010	20*	0	0	Puyo
EGV1283	07/05/2010	40*	0	0	Puyo
EGV1284	07/05/2010	40*	0	0	Puyo
EGV1717	22/10/2010	40**	0	20	Puyo
EGV1718	22/10/2010	80**	0	20	Puyo
EGV1719	22/10/2010	40*	0	0	Puyo
EGV1720	22/10/2010	20*	0	0	Puyo
EGV 2149	01/09/2011	20*	0	0	Puyo
EGV653	11/09/2009	0	0	20*	Puyo
EGV655	11/09/2009	0	0	40*	Puyo
EGV658	11/09/2009	0	0	20*	Puyo
EGV1722	22/10/2010	0	0	20*	Puyo
EGV1744	16/02/2011	0	0	80*	Puyo
EGV1745	16/02/2011	0	0	40*	Puyo
EGV1837	17/02/2011	0	0	20*	Puyo

**Table S5.** Number of mosquitoes per genus and species captured in the locations studied

<b>Species</b>	<b>Vinces</b>	<b>Manglaralto</b>	<b>Puyo</b>	<b>Total</b>
<i>Trichoprosopon digitatum</i>	0	0	7	7
<i>Culex (Mel) spp.</i>	62,732	293	32	63,057
<i>Ochlerotatus serratus</i>	2,325	507	27	2,859
<i>Aedeomyia squamipennis</i>	261	1	0	262
<i>Psorophora (Jan) ferox</i>	1	0	5	6
<i>Mansonia pseudotitilians/indubitans</i>	4,234	72	0	4,306
<i>Coquillettidia venezuelensis</i>	1,476	0	160	1,636
<i>Aedes (Stg) aegypti</i>	0	912	0	912
<i>Anopheles (Nys) albimanus</i>	32	1	4	37
<i>Anopheles (Nys) triannulatus</i>	319	14	1	334
<i>Anopheles (Nys) pseudopunctipennis</i>	0	85	0	85
<i>Anopheles (Nys) nuneztovari</i>	0	4	0	4
<b>Total</b>	<b>71,380</b>	<b>1,889</b>	<b>236</b>	<b>73,505</b>

**Table S6.** Spearman correlation of the number of mosquitoes captured by species and the proportion of human samples showing IgG Abs reactive against DENV, SLEV, WNV, YFV, EEV, WEEV, and VEEV

