

EPIDEMIOLOGICAL EVALUATION OF RUBELLA VIRUS INFECTION AMONG PREGNANT WOMEN IN IBADAN, NIGERIA

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□ Rubella is a vaccine-preventable, mild rash-inducing viral disease with complications that include a spectrum of birth defects in the developing fetus, especially if the infection is acquired in the early months of pregnancy. Consequently, the primary objective of global rubella control programs is prevention of congenital rubella infection and associated birth defects. Despite the availability of safe and effective vaccines, and the elimination of the rubella virus in many developed countries, substantial commitment to rubella control has not been demonstrated in developing countries. This study appraises immunity to rubella, and consequently makes appropriate recommendations aimed at facilitating effective control. A cross-sectional sero-surveillance study was carried out among defined 272 consenting ante-natal clinic attendees in south-western, Nigeria. Prevalence rates of 91.54% and 1.84% were recorded for the anti-rubella virus (anti-RV) IgG and IgM, respectively. Also, 90.7% and 92.3% of the women aged ≤ 30 years and > 30 years, respectively, had detectable anti-RV IgG. No significant association ($p = 0.94$) was recorded between anti-RV IgG detection and age of the women. Previous exposure and susceptibility of significant fraction of the population to rubella infection were confirmed. Considerable political commitment and promotion of free rubella immunization specifically for women with childbearing potential were recommended.

Keywords anti-rubella, CRS, Nigeria, rubella, rubella virus, vaccine-preventable

INTRODUCTION

The rubella virus is a member of the *Rubivirus* genus in the family *Togaviridae*.^[1] It is a cubical, medium-sized (60–70 nm), lipid-enveloped virus with a positive-sense, single-stranded RNA genome. It is the only non-arthropod borne virus in the family and the aetiologic agent of rubella.

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Rubella is a vaccine-preventable, mild rash-inducing viral disease with complications^[2,3] that include a spectrum of birth defects in the developing fetus, especially if the viral infection is acquired in the early months (first trimester) of pregnancy.^[3–6] Birth defects associated with the rubella virus infection range from blindness, deafness, and congenital heart disease, to mental retardation and central nervous system (CNS) complications which are often collectively referred to as congenital rubella syndrome (CRS).^[4,7,8] Furthermore, in extreme cases, in-utero infection of a fetus with the rubella virus can cause fetal demise.^[9] Consequently, the primary objective of rubella-control programs is prevention of congenital rubella virus infection, and by association CRS.^[10]

Despite the development and administration of effective vaccines for prevention and control of the rubella virus infection since the late 1960s, and prevention as well as feasibility of, or elimination of, the causative agent in many developed countries,^[11,12] the infection is still endemic in Nigeria. In fact, it was noted in a recent study^[13] that a significant number of non-immunized women of childbearing age remain susceptible to the rubella virus infection in the country. Also, subclinical or clinical infections as well as continuous circulation of the rubella virus have previously been reported in Nigeria.^[13–18]

Efforts to realize significant political commitment and investment in rubella control and possible virus elimination in Nigeria has not yielded significant result. For example, to date the rubella vaccine is only accessible at a cost to the informed few in the population. Also, most vaccinees receive monovalent measles rather than rubella-containing vaccines (RCVs) like trivalent measles-mumps-rubella (MMR) vaccine, although the later is advertised on the platform of National Immunization Programme (NIP).

Although previous studies have documented rubella infection among selected populations in different regions of the country,^[13–18] this study was designed to specifically appraise the rubella infection among pregnant women with rubella-like clinical presentations in the population. Findings from the study will add to existing information about rubella in the region and further strengthen the drive for effective prevention and elimination of rubella in Nigeria. Therefore, to achieve the aforementioned, this study was designed and conducted to evaluate anti-RV IgM and IgG among antenatal clinic attendees with rubella-like clinical presentations in the selected facilities.

