Screening for rubella immunity in an antenatal population: a multivariate analysis.

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ABSTRACT

Rubella causes disease in the fetus. Immunity to rubella is therefore routinely screened in pregnant women. In this retrospective observational study, we assessed both the effectiveness of screening for rubella and the levels of immunity to rubella in the population of a north London antenatal clinic. Risk factors for non-immunity to rubella and changes in levels of immunity over time were studied. Almost all women were screened for rubella immunity (99.8%). The majority were immune (92.8%). Women booking in earlier years within the study period showed higher levels of immunity. Every increasing year gave women an odds ratio of 0.86 (CI:0.80,0.92) of being immune. Age was associated with immunity to rubella, with a 5.4% (CI:4.0%,6.8%) increased likelihood of immunity for every year older. Country of birth was associated with differences in susceptibility, with those from countries that do not offer MMR vaccine having an odds ratio of 0.710 (CI:0.514,0.906) of being immune to rubella. This study suggests there has been significant decline in immunity to rubella over the past 5 years. Age and country of birth are significant risk factors for non-immunity to rubella. Those at risk should be screened for rubella immunity and offered vaccine prior to conception, including post-partum.

Keywords rubella screening vaccination immunisation antenatal

INTRODUCTION

Rubella is an important pathogen for the fetus. Primary infection in the first trimester can cause congenital heart disease, sensorineural hearing loss and cataracts (Public Health England, 2013). Prior to the UK vaccination programme, around 1 in 5 women remained susceptible to rubella with hundreds of cases of congenital rubella syndrome (CRS) (Tookey, 2004). Rubella infection and CRS are now rare in the UK, with twenty infants diagnosed with CRS since 2000 (Tookey, 2014). In the majority of these cases rubella was acquired abroad (Tookey, 2014). Worldwide, however, an estimated 110,000 babies are born with CRS every year (Cutts & Vynnycky, 1999).

In the UK, a national immunisation programme for adolescent girls began in 1970 using measles/rubella vaccine (Public Health England, 2013). This was later replaced by universal measles/mumps/rubella (MMR) vaccine for toddlers in 1988 (Public Health England, 2013). Since the 1970s, women at antenatal booking clinics have been routinely screened for rubella immunity through testing for IgG antibodies specific for rubella. Any found to be non-immune are recommended to receive MMR vaccine postpartum. This activity will shortly cease, because the Joint Committee on Vaccination and Immunisation has recently recommended that immunity to rubella should be assessed solely by MMR vaccination history (Public Health England, 2016).

Because of this planned change, we took the opportunity to review our screening programme for antenatal testing of rubella immunity before screening ceases. Levels of immunity to rubella in the antenatal population of a north London hospital were determined. Risk factors for non-immunity to rubella and changes in levels of immunity over the study period were assessed.

MATERIALS AND METHODS

We undertook a retrospective analysis of attendances to antenatal booking appointments at the Royal Free Hospital. Attendances between January 2010 and September 2014 were included in the study. At each booking appointment midwives collected information on a standard electronic pro forma. This information was self-reported and included age, country of birth, ethnic origin, previous pregnancies and estimated gestational age of the fetus.

At booking, every woman was offered screening for immunity to rubella. Rubella IgG was measured using the Abbott ARCHITECT.[1] Positivity for rubella IgG was defined as detection of IgG antibodies at a level of greater than or equal to 10 IU/mL, as recommended by the UK National Screening Committee (UK National Screening Committee, 2003). Results were matched with antenatal booking information using hospital number and date of test.

Demographic data from electronic booking appointment records were analysed to identify risk factors for rubella non-immunity. Individuals were defined as being from a high-risk country for rubella non-immunity if their country of origin was listed as a WHO target country for rubella vaccination (World Health Organsiation, 2012) or if the country of origin does not have rubella vaccination as part of the routine childhood immunization schedule (World Health Organisation, 2016).

Previous pregnancy was defined as previous live births, stillbirths, miscarriage or terminations of pregnancy as reported by the women at their first booking appointment in the study period.

