

BMJ Open Rubella virus infection and associated factors among pregnant women attending the antenatal care clinics of public hospitals in Hawassa City, Southern Ethiopia: a cross-sectional study

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Objective To assess the seroprevalence of recent/acute and past exposure to rubella virus infection and associated risk factors among pregnant women.

Design A hospital-based cross-sectional study.

Setting The study was conducted in two public hospitals in Hawassa City, Southern Ethiopia.

Participants A total of 422 pregnant women attending antenatal care clinics were selected using a systematic random sampling technique from March to June 2016.

Outcome measures Data on sociodemography and related factors were collected using a structured questionnaire. Blood samples were also collected from each study participant and tested for antirubella IgM and IgG antibodies using ELISA. IgG seropositivity indicates past exposure to rubella (protective immunity). IgM seropositivity indicates recent exposure to rubella (or reinfection).

Results The seroprevalence of antirubella IgM and IgG antibodies was 2.1% and 86.3%, respectively. Thus, the rate of susceptibility to rubella virus infection among pregnant women was found to be 13.7%. A significant association between residence site and IgG seropositivity was observed, where urban dwellers had higher past rubella exposure compared with rural residents (crude OR 6.3; 95% CI 3.29 to 12.14, $p < 0.001$).

Conclusion The high rate of rubella exposure and its similar distribution by sociodemography (except residence site) suggests the continuous transmission and endemicity of the infection in the study area. These findings emphasise the importance of introducing rubella-containing vaccine into routine childhood immunisation programme and vaccinating susceptible women of childbearing age.

INTRODUCTION

Rubella is an acute, usually mild, contagious disease caused by rubella virus. Transmission of postnatal rubella is mainly through the respiratory route and commonly occurs in children and young adults. The infection may remain subclinical or cause self-limiting

Strengths and limitations of this study

- The study showed the magnitude of recent and past exposure to rubella infection in a randomly selected high-risk population in contrast to case-based surveillance, which informs only about recent infection in people with clinical presentation of measles.
- As with any institution-based study, results may not be generalised to all pregnant women in the study area.
- The ability of the study participants to recall past events and/or their willingness to provide genuine information may have introduced recall and/or information bias.

illness with clinical features such as low-grade fever, lymphadenopathy and skin rash.¹ Rubella can also be transmitted from infected pregnant women to their unborn babies. Congenital rubella infection (CRI) has outcomes including miscarriage, stillbirth, abortion, congenital rubella syndrome (CRS) or asymptomatic infection in the infant. Manifestations of CRS encompasses cardiac, cerebral, ophthalmic and auditory defects.² The risk of congenital defects varies from 10% to 90% depending on the gestational age of the fetus at the time of infection. The occurrence of rubella earlier in gestation, particularly during the first 12 weeks, increases the risk of more severe outcomes. Congenital anomalies are rare if infection occurs after 20 weeks of gestation even though fetal infection may occur throughout pregnancy.³

Rubella is a vaccine-preventable infection, and considered to be potentially eradicable. As a result of the vaccination programme in many high-income and in some low-income

and middle-income countries, the estimated number of CRS cases globally decreased from about 1 190 000 cases in 1996 to about 1 050 000 cases in 2010.⁴ The large-scale vaccination programme in Americas and Europe has achieved a drastic reduction or elimination of both the virus and CRS. In contrast, the highest risk of CRS is found in countries where the rubella-containing vaccine (RCV) has not been introduced to the national immunisation programme or the vaccine coverage is low.¹ Africa and South East Asia regions, with the respective estimated incidence of 116 and 211 per 100 000 live births in 2010, have the highest rates of CRS. In Ethiopia, estimates of the rate of CRS range from 24 to 112 per 100 000 live births in urban Addis Ababa and rural Ethiopia, respectively.⁴

With the goal of extending the full benefits of immunisation to all persons, the Global Vaccine Action Plan (GVAP) 2011–2020 was outlined and endorsed by the World Health Assembly in 2012.⁵ Towards achieving GAVP goals, rubella vaccine had been introduced in 149 (77%) of 194 WHO member countries as of September 2016.⁶ However, Ethiopia has planned to introduce measles-rubella vaccine into the routine expanded programme for immunisation schedule for children under 1 year of age in 2019.¹

