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Brief Original Article

Factors associated with increased odds of sensorineural hearing loss in infants exposed to the Zika virus during pregnancy

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Abstract

Introduction: The Zika virus (ZIKV) infection in pregnant women has been associated with an increased risk of birth defects. We aimed to estimate the prevalence of sensorineural hearing loss (SNHL) in infants exposed to the ZIKV during their gestation and evaluate the factors associated with its increased odds.

Methodology: A cross-sectional study was performed from July 2016 to June 2019 in a Western state of Mexico and data from 61 infants that presented with laboratory-positive (RT-qPCR) evidence of *in utero* exposure to ZIKV were analyzed. Brain stem auditory evoked potentials were used.

Results: Hearing loss was documented in 6 (9.8%) of infants. The prevalence of SNHL in children with microcephaly was 75.0%, as compared to 5.3% in those without anomalies (odds ratio, OR = 14.31, 95% CI = 2.54 – 19.12). Half of children with SNHL had no physical manifestations of gestational ZIKV exposure.

Conclusions: Hearing loss was a frequent event in ZIKV-exposed children, particularly among those with microcephaly. Our results highlight the relevance of systematic hearing screening.

Key words: Zika virus; pregnancy; brain stem; hearing loss; sensorineural.

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Introduction

Zika virus (ZIKV) infection in pregnant women has been associated with an increased risk of birth defects, defining the congenital Zika syndrome (CZS) that includes severe microcephaly, subcortical calcifications, and hypertonia [1]. An increased risk for sensorineural hearing loss (SNHL) has been documented in infants with microcephaly and *in utero* ZIKV exposure [2,3].

The ZIKV burden in Mexico has been high and, within the first nine months from the local outbreak start, the computed incidence rate among pregnant women was 66 per 100,000 pregnancy-months [4].

Published studies evaluating SNHL in Latin-American among Latin-American children that have no manifestations of gestational ZIKV exposure are scarce and results are heterogeneous [5,6]. The present study aimed to estimate the risk of SNHL in infants born to women with laboratory-confirmed ZIKV infection during pregnancy. The factors associated with the risk of hearing impairment were also evaluated.

Methodology

A cross-sectional study was conducted in Colima, a subtropical western state of Mexico, from July 2016 to June 2019. The state of Colima is a subtropical area located in the Western region of Mexico and high *Aedes aegypti* indices, and related vector-borne diseases, have been documented [7].

The eligible subjects were randomly selected from full-term infants, born to women with laboratory-confirmed exposure to ZIKV (RT-qPCR, reverse transcription quantitative polymerase chain reaction) (TaqMan™ Zika Virus Triplex Kit, catalogue code A31747, Applied Biosystems™, U.S.A) [8] during pregnancy. The Mexican Institute of Social Security

(Instituto Mexicano del Seguro Social - IMSS), where this study took place, has a Laboratory Network for Epidemiological Surveillance integrated by four highly specialized settings. Their analytical procedures are endorsed by the Institute for Epidemiological Diagnosis and Reference, the highest related-authority in Mexico. Eligible children were identified from data belonging to the National System for Epidemiological Surveillance (Sistema Nacional de Vigilancia Epidemiológica - SINAVE). Known infectious or traumatic causes (cranioencephalic trauma) of hearing loss were the exclusion criteria. The communicable diseases were evaluated by reviewing the laboratory results of prenatal tests (TORCHS screen) that are mandatory according to national normative standards guiding the prenatal and postnatal care to mother and child [9]. Subjects with documented neonatal jaundice or kernicterus were also excluded, as wells as those with reported intra-/extra-uterine exposure to ototoxic drugs.

After written statements of informed consent to participate in the study obtained from the parents/legal guardians of the infants, brain stem auditory evoked potential (BAEP) (Nihon Kohden Neuropack S1 MEB-9400, Japan) tests were performed by a trained health professional (doctor of audiology). Prior to be evaluated, all children were physically examined (otoscopy) by the same professional that performed the BAEP in order to discard causes of conductive hearing impairment (i.e. wax in the ear canal or middle ear pathology). Infants with persistent wave V after a 40 dB stimuli tested positive for SNHL.

