



Neurological evaluation of microcephalic children with Zika syndrome and congenital cytomegalovirus infection

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ABSTRACT

Introduction: The association between the virus prenatal infection by Zika virus (ZIKV) and central nervous system disorders has been well established and it has been described as the Congenital Syndrome Associated to the Zika Virus (CSZ). However, the neurological development in those patients is still an object of study. The main differential diagnosis is the Cytomegalovirus (CMV).

Objective: Describe the involvement of microcephalic patients affected by the congenital infection by the Zika Virus or CMV.

Methodology: Data has been collected from microcephalic patients whose birth took place after 2016 and which also had the congenital infection confirmed or presumed. The researched data consists in: congenital infection, head circumference from birth, presence of epilepsy, treatment by mono or polytherapy, electroencephalographic patterns, neurological physical examination and evaluation of gross motor development.

Results: 21 microcephalic children have been included showing the following congenital infectious syndromes: 9 were affected by cytomegalovirus (43%), 6 by the Zika virus (29%) and 6 ones by presumed infection due to the Zika virus (29%). From those ones, 13 (62%) presented epilepsy diagnosis including generalized crises and 9 (69%) were in current use of polytherapy. All of them also showed disorganized and asymmetrical base rhythms. Concerning the epileptiform activity, 5 presented multifocal activity and 3 ones hypsarrhythmia. All of the patients went under neuroimaging: 12 (57%) of them presented calcifications and 5 (24%) hydrocephalus. On the neurological exam, 17% presented a decreased axial tone and an enlarged appendicular. Smaller head circumference children had greater motor impairment and severity in the epilepsy. There was no difference in the frequency of epilepsy between children with CSZ and CMV.

Conclusion: Epilepsy is confirmed as one of the most important complications of congenital infections by CSZ and CMV.

1. Introduction

The infection caused by Zika (ZIKV) had been considered a benign disease until October 2015 when it was observed an increase in the number of neonates with microcephaly in Brazil northeast region [1]. From this situation, several studies were published associating casually the ZIKV outbreak in several countries, including Brazil, and a congenital infection caused by the Zika Virus (CSZ) with acute microcephaly presented at birth.

In November 2015, the Ministry of Health Public Health Emergency of National Importance developed an electronic instrument called

Events Record in Public Health (ERPH - microcephaly) developed by the Computer Department of the National Health Service (DATASUS) to conduct the notifications, monitoring, investigations, and the classification of the new cases of microcephaly [2].

In children presenting microcephaly, beyond the acute delay in development, epilepsy was also often found. The brain involvement usually causes epileptic seizures and it can be one of the first symptoms existing in many cases of congenital infection [3]. Studies show that epilepsy in the congenital syndrome by the CSZ manifests itself earlier and with more prevalence than other congenital infections [4]. The prenatal infection by the ZIKV may have a vertical transmission and can

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damage the brain in development, interfering in the multiplication and migration of the nervous system cells, accelerating the apoptosis, the formation due to clinical resemblance. The CMV (Cytomegalovirus) is the main differential diagnosis of the ZIKV.

The SCZ leads to the formation of a reduced brain size, low differentiated cortex and with less gyri [5]. It can also cause juxtacortical calcifications, cortical malformations, ventriculomegaly, epileptical seizures, delayed neuro-psychomotor with spasticity, hyperexcitability and dysphagia [6]. Affected children may have exacerbated primitive reflexes which disappear later than expected [7]. Unlike other viral congenital infections which affect multiple organs, the ZIKV has a peculiar neurotropism [8], reaching the neuronal cells and glial cells in all maturation stages, including their progenitors. Moreover, it causes an acute destructive-disruptive process to brains which would have developed as usual during the embryogenesis period, and, therefore causing a viral cerebellitis with neuronal apoptosis resulting in microcephaly [1,9].

Children affected by microcephaly are at risk of cerebral palsy, being this one a common incapacity [10,11]. According to the study of Watemberg et al. [19], it was verified that cerebral palsy occurred in 21,4% of the 216 children affected by microcephaly when compared to 8,8% of the 1.159 normocephalic children ($p < 0,001$). Previous studies have indicated that cerebral palsy is part of the clinic of CSZ [10,11].

This article aims to contribute to the description of the neurological phenotype of children affected by secondary congenital microcephaly in relation to the CSZ and the CMV, comparing the motor development, epileptic pattern, electroencephalographic characteristics and the presence of cerebral palsy. This neurological phenotype contributes to doctors who will be able to precociously identify the consequences enabling early treatment and rehabilitation.