Most African countries have established measles case-based surveillance, in which suspected cases are screened with laboratory tests. Rubella is also integrated into the measles case-based surveillance where specimens found to be negative or indeterminate for antimeasles IgM are tested for antirubella IgM.⁷ Analysis of such surveillance results for the period 2002–2009 in Africa showed a 5% seropositivity rate of antirubella IgM among women of reproductive age, indicating the risk of having an infant with CRS.⁸ Published surveillance reports from Ethiopia indicate the rates of antirubella IgM cases were 12.1% in the period 2004–2009⁹ and 15.3% in the period 2009–2015.¹⁰ These findings demonstrate the endemicity of rubella virus infection in the country.

Measles case-based surveillance has limitations in depicting the epidemiology of rubella virus infection. The clinical features of rubella may not be the same or may not satisfy the case definition of measles. Moreover, surveillance data, which is generated through investigations of outbreak situations or suspected persons might not represent a defined population. In Ethiopia, serosurvey is not conducted to determine the current status of rubella virus infection or rate of susceptibility either in the general population or different subpopulations. Information is particularly scarce about recent and past exposure to rubella virus infection among women of childbearing age and pregnant women despite the associated risk of CRI in these subpopulations. No system has been in place to track the magnitude of CRS in the country. Therefore, this study aimed to assess the seroprevalence and associated risk factors of rubella virus infection among pregnant women attending the antenatal care (ANC) clinics in Southern Ethiopia where outbreaks of measles and rubella are common. The findings may inform decision

makers and other concerned bodies regarding the rate of susceptibility to rubella virus infection among pregnant women and the magnitude of recent infection so that the best intervention approach can be defined. The findings of this investigation may also help estimate the incidence of CRS and provide an indirect measure CRS burden.

METHODS

Study area and period

An institution-based cross-sectional study was conducted from March to June, 2016 in Hawassa City, Southern Ethiopia. The city is established on the shore of Lake Hawassa, and is the capital of the Southern Nation and Nationalities People's Region. ANC clinics in two public hospitals, Adare Hospital and Hawassa University Comprehensive Specialized Hospital, were the study sites. The latter hospital is the largest in the administrative region, and ANC clinics in both hospitals provide clinical and diagnostic services for large number of pregnant women. ANC attendees are routinely screened for HIV and syphilis; however, they are not screened for rubella virus infection.

Population

All pregnant women who attended the ANC clinics of the two hospitals during the study period were the source population. Pregnant women 15–49 years of age who consented to participate AND had no major sickness that would prevent them from being interviewed were eligible. Major sickness was defined based on clinical assessment or the presence of any general signs of critical illness such as impending airway obstruction, active or recent history of seizures or unconsciousness.

Sample size and sampling technique

The sample size was determined using a single population proportion formula. As seroprevalence of antirubella antibody among pregnant women in Ethiopia is unknown, we assumed a 50% seropositivity rate, a 5% margin of error and 95% level of confidence. Further considering a 10% non-response rate, the sample size was calculated to be 422. Systematic random sampling method was used to recruit pregnant women attending the ANC clinics. Based on the hospitals' plan and performance within 3 months prior to the time of data collection, an estimated 845 pregnant women were expected to visit ANC of the two hospitals during the study period. This estimate was divided by the sample size to determine the sample interval (*k*-value), which would be 2. The first served pregnant woman and every second woman thereafter were invited to participate in the study until the required sample size was obtained. The desired number of study participants for each hospital was determined considering their respective ANC coverage using proportionate sampling. The target recruitment at Adare Hospital and Hawassa University Comprehensive Specialized Hospital was calculated to be 286 and 136, respectively.

Data collection

Sociodemographic and reproductive characteristics

Trained nurses collected data on sociodemography (age, occupation, educational level, residence and marital status) and reproductive characteristics (gravidity, parity, gestational age and histories of stillbirth, spontaneous abortion and fetal deaths) using structured questionnaires.