MATERIALS AND METHODS

Study Location

This study was carried out among pregnant women attending antenatal clinics in University College Hospital and Ade-Oyo State Hospital

in Ibadan, south-western, Nigeria. University College Hospital is a tertiary health care facility of the University of Ibadan. The hospital is equipped with facilities for teaching of medical students, research, and provision of clinical services to the community. Attendees in the hospital are majorly residents of average economic and educational status. On the other hand, Ade-Oyo State Hospital is a secondary health care facility located in the aboriginal nerve of the city, densely populated by indigenes and serving wide range of people with spectra of social, economic, and educational background. The hospital serves pregnant women of varied economic and educational status from different parts of the city. The ante-natal clinic records an average of 380 ± 20 new pregnant women per week. Pregnant women from the University College Hospital and Ade-Oyo State Hospitals were subsequently referred to as RUC (rubella study pregnant women in UCH) and RAD (rubella study pregnant women in Ade-Oyo) study groups, respectively. The two hospitals selected for the study attend to pregnant women of varied educational and socio-economic backgrounds.

Enrollment of Pregnant Women

To achieve our aim and objectives, consenting ante-natal clinic attendees were enrolled from the two selected hospitals described above. Pregnant women were enrolled between September 2012 and June, 2013. During each visit a short presentation was made to educate the ante-natal clinic attendees on rubella. Afterwards, consenting pregnant women with any of observable rubella-like rash, fever, lymphadenopathy, or arthralgia were enrolled for the study. Pregnant women without any of the clinical presentations were excluded from the study. Demographic and other relevant information were obtained using a structured questionnaire.

Research Methodology

Blood sample was collected from a total of 272 (median age = 31 years, age range = 17–43 years [RUC: n = 182; age range = 17–43 years; RAD: n = 90; age range = 19–42 years]) pregnant women who consented verbally, and were subsequently enrolled based on presence of any rubella-like clinical presentation at the point of registration and routine examination for ante-natal clinic. Ethical approvals for the study were granted by the UI/UCH Ethics Committee (UI/EC/11/0058) and Oyo State Ministry of Health (AD3/479/349).

Sample Collection

About 5 mL of blood sample was collected via venepuncture of each pregnant woman into an appropriately labeled sterile container free of anti-coagulants or preservatives. Thereafter, samples were transported to the laboratory immediately in a cold box with frozen ice packs to maintain a condition of about 4–8°C. Serum samples were separated by low-speed centrifugation at 500 g for 5 min, or direct removal of the serum using a sterile disposable pipette after retraction of the clot. Then, two aliquots of serum were prepared and transferred into labeled sterile cryovials and stored at –20°C until ready for analysis, while the coagulated cells were stored at –20°C in the sterile container.

Laboratory Analysis

Laboratory analysis was carried out in the Department of Virology, College of Medicine, University College Hospital, Ibadan. The samples were analyzed for qualitative and quantitative detection of anti-rubella IgM and stable memory IgG using DIA.PRO[®] Diagnostic Bioprobes srl (Sede legale: Via Lucio Giunio Columella, 31-20128-Milano, Italy) Enzyme Immunoassay in accordance with the manufacturer's description. Results of the anti-IgG assay was interpreted with antibody titer ≥ 15 IU/mL as the cut-off point. Both test kits used have diagnostic sensitivity and specificity performance of >98%.

Statistical Analysis

Results of the study were analyzed with t-test and χ^2 statistical tests using Statistical Package for the Social Sciences (SPSS) version 15.0 for Windows. P-value ≤ 0.05 was used as indicator of statistical significance. Also, demographic features and other relevant information about the study populations were compared (Table 1).

RESULTS

Overall, prevalence rate of 91.54% (249/272) and 1.84% (5/272) were recorded for anti-rubella virus (anti-RV) IgG and IgM, respectively. Further analysis of the results showed that 83 (92.2%) and 1 (1.1%) of the women in RAD had anti-RV IgG and IgM, respectively, (Table 2). Also, 166 (91.2%) and 4 (2.2%) of the women in RUC had anti-RV IgG and IgM, respectively (Table 2). One of the 5 pregnant women with detectable anti-RV IgM was in her first trimester. Also, 3 and 4 of the women with detectable anti-RV IgM presented with fever and arthralgia respectively (Table 3). Overall,

TABLE 1 Profile of the RAD and RUC ante-natal clinic attendees enrolled for the rubella epidemiology study in Ibadan, Nigeria