2. Methodology

This study has a contemporary, cross-sectional study, descriptive-analytical-exploratory outline. Birth data were collected retrospectively, as patients were referred from other cities in the state of Rio Grande do Sul. Children under medical follow-up at the microcephaly outpatient clinic between January 2016 and December 2019 have been included. This outpatient clinic was recognized as a reference to Rio Grande do Sul state by Public Health Event Registration (PHEV).

It includes neonates or children affected by congenital microcephaly presenting the serological and clinic diagnosis confirmed or who are either highly suspect of being infected by the ZIKV or presenting a CMV diagnosis and who were born in the State of Rio Grande do Sul. It had been excluded from the study children who presented microcephaly due to other infectious or noninfectious causes, such as genetic syndromes, teratogens and patients born before January 2016. For the purpose of this study, congenital microcephaly was defined as a condition which causes the head circumference to be lower to Z scores below 2 standard deviations from the mean for sex and gestational age during the birth moment and 24 h after, using the *Intergrowth 21st* scales [12]. None of the participants included have any other positive congenital infections.

The congenital microcephaly diagnosis by ZIKV or CMV was either based on laboratory results as well as clinical-epidemiological following the "Integrated orientations of surveillance and attention to health in the field of Public Health Emergency of National Importance" [13]. It was considered confirmed cases of congenital infection by ZIKV neonates with positive results or reactive to Zika on a more precise test but with a negative or inconclusive result in at least 1 STORCH (syphilis, toxoplasmosis, rubella, CMV or herpes simplex) on sample taken from the NB or the mother (during gestation) and two or more of the signals and symptoms (on image scans or clinical exam) presented on the table below (Table 1) [13]. It was considered suspected cases of congenital infection by ZIKV those neonates whose clinical exams were suggestive of SCZ (microcephaly with craniofacial disproportion and, at least, two neuroimaging and neurological changes according to the Table 1) but

Table 1

Most common changes identified during birth and within the 1st month of life.

Neuroimaging Findings	Clinical Findings
Brain calcifications	Changes in muscle tone
Cerebral cortical developmental disorder	Posture change
Frontal parietal predominance of cortical thickening	Primitive reflexes exaggeration
Polymicrogyria	Hyperexcitability
Simplification of the brain gyri patterns / sulci patterns	Irritability
Ventriculomegaly/ Ventricular dilation	Epileptic seizures
Changing of the posterior fossa pattern	Sucking and swallowing difficulties
	Dysphagia
Brainstem Hypoplasia, cerebellum, callosal commissure	Fundoscopy changes (retina and optic nerve)
	Abnormal eye movements

Source: Adapted from Brazil (2017).

the Real-time Polymerase Chain (PCR) was negative or not realized and having negative serology or not realized [13]. It were considered confirmed cases of congenital infection by CMV those neonates whose PCR results were positive to CMV in the urine test and either having a negative result or inconclusive result to ZIKV on samples of the neonate as well as having two or more symptoms and signals presented in the Table 1 [13].

In the initial appointment, demographic and clinical data from the patients as well as the gynecologic-obstetric history and familiar history were collected [13]. The complete physical examination was realized by a geneticist doctor and by a pediatrician while the neurological exam was realized by a neuropsychiatrist. Imaging exams were realized in all children according to the disponibility. In the Imaging exams, the anomalies described in Table 1 were searched.

An electroencephalogram was run in all individuals of the sample. The classification of the type of epileptical seizures and of the epilepsy was determined according to the nomenclature proposed by the International League Against Epilepsy in 2017 [14]. The acquisition of electroencephalograms occurred by the NEUROTEC (Neuromap)[®] machine. It were used 21 scalp electrodes which were placed according to the norms of the 10–20 International System [15]. The exams had been run under spontaneous sleep and with at least 30 min of duration.

The clinical evaluation of the neuromotor development was realized through the GMFCS scale [16]. The patients were always evaluated by the same care team during the clinical appointment. The evaluators were not blinded to the type of pathology of the patient.

The motor development had been realized according to the GMFCS (*Gross Motor Function Classification System*) scale which is one of the tools utilized to classify the functional capacity of children presenting with cerebral palsy along with the establishing of long-term development expected [16]. This tool is widely utilized to classify the gross motor function and it has five levels of classification based on the abilities and on the initiative of the movement (the level I represents the best abilities whereas the level V the less developed ones).

The following variables were described: sex, gestational age (GA) from the birth in weeks (if pregnant and with early ultrasound, before the 12th week, this was the preferential method or in case of not being performed the Capurro technique at the birth moment, according to the hospital of origin), number of prenatal appointments (it was considered a completed prenatal if there were at least 6 appointments realized [17]), realization of the laboratorial exam for the ZIKV during the pregnancy, head circumference in centimeters and millimeters at birth, classification of head circumference according to birth GA in Z score (being microcephaly with two or more standard-deviations below the mean for the gestational age, sex and acute microcephaly defined by the Z score of the cephalic perimeter < -3), classification according to the pathology (confirmed CSZ; presumed CSZ; CMV), presence of epilepsy, use of medications (type and number) and in the GMFCS (I to V) classification.

