ISSN: 2690- 909X

Case Report

Advances in Neurology and Neuroscience

Congenital Rubella syndrome cases (based on WHO criteria) one decade after Measle and Rubella vaccination campaign in Tehran; Iran.

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Submitted: 10 Mar 2022; **Accepted:** 18 Mar 2022; **Published:** 26 Mar 2022

Citation: *Mohammad Vafaee-Shahi, Samileh Noorbakhsh, Leila Tahernia, Sarvenaz Ashouri, Aina Riahi (2022). Congenital Rubella syndrome cases (based on WHO criteria) one decade after Measle and Rubella vaccination campaign in Tehran; Iran, Adv Neur Neur Sci, 5(1), 40-44.*

Abstract

Background: *Active CRS surveillance is recommended strongly even in country with full rubella vaccination.*

Objective: *Searching the CRS based on WHO criteria one decade after MR vaccination campaign (2003)*

Methods: *a prospective case control study carried out in a third referral educational hospital (Rasoul Akram hospital) in Tehran, IRAN during five years (2013 -2018). From 89 CRS suspected infants (according to WHO criteria); serum samples were tested for rubella-specific IgG and IgM by ELISA method. Selected samples (positive Rubella IgM) were tested for rubella RNA by reverse transcriptase–polymerase chain reaction (RT–PCR).*

Findings: *"Confirmed CRS" based on positive IgM was found in 5.6% (5/89) all were < 4 months old. Positive RT–PCR detected in 1.2 % (1/89) of "CRS suspected" infants.Except a good correlation between abnormal neurologic findings and positive IgM in "Confirmed CRS "cases, other clinical findings were not related to serologic tests. The "Compatible CRS" cases in the 31 patients who had primary negative IgM serology and did not have sufficient laboratory evidence to confirm CRS, but clinical symptoms favoured CRS.*

Conclusion: *Despite MR vaccination in Iran, after one decade "confirmed CRS" and "compatible CRS" were diagnosed in 5 and 31 from 89 CRS suspected cases. The incidence of "confirmed CRS" in every 100 CRS suspected infants (after campaign) is 5.6 %; and 31 CRS Compatible cases are so important. Without active CRS surveillance; mild infection such as IUGR, hearing loss,heart abnormalities, impaired vision, and mental retardation even in developed country might be missed.Fetal infection is persistent which impose additional costs on the country..Another mass vaccination in women and girls is needed. Also, the anti-rubella IgG testing before pregnancy in women who were not vaccinated; vaccination of women before marriage /preganacy should be obligatory in order to prevent the CRS.*

Keywords: Rubella, Congenital Rubella Syndrome, Surveillance, Immunization Programs

Introduction

First, Gregg's (1941) reported the classical clinical manifestations of congenital rubella syndrome (CRS) with triad of deafness, cataracts and cardiac disease (1) Rubella infection during pregnancy, especially during the first 12 weeks of gestation, can lead to miscarriage, intrauterine fetal death, and a variety of birth defects associated with CRS (intrauterine growth retardation, hearing loss, impaired vision, and mental retardation) [1-3]. Complete evaluation of rubella infection should be performed in any newborn who is born of a mother who has rubella or even suspected to have rubella at any age of pregnancy [1,3] Specific prenatal tests are available to diagnose maternal rubella infection [1,2,3] Maternal antibody at the time of delivery usually is composed entirely of IgG. In contrast, the infant titer consists of fetal IgM, presumably fetal IgG, and occasionally fetal IgA and transplacentally derived maternal IgG [1,4]. In suspected pregnant mothers, serum IgG should be checked at 3, 5 and 6 months and followed with another sample if necessary. Stable high IgG levels at this age indicates intrauterine rubella infection [1, 2 ,4]. Maternal IgM antibody normally is not passed transplacentally, but a fetus challenged in utero with a virus can have specific IgM antibodies against the rubella virus. Often a fetus with detectable virus-specific IgM will have severe in utero disease caused by the virus. Because acquired rubella infection before first year is unlikely, CRS can be demonstrated by looking for the specific virus antibody (IGM) in patient's serum, or a urine culture for rubella infection [2,4]. The use of PCR in amniotic fluid samples (100% sensitivity) and/ or in chorionic villus samples (83%sensitivity) is definitive. The specific IgG titer in cord blood is not valued due to incomplete deletion of IgG (most of which are of maternal origin) or rheumatoid factor (false positive) [1, 4]. The second approach is to monitor whether IgG levels remain permanent or not. Positive IgG after 6 months of age may indicate either prenatal or postnatal infection (4) If the infant's value is the result solely of transplacentally acquired antibody, it should drop fourfold to eightfold by the time the infant reaches 3 months of age and continue to fall to nondetectable values by 6 to 8 months of age. Because the antibody value in some congenital infections also may fall, disappearance of antibody in serum does not rule out in utero infection completely. In questionable cases, the study of IgG rubella antibody avidity may be useful. The detection of IgG1 (which is low) is diagnosed as prenatal infection [1,2,4].