Parameters	RAD (%)	RUC (%)	Remarks
Mean Age (Year)	30.07	31.57	Significantly different ($p = 0.02$)
Age at first marriage (Year)	25.1	27.67	Significantly different ($p = 0.0005$)
Mean parity	1.36	0.97	Significantly different ($p = 0.03$)
Vaccination record	23 (25.6)	56 (30.8)	No association between location and vaccination record ($p = 0.37$)
Education			Significant association between location and educational status ($p = 0.0005$), that is a woman with tertiary education was likely to be in RUC.
Primary	8 (8.9)	1 (0.6)	
Secondary	39 (43.3)	19 (10.5)	
Tertiary	43 (47.82)	162 (89.0)	
Fever	44.4%	51.1%	Fever not associated with location ($p = 0.3$)
Lymphadenopathy	9 (10.0)	27 (14.8)	Lymphadenopathy not associated with location ($p = 0.27$)
Arthralgia	63 (70.0)	106 (58.2)	Arthralgia not associated with location ($p = 0.06$)
Ever had rash	18 (20)	46 (25.3)	Ever had rash not associated with location ($p = 0.34$)
Rash (2 weeks before enrolment)	15 (16.7)	40 (22.0)	Rash not associated with location ($p = 0.31$)

RAD: Rubella study subjects in Ade-Oyo Maternity Hospital; RUC: Rubella study subjects in University College Hospital.

90.7% (117/129) of women aged ≤ 30 years and 92.3% (132/143) of those aged > 30 years, respectively, had detectable anti-RV IgG. Further analysis of the results for RAD showed that 46 (90.2%) of the women aged ≤ 30 years and 37 (94.4%) of those aged > 30 years, respectively, had detectable anti-RV IgG, these rates were statistically comparable (Table 2). Also, results for RUC showed that 71 (91.0%) of the women aged ≤ 30 years and 95 (91.3%) of those aged > 30 years had detectable anti-RV IgG (Table 2). No significant association ($p = 0.94$) was recorded between the presence of anti-RV IgG and age of pregnant women (Table 2). Significant difference ($p = 0.0005$) was recorded in educational status of the women by location (woman with tertiary education were more likely to be in RUC); however, similar anti-RV IgG prevalence rates were observed in both locations. Pregnant women enrolled for the study had comparable presentations of fever, lymphadenopathy, arthralgia, and rash (Table 1). Also, Chi square analysis showed no association ($p = 0.78$) between location and previous exposure to RV (presence of anti-RV IgG) (Table 2).

TABLE 2 Relationship between the anti-rubella virus antibody prevalence rates and variables of RAD and RUC ante-natal clinic attendees in Ibadan, Nigeria

Variables	No. tested among RAD (% positive)	p value	No. tested among RUC (% positive)	p value
Anti-RV IgG by age				
≤ 30 years	51 (90.2)	Not valid	78 (91.0)	0.94
> 30 years	39 (94.4)		104 (91.3)	
Overall anti-RV IgG	90 (92.2)		182 (91.2)	0.78
by location				
Overall anti-RV IgM	90 (1.1)		182 (2.2)	Not valid
by location				

TABLE 3 Profile of the five ante-natal clinic attendees with detectable anti-RV IgM

Sample ID	Age (Year)	Gestational Age (Months)	Rash	Fever	Arthralgia	Lymphadenopathy
RAD/13/023	27	6	No	No	Yes	No
RUC/12/031	27	9	Yes	No	No	No
RUC/13/039	27	2	No	Yes	Yes	No
RUC/13/068	36	3.5	No	Yes	Yes	No
RUC/13/076	24	6	No	Yes	Yes	Yes
Total			1	3	4	1

RAD: Rubella study subjects in Ade-Oyo Maternity Hospital; RUC: Rubella study subjects in University College Hospital; Yes: Presence of symptom; No: Absence of symptom

DISCUSSION

A high prevalence rate of anti-RV IgG was observed in the study. This suggests previous exposure to the rubella virus by a significant number of the women who were rubella vaccine inexperienced. It also implies previous subclinical or clinical infections with the rubella virus. Anti-RV IgM was not detectable in most of the women despite the enrolment technique (based on presentations of clinical symptoms suggestive of rubella). Thus, it implies that the clinical presentations though suggestive of rubella infection might have resulted from other infections. However, detection of anti-RV IgM in a fraction of the study population confirms recent infection and continuous circulation of the virus. More importantly, detection of anti-RV IgM in a pregnant woman in her first trimester implies higher risk and possibility of occurrence of unreported cases of CRS in the population. Furthermore, the presence of serologically naive pregnant women (8.46%) in the population demonstrates susceptibility of a significant fraction of the population to the rubella virus infection.