The symmetry of the variables was verified with the Shapiro Wilk's test. The quantitative variables with symmetric distribution were described by the mean and the standard-deviation. The categorical variables were compared among themselves by the Fisher Exact test, considering a significance level of 5%.

This article is part of a study entitled "Characterization of Embryopathy Phenotype by ZIKA" approved by the Ethics and Research. The parents or guardians signed an Informed Consent Form (ICF) and the researchers signed the Data Use Commitment Term (DUCT).

3. Results

In this study 21 children were accompanied, 10 female and 11 male children. The clinical, epileptical and EEG characteristics these children were described in Table 2. The average gestational age was 36,69 weeks. According to Table 3, twenty mothers' patients (95,2%) completed the prenatal appointments. Ten pregnant (47,6%) have not been tested previously to ZIKV, 9 (42,9%) were negative and 2 (9,5%) were positive to the test. The pregnant have not collected the PCR test to CMV during the pregnancy period.

The main for the cephalic perimeter for girls was $29,2 \pm 1,73$ cm and for boys $29,61 \pm 0,80$ cm. On the curve of Z score for the cephalic perimeter for weight and sex, 10 patients (47,6%) were classified as below 2 standard-deviations and 11 patients (52,4%) below 3 standard-deviations.

Among the 21 patients studied, 9 patients had the CMV diagnosis (42,9%) confirmed by positive urine CMV PCR, 6 to CSZ (28,6%) and another 6 to presumed infection by ZIKV (28,6%). The presumed infection diagnosis was determined through the characteristic phenotype of CSZ however, without laboratory diagnosis due to the difficulty of detecting the virus among the infected pregnant retrospectively.

Among those children, 61,9% ($N = 13$) presented the epilepsy diagnosis, and within those 13, 69,2% ($N = 9$) were using polytherapy in their treatment. In the evaluation of the types of epilepsy, they were all considered as generalized. Between the 13 children, 53,8% ($N = 7$) presented spasms. Among the children who presented epilepsy, the electroencephalographic analysis evidenced that all of them demonstrated asymmetrical disorganized background activity. As for the epileptiform activity, 7,7% ($N = 1$) presented focal activity, 38,5% multifocal activity ($N = 5$), 30,8% generalized activity ($N = 4$) and 23,1% hypsarrhythmia ($N = 3$). In Table 2, we observed that there was not a difference between the frequency of the epilepsy among children with microcephaly by CSZ or CMV (Fisher Exact test $p = 0,673$).

Analyzing the electroencephalographic profile, presence of epilepsy, type of crisis and use of antiepileptic only by patients with CSZ confirmed and presumed, it has been observed that the presence of epilepsy was 66,7% ($N = 8$), being the generalized crisis the most frequent type. Epileptic spasms were presented in 37,5% ($N = 3$). The patients using more than one medication were 62,5% ($N = 5$). The background activity of patients presenting epilepsy were disorganized and asymmetrical. When present, the most frequent epileptiform activities were multifocal and generalized. In Table 4, the data referring to the cerebral involvement between all the congenital infections included in the study was compared.

The cerebral palsy was found in 91,7% of patients with CSZ confirmed or presumed and in 66,7% of patients with CMV. The GMFCS classification which represents a greater severity had a similar percentage in both groups, 55,6% in the CMV and 58,3% in the CSZ confirmed and presumed group. The GMFCS scale was applied during the evaluations in all patients. A major part of them, 66,6% were classified in IV and V, 14,3% in III and 14,3% in I.

A comparison between the Z score of cephalic perimeter and the GMFCS classification was realized and described in Table 5. It's noticeable that 100% of children classified as GMFCS I and I had the cephalic perimeter between -2 and -3 standard-deviations, and 66,7% of patients with scores below -3 standard-deviations presented the most

severe classification in the GMFCS. The result of the Fisher Exact Test was 0,273.

In the association between epilepsy with the GMFCS classification, the result of the Fisher Exact Test was 0,007, thus showing that there was a direct association between the two variables. Through the adjusted standardized residual analysis, patients with GMFCS IV (100%) and V (75%) have more predisposition to present epilepsy whereas patients with GMFCS I have less. When compared, the etiology of patients with epilepsy according to the classification, a p significant value was not obtained. The association is described in Table 6.