Serological analysis

About 5 mL of venous blood sample was collected from each study participant and separated sera were stored at 2°C–8°C for 5 days. Samples were transported to Hawassa Regional Laboratory using a cold box and stored at –70°C until a sufficient number was obtained for batch testing. All sera were tested for antirubella IgM and IgG using ELISA kits. The Enzygnost Anti-Rubella Virus/IgM test (Siemens Healthcare Diagnostics, Marburg, Germany) and Rubella IgG EIA test kit (DIALAB Diagnostics, Austria) were used and performed according to the instruction of the respective manufacturers.

Definitions

Stillbirth: delivery of fetus showing no signs of life after 28 completed weeks of gestation.

Fetal death: in utero fetal death after 28 completed weeks of gestation.

Spontaneous abortion: a clinically recognised spontaneous pregnancy loss before the 28th week of gestation.

Past exposure to rubella virus infection: pregnant women whose blood is tested positive for IgG antibody; thus, a protective immunity against the infection.

Recent rubella virus infection: pregnant women who tested positive for IgM antibody.

Data quality assurance

The questionnaire was pretested with 5% of study subjects in Hawassa Millinium Health Center before the actual data collection to assess its validity and completeness. Training was given for data collectors and the principal investigator closely monitored the process of data collection. Questionnaire data obtained from each study participant was reviewed immediately to check accuracy and completeness. Samples were processed and tested by an experienced laboratory professional and according to the specifications of the manufacturers. The respective sensitivity and specificity was 98% and 97.3% for Siemens Enzygnost Anti-Rubella Virus/IgM test, and 96.4% and >99.9% for DIALAB Rubella IgG EIA test kit. Controls and calibrators were run each time samples were analysed in order to make sure technical procedures were carried out correctly and test kits were working properly.

Data analysis

Data entry and analysis was performed using SPSS V.20 software. Descriptive statistics including proportion, mean and absolute figure were calculated and presented using tables. Binary logistic regression analysis was performed to assess the association between seroprevalence of

rubella with various sociodemographic and reproductive characteristics. OR with its 95% CI was used to measure the strength of the association. A p value <0.05 was considered to be statistically significant. Multivariable analysis was planned but was not done since only one independent variable showed significant association (p<0.05) with the outcome variable in bivariate analysis.

Ethical clearance

Ethical approval was obtained from the Institutional Review Board of the College of Medicine and Health Sciences, Hawassa University. Permission to conduct the study was also obtained from the management of Adare Hospital and Hawassa University Comprehensive Specialized Hospital. Study participants were given adequate information regarding the purpose, risk, benefit and confidentiality of the study. Participation was fully voluntary and informed written consent was obtained from each participant. Code numbers were used in place of identifiers to maintain the confidentiality of participant's information. The study incurred no cost to the study participants and testing for rubella was performed free of charge. Laboratory test results were given to the ANC clinics for possible follow-up and management.

RESULTS

Sociodemographic characteristics

A total of 422 pregnant women who attended the ANC of Adare Hospital and Hawassa University Comprehensive Specialized Hospital were approached, and all women met the inclusion criteria and completed the survey, making the response rate 100%. The mean age of participants was 25.1 years (range 17–42 years; SD 4.4). Women in the age group of 20–24 years and 25–29 years accounted for 36.7% and 37.9%, respectively. The majority of respondents were urban residents (87.9%) and married (98.1%). The proportion of pregnant women who were housewives or had completed a secondary school level education were 41.5% and 56.4%, respectively (table 1).

Seroprevalence of rubella

Serological analysis found that 2.1% of the study participants were positive for antirubella IgM, indicating a recent rubella virus infection (or reinfection). The seropositivity rate of antirubella IgG was 86.3%, which indicates past exposure/infection with development of protective immunity. Thus, the rate of susceptibility to rubella among the pregnant women was calculated to be 13.7%. Overall, 1.9% of participants were found to be positive for both IgM and IgG. The positivity rates for antirubella IgM were 5.4% in the age range 15–19 years, 2.5% among those who had a secondary level education, 2.4% among merchants and 2.2% among urban residents or married. The rate of antirubella IgG peaked in the age range 20–24 years (89%), and decreased thereafter with increasing age. The seropositivity of IgG was higher among urban residents (90%), merchants (90.5%), women with a primary level