Virus and statistics estimates			Mosquitoes especies												
			Td	C_sp	O_s	A_s	Ps_sp	M_sp	Cq_v	Ae_a	An_a	An_t	An_pse	An_nun	
Spearman's rho	DENV	Correlation coefficient	-0.866	0.500	0.500	0.500	-1,000**	0.500	-0.500	0.866	-0.500	0.500	0.866	0.866	
		Sig. (2-tailed)	0.333	0.667	0.667	0.667		0.667	0.667	0.333	0.667	0.667	0.333	0.333	
		N	3	3	3	3	3	3	3	3	3	3	3	3	
	Sampling simulation	Bias	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	
		Standard error	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	
	Confidence interval 95%	Lower limit	-0.866	0.500	0.500	0.500	-1.000	0.500	-0.500	0.866	-0.500	0.500	0.866	0.866	
		Upper limit	-0.866	0.500	0.500	0.500	-1.000	0.500	-0.500	0.866	-0.500	0.500	0.866	0.866	
	SLEV	Correlation coefficient	Sig. (2-tailed)	-0.866	0.500	0.500	0.500	-1,000**	0.500	-0.500	0.866	-0.500	0.500	0.866	0.866
			Sig. (2-tailed)	0.333	0.667	0.667	0.667		0.667	0.667	0.333	0.667	0.667	0.333	0.333
			N	3	3	3	3	3	3	3	3	3	3	3	3
Sampling simulation		Bias	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	
		Standard error	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	
Confidence interval 95%		Lower limit	-0.866	0.500	0.500	0.500	-1.000	0.500	-0.500	0.866	-0.500	0.500	0.866	0.866	
		Upper limit	-0.866	0.500	0.500	0.500	-1.000	0.500	-0.500	0.866	-0.500	0.500	0.866	0.866	
WNV		Correlation coefficient	Sig. (2-tailed)	-0.866	0.500	0.500	0.500	-1,000**	0.500	-0.500	0.866	-0.500	0.500	0.866	0.866
			Sig. (2-tailed)	0.333	0.667	0.667	0.667		0.667	0.667	0.333	0.667	0.667	0.333	0.333
			N	3	3	3	3	3	3	3	3	3	3	3	3
	Sampling simulation	Bias	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	
		Standard error	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	
	Confidence interval 95%	Lower limit	-0.866	0.500	0.500	0.500	-1.000	0.500	-0.500	0.866	-0.500	0.500	0.866	0.866	
		Upper limit	-0.866	0.500	0.500	0.500	-1.000	0.500	-0.500	0.866	-0.500	0.500	0.866	0.866	
	YFV	Correlation coefficient	Sig. (2-tailed)	0.000	-0.500	-0.500	-0.500	-0.500	-0.500	-1,000**	0.866	-1,000**	-0.500	0.866	0.866
			Sig. (2-tailed)	1.000	0.667	0.667	0.667	0.667	0.667	0.667	0.333		0.667	0.333	0.333
			N	3	3	3	3	3	3	3	3	3	3	3	3
Sampling simulation		Bias	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	
		Standard error	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	
Confidence interval 95%		Lower limit	0.000	-0.500	-0.500	-0.500	-0.500	-0.500	-1.000	0.866	-1.000	-0.500	0.866	0.866	
		Upper limit	0.000	-0.500	-0.500	-0.500	-0.500	-0.500	-1.000	0.866	-1.000	-0.500	0.866	0.866	
EEEV		Correlation coefficient	Sig. (2-tailed)	-0.866	0.500	0.500	0.500	-1,000**	0.500	-0.500	0.866	-0.500	0.500	0.866	0.866
			Sig. (2-tailed)	0.333	0.667	0.667	0.667		0.667	0.667	0.333	0.667	0.667	0.333	0.333
			N	3	3	3	3	3	3	3	3	3	3	3	3
	Sampling simulation	Bias	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	
		Standard error	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	
	Confidence interval 95%	Lower limit	-0.866	0.500	0.500	0.500	-1.000	0.500	-0.500	0.866	-0.500	0.500	0.866	0.866	
		Upper limit	-0.866	0.500	0.500	0.500	-1.000	0.500	-0.500	0.866	-0.500	0.500	0.866	0.866	
	WEEV	Correlation coefficient	Sig. (2-tailed)	-0.866	0.500	0.500	0.500	-1,000**	0.500	-0.500	0.866	-0.500	0.500	0.866	0.866
			Sig. (2-tailed)	0.333	0.667	0.667	0.667		0.667	0.667	0.333	0.667	0.667	0.333	0.333
			N	3	3	3	3	3	3	3	3	3	3	3	3
Sampling simulation		Bias	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	
		Standard error	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	
Confidence interval 95%		Lower limit	-0.866	0.500	0.500	0.500	-1.000	0.500	-0.500	0.866	-0.500	0.500	0.866	0.866	
		Upper limit	-0.866	0.500	0.500	0.500	-1.000	0.500	-0.500	0.866	-0.500	0.500	0.866	0.866	
VEEV		Correlation coefficient	Sig. (2-tailed)	-0.866	1,000**	1,000**	1,000**	-0.500	1,000**	0.500	0.000	0.500	1,000**	0.000	0.000
			Sig. (2-tailed)	0.333				0.667		0.667	1.000	0.667		1.000	1.000
			N	3	3	3	3	3	3	3	3	3	3	3	3
	Sampling simulation	Bias	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	
		Standard error	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	
	Confidence interval 95%	Lower limit	-0.866	1.000	1.000	1.000	-0.500	1.000	0.500	0.000	0.500	1.000	0.000	0.000	
		Upper limit	-0.866	1.000	1.000	1.000	-0.500	1.000	0.500	0.000	0.500	1.000	0.000	0.000	

\*\* Correlation is significant at 0,01 Sig. (2-tailed).  
Simulation was based on 1,000 replicates.

**Table S7.** Mosquito species identified in this study and the arboviruses they harbor according to viral isolation studies elsewhere\*

Mosquitos species	Arbovirus carried	Reference
<i>Trichoprosopon digitatum</i>	Pixuna virus and Wyeomyia virus. Bussuquara, SLEV and ILHV have been isolated from mixed pools that included this species.	(1-2)
<i>Culex (Mel) spp.</i>	WNV, VEEV, VESL, Japanese Encephalitis virus	(3)
<i>Ochlerotatus serratus</i>	OROV, YFV, ILHV	(4-6)
<i>Aedeomyia squamipennis</i>	Gamboa virus, VEEV	(7,8)
<i>Psorophora (Jan) ferox</i>	Rocio virus	9
<i>Mansonia pseudotitilians/indubitans</i>	VEEV	10
<i>Coquillettidia venezuelensis</i>	MAYV, OROV y SLEV, WNV	(11-12)
<i>Aedes (Stg) aegypti</i>	DENV, YFV, ZKV, CHKV	13
<i>Anopheles (Nys) albimanus</i>	Not known	
<i>Anopheles (Nys) triannulatus</i>	Not known	
<i>Anopheles (Nys) pseudopunctipennis</i>	Not known	
<i>Anopheles (Nys) nuneztovari</i>	Not known	
TOTAL	Not known	

\*This is not an exhaustive list of arboviruses carried by mosquitoes

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