4. Discussion

The congenital infections are implicated in the development of epilepsy, changing in the neurologic physical exam and in motor development. As a result of ZIKV, the microcephaly outbreak highlighted the importance of studies which could establish the relation between the microcephaly and cerebral involvement. This study included 21 patients with a microcephaly diagnosis due to congenital CMV or either CSZ confirmed or presumed. The majority of the patients of this study had the epilepsy diagnosis, 61,9% ($N = 13$) and 69,2% ($N = 9$) demonstrated that epileptic seizures may be difficult to control. The background activities of patients who presented epilepsy are disorganized and asymmetrical, thus showing the immaturity of the NCS. From the 13 children diagnosed with epilepsy, 50,8% ($N = 7$) presented spasms. All children started with epileptic seizures before reaching 1 year of age and there was not difference in this timepoint between children with CSZ and CMV.

This study compared the neurological clinic and the electroencephalographic patterns of 21 patients with microcephaly and diagnosis of CMV and either CSZ confirmed or presumed. It was found that there is a prevalence of epilepsy in this study similar to the consulted literature (66,7% of children with infection by ZIKV or probable and 61,9% in the total sample). The form in which the epilepsy behaves was also similar, since the polytherapy is presented in 62,9% of children with microcephaly which was caused by either an infection or epilepsy. Moreover, all the patients with epilepsy presented disorganized and asymmetric patterns, thus showing the immaturity of the nervous central system. There have not been identified a significant difference when comparing the manifestations in neither the epilepsy profile nor in the electroencephalographic characteristics in patients with CSZ and CMV, probably due to the fact that both the pathogens are causative of microcephaly.

The Carvalho et al. [18]. included 91 children with microcephaly caused by ZIKV. Among these, 65 (71,4%) developed epilepsy during the following, similarly to this study (66,7%). While the epileptic spasms ($N = 55$) were the most common type of convulsion in the first year of life in Carvalho et al.'s studies [18], the current study has found a lower incidence of 37,5%. Among the 65 epileptic participants evaluated by the electroencephalogram, the most frequent patterns observed were focal epileptiform abnormalities (53,2%) as the epileptiform main activity, whereas the current study has shown the multifocal activity and generalized ones as being most prevalent [18].

In the study of Kwak et al. [19], which included patients with microcephaly caused by CMV, 12 out of the 31 (38,7%) developed epileptic seizures, similarly to this study (44,4%). The study of Suzuki et al. [20] has found an incidence of epilepsy associated with the congenital CMV which varied between 10% and 56% as well as this study (55,6%).

In the study of Melo et al. [21], during the motor evaluation of the 59 patients included in the study, a child (2%) was classified as GMFCS level I, three (5%) as GMFCS level II, seven (12%) as GMFCS level IV and 48 (81%) as GMFCS level V. Similarly to our study, these results exemplify that the acute impairment of the motor function (GMFCS level V) occurred in most of the cases, mainly when related to the lowest score of the cephalic perimeter from birth. The level V on the GMFCS scale indicates that there is an acute motor impairment and that the children has an impairment of functional capacity, thus needing the assistance in

Table 2
Clinical, epileptic, and EEG characteristics of the sample.