Ethnicity was self-reported and categories included ‘African’, ‘Any other Asian background’, ‘Any other Black background’, ‘Any other ethnic group’, ‘Any other mixed background’, ‘Any other White background’, ‘Bangledeshi’, ‘British’, ‘Caribbean’, ‘Chinese’, ‘Indian’, ‘Irish’, ‘Not Known’, ‘Not Stated’, ‘Pakistani’, ‘Unknown’, ‘White and Asian’, ‘White and Black African’ and ‘White and Black Caribbean.’ For simplicity of analysis, these were grouped into White (Any other White background, British and Irish), ‘Non-white or mixed’ (‘African’, ‘Any other Asian background’, ‘Any other Black background’, ‘Any other ethnic group’, ‘Any other mixed background’, ‘Bangledeshi’, ‘Caribbean’, ‘Chinese’, ‘Indian’, ‘Pakistani’, ‘White and Asian’, ‘White and Black African’ and ‘White and Black Caribbean’) and Unknown (‘Not Known’, ‘Not Stated’, ‘Unknown’).

Age of women was stratified into six cohorts <20, 20-24, 25-29, 30-34, 35-39, >39 years old. Year of booking was taken as the date of the electronic maternal booking form.

Analysis was carried out in IBM SPSS Statistics v20. The frequency of rubella non-immunity was calculated by year of first booking, age, ethnicity, previous pregnancy, country of birth and number of bookings within the study period. Differences in proportions between groups were compared using Pearson’s chi-squared test. Logistic regression modeling was used to investigate the relationships between the effects on rubella immunity of

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year of first booking, age, ethnicity, previous pregnancy, country of birth and number of bookings within the study period

RESULTS

16,250 booking appointments from 14,118 individuals were identified during the study period. Demographic details of these individuals are shown in Table 1. Approximately half (7,840/14,118, 55.5%) of individuals' reported their ethnic background as white. The mean age at their first booking appointment in the study period was 31 years (range 14-54). Just over half of individuals reported a previous pregnancy at their first booking appointment in the study period (8,101/14,118, 57.3%).

14,100/14,118 (99.8%) of individuals had rubella IgG tested on first booking within the study period. 13,088/14,100 (92.8%) were immune to rubella on their initial booking.

Univariate analysis to investigate factors for non-immunity to rubella.

Age

Age was a significant predictor of immunity to rubella. Those aged under 20 showed the lowest levels of rubella immunity (80.8%) compared with those aged 20 to 24 (89.4%), 25 to 29 (92.9%), 30 to 34 (94.5%), 35-39 (94.2%) and over 39 (92.8%) ($p<0.001$). See Figure 1 and Table 2.

Ethnicity.

92.1% (13,003/14,118) of women declared an ethnicity at booking. Those identifying as a 'white' ethnicity were more likely to be immune to rubella (7,340/7,830, 93.7%) compared to those of other ethnicities (4,743/5,173, 91.7%) $p<0.001$).

Year of booking

Differences in rubella immunity in different years of booking within the study period was analysed using univariate analysis. Women booking in 2010 showed significantly higher rates of immunity (94.9%), which declined consistently in subsequent years: 2011, 94.0%; 2012, 92.1%; 2013, 90.7%; 2014, 91.2%. ($p<0.001$). See Table 3.

Number of bookings in study period

Number of booking appointments in the study period was associated with rubella immunity in univariate analysis. Immunity in those with only one booking was significantly lower (n 11,255/12,154; 92.6%) than in those with multiple bookings (n=1,835/1,946 94.2%) (p=0.01).

Previous pregnancy

At individuals' first booking within the study period 6017/14,118 (42.6%) reported no previous pregnancies. Previous pregnancy was not identified as significant risk factor for rubella non-immunity at booking in univariate analysis. (468/6,010 7.8% vs 544/8,010 6.7%, p=0.111)

Country of birth

Country of birth was investigated as a risk factor for non-immunity to rubella IgG. 10,565/14,118 (74.8%) of individuals had both a country of birth recorded and a rubella IgG test performed. The rate of non-immunity in those with a country of birth recorded was not significantly different from those who did not have a country of birth recorded (7.4% vs 8.2%, p=0.12). Being from a country at risk of rubella non-immunity significantly increased the risk of being rubella non-immune at booking (221/2,226, 9.9% vs 564/8,339, 6.8% p<0.001).

Multivariate analysis

Logistical regression was used to investigate the effect size of these individual factors and to minimise confounding. Age, ethnicity, previous pregnancy, year of booking, number of bookings within the study period and whether born in a high risk country for rubella non-immunity were analysed using a multivariate binary regression model (Table 4).