Table 1 Distribution of antirubella IgM and IgG by sociodemography of pregnant women attending antenatal care clinics of public hospitals in Hawassa City, Southern Ethiopia, 2016

Characteristics	Antirubella IgM			Antirubella IgG			
	No. (%) tested	No. (%) pos.	COR (95% CI)	p Value	No. (%) pos.	COR (95% CI)	p Value
Age (in years)							
15–19	37 (8.8)	2 (5.4)	9.0 (0.8 to 103.0)	0.075	31 (83.8)	1.4 (0.3 to 6.6)	0.664
20–24	155 (36.7)	4 (2.6)	4.2 (0.5 to 38.1)	0.201	138 (89.0)	2.2 (0.6 to 8.7)	0.256
25–29	160 (37.9)	1 (0.6)	1		138 (86.3)	1.7 (0.4 to 6.6)	0.437
30–34	56 (13.3)	2 (3.6)	5.9 (0.5 to 66.2)	0.151	46 (82.0)	1.3 (0.3 to 5.3)	0.759
≥35	14 (3.3)	0			11 (78.6)	1	
Residence							
Urban	371 (87.9)	8 (2.2)	1.1 (0.1 to 9.0)	0.928	334 (90.0)	6.3 (3.2 to 12.1)	<0.001*
Rural	51 (12.1)	1 (2.0)	1		30 (58.8)	1	
Marital status							
Married	414 (98.1)	9 (2.2)	-	-	358 (86.5)	3.2 (0.3 to 35.8)	0.346
Single	5 (1.2)	0 (0)	-	-	4 (80.0)	2.0 (0.1 to 51.6)	0.676
Widowed and divorced	3 (0.7)	0 (0)	-	-	2 (66.7)	1	
Educational status							
No formal education	42 (10.0)	1 (2.4)	1.7 (0.2 to 19.3)	0.666	35 (83.3)	1	
Primary education	142 (33.6)	2 (1.4)	1		125 (88.0)	1.5 (0.6 to 3.8)	0.429
Secondary and above	238 (56.4)	6 (2.5)	1.8 (0.4 to 9.1)	0.471	204 (85.7)	1.2 (0.5 to 2.9)	0.688
Occupation							
Student	45 (10.7)	1 (2.2)	1		39 (86.7)	1	
Employed	135 (32.0)	3 (2.2)	1 (0.1 to 9.9)	0.961	116 (85.9)	0.9 (0.4 to 2.5)	0.901
Merchant	42 (10.0)	1 (2.4)	1.1 (0.1 to 17.7)	0.980	38 (90.5)	1.5 (0.4 to 5.6)	0.579
Housewife	175 (41.5)	4 (2.3)	1.02 (0.1 to 9.4)	0.998	149 (85.1)	0.8 (0.3 to 2.3)	0.796
Others	25 (5.9)	0 (0)	-		22 (88)	1.1 (0.3 to 4.9)	0.873

*Statistically significant association.

COR, crude OR; No., number; Pos., positive.

education (88%) and women who were married (86.5%). However, the rate of past exposure to rubella was found to be significantly influenced only by residence site where urban dwellers had more exposure than rural dwellers (crude OR 6.3; 95% CI 3.29 to 12.14, $p < 0.001$) (table 1).

Reproductive characteristics

More than half of the pregnant women were in their second trimester (51.2%), were multigravida (61.4%) and were multiparous (54%). Histories of stillbirth, spontaneous abortion, and fetal death were reported by 8.8%, 17.5% and 5.7% of participants, respectively. The seropositivity rate of antirubella IgM was higher among women who were in their first trimester (4.2%), primigravida (3.7%) and nulliparous (3.1%). A higher rate of antirubella IgG was observed among primigravida women (89.6%), nulliparous (89.2%) and those in their third trimester (88.2%). In bivariate analysis, none of these factors was found to be significantly associated with either IgM or IgG seropositivity status (table 2).