In previous studies, [14–18] varied anti-RV IgG prevalence rates have been reported among women of childbearing age and pregnant women in different regions of Nigeria. It is, however, pertinent to note that findings from

this study corroborate previous reports of subclinical or clinical infection as well as continuous circulation of the rubella virus in Nigeria.^[13–18] It is also in congruence with preliminary report^[13] of high prevalence rate (89.4%) of anti-RV IgG among vaccine naïve pregnant women attending ante-natal clinic in one (Ade-Oyo State Hospital) of the study locations. Previously, anti-RV IgG prevalence rate of 54.1% was reported among rubella vaccine naïve pregnant women in a study conducted in northeastern Nigeria.^[16]

Specifically, detection of anti-RV IgM (Tables 2 and 3) and record of rubella antibody naivety among a fraction of the study population depict CRS situation in the country. Cutts and Vynnycky,^[19] in a review of the literature on the prevalence of anti-rubella antibodies from developing countries, concluded that CRS is an under-recognized public health problem and that appropriate data need to be collected to estimate the cost-effectiveness of a potential global rubella control program. Furthermore, it had been shown that determination of the incidence of rubella and CRS remain important steps to achieve effective prevention and control program.^[11]

Comparable prevalence rates of anti-RV IgG were recorded despite varied age, age at first marriage, and mean parity (Table 1) of the studied pregnant women. This observation might imply that women in the community possibly become exposed and infected with the rubella virus early in life, before reaching childbearing age. However, there is the need for more extensive study on specific variables to facilitate appropriate conclusion. Similar anti-RV IgG prevalence rates were recorded among the women irrespective of their educational status or location of residence (Table 1). Also, comparable rates of presentations of probable symptoms of rubella infection including fever, lymphadenopathy, arthralgia, and rash were observed among the studied population. These observations might suggest comparable risks of infection irrespective of persons' educational and economic status in the region. Also, it might indicate continuous and consistent circulation of the rubella virus in the population. It is noteworthy that higher rates of arthralgia and fever were documented among the women with detectable anti-RV IgM (Table 3). However, extrapolation of the outcome may be limited by the small size of the group.

The rubella vaccine is not included in the childhood immunization program neither is there provision for selective immunization of women of childbearing age in Nigeria. However, it is only available to informed few at a cost, thus high prevalence rates of anti-RV IgG detection in the population confirms previous exposure and infection by the virus. It is pertinent to note that the World Health Organization (WHO) recommended the use of rubella-containing vaccine (RCV) in all countries with national childhood immunization schedules to prevent congenital rubella infection, including CRS in 2000.^[20] The number of WHO member states using RCV increased from 83 (43%) in 1996 to 130 (67%) in 2009. Consequently, the number

of rubella cases reported dramatically decreased from 670,894 in 2000 to 121,344 in 2009.^[21] However, despite the WHO recommendation and subsequent accomplishments in different parts of the world, rubella vaccine is still available to Nigerians at a cost.

It has been recognized,^[22] with confirmations,^[23–25] that high childhood immunization rates are essential to achieving effective prevention of CRS.^[22] Accordingly, WHO advises a minimum target rate of 80% for childhood immunization programs.^[20] However, considering the practicability of achieving 80% success rates in childhood immunization in Nigeria, vis-a-vis documented success in prevention of CRS with selective immunization of all women of childbearing age^[2] we recommend selective vaccination of women with childbearing potential in the country.

CONCLUSIONS

Specifically, the study shows serologic evidence of previous exposure and/or recent infection by the rubella virus among the studied women. It also shows that a proportion of the pregnant women were newly infected, even as some of them remained susceptible to the virus. Findings from the study corroborate reports of previous studies in the country and further approve that elimination of the rubella virus in Nigeria is feasible since the definite susceptible population is defined. Therefore, to facilitate effective rubella control in Nigeria we recommend substantial political commitment and institution of health policy that promotes awareness and free rubella virus immunization program especially for women of childbearing age.

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