Rate ratios with their 95% confidence intervals are presented (univariate analysis). Odds ratios (OR) with 95% confidence intervals (CIs) and estimated through logistic regression models were used to evaluate the association between the exposures analyzed and the odds for SNHL. A multivariate model was built by following a purposeful selection procedure [10]. In order to obtain a better approximation to risk ratios, the

Table 1. Characteristics of the study sample for the selected variables, Mexico 2016 – 2019.

Characteristics	Overall (n = 61)		SNHL Yes (n = 6)		RR (95% CI)	
Sex						
Female	38	(62.3%)	2	(5.3%)	1.00	
Male	23	(37.7%)	4	(17.4%)	3.28	(0.93 - 7.32)
First-degree family history of hearing loss						
No	58	(95.1%)	6	(10.3%)	NC	
Yes	3	(4.9%)	0	(0%)	NC	
Trimester of pregnancy[a]						
First	19	(31.2%)	1	(5.3%)	1.00	
Second	15	(24.5%)	3	(20.0%)	3.77	(0.82 - 9.07)
Third	27	(44.3%)	2	(7.4%)	1.40	(0.17 - 4.58)
Folate intake during pregnancy						
No	3	(4.9%)	1	(33.3%)	1.00	
Yes	58	(95.1%)	5	(8.6%)	0.26	(0.09 - 0.57)
Delivery mechanism						
Vaginal	32	(52.5%)	3	(9.4%)	1.00	
Cesarean section	29	(47.5%)	3	(10.3%)	1.10	(0.23 - 2.91)
Birth weight for gestational age						
Low	6	(9.8%)	1	(16.7%)	1.00	
Normal	55	(90.2%)	5	(9.1%)	0.54	(0.18 - 1.19)
Clubfoot						
No	59	(96.7%)	4	(6.8%)	1.00	
Yes	2	(3.3%)	2	(100%)	14.71	(2.33 - 27.09)
Microcephaly[b]						
No	57	(93.4%)	3	(5.3%)	1.00	
Yes	4	(6.6%)	3	(75.0%)	14.15	(3.66 - 18.75)
Cleft lip/palate						
Yes	4	(6.6%)	0	(0%)	NC	
No	57	(93.4%)	6	(10.5%)	NC	

1) The absolute and relative (%) frequencies are presented, as well as the p-value from the chi-square tests; 2) The brain stem auditory evoked potential (BAEP) Nihon Kohden Neuropack S1 MEB-9400 EMG EP test was used to determine the functional integrity of the auditory system; the absence of waves or the presence of wave V after 80 dB or higher stimuli were considered as positive for sensorineural hearing loss. [a] At the medical diagnosis of acute Zika virus disease; [b] More than 2 standard deviations below the mean for age and sex. SNHL: sensorineural hearing loss; RR: rate ratio; CI: confidence interval; NC: non-computable.

ORs were corrected through the method suggested by Zhang and Yu [11]. The present study was approved (R-2019-601-006) by the Local Health Research Ethics Committee of the IMSS.

Results

Data from 61 infants were analyzed and the overall prevalence of SNHL was 6 (9.8%). All mothers reported ZIKV-suggestive symptoms during pregnancy. Occurrence of hearing loss in children with microcephaly (> 2 standard deviations below the mean for age and sex) was 75%, as compared to 5.3% in those without anomalies Half (n = 3/6; 50%) of children with SNHL had no physical manifestations of gestational ZIKV exposure. All children with hearing impairment recorded no waves or wave V after 80 dB or higher stimuli in BAEP testing. Mean age of participants upon the neurosensitive test was 4.9 ± 0.8 months. Table 1 shows the characteristics of the study sample for the selected variables.