Name	Sex	Cephalic Perimeter Measure (cm)	Cephalic Perimeter at birth	Resulted of collected ZIKV test	Congenital Infection	Epilepsy	Types of epilepsy	Spasms	Antiepileptic usage	Medications	Background Activity -Asymmetrical	Background Activity - Disorganized	Epileptiform activity	GMFSC -Classification	Cerebral Palsy
EGCM	Male	31	<2SD	Negative	CMV	No	-	-	-		-	-	-	2	No
TDPD	Male	28,5	<3SD	Negative	CMV	Yes	Generalized	Yes	Monotherapy	Valproate	Yes	Yes	Generalized	5	Yes
ILP	Male	29	<3SD	Not tested	CMV	Yes	Generalized	Yes	Polytherapy	Phenobarbital and Valproate	Yes	Yes	Multifocal	5	Yes
JMM	Male	30,8	<2SD	Not tested	Presumed Infection by ZIKV	Yes	Generalized	No	Polytherapy	Clonazepam and Valproate	Yes	Yes	Generalized	4	Yes
YVMW	Female	26,5	<3SD	Positive	ZIKV	Yes	Generalized	No	Polytherapy	Clonazepam and Phenobarbital	Yes	Yes	Multifocal	5	Yes
JVA	Male	29.5	<2SD	Not tested	Presumed Infection by ZIKV	No	-	-	-		-	-	-	5	Yes
VIC	Female	27	<3SD	Not tested	ZIKV	Yes	Generalized	No	Monotherapy	Phenobarbital	Yes	Yes	Generalized	4	Yes
ILOC	Female	30,5	<2SD	Negative	CMV	No	-	-	-		-	-	-	1	No
LCB	Female	32	<2SD	Positive	ZIKV	No	-	-	-		-	-	-	1	No
KAJPM	Male	30	<2SD	Not tested	ZIKV	Yes	Generalized	No	Monotherapy	Valproate and Phenobarbital	Yes	Yes	Generalized	3	Yes
EGMS	Male	30	<2SD	Not tested	ZIKV	Yes	Generalized	No	Monotherapy	Valproate	Yes	Yes	Multifocal	5	Yes
HKSR	Female	30	<3SD	Negative	CMV	Yes	Generalized	No	Polytherapy	Valproate and Phenobarbital	Yes	Yes	Multifocal	5	Yes
LNP	Female	28	<3SD	Negative	Presumed Infection by ZIKV	No	-	-	-		-	-	-	3	Yes
MLSL	Female	30	<2SD	Negative	CMV	Yes	Generalized	Yes	Polytherapy	Valproate and Vigabatrin	Yes	Yes	Hipsarrhythmia	5	Yes
HAB	Female	28	<3SD	Not tested	ZIKV	No	-	-	-		-	-	-	5	Yes
LHLM	Female	30	<3SD	Negative	Presumed Infection by ZIKV	Yes	Generalized	Yes	Polytherapy	Valproate, Clobazam and Vigabatrin	Yes	Yes	Hipsarrhythmia	5	Yes
ALP	Female	30	<2SD	Not tested	CMV	No	-	-	-		-	-	-	1	No
LRFSP	Male	29	<3SD	Not tested	Presumed Infection by ZIKV	Yes	Generalized	Yes	Polytherapy	Valproate, Phenobarbital and Levetiracetam	Yes	Yes	Multifocal	5	Yes
AA	Male	29	<3SD	Negative	CMV	No	-	-	-		-	-	-	3	Yes
BGR	Male	30	<3SD	Negative	Presumed Infection by ZIKV	Yes	Generalized	Yes	Polytherapy	Valproate and Clobazam	Yes	Yes	Focal	5	Yes
AFR	Male	29	<2SD	Not tested	CMV	Yes	Generalized	Yes	Polytherapy	Valproate, Phenobarbital and Levetiracetam	Yes	Yes	Hipsarrhythmia	5	Yes

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Table 3
Samples' description.

Sex	Female	10 (47,6%)
	Male	11 (52,4%)
Completed Prenatal	Yes	20 (95,2%)
	No	1 (4,8%)
ZIKV's collect during pregnancy	Yes	11 (52,4%)
	No	10 (47,6%)
Resulted of collected ZIKV test	Positive	2 (9,5%)
	Negative	9 (42,9%)
	Not tested	10 (47,6%)
Cephalic Perimeter Measure	Females	29,2 ± 1,73 cm
	Males	29,61 ± 0,80 cm
Cephalic Perimeter at birth	Z < -2	10 (47,6%)
	Z < -3	11 (52,4%)
Congenital Infection	CMV	9 (42,9%)
	ZIKV	6 (28,6%)
	Presumed Infection by ZIKV	6 (28,6%)

Table 4
Cerebral Involvement in patients with CMV, CSZ confirmed or presumed.

	CMV Number/ Total (%)	CSZ confirmed or presumed Number/ Total (%)	Fisher Exact Test
Epilepsy			0,673
Yes	5 (55,6%)	8 (66,7%)	
No	4 (44,4%)	4 (44,4%)	
Spasms	4/5 (80%)	3/8 (37,5%)	0,266
Antiepileptic usage			
Monotherapy	1/5 (20%)	3/8 (37,5%)	
Polytherapy	4/5 (80%)	5/8 (62,5%)	
Background Activity			
Asymmetrical	5/5 (100%)	8/8 (100%)	
Disorganized	5/5 (100%)	8/8 (100%)	
Epileptiform activity			
Focal	0/5 (0%)	1/8(12,5%)	
Multifocal	2/5 (40%)	3/8 (37,5%)	
Generalized	1/5 (20%)	3/8 (37,5%)	
Hipsarrhythmia	2/5 (40%)	1/8 (12,5%)	
Cerebral Palsy	6/9 (66,7%)	11/12 (91,7%)	0,355
GMFCS 5	5/9 (55,6%)	7/12 (58,3%)	0,077

performing tasks as well as adapted equipment.

