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RESEARCH ARTICLE

A STUDY TO DETECT RUBELLA SPECIFIC IgG ANTIBODIES AMONG ANTENATAL MOTHERS IN AND AROUND ARIYUR, PUDUCHERRY

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ABSTRACT

Introduction: Rubella or German measles is an acute childhood infection, usually mild and of short duration, accompanied by low grade fever, lymphadenopathy and maculopapularrash. Infection during the first trimester of pregnancy leads to still births and spontaneous abortion.

Material and method: This is a hospital based study. About 102 pregnant women irrespective of the age, immunization status, previous bad obstetric history and family history of CRS were included. About 2-3 ml of venous blood was collected after informed consent and questionnaires filled by the patient. Test was carried out using ENZYME LINKED IMMUNOSORBENT ASSAY METHOD (ELISA) using commercially available ELISA kit.

Results: A total of 102 subjects were included in the study. All the 102 subjects, serum sample were tested for the presence of Rubella specific IgG. 84 (82.3%) subjects were positive IgG i.e. they were immune to rubella infection, 11 (10.7%) subjects were not immune to Rubella infection, 7 (6.8%) tested sample were equivocal.

Conclusion: Testing for Rubella antibodies should be advised to the women in the child bearing age group who plan to conceive following marriage. If the results come out to be negative, they should be administered a dose of Rubella vaccine and advised to plan pregnancy after 3 months. Regular antenatal screening for Rubella antibodies should be advised to prevent adverse pregnancy outcomes, by the consulting Obstetrician.

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INTRODUCTION

Rubella is a disease characterized by lymphadenopathy, low grade fever and a mobiliform rash that is short lived. It is a benign, acute and mild exanthematous disease. Rubella is prevalent all over the world except where it has been eliminated and has been added in their national immunization schedule. It occurs mostly in late winter and spring exhibiting a seasonal pattern in the temperate zones and its epidemics occur every five to nine years but there extent and periodicity varies in developed and developing countries (Hobman *et al*., 2007). Rubella can cause in utero infection and post natal infection. If the infection occurs during the first trimester of pregnancy it leads to still births and spontaneous abortions. A postnatal infection is less serious and is associated with less severe complications but an in utero infection following transplacental transmission has grave consequences leading to Congenital Rubella Syndrome (CRS) (Hobman *et al*., 2007). Outbreaks occur involving school children and young adults living in groups but are uncommon in preschool children.

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Most common source of infection for the women of child bearing age (CBA) is children in their household and occupational exposure. If the susceptibility rate is high in the women of child bearing age group there is a higher risk of CRS (Bushra Al-rubaii *et al*., 2010). According to WHO around 100,000 children are born with CRS every year and maximum cases are from the developing countries (Vijaylakshmi *et al*., 2004). Immunity to Rubella in pregnancy indirectly points towards the risk of acquiring CRS. Seroepidemiological surveys of Rubella are the most important tool to know about the proportion of population that is susceptible to Rubella and the risk of acquiring CRS (Ekta *et al*., 2006). Only a few studies have been conducted in India and the estimated prevalence of Rubella is based on the worldwide data that comes out to be 100-200 per 100,000 populations (Panda and Panigrahi, 2009). Various studies conducted show that the women gain immunity either due to a natural infection with Rubella or post vaccination in childhood but by the time they reach child bearing age group they either have waning immunity or no immunity at all. It has been documented that the antibody level decreases over time in the vaccinated and seropositivity to Rubelladecreases with increasing age (Ekta *et*

al., 2006). There is a lot of variation in the prevalence of Rubella antibodies among the women of child bearing age group globally. European women have a higher prevalence of immunity to Rubella (92.2%) as compared to women of Africa (86.7%) and Asian origin (78.4%). Serosurveys conducted in India show that 45% of women in the child bearing age group are susceptible to Rubella infection and are at a considerable risk of acquiring infection during pregnancy (Gandhoke *et al*., 2005). As this condition is completely preventable by appropriate vaccination, a fierce research will help in identify subjects at risk and in turn provide a safe environment for their off-springs. Hence this study was undertaken to know the immune status of pregnant women in the given region.

MATERIALS AND METHODS

This prospective study was conducted over a period of one year from November 2012 to December 2013 at Sri Venkateshwaraa Medical College Hospital and Research Centre Ariyur, Puducherry. Institional Ethical Committee approval was sought prior to the collection of samples.Women visiting the Obstetrics and Gynaecology (OBG) Out Patient Department (OPD) for regular antenatal checkup were explained about the study and informed consent was taken. Detailed history was taken that contained information regarding patient's Name, Age, Sex, Occupation, Communication address, Socioeconomic Status, Parity, and Immunization Status. Patients with a history of rash, fever, lymphadenopathy or arthralgia in the past were given special reference. Cases with bad obstetric history (BOH) in terms of two or more consecutive spontaneous abortion, history of intrauterine fetal death, intrauterine growth retardation, still births, early neonatal death and/or congenital anomalies were also given importance. LMP and EDD of the subject were also noted.Socioeconomic status of the subjects was scored based on the Kuppuswamy's Criteria. Ssample collection and storage: Blood samples were collected from 102 pregnant women from November 2012 to December 2013 who were asymptomatic and women with BOH irrespective of their gestational age, immunization status. Complete aseptic measures were implied along with the use of personal samples reached the microbiology laboratory where they were centrifuged; serum was separated and stored at -200C until it was tested. Freezing and thawing were restricted to the minimum. Processing of samples: Rubella specific IgGwere purchased from Delta biological and Diesse Diagnostica Senese Viadelle Rose 10. Manufacturers guidelines were followed strictly while doing the procedure and during reporting. Samples were tested for Rubella specific IgG. The O.D. of each sample was read at 450 nm using a dual wavelength ELISA reader with an automatic washer attached. Statistical Analysis:Variables like age, parity, and antibody index, outcome of pregnancy, socioeconomic status, occupation, and immunization status were compared. Properties of outcome were expressed as percentages.Mean, standard deviation and T test were calculated for the data, p value of <0.05 was considered significant.