In 1969, a weakened live rubella vaccine introduced and used worldwide in all children and young adults, especially girls to prevent CRS. Due to teratogenic effect of rubella virus; the high cost–benefit ratio of immunizations reduces public health costs [5]. Active detection of CRS cases is required even with a major symptom (according to WHO classification). With estimation of births, it could be possible to estimate the number of congenital rubella cases per 1000 live births [5,6].WHO reported that in the absence of widespread rubella vaccination, the incidence of CRS varies between 0.1–0.2 per 1,000 live births, with higher rates per 1,000 live births during epidemics [5,6] During rubella outbreaks, rates of CRS per 1000 live births were at least 1.7 in Jamaica, 0.7 in Oman, 2.2 in Panama, 1.5 in Singapore, 0.9 in Sri Lanka, and 0.6 in Trinidad and Tobago [6]. These rates are similar to those reported from industrialized countries during the pre-vaccine era [6,7] For example, results of CRS study in Brazil defined, the cost of first year of follow-up for CRS cases was US $ 61,824, and it will continue for the following years and until the end of life [8] Routine CRS surveillance should be done according to the guideline of World Health Organization and CDC. However, some symptoms may be late onset. After one year the diagnosis would be very difficult [6,7, 9]. The number of CRS cases per 1000 live births should be reported annually [6, 9]. Active detection of cases of congenital rubella is required even with a major symptom (according to WHO classification). With estimation of births, it could be possible to estimate the number of CRS cases per 1000 live births [6 ,9].

The prevalence of CRS in Iran calculated approximately 0.2/1000 before the MR vaccination campaign in Iran (2003) [10]. During last decade except serologic studies of population [11-15], insufficient studies for the active CRS surveillance system in our country were reported. Rubella vaccine are used in IRAN and a routine surveillance for CRS was organized nationwide from 2003; mass rubella vaccination program is introduced in our system since 2004 [16 , 17]. Now, due to limited CRS surveillance and reporting systems, data on the incidence of rubella and CRS in Iran are scant [18-23]. This study provides data on the prevalence of CRS in Tehran (Capital city); IRAN one decade after MR vaccination campaign (2003). This data could potentially be used as baseline data, which in conjunction with an appropriate method, to establish a surveillance system for rubella vaccination in Iran to establish a proper time for massive vaccination in Iran.

Methods

A prospective case control study was carried out in a third referral educational hospital (Rasoul Akram hospital) affiliated by IUMS in Tehran, IRAN during five years (2014 -2018). All project parents adhers to the principles of the Helsinki Declaration. Written informed consent was obtained from the parents of all children enrolled in the study. Performing all clinical examinations and diagnostic tests will be at the expense of the plan.

A preliminary tests and standard examination were requested after initial examinations by a paediatrician for excluding other etiologic causes except intrauterine infection Diseases such as chromosomal and metabolic diseases, sepsis, hyperamonemia, other diagnosed CNS disorders except CRS, etc. were done and all were excluded from this study.