Lage et al.'s study [22] has shown that the neurological abnormalities were found in the most of cases: hypertonia/ spasticity (97%), delayed neurological development from the beginning (92,8%) and hyperreflexia (73,3%), these suggest early signs of acute motor impairment. These findings are consistent with the clinical diagnosis for cerebral palsy which describes a group of permanent disturbances on the development of movement and posture, causing activity limitation. It was also noticed that the cephalic perimeter is likely to be a prognostic marker of a worst development result at this age.

Table 5
Comparison between the GMFCS score and the Z score of the Cephalic Perimeter.

GMFCS Classification	I Number/ Total (%)	II Number/ Total (%)	III Number/ Total (%)	IV Number/ Total (%)	V Number/ Total (%)	Total	p Value
CP < -2	3/10(100%)	1/10(100%)	1/10(33,3%)	1/10 (50%)	4/10(33,3%)	10	0,273
CP < -3	0/11 (0%)	0/11 (0%)	2/11(66,7%)	1/11 (50%)	8/11(66,7%)	11	

Table 6
Comparison between the GMFCS and the epilepsy prevalence and its cause.

GMFCS Classification	I Number/ n = 3(%)	II Number/ n = (%)	III Number/ Total (%)	IV Number/ Total (%)	V Number/ Total(%)	P value
With Epilepsy	0/3 (0%)	0/1 (0%)	1/3(33,3%)	2/2(100%)	10/12(83,3%)	0,007
CMV	2/3 (66,7%)	1/1 (100%)	1/3(33,3%)	-	5/12 (41,7%)	0,622
ZIKV + probable ZIKV	1/3 (33,3%)	-	2/3 (66,7%)	2/2(100%)	7/12 (58,3%)	

In Quilião et al. 's study [23], among the 11 evaluated children, 8 presented acute microcephaly associated with motor disability and/or epilepsy. It was observed that children with less motor impairment had minor relative change in head circumference, normal electroencephalogram tracing and absence of seizures [23]. In this study, 100% of children with GMFCS classification I and II had lower values of Z score of cephalic perimeter when compared to other classifications and the patients with more acute GMFCS levels were more likely to be epileptic.

Taking into consideration the few numbers of participants in this study as well as the clinical and physiopathological diversity of CSZ, these results must not be generalized. The monitoring of these children is crucial to better comprehend the real impact of the ZIKV on motor development and its abilities. This represents the first step to understand the motor function in children with CZS using a validated scale and also to define necessary interventions to minimize the motor sequels and functional limitations of these children. Another obstacle was the difficulty in diagnose the infection by ZIKV retrospectively in asymptomatic women, this is because the exposure to the ZIKV is extraordinarily difficult to determine if the patients were not tested using the PCR during the acute infection.

Comprehending the perspective of the gross motor function and promptly identifying possible deficiencies helps rehabilitation teams create better treatment plans based on the motor prognoses. Better plans will give children better support so that they may more efficiently reach all their motor potential.

To conclude, we emphasize the importance of investigation of the CSZ specially when a child was born from an asymptomatic mother presents microcephaly from birth or during the postnatal period. The child monitoring, associated with electroencephalogram, may help to produce a more precise diagnosis, and therefore anticipate a neurological rehabilitation of these patients as well as raise parents' awareness to a possible prognosis. Future works are necessary to (i) predict the evolvement of the ZIKV in relation to the electroencephalographic pattern and to the epilepsy profile, (ii) optimize the treatment of these patients, (iii) better understand how the ZIKV infection behaves in short and long term and (iv) to evaluate the persistency of the neurological sequels over time.

Data sharing

All data are publicly available and listed in this article.

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CRediT authorship contribution statement

Lavinia Schüller Faccini: Conceptualization, Investigation, Writing

– original draft, Writing – review & editing, Supervision. **Luciana Friedrich:** Conceptualization, Methodology, Investigation, Writing – original draft, Writing – review & editing, Supervision. **Sara Kvitko de Moura:** Conceptualization, Methodology, Formal analysis, Investigation, Writing – original draft, Writing – review & editing. **Fernanda Diffini Santa Maria:** Resources, Data curation, Investigation, Data curation. **Steice da Silva Inácio de Bone:** Resources, Data curation, Investigation, Visualization.

Declaration of Competing Interest

We declare there are no competing interests.