Age remained a significant predictor of immunity to rubella. For every year older there was a 5.4% increased chance of being immune to rubella (CI: 4.0%, 6.8%, p<0.001).

Being born in a high risk country for rubella non-immunisation was a significant predictor of non-immunity. Those born in a high risk country were around 30% more likely to lack rubella immunity (OR 0.710 CI: 0.514, 0.906, p=0.001).

Year of initial booking within the study period was associated with differences in immunity to rubella. Each successive year of the study period was associated with a 14.4% increased chance of non-immunity (OR: 0.856, CI: 0.797, 0.915, p<0.001).

Ethnicity and number of bookings within the study period failed to reach significance in the multivariate analysis as predictors of rubella non-immunity in antenatal women. Previous pregnancy remained a non-significant predictor of non-immunity to rubella in the multivariate analysis.

DISCUSSION

The rubella screening programme exists to assess the susceptibility of pregnant women to rubella infection. In this study over 99% of the antenatal population was screened for rubella immunity, confirming good implementation of screening in our hospital.

This study confirms that the vast majority of women seeking antenatal care in North London are immune to rubella. Screening showed an overall immunity to rubella of 92.8% amongst women at first booking.

The range of herd immunity seen in this study is above minimum estimates of critical prevalence of rubella immunity required to prevent an outbreak (Plans, 2013). However, it is below the 95% immunity level recommended for childbearing women by the 2003 WHO European strategic plan (World Health Organisation, 2003). As such, clinicians should be vigilant for women at risk of rubella susceptibility and offer screening or vaccination as appropriate.

Immunity across the study period decreased consistently in later years. Between 2010 and 2014 immunity in our cohort significantly waned from 94.9% to 91.2%. The decrease in immunity over time in our antenatal population supports the findings of other studies. Between 2004 and 2009, the susceptibility of childbearing women was found to increase by 60% in England (Byrne *et al*, 2012). Over a similar time period in the West Midlands, susceptibility in pregnant women increased around five times to around 7% (Skidmore *et al*, 2014). These figures are broadly similar to our findings. Lower levels of rubella immunity in later years may reflect the fact that rubella is no longer circulating in the UK. Alternatively it may reflect decreasing vaccine uptake. This emphasizes the importance of maintaining herd immunity through screening at-risk women and offering universal MMR vaccine.

Country of birth was associated with differences in immunity to rubella. Women born in countries where vaccination programmes are known to be inadequate or where vaccination is not universally recommended were at increased risk of rubella non-immunity. North London has a highly mobile population and high levels of immigration. Recent waves of immigration may explain declining immunity in our population. This hypothesis is supported by our findings that country of birth is a significant predictor of non-immunity. Other studies have previously identified women of childbearing age from non-white ethnic groups as significantly more likely to lack immunity to rubella (Byrne *et al,* 2012; Tookey *et al*, 2002).

Risk factors for non-immunity to rubella also include age, with younger women being significantly more at risk than older women. Indeed women under 20 at first booking had the lowest levels of immunity, with rubella IgG positivity at just 80.8%. The levels of susceptibility to rubella in this age group are around 2.5 times higher than in those aged 40 and over.

The reasons for decreases in immunity with younger women and in later years is unclear. Decreases in immunity may relate to declining rates of vaccine uptake, explaining why levels of immunity are lower in younger women and in later years. Unfortunately, our dataset did not include information on vaccination history. Previous, widely reported data showed that vaccine uptake for the MMR vaccine fell after 1998 associated with the MMR-autism scandal (Jansen *et al*, 2003). The youngest women in our cohort, aged between 14 and 25 and with the lowest levels of rubella immunity would have been vaccinated between 1990 and 2002. Thus if low immunity in our sample was due to poor uptake, this may be in part attributable to the Wakefield scandal.