DISCUSSION

Despite the public health significance of rubella in Ethiopia, studies are scarce regarding the epidemiology of the infection in various subpopulation. The lack of information regarding the burden of infection, particularly among women of childbearing age, limits intervention efforts that might reduce the consequences of vertical transmission. This study aimed to generate data on recent and past exposure to rubella virus infection among pregnant women. IgM positivity indicates more recent infection (or reinfection), while IgG positivity indicates past exposure/infection with development of protective immunity. The seroprevalence of antirubella IgM and IgG was 2.1% and 86.3%, respectively.

The rate of IgG positivity in this study was comparable with results in pregnant women in Mwanza, Tanzania (92.6%),¹¹ in Osogbo, Nigeria (87.5%)¹² and in Cameroon (88.6%).¹³ The rate in Zaria, Nigeria (97.9%)¹⁴ was relatively higher than our finding despite the absence of a vaccination programme in both contexts. Contrasting lower rates were also reported in Western Sudan (65.3%)¹⁵ and in Algeria (68.6%).¹⁶ The difference in rate of past exposure to rubella between countries may indicate a varying epidemiology of the infection in different settings. However, the observed rates of past exposure in the current study or other reports in different African countries were generally high and indicated a sustained circulation of the infection in the region. The case-based surveillance data in Africa suggest that most children are exposed to the virus by the age of 15 years and developed immunity as a result of natural infection.⁸ However, the current study showed that 13.7% of the pregnant women were negative for antirubella IgG and did not have immunity against the infection. Thus, the observed intermediate level of susceptibility (10%–20%)

is likely to indicate a medium risk of CRS in the study area.¹⁷

The seropositivity of antirubella IgM in this study was similar to the rate among pregnant women in Turkey (2%), where the rubella vaccination programme reduced the susceptibility rate to 3.6%.¹⁸ On the other hand, a study from Bangladesh showed a comparable susceptibility rate (15.7%) and IgM seropositivity rate (0.75%)¹⁹ to that reported in this paper. It has been suggested that factors including population density, immunisation status and level of herd immunity at the time of virus introduction influences rate variability in different localities.²⁰ A 5% rate of antirubella IgM among women of reproductive age with clinical presentations of measles in Africa during 2002–2009⁸ was higher than the current data. This may highlight the significance of conducting serosurveys to avoid the risk of overestimating the burden of CRS based on results of case-based surveillances. It was observed that eight cases were IgM positive and IgG positive and only one case was IgM positive and IgG negative. This may be due to the fact that IgM is produced earlier in the course of infection during which antirubella IgG has not yet been produced. Moreover, reinfection with rubella virus or the presence of rheumatoid factors and other viral infections may lead to a positive IgM test result in the absence of IgG.²¹ Overall, the observed rate of IgM positivity among the participants in this study is of concern as they were randomly selected pregnant women with no apparent clinical presentations.

Regarding the distribution of rubella exposure by residence, pregnant women in urban areas had 6.3-fold greater odds of protective immunity to rubella compared with those who were rural dwellers. This finding is supported by reports from Algeria,²² Burkina Faso²³ and Iraq.²⁴ However, a contrasting result was shown in India,²⁵ which reported a higher prevalence among pregnant women dwelling in rural areas. The higher rate of rubella exposure in urban pregnant women in the current study may be due to the fact that urban populations are more likely to live in crowded situations and become susceptible to a rapid spread of the infection. The rate of rubella exposure was not found to be significantly influenced by age as was also shown in a report in Kenya.²⁶ Nonetheless, a contrasting result was reported in a study conducted in Burkina Faso,²³ where pregnant women in the age range 40–42 years had a higher exposure rate.

In the current study, the rates of recent and past exposure to rubella were not influenced by trimester as previously reported in Nigeria.^{27,28} However, the occurrence of rubella during the first trimester of pregnancy is generally a concern since the risk of vertical transmission to fetus is much higher (up to 90%).²⁹ Although no treatment is available for mothers with active rubella, a routine prenatal screening would help monitor those with the infection for adverse pregnancy outcomes including CRS.