Exclusion Criteria Included

no consent to participate in the study - evidence of other causes describing clinical symptoms. Around 2 mililiters of blood sample was taken from patients and centrifuged and stored at Rasool Akram Hospital Research Laboratory. In control group, 70 children were selected in the same age group who were referred to

the outpatient clinic for examinations, or those who had undergone elective surgery in the surgical ward (without any deficits). Blood serum was first separated and centrifuged at this center. These samples were stored at -30 ° C. Then the presence of rubella IgM and IgG antibodies in samples was evaluated by ELISA (RADEM-K kit made in Italy). Then frequency of seropositivity in these antibodies was reported for these patients. Information on infants and antibody levels and seropositivity was collected in a checklist containing study variables. Rubella IgM samples were tested for rubella RNA by reverse transcriptase–polymerase chain reaction (RT–PCR); and recorded in the supplemental information forms. The collected data and laboratory results were statistically analyzed by SPSS software version 13.5. Chi-square and student's t-tests were used to compare the mean between the two groups. p-values less than 0.05 were considered valuable.

Cases Definition

Inclusion criteria as follows: A suspected case is any infant <1 year of age in whom a health worker suspects CRS (Based on WHO criteria) figure-1:

1. Suspected CRS: Having at least one major symptom or 2 minor symptoms.

a. Major symptoms included: Congenital heart disease, and/or suspicion of deafness, and/or one or more ophthalmic signs.

b. Minor Symptoms: Jaundice on birth day of hepatomegaly, splenomegaly, steatosis, petechiae or purpura, muffin skin lesions, developmental disorders, seizures,

2. Compatible CRS: who had primary negative IgM serology and did not have sufficient laboratory evidence to confirm CRS, but 2 Complications of the first symptoms or one of first and one of the second symptoms were observed in patients. The first symptoms included congenital cataract, glaucoma, congenital heart disease, hearing loss, pigmented retinopathy, and the second symptoms were purpura, splenomegaly, microcephaly, mental retardation, meningoencephalitis, radiolucent bone disease, jaundice that appears within the first 24 hours of birth, but laboratory evidence is not sufficient to prove congenital rubella.

3. Confirmed CRS: Confirmed congenital abnormalities with laboratory evidence positive IgM /or PCR.

Results

During 5 years; 186 infants (< 1-year-old) with Intra uterine infection suspected infants referred to our hospital, eight infants (< 3 months) died due to severe organ (heart or brain) diseases before complete preliminary and specific serologic tests and excluded.

In the first step, 89 patients were excluded after initial examinations and Lab test with other diagnosis: chromosomal, metabolic diseases, sepsis, hyperammonaemia, CNS disorders, asphyxia, subdural hematoma, etc) and were not followed for CRS.

From 98 cases were eligible for intrauterine infection screening (Mean age: 4.74 + 3 3.78 months), 55.2% were male and 44.8% were female. Serologic intrauterine infections (TOCH) including rubella antibodies (IgM&IgG) were surveyed by ELISA method in sera of all cases and age matched controls. Congenital toxoplasmosis in 12 and congenital CMV in 20 infants confirmed retrospectively and excluded from study.

Overall 87% of TORCH suspected cases were positive for anti-rubella IgG, negative IgM and CRS were not the final diagnosis in them.

89 CRS suspected infants studied. Positive Rubella-IgM and IgG found in 5.6 % and 33.3% of infants. The positive Rubella IgM in cases was 2.5 times more than control (healthy infants) but it was not significant. (P-value = 0.1) In contrast, previous immunity in control group was significantly higher (80%) than case group (80 % vs 33%; p=0.0000). All clinical signs between groups with positive and negative IgM was significant (P=0.000). But not different for isolated signs (icter, hepatosplenomegaly, mental retardation, eye involvement, deafness, IUGR, CNS involvement) except for congenital heart disease. Positive IgM detected just in 5.6% (5/89) of infants. Positive RT–PCR detected in 1.2 % (1/89) of CRS suspected cases.