References

- [1] L. Schuler-Faccini, E.M. Ribeiro, I.M. Feitosa, et al., Possible Association Between Zika Virus Infection and Microcephaly. Brazil, 2015. *MMWR Morb Mortal Wkly Rep* 65, 2016, pp. 59–62 (DOI: <https://doi.org/10.15585/>).
- [2] Brasil. Ministério da Saúde, Secretaria de Vigilância em Saúde, Registro de Eventos em Saúde Pública. (RESP-Microcefalia) [Internet], 2015 (Disponível em: <http://www.resp.saude.gov.br/microcefalia/#/painel>. Acesso em: 22 abr. 2020).
- [3] P.A.M. Kanda, A.A.X. Aguiar, J.L. Miranda, A.L. Falcao, C.S. Andrade, L.N.S. Reis, E.W.R.B. Almeida, Y.B. Bello, A. Monfredinho, R.G. Kanda, Sleep EEG of microcephaly in Zika outbreak, *Neurodiagnostic J.* 58 (2018) 11–29, <https://doi.org/10.1080/21646821.2018.1428461>.
- [4] M.L. Nunes, C.R. Carlini, D. Marinovic, F.K. Neto, H.H. Fiori, M.C. Scotta, et al., Microcephaly and Zika virus: a clinical and epidemiological analysis of the current outbreak in Brazil, *J. Pediatr.* 92 (2016) 230–240, <https://doi.org/10.1016/j.jpeds.2016.02.009>.
- [5] T. Saad, et al., Neurological manifestations of congenital Zika virus infection, *Childs Nerv. Syst.* 34 (2018) 73–78.
- [6] Z.A. Klase, S. Khakhina, B. Schneider Ade, M.V. Callahan, J. Glasspool-Malone, R. Malone, Zika fetal neuropathogenesis: etiology of a viral syndrome, *PLoS Negl. Trop. Dis.* 10 (8) (2016), <https://doi.org/10.1371/journal.pntd.0004877> e0004877. Aug 25. (PMID: 27560129; PMCID: PMC4999274).
- [7] S. Cavalheiro, et al., Microcephaly and Zika virus: neonatal neuroradiological aspects, *Child's Nervous Syst.* 32 (6) (2016) 1057–1060. June.
- [8] M. Del Campo, I.M. Feitosa, E.M. Ribeiro, D.D. Horovitz, A.L. Pessoa, G.V. França, A. Garcia-Alix, M.J. Doriqui, H.Y. Wanderley, M.V. Sanseverino, J.I. Neri, J. M. Pina-Neto, E.S. Santos, I. Verçosa, M.C. Cernach, P.F. Medeiros, S.C. Kerbage, A. A. Silva, V. van der Linden, C.M. Martelli, M.T. Cordeiro, R. Dhalia, F.S. Vianna, C. G. Vitoria, D.P. Cavalcanti, L. Schuler-Faccini, Zika Embryopathy task force- Brazilian Society of Medical Genetics ZETF-SBGM. The phenotypic spectrum of congenital Zika syndrome, *Am. J. Med. Genet. A* 173 (4) (2017) 841–857, <https://doi.org/10.1002/ajmg.a.38170>. Apr. (PMID: 28328129).
- [9] A. Costello, T. Dua, P. Duran, M. Gülmezoglu, O.T. Oladapo, W. Perea, J. Pires, P. Ramon-Pardo, N. Rollins, S. Saxena, Defining the syndrome associated with congenital Zika virus infection, *Bull. World Health Organ.* 94 (6) (2016), <https://doi.org/10.2471/BLT.16.176990>, 406–406A. Jun 1. (PMID: 27274588; PMCID: PMC4890216).
- [10] A.L. Carvalho, P. Ventura, T. Taguchi, I. Brandi, C. Brites, R. Lucena, Cerebral palsy in children with congenital Zika syndrome: a 2-year neurodevelopmental follow-up, *J. Child Neurol.* 35 (3) (2020) 202–207, <https://doi.org/10.1177/0883073819885724>. Mar. (Epub 2019 Nov 13. PMID: 31718421).
- [11] A. Carvalho, C. Brites, G. Mochida, P. Ventura, A. Fernandes, M.L. Lage, T. Taguchi, I. Brandi, A. Silva, G. Franceschi, P. Lucena, R. Lucena, Clinical and neurodevelopmental features in children with cerebral palsy and probable congenital Zika, *Brain and Development* 41 (7) (2019) 587–594, <https://doi.org/10.1016/j.braindev.2019.03.005>. Aug. (Epub 2019 Mar 23. PMID: 30914212).
- [12] International Fetal and Newborn Growth Consortium for the 21st century (INTERGROWTH-21st). Sobre INTERGROWTH-21st. c2009–2016 [Internet] [citado 2020 nov 24]. Disponível em, <https://intergrowth21.tghn.org/about/sobre-intergrowth-21st-portuguese/>, 2016.