RESULTS

The subjects in the study were from in and around Ariyur which belongs to the rural area of Puducherry. In table-1 subjects were grouped into different age groups between 17 - 32 and their IgG immune status compared. Subjects were immune to Rubella infection. It was noticed maximum seropositivity was seen in the age group 21-24 years and maximum seronegativity seen in age group 17-20 years. It was also noticed that as age advanced the seronegativity decreased. The P value was 0.542 and was statistically not significant. In table 2 the percentage of IgG antibody index (> 1.3) was higher in the 21-24 age group, it was seen that antibody index decreased with age. The numbers of subjects with an antibody >50IU were also seen in this group. Table 3 Subjected with BOH were grouped into A1, A2, and A>2 depending on the number of abortions. Percentage of positivity was higher in A2 and A>2. Maximum seronegativity was seen in A1 group 21-24 year age group. Table 4 When the IgG antibody index were compared among A1, A2, and A4 the percentage of subject with an IgG antibody index of > 1.3 was seen to be higher in the A1 group when compared to the other groups. P value was not significant. Table no. 5. In this study total numbers of primis (G1) were 49 and subjects with multiparity were 53. 5 of the primi and 7 of the multiparous subjects were susceptible to Rubella. Maximum percentages of IgG positivity were seen among primigravida 87.8%. Maximum seronegativity was seen in G2. Table- 6 showed maximum IgG positivity and antibody index among G1 when compared with the others. P value was 0.235. Table 7 78/102 (76.47%) subjects in the study were from the lower socio economic status and when the subjects in the middle upper and lower socioeconomic status were also taken into account their immune status to Rubella was 100% 100% and 76.47% respectively. Table 8. In this table positivity of IgG is more in working community in comparison to house wife. Maximum numbers of seronegativity were seen in housewife group.

Table 1. Seroprevalence of *Rubella* specific IgG antibodies in different age groups

Age in years	IgG		Equivocal n (%)	χ2	p value
	Positive n (%)	Negative n (%)			
17-20 (n=20)	16(80)	4(20)	0(0)	3.683	0.1586
21-24 (n=49)	42(85.7)	5(10.2)	2(4.1)	1.222	0.5429
25-28 (n=18)	15(83.3)	2(11.1)	1(5.6)	0.059	0.9709
29-32 (n=15)	11(73.3)	0(0)	4(26.7)	12.12	0.0023*
TOTAL (N= 102)	84(82.3)	11(10.7)	7(6.9)		

Table 2. Comparison of IgG antibody index among different age groups

AGE(years)	> 1.3	1.3-0.7	< 0.7	χ	p value
17-20 (n=20)	16	0	4	3.683	0.1586
21-24 (n=49)	42	2	5	1.222	0.5429
25-28 (n=18)	15	1	2	0.059	0.9709
29-32 (n=15)	11	4	0	12.12	0.0023*
TOTAL (N=102)	84	11	7		

Table 3. Comparison of IgG Sero prevalence in BOH subjects

BOH	IgG positive	Equivocal	IgG negative	χ	p value
A 1 (n=12)	7	4	1	1.296	0.523
A 2 (n=1)	1	0	0	0.598	0.743
A 4 (n=1)	1	0	0	0.598	0.743
TOTAL (N=14)	9	4	1		

Table 4. Comparison of IgG antibody index in BOH subjects

BOH	> 1.3	1.3 - 0.7	< 0.7	χ	P value
A1 (n=12)	7	4	1	1.296	0.523
A2 (n=1)	1	0	0	0.598	0.743
A4 (n=1)	1	0	0	0.598	0.743
TOTAL (N=14)	9	4	1		

Table 5. Comparison between parity and IgG Sero prevalence

Parity	IgG positive	Equivocal	IgG negative	χ	P value
G1 (n=49)	43	1	5	2.895	0.235
G2 (n=39)	31	2	6	0.827	0.661
G3 (n=9)	6	2	1	4.779	0.092
>G4 (n=5)	4	1	0	3.104	0.212
TOTAL (N=102)	84	6	12		