Furthermore, wild-type rubella virus no longer circulates in the UK. Immunity in more recent years is therefore solely attributable to vaccination. Vaccination may not produce antibody titres as high as natural infection. Furthermore, lack of exposure to circulating wild-type rubella will prevent antibody boosting via secondary immune responses. Previous data supports this, showing IgG levels in vaccinated individuals decrease in relation to the time elapsed since vaccination. In Canada, a retrospective study showed 1/3 of vaccinated individuals had IgG levels that were below the designated threshold of immunity after 15 years. (Lai *et al*, 2015) Given sustained elimination of rubella virus in Canada, it is argued low levels of rubella-specific antibodies below the designated threshold for immunity may be sufficient to prevent spread of infection. *In vitro* data supports this, with college age students with antibody levels below

As our data did not include information on individuals' vaccination history these effects could not be assessed, especially in the knowledge that many

foreign-born women may have received different vaccination schedules. Our study used country of birth as a proxy measure for likelihood of vaccination in childhood. In the absence of individuals' vaccination records, a more accurate measure may be length of residence in the UK. This study could also not investigate the effect of other factors such as socioeconomic class due to lack of data.

This study is a topical assessment of immunity to rubella in an at-risk population just before routine screening ceases. It confirms that the vast majority of antenatal women attending our clinic show immunity to rubella. As such, screening is unlikely to have benefit for these women. There exists a small but significant proportion of women potentially susceptible to rubella infection. By stopping routine screening these women are unlikely to be identified as potentially susceptible. We have identified risk-factors which may aid identification of those potentially susceptible to rubella infection.

One potential strategy for increasing herd immunity would be to selectively screen patients in high-risk categories and offer post-partum vaccination if they were found to be potentially susceptible. The strategy of identifying those at risk of non-immunity using risk factors is unclear given the assertion that previously vaccinated individuals with antibodies below the threshold may in fact have sufficient immunity to prevent infection

Stopping routine screening of rubella immunity in antenatal women will also mean a loss of valuable data with which to analyse changes in immunity over time and demographics. This is especially pertinent given the changes in immunity demonstrated in this study.

This study is limited by technical problems with assessing rubella immunity and the definition of susceptibility. The accepted and recommended practice of using a 10 IU/mL threshold for immunity has been reported to lack sensitivity.(ref) This may lead to some individuals with a concentration of rubella IgG below the designated immunity threshold being incorrectly labelled as 'susceptible' to rubella infection. Unfortunately, for individual's falling below the 10 IU/mL threshold for immunity a second, gold-standard reference assay was not uniformly employed to assess immunity. As such we cannot ascertain which proportion of our population were falsely deemed 'potentially susceptible' to rubella infection due to poor sensitivity of the assay.

In a recent series, over half of those samples falling below immunity threshold when tested using the Abbott Architect assay tested positive for antibodies by immunoblot and neutralization assays. (Bouthry *et al*, 2016).

Around 2/3 of these false-negatives fell into the equivocal category (5-10 IU/mL). This supports previous modelling which has suggested antibody levels <5 IU/mL may more accurately represent women susceptible to infection (ai *et al*, 2015). This may mean our analysis overestimates susceptibility substantially despite calibration against an international standard. Provide equivicol 5-10

It does not, however, describe rubella immunity in the general population, which is also important for providing the protection of herd immunity to the 7.7% of our antenatal population susceptible to rubella infection.

Younger women are also significantly more likely to be susceptible to rubella infection. Further efforts must be made to offer vaccination to young women, who have been consistently shown to have higher levels of susceptibility to rubella.

At present, seronegative women are protected while they remain in the UK, because there is no evidence of rubella circulation. However, they could be exposed through travel abroad or by contact with imported cases of rubella. An important practice point is that healthcare professionals, especially GPs and obstetricians, should actively offer screening for rubella immunity to women born outside the UK and administer vaccine to those who are found to be seronegative. Decreases in immunity over time and in younger women require special efforts to boost herd immunity.

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None.

DISCLOSURE OF INTERESTS

The authors had no relevant interests to disclose

DETAILS OF ETHICS APPROVAL

The study was registered with the local Governance department and subject to their standards. Use of clinical data was approved by the local NHS Trust Governance Committee, including retrospective use of women's demographic data and test results. Approval was granted on 9th September 2014.

FUNDING

No funding was received for this study.