The lack of association between rubella exposure and most of the sociodemographic characteristics or other factors such as parity and gravidity in this study has also

Table 2 Distribution of rubella virus infection among pregnant women attending antenatal care clinics in public hospitals in Hawassa City, Southern Ethiopia, 2016

Variables	Antirubella IgM			Antirubella IgG			
	No. (%) tested	No. (%) pos.	COR (95% CI)	p Value	No. (%) pos.	COR (95% CI)	p Value
Trimester							
First	96 (22.7)	4 (4.2)	2.3 (0.4 to 13.1)	0.331	79 (82.3)	1	
Second	216 (51.2)	3 (1.4)	0.7 (0.1 to 4.6)	0.766	188 (87.0)	1.4 (0.8 to 2.8)	0.273
Third	110 (26.1)	2 (1.8)	1		97 (88.2)	1.6 (0.7 to 3.5)	0.235
Gravidity							
Primigravida	163 (38.6)	6 (3.7)	0.3 (0.1 to 1.2)	0.098	146 (89.6)	1	
Multigravida	259 (61.4)	3 (1.2)	1		218 (84.2)	0.6 (0.3 to 1.1)	0.119
Parity							
Null parity	194 (46)	6 (3.1)	2.3 (0.6 to 9.7)	0.222	173 (89.2)	1	
Multiparity	228 (54)	3 (1.3)	1		191 (83.8)	0.62 (0.4 to 1.1)	0.110
Stillbirths							
Yes	37 (8.8)	2 (5.4)	3.1 (0.6 to 15.4)	0.170	34 (91.9)	1.8 (0.6 to 6.4)	0.305
No	385 (91.2)	7 (1.8)	1		330 (85.7)	1	
Spontaneous abortion							
Yes	74 (17.5)	1 (1.4)	0.6 (0.1 to 4.7)	0.613	60 (81.1)	0.62 (0.3 to 1.2)	0.157
No	348 (82.5)	8 (2.3)	1		304 (87.4)	1	
Fetal deaths							
Yes	24 (5.7)	1 (4.2)	0.5 (0.1 to 3.9)	0.488	21 (87.5)	1.12 (0.3 to 3.8)	0.855
No	398 (94.3)	8 (2.0)	1		343 (86.2)	1	

COR, crude OR; No., number; Pos, positive.

been reported in various studies in Africa.^{30–32} This may be due to the fact that most people in endemic settings are exposed to rubella virus infection at an early age.

Introducing RCV into countries' routine childhood immunisation schedule has been suggested as a cost-effective strategy to prevent CRI including CRS.³³ However, this strategy should be limited to countries that have achieved >80% coverage with the first-dose measles containing vaccine. Otherwise, suboptimal vaccine coverage could result in a paradoxical increase in susceptibility rate among older age groups, which in turn shifts the average age of rubella infection for females from childhood to the childbearing years.⁸ Therefore, in countries with lower vaccination coverage, mass immunisation of everyone aged under 40 years with measles-rubella vaccine is recommended to protect women of childbearing age from giving birth to babies with CRS.¹ Ethiopia has to exert every effort to achieve and maintain high (>80%) measles vaccine coverage to succeed in effective introduction of measles-rubella vaccine in 2019.

This study has some limitations which needs to be taken into consideration. Similar to any study conducted in a health setting, results may not be generalised to all pregnant women in the study area. Also, this study might not be adequately powered to identify risk factors for rubella seropositivity. Data on factors such as histories of abortion, stillbirth and fetal death depends on the recall ability of the study participants and/or their commitment to provide genuine information; thus, a recall and/or information bias might be introduced.

In conclusion, the high rate of exposure to rubella and similar distribution by reproductive and sociodemographic characteristics (except residence site) suggests the continued circulation and endemicity of the infection in the study area. The observed rate of recent infection among randomly selected pregnant women, particularly in those in their first trimester is of concern and may hint at the significance of CRS in our context. The susceptibility rate to rubella in this study calls for interventions that reduce the incidence of the infection in women of childbearing age and the risk of giving birth to babies with CRS. In this regard, introducing RCV in routine childhood immunisation programme and vaccinating susceptible women of childbearing age should be considered.

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Patient consent Obtained.

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