Confirmed CRS: congenital abnormalities with laboratory evidence (positive serology/PCR) were diagnosed in 5.6 % (5/89) of CRS suspected cases, Positive RT–PCR detected in 1.2% (1/89) of CRS suspected cases; All confirmed CRS cases were less than 4 months old. Except a good correlation between abnormal neurologic findings and positive IgM in Confirmed CRS cases; other clinical findings were not related to serologic tests.

Compatible CRS diagnosed in the remaining in 34.8% (n=31) patients (who had primary negative IgM serology and did not have sufficient laboratory evidence to confirm congenital rubella), but clinical symptoms favoured congenital rubella (two complications of first symptoms or one from. second symptoms).

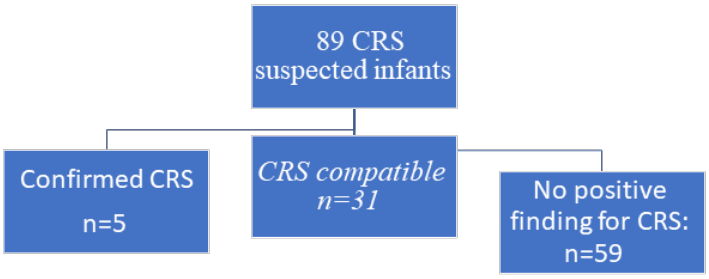

Figure 1: Schematic cases were eligible for CRS and final diagnosis.

Discussion:

Here, 89 CRS suspected infants (having at least one major symptom or 2 minor symptoms in favour of CRS) were studied. Confirmed CRS (positive IgM) was found in 5.6% (5/89); Positive RT–PCR detected in 1.2 % (1/89) of cases. All "confirmed CRS" cases were young (< 4 months old). The "Compatible CRS" cases

diagnosed in 34.8% (n=31) of cases who had primary negative IgM and RNA, and did not have sufficient laboratory evidence to "confirm CRS", but clinical symptoms favoured CRS (Bases on WHO criteria). Some previous studies [1-5] determined the rubella virus may be eliminated before they were delivered. Additionally, rubella IgM in congenitally infected infants may also become negative before delivery. So, the diagnosis of CRS based on the positive results of IgM and RNA may underestimate the infection rate and 31 CRS Compatible cases are acceptable in present study. Except a positive correlation between abnormal neurologic findings and positive IgM in Confirmed CRS cases, other clinical findings were not related to serologic tests. Like other references [2-3] This indicated the importance of having at least one other symptom except heart disease, as an adjunct to the diagnosis of CRS in "CRS suspected" infants. [2-3].

Overall 87% of 98 cases were eligible for intrauterine infection (Mean age: 4.74 + 3 3.78 months) were positive for anti-rubella IgG (negative IgM and RNA) and CRS were ruled out in them. Probably, those had previous immunity, (trans placental immunity). Near 33.3% of CRS suspected infants (n= 89) were positive for anti-rubella IgG, and had protection. Positive IgM found in 5.6 % of them, which was 2.5 times more than healthy controls (no significant) In contrast, previous immunity in control group was significantly higher (80% vs 33%; p=0.0000). Here, the rate of rubella immunity in studied infants was above 80%; which is similar to that in other countries during the rubella pandemic of the 1960's [7-9]. The above findings are close to Sadigh et al [11] study; they show vulnerability to rubella infection is persistent in the at-risk population, which can increase fetal infection and impose additional costs on the country [11]. The seroposivity in studied cases is very close to Marashi et al study [18]. Like us, Marashi et al (2011) found anti-rubella IgG antibody in 85% and rubella IgM seropositivity rate in 0.5% OF NEONATES WITH CONGENITAL DEFECTS IN 4 PROVINENCE OF IRAN [18]. The seroposivity results is higher than 72.7% reported by Almasi et al in umbilical cord blood samples [19]. Just, 13% of our studied infants were non-immune, there was still a risk of infection in pregnant women, so eliminating CRS is impossible at present time.