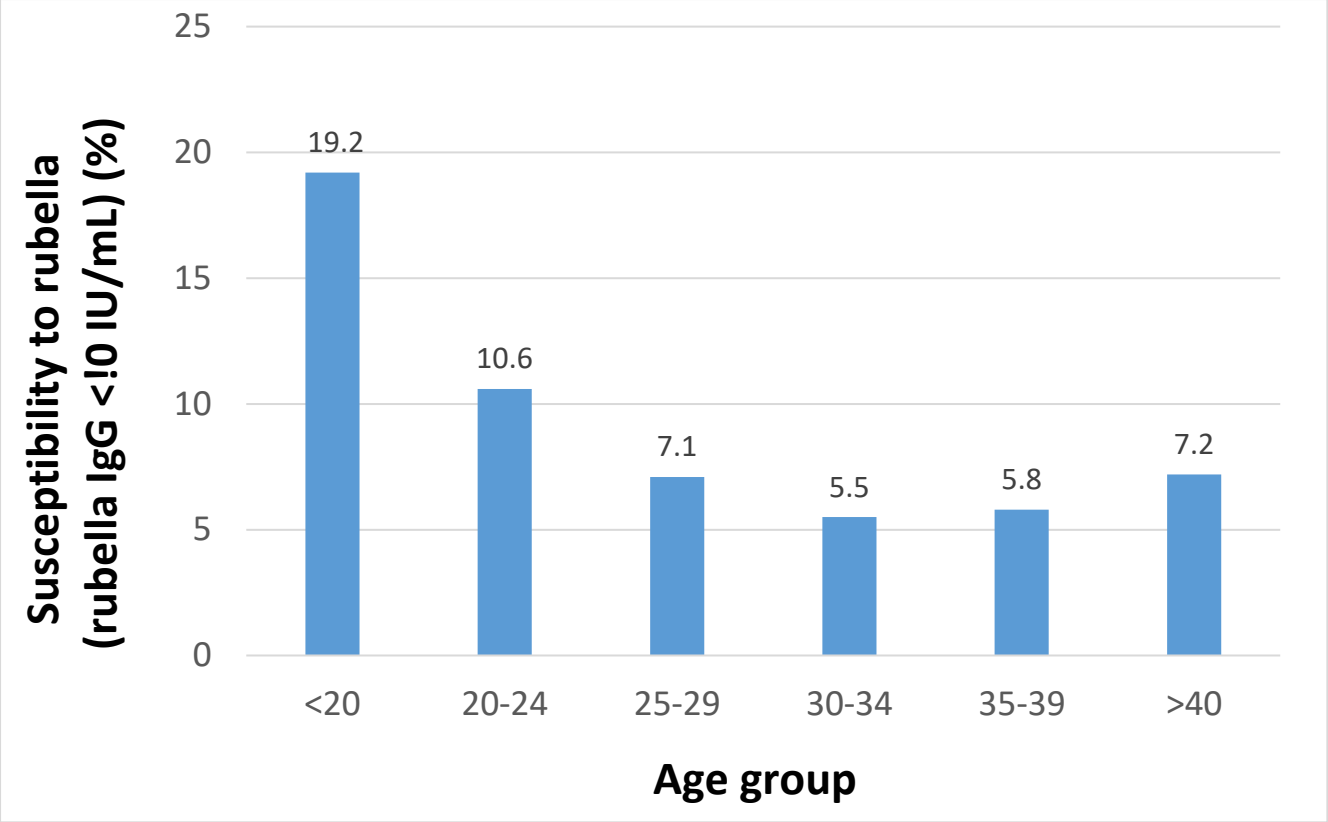

Figure 1: Age-stratified susceptibility to rubella at first booking appointment within the study period

	Women (n) ∑=14,118	Women tested for rubella immunity (n) ∑=14,100	Women tested for rubella immunity (%)	Women immune to rubella (n) ∑=13,088	Susceptibility of women to rubella(%)
ETHNIC BACKGROUND					
White	7840	7830	99.9	7340	6.3
Non-white or mixed	5181	5173	99.8	4743	8.3
Unknown	1097	1097	100%	1005	8.4
YEAR OF BOOKING					
2010	3530	3521	99.7	3340	5.1
2011	3208	3204	99.9	3011	6.0
2012	2998	2993	99.8	2756	7.9
2013	2750	2750	100	2493	9.3
2014	1632	1632	100	1488	8.8
PARITY					
0	6017	6010	99.9	5542	7.8
1+	8101	8090	99.9	7546	6.7
AGE					
<20	294	293	99.7	237	19.1
20-24	1756	1753	99.8	1539	12.2
25-29	3816	3813	99.9	3543	7.1
30-34	4860	4855	99.8	4589	5.5
35-39	2677	2673	99.9	2517	5.8
>39	715	713	99.7	662	7.2

Table 1: Summary of results of screening for rubella immunity in women between 2010-2014 in an antenatal clinic, with breakdown of demographic characteristics.