Table 6. Comparison between parity and IgG antibody index

Parity	>1.3	1.3 – 0.7	<1.3	χ	P value
G1 (n=49)	43	1	5	2.895	0.235
G2 (n=39)	31	2	6	0.827	0.661
G3 (n=9)	6	2	1	4.779	0.092
G4 (n=4)	3	1	0	3.104	0.212
G5 (n=1)	1	0	0	0.216	0.897
TOTAL (102)	84	6	12		

Table 7. Comparision between IgGsero prevalence with socioeconomic status

Socioeconomic status(SE)	IgG positive	Equivocal	IgG negative	χ	P value
Upper socio economic (n=1)	1	0	0	0.216	0.897
Middle upper (n=2)	2	0	0	0.437	0.804
Middle lower (n=3)	3	0	0	0.662	0.718
Lower socio economic (n=96)	78	7	11	1.366	0.505
Total (102)	84	7	11		

Table 8. Comparison of IgGsero prevalance with occupation

Occupation	IgG positive	Equivocal	IgG negative	χ	P value
House wife (HW) (n=96)	78	7	11	1.366	0.505
Professional (n=5)	5	0	0	1.127	0.569
Medical Student (n=1)	1	0	0	0.216	0.897
Total (102)	84	7	11		

DISCUSSION

Rubella and Congenital Rubella Syndrome are not yet notifiable diseases in India. Syndrome of congenital abnormalities following maternal Rubella infection has been recognized for over 50 years from now. Despite this, the immunization rates for Rubella are not optimal and infections during pregnancy still occur. Rubella vaccine is not incorporated in the immunization schedule of many countries. Therefore, 5 to 25% of women in the childbearing age lack Rubella IgG antibodies and are susceptible to primary infection. Extrapolated data has related Rubella to the etiology of 26% cataracts, 17-12% of congenital malformations and up to 29% of sensori-neural hearing loss in infants in India (Panda and Panigrahi, 2009) One of the causes of BOH is Rubella and seroconversion of pregnant women has been considered as one of the reasons for medical termination of pregnancy. The infection is mild and the rash and lymphadenopathies are transient thus the diagnosis of Rubella is often missed. The most useful and reliable method to detect the infection is serodiagnosis (Vijaylakshmi *et al*., 2004). A single cross-sectional survey of IgG antibody seroprevalence in women of childbearing age is of limited usefulness in demonstrating disease burden. High risk of CRS is indicated by a high level (>20%) of susceptibility but a low level of susceptibility cannot be taken to imply absence of risk of CRS. CRS can occur even when the susceptibility levels in women are below 10% (Jorge Barreto *et al*., 2006) In India, the National Immunization Programme (NIP), also known as the Universal immunization programme (UIP) advises only measles vaccination at 9 months of age and Rubella vaccination is optional. Although MMR has not been included in the NIP, it has gained widespread usage in the private sector. A dose of MMR is recommended to all children at the age of 12-15 months by the Indian Academy of Paediatrics (IAP) in the schedule and it emphasizes on its inclusion in the NIP. Usage of MMR in the private sector has made it available to the elite section of the society, but it remains out of reach for the common man due to its prohibitive cost and unavailability in the govt. hospitals. Puducherry is the largest urban agglomeration in Puducherry Union Territory and the health system here is the same as anywhere in India. The rural population of Puducherry is 12, 48,000.(2011 census of India).

So far, only a few studies have been conducted in India on the epidemiology of Rubella and CRS. These studies have helped us infer that Rubella and CRS are not uncommon in our country. But the extent and magnitude of the problems yet to be determined. The fact that the condition is completely vaccine preventable makes it necessary to study and research about the disease further. The US has had its devastating bout of Rubella when 60,000 children were born deaf. The disease was controlled by widespread use of Rubella vaccine. Immunization against Rubella is rare in India which is ironic as it is the home of the Serum Institute in Pune, the producer of half of the world's Rubella vaccine requirements! Consultations between infectious disease experts in India and the US suggest that administering the vaccine to preadolescent girls could drastically reduce the incidence of Rubellaand related birth defects. The circulation of Rubella in the population can be reduced if infants are vaccinated. For this to be possible, Government must ensure that 80% of the child population is covered by the programme or else something like in the Greek epidemic would be repeated which occurred due to a sustained poor coverage of less than 50% with MMR vaccine in 1 year old children there.

Conclusion

Rubella and CRS can be eradicated only by implementing proper surveillance programmes, making the disease notifiable and vaccination mandatory. National Immunization Schedule should incorporate Rubellavaccination as per the recommendations of Indian Academy of Peadiatrics. The first dose should be administered at 12-15 months of age and a second dose should follow at the time of school entry. The goal should be, – maintaining the vaccine coverage above 80% to prevent outbreaks. Antenatal cases with a history of rash or contact with Rubella like rash should be tested for IgMRubellaantibody and if the diagnosis is confirmed, therapeutic abortion should be advised. Testing for Rubellaantibodies should be advised to the women in the child bearing age group who plan to conceive following marriage. If the results come out to be negative, they should be administered a dose of Rubellavaccine and advised to plan pregnancy after 3 months. Regular antenatal screening for Rubellaantibodies should be advised to prevent adverse pregnancy outcomes, by the consulting Obstetrician.

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