First report for Rubella immunity in Iran (1974) by Saidi (10) published in WHO bultin (not available). The rate of rubella immunity in Iranian women of childbearing age was flactuated between 70% and 95% from 1968 to 1995 [11-13]. The immunity of children and adult females to rubella virus infection in Tehran by Modarres et al (1996) determined the Rubella immunity rate 79-84% [13]. Then, it decreased significantly to 78.4% (2001) in Doroudchi et al study [15]. Role of National Immunization Technical Advisory Group on improvement of immunization programmes in the Islamic Republic of Iran published (2010) by Zahraei et al. Efficacy of measles and rubella vaccination 1 year after the nationwide campaign in Shiraz, Iran studied by Pourabbas et al. and in Tehran (capital city) in Iran by Soleimanjahi et al. Namaei et al. surveyed the CRS in infants of women vaccinated during or just before pregnancy with measles-rubella vaccine [20, 17, 21, 22]. Hamkar et al. reported the distinguishing between primary infection and reinfection with rubella vaccine virus by IgG avidity assay in pregnant women; 98% of the susceptible group (723/738) acquired immunity against rubella after vaccination [23].

According to the WHO recommendation in countries where Rubella has not been eradicated, assessment of Rubella epidemics should be used to activate CRS surveillance. The number of CRS cases per 1000 live births should be reported annually [6, 9]. Active detection of cases of CRS is required even with a major symptom (according to WHO classification). With estimation of births, it could be possible to estimate the number of CRS per 1000 live births. WHO had the reports of CRS prevalence in various regions of world [6, 9]. Before the MR vaccination campaign (2003) in Iran, the epidemiology of rubella and CRS was not clear. In spite of this unclear epidemiology, mass rubella vaccination was launched in Iran since 2004. The incidence of "confirmed CRS" in present study after campaign was 5.6%; lower than incidence in Tanzania (12%). The prevalence of CRS in Iran calculated approximately 0.2/1000 before the MR vaccination campaign (2003) in Iran [11].

Conclusion

Despite MR vaccination in Iran, after one decade "confirmed CRS" and "compatible CRS" were diagnosed in 5 and 31 (from 89 CRS suspected cases). The incidence of "confirmed CRS" in every 100 CRS suspected infants in present study (after campaign) is 5.6 %. Positive rubella IgM test in blood samples (infants < 6 month) is diagnostic for CRS. In addition, due to underestimate the infection rate based on positive results of IgM and RNA, 31 CRS Compatible cases are important. Above finding showed that vulnerability to rubella infection is persistent in the at-risk population, which can increase fetal infection and impose additional costs on the country. Without active CRS surveillance; CRS especially mild infection (not classic form) such as IUGR (intrauterine growth retardation), hearing loss, heart abnormalities, impaired vision, and mental retardation even in developed country might be missed It is recommended to conduct another mass vaccination for rubella in women and girls aged between 5 to 25 years; continuation of routine vaccination of infants. Also, the anti-rubella IgG testing before pregnancy in women who were not vaccinated; vaccination of women before marriage /pregnancy should be obligatory in order to prevent the CRS.

Declaration

Ethics approval and consent to participate

This study was accredited by Ethical Committee of Iran University of Medical Sciences. Helsinki Declaration was respected across the study and the informed consent form was signed by the parents.

Competing interests

The authors declare no conflict of interest in preparing this study.

Funding

This study received no specific grant from any funding agency in the public, institutions or not-for-profit sectors.

All authors confirm Consent for publication

All authors confirm Availability of data and materials

Authors' contributions:

NS designed and supervised the study, writing the initial report.
VM visited and interpreted the patient's data;
TL were major contributors in rewriting and English editing the manuscript.
SA: methodology and statistics
AR: Analysis the results

Acknowledgment

We thank for support of the Infectious Diseases Research Center of Iran University of Medical Sciences

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