- [13] Brasil. Ministério da Saúde, Secretaria de Vigilância em Saúde, Secretaria de Atenção à Saúde, Orientações integradas de vigilância e atenção à saúde no âmbito da Emergência de Saúde Pública de Importância Nacional : procedimentos para o monitoramento das alterações no crescimento e desenvolvimento a partir da gestação até a primeira infância, relacionadas à infecção pelo vírus Zika e outras etiologias infecciosas dentro da capacidade operacional do SUS [recurso eletrônico] / Ministério da Saúde, Secretaria de Vigilância em Saúde, Secretaria de Atenção à Saúde. – Brasília : Ministério da Saúde, 2017 (158 p. : il).
- [14] R.S. Fisher, J.H. Cross, J.A. French, N. Higurashi, E. Hirsch, F.E. Jansen, L. Lagae, S.L. Moshé, J. Peltola, E. Roulet Perez, I.E. Scheffer, S.M. Zuberi, Operational classification of seizure types by the international league against epilepsy: position paper of the ILAE commission for classification and terminology, *Epilepsia.* 58 (4) (2017) 522–530, <https://doi.org/10.1111/epi.13670>. Apr. (Epub 2017 Mar 8. PMID: 28276060).
- [15] G.L. Holmes, C.T. Lombroso, Prognostic value of background patterns in the neonatal EEG, *J. Clin. Neurophysiol.* 10 (3) (1993) 323–352, <https://doi.org/10.1097/00004691-199307000-00008>. Jul. (PMID: 8408599).
- [16] R. Palisano, P. Rosenbaum, S. Walter, D. Russell, E. Wood, B. Galuppi, Development and reliability of a system to classify gross motor function in children with cerebral palsy, *Dev. Med. Child Neurol.* 39 (4) (1997) 214–223, <https://doi.org/10.1111/j.1469-8749.1997.tb07414.x>. Apr. (PMID: 9183258).
- [17] Brasil, Ministério da Saúde, Atenção ao pré-natal de baixo risco, Brasília: Ministério da Saúde, *Cadernos de Atenção Básica*, 2012, p. 32.
- [18] M.D.C.G. Carvalho, R.A.A. Ximenes, U.R. Montarroyos, et al., Early epilepsy in children with Zika-related microcephaly in a cohort in Recife, Brazil: characteristics, electroencephalographic findings, and treatment response, *Epilepsia.* 61 (2020) 509–518, <https://doi.org/10.1111/epi.16444>.
- [19] M. Kwak, M.S. Yum, H.R. Yeh, H.J. Kim, T.S. Ko, Brain magnetic resonance imaging findings of congenital cytomegalovirus infection as a prognostic factor for neurological outcome, *Pediatr. Neurol.* 83 (2018) 14–18, <https://doi.org/10.1016/j.pediatrneurol.2018.03.008>. Jun. (Epub 2018 Mar 21. PMID: 29681488).
- [20] Y. Suzuki, Y. Toribe, Y. Mogami, K. Yanagihara, M. Nishikawa, Epilepsy in patients with congenital cytomegalovirus infection, *Brain and Development* 30 (6) (2008) 420–424, <https://doi.org/10.1016/j.braindev.2007.12.004>. Jun. (Epub 2008 Jan 22. PMID: 18215482).
- [21] A. Melo, G.L. Gama, R.A. Da Silva Júnior, P.L. De Assunção, J.S. Tavares, M.B. Da Silva, K.N.F.S. Costa, M.L. Vânia, M.A. Evangelista, M.M.R. De Amorim, Motor function in children with congenital Zika syndrome, *Dev. Med. Child Neurol.* 62 (2) (2020) 221–226, <https://doi.org/10.1111/dmnc.14227>. Feb. (Epub 2019 Apr 4. PMID: 30945276).
- [22] Lage CML, A.L. Carvalho, P.A. Ventura, T.B. Taguchi, A.S. Fernandes, S.F. Pinho, O. T. Santos-Junior, C.L. Ramos, C.M. Nascimento-Carvalho, Clinical, neuroimaging, and neurophysiological findings in children with microcephaly related to Congenital Zika Virus infection, *Int. J. Environ. Res. Public Health* 16 (3) (2019) 309, <https://doi.org/10.3390/ijerph16030309>. Jan 23. (PMID: 30678125; PMCID: PMC6388186).
- [23] M.E. Quilão, F.A. Venancio, L.K. Mareto, S.A. Metzker, A.I.D. Nascimento, D. C. Vitorelli-Venancio, C.D.B. Santos-Pinto, E.F. de Oliveira, Neurological development, epilepsy, and the pharmacotherapy approach in children with congenital Zika syndrome: results from a two-year follow-up study, *Viruses.* 12 (10) (2020) 1083, <https://doi.org/10.3390/v12101083>. Sep 25. (PMID: 32992985; PMCID: PMC7601787).