Age	Number of women	Number tested for rubella IgG	Number IgG positive	Number IgG negative	Rubella IgG positive (%)
<20	294	293	237	56	80.9%
20-24	1756	1753	1539	214	87.8%
25-29	3816	3813	3543	270	92.9%
30-34	4860	4855	4589	266	94.5%
35-39	2677	2673	2517	156	94.2%
>39	715	713	662	51	92.8%

Table 2: Rubella IgG positivity in antenatal women at first booking appointment within the study period stratified by age

Year	Number of women (first booking within study period)	Number of women tested for rubella IgG	Number positive for rubella IgG	Number negative for rubella IgG	Rubella IgG poitivity (%)
2010	3530	3521	3340	181	94.9%
2011	3208	3204	3011	193	94.0%
2012	2998	2993	2756	237	92.1%
2013	2750	2750	2493	257	90.7%
2014	1632	1632	1488	144	91.2%

Table 3: Rubella IgG positivity in antenatal women stratified by year of first booking appointment in the study period

REFERENCES

Bouthry E., Furione M., Huzly D, Ogee-Nwanko A., Hao, L. Adebayo A, Icenogle, J., Sarasini, A., Grazia Revello, M., Grangeot-Keros, L., Vauloup-Fellous, C (2016) Assessing Immunity to Rubella Virus: a Plea for Standardization of IgG (Immuno)assays Journal of Clinical Microbiology: 54(7):1720-1725

Byrne L., Brant L, Reynolds C, Ramsay M. (2012) Seroprevalence of low rubella IgG antibody levels among antenatal women in England tested by NHS Blood and Transplant: 2004-2009. Is rubella susceptibility increasing? Vaccine 5(30):161-7

Cutts F.T., Vynnycky E. (1999) Modelling the incidence of congenital rubella syndrome in developing countries. Int J of Epidemiology 28:1176-1184

Jansen V.A., Stollenwerk N., Jensen H.J., Ramsay M.E., Edmunds WJ, Rhodes CJ (2003) Measles outbreaks in a population with declining vaccine uptake Science, 301 p. 804

Lai FY., Dover DC, Lee B, Fonseca K, Solomon N, Plitt SS, Jaipaul J, Tipples GA, Charlton CL. (2015) Determining rubella immunity in pregnant Alberta women 2009-2012. Vaccine 33(5):635-41

Plans P. (2013) New preventive strategy to eliminate measles, mumps and rubella from Europe based on the serological assessment of herd immunity levels in the population Eur J Clin Micro & ID 32:961-966

Public Health England. (2013) Rubella, in: Salisbury D, Ramsay M, editors . The Green Book 2nd edition, Public Health England 2013. p. 343-365

Public Health England. (2016) Press release: Rubella susceptibility screening in pregnancy to end in England Cited: 27 January 2016 Available from: https://www.gov.uk/government/news/rubella-susceptibility-screening-in-pregnancy-to-end-in-england

Skidmore S., Boxall E, Lord S. (2014) Is the MMR vaccination programme failing to protect women against rubella infection? Epidemiol Infect 142:1114-7

Tookey P.A., Cortina-Borja M., Peckham C.S. (2002) Rubella susceptibility among pregnant women in North London. J of Pub health Med 24(3):211-216

Tookey P.A. (2004) Rubella in England, Scotland and Wales. Euro Surveill 9(4):21–3.

Tookey P.A. (2014) Congenital rubella in British Paediatric Surveillance Unit. Annual report 2013-14. p. 10 Available from: http://www.rcpch.ac.uk/bpsu/annualreports

UK National Screening Commitee. (2003) Infectious diseases in pregnancy screening programme. handbook for laboratories; 2003.

World Health Organisation Europe. (2003)Strategic plan for measles and congenital rubella infection in the european region of WHO Geneva: p4.

World Health Organization. (2012) Global measles and rubella strategic plan: 2012-2020. Geneva: 2012. 42 p.

World Health Organisation. [Internet] 2016 WHO immunization schedule by country Available from: http://www.who.int/immunization/monitoring_surveillance/data/en/