None of the infants with hearing loss had a family history of hypoacusia neither the use of any ototoxic drugs during pregnancy or extra-uterine life. The prevalence of SNHL among children with clubfoot (n = 2) was 100%. Therefore and as it is presented in Table 1, a 14-fold increase in the rate of hearing impairment was documented in children with clubfoot (RR = 14.71, 95% CI = 2.33 – 27.09) or microcephaly (RR = 14.15, 95% CI = 3.66 – 18.75). In multivariate analysis (Table 2), infants with microcephaly were at increased odds of deafness (OR = 14.31, 95% CI = 2.54 – 19.12).

Discussion

Our findings suggest that hearing impairment is a frequent event among children exposed to ZIKV during pregnancy and SNHL was confirmed in about 1 out 10 infants analyzed. While microcephaly was clearly related to hearing dysfunction with a more than fourteen-fold prevalence than in those without microcephaly, hearing loss was also documented in infants that did not present with microcephaly or a CSZ-suggestive (namely clubfoot or microcephaly) phenotype, which highlights the importance of systematic hearing screening in all exposed newborns.

The mechanism of ZIKV-induced microcephaly has not been elucidated. Animal models have documented a large number of alterations in brain development after ZIKV infection and include abnormal gene expression and immune response, lysosome circuits among others [12].

The overall occurrence of SNHL observed in our study was lower than that estimated in a sample of Brazilian children (9.8% *vs*. 17.3%) [2]. Only children with microcephaly were included in the South American study In our analysis, the prevalence SNHL among children with microcephaly was 75.0%.

No hearing loss was documented in Colombian children born to women exposed to ZIKV during pregnancy [13]. However, in that analysis, exposure to the pathogen was suggestive, with no confirmatory tests performed on the majority of the study subjects. In addition, evoked potentials were only utilized in a subset of infants in the Colombian research.

The pathogenic mechanism of hearing impairment after in utero exposure to ZIKV remains poorly understood. Animal models have evidenced that the virus can directly infect and damage the auditory and vestibular components of the embryonic chicken inner ear [14]. Besides, ZIKV infection induces maternal immune activation, which is associated with the occurrence of neuroanatomic and other fetal abnormalities [15]. Nearly 7% of infants in our study presented with cleft lip or cleft palate, but no association with hearing loss was found. That frequency is higher than the general incidence documented in Mexico (1:1,000 live births) [16]. The association between ZIKV infection and the risk of cleft lip/palate

Table 2. Factors associated with the odds of sensorineural hearing loss in the study sample, Mexico 2016 – 2019.

	Univariate analysis			Multivariate analysis		
	OR	**95% CI**	**p**	**OR**	**95% CI**	**p**
Sex						
Female	1.00			1.00		
Male	1.34	(0.84 - 1.49)	0.151	1.37	(0.82 - 1.50)	0.134
Folate intake during pregnancy						
No	1.00			1.00		
Yes	0.87	(0.22 - 1.02)	0.203	0.91	(0.22 - 1.03)	0.481
Microcephaly[a]						
No	1.00			1.00		
Yes	14.25	(2.53 - 18.88)	< 0.001	14.31	(2.54 - 19.12)	< 0.001

1) The odds ratios (OR) and 95% confidence intervals (CI) were estimated through logistic regression models. 2) The multiple analysis estimators were adjusted by the variables presented in the table. [a] The following were included: microcephaly (more than 2 standard deviations below the mean for age and sex).

has been previously described in previous studies, specifically in Brazilian children [17].

The inclusion of children with laboratory-conclusive evidence of *in utero* exposure to ZIKV, together with the performance of BAEPs in all the subjects analyzed, are major strengths of the present study. Its potential limitations include the limited sample size the fact that we lacked neuroimaging data that would have enabled the radiographic identification of CZS cases and that the sample size was relatively small. Nevertheless, only infants with laboratory-conclusive evidence of *in utero* exposure to ZIKV were enrolled and all the study subjects underwent BAEP testing.

Conclusions

Hearing impairment was a frequent event in the individuals analyzed in the present study, and those with microcephaly were at an 14-fold increased risk for SNHL. That neurosensory disorder was also documented in infants that did not present with phenotypic alterations, highlighting the benefits of systematic hearing screening of all exposed newborns.

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