Rubella seroprevalence in childbearing age women: a cross sectional study in the province of Frosinone, Central Southern Italy

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Abstract

Background: Congenital rubella infection can be prevented by protecting women of reproductive age through vaccination. The aims of this study were to estimate the seroprevalence of rubella virus antibodies among females aged between 15 and 45 years in the province of Frosinone and to assess knowledge about Rubella and Congenital Rubella Syndrome (CRS) and the association between rubella immunization status and various risk factors.

Methods: A cross sectional study was carried out involving 1242 females recruited by random selection. Enzyme immunoassay method was used to detect and quantify human anti IgG antibodies for rubella virus in 1242 anonymously females aged between 15 and 45 years. A questionnaire was used to collect demographic data including age, sex, previous history, previous vaccination and knowledge about rubella. Statistical analysis was carried out using the program EpiInfo 3.3.

Results: The majority of the females recruited to the study were 15-24 years old (66%), unmarried (71.2%), students (61%) who had not previously given birth (87.9%). Rubella Seroprevalence was 90.3% in women aged 40-45 years old and 68.8% in those aged 15-19 (p=0.0001). Only 137 participants reported having received rubella vaccination. Generally knowledge about rubella and congenital rubella syndrome was lacking.

Conclusions: In the province of Frosinone, women in each age group were in need of active immunization. Furthermore, the susceptibility rates for rubella are far from the value of national serosurveys for similar age groups (8%) and even further from the target set by the National Plan for Measles and Congenital Rubella Elimination launched in 2003. For females of reproductive age, immunization is an effective but underutilized method of CRS prevention and although rubella vaccine is free charge for these women and pre-conception screening is available, these opportunities remain substantially underutilised.

*Key words: rubella, seroprevalence, childbearing women, Italy*

Introduction

Rubella elimination and Congenital Rubella Syndrome (CRS) prevention have been identified as priorities by the WHO Regional Office for Europe since the 1980's when vaccination programmes against measles, mumps and rubella were established according to the Expanded Programme on Immunization of the World Health Organization (WHO). In 2002 the WHO Regional Office for Europe implemented a strategic plan with the specific aim of reducing the number of CRS cases to less than case/100,000 live births by 2010 [1,2].

Despite improvements in vaccination programmes the eradication of Rubella regionally remains a rather distant goal; several countries report high proportions of childbearing age women remain susceptible to rubella virus infection. The number cases of cases of CRS in 2006, reported by 25 European countries, equated to 4,367 confirmed rubella cases from a total of 25,026 cases, giving an overall notification rate of 1.3 x 100,000. Only Denmark and Iceland have achieved uninterrupted 'zero reporting' for at least three years (2004-2006) [3,4].

Reliable data for CRS are difficult to obtain because of the weakness of the surveillance system, but also because rubella infection can be
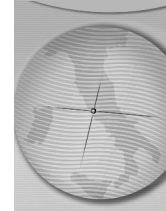
asymptomatic or clinical signs and specific symptoms may appear late in the infection. In 2004 17 cases of CRS across Europe were reported, however rates of CRS, as high as 350 x100,000 live births, have been described during outbreaks in the Russian Federation between 2002-2004 [1,5].

In Italy, vaccination for rubella was introduced during the1970s although no specific target for voluntary vaccinations was identified and no organic policy for offering vaccination existed until 1995 when the Ministry of Health established the Measles Mumps Rubella (MPR) combined vaccination programme [6,7]. In 2003 the National Plan to Elimination of Measles and CRS was approved along with the introduction of CRS surveillance, immunization status evaluation and vaccination for susceptible women of childbearing age. The goals was the reduction of the incidence of CRS to 1 case/100,000 live birth by 2007 [8-10].

All of the described programmes have resulted in the reduction of Rubella incidence in Italy, however cases are still recurrent in non immune subjects and no single Region achieved the target immunization coverage of 95% with the first MMR dose in 2006 [11,12]. Furthermore, homogeneous results are yet to be achieved in Italy [13,14] and there are great variations in immunization coverage not only among the Regions but also among Local Health Units within the same Region.

The last rubella epidemic in Italy occurred in 2002 when 6,224 cases reported , since then a marked decrease in the incidence has been observed, with a historical minimum of 139 reported cases being reported in 2005 [10]. From January 2005 to May 2008, 5 Italian Regions reported 37 notifications of suspected CRS, 5 of which were laboratory confirmed (2 in 2008). In the same period 11 Regions reported 110 suspected cases of Rubella during pregnancy (48 laboratory confirmed cases, 38 of which were in 2008) The average age of the cases was 28 years [15]. In the province of Frosinone (Lazio Region – Central Italy,) MMR vaccination coverage was 62.9% in 2000 and an increase in the percentage of rubella cases in those aged between 15-19 years was observed from 1995 to 2003 [16-18].

Although immunization programs have led to a slow-down in the movement of the Rubella virus, values that indicate the prevention of its spread have not yet been reached [19], explaining the shift of the average age of cases and exposing adolescents and young adults to a greater susceptibility to infection [11,16].

The aims of this study were as follows:

- to estimate the seroprevalence of rubella virus antibodies among females aged between 15 and 45 years in the province of Frosinone;
- to assess the association between rubella immunization status and some risk factors;
- to examine the knowledge of these women about rubella disease and its prevention.

Methods

A cross sectional study was carried out on 1,242 females recruited by random selection using the simple random sampling method. Participation (approximately 12%) was calculated by dividing the total number of participants by the total number of women of the same age, living in the Italian province of Frosinone (Lazio region). Informed consent for taking blood specimens and the use of personal data was obtained from the individuals or their parents (for those under the age of 18), with data being collected using a specific form. A questionnaire was used to collect demographic information including age, marital status, educational level, history of rubella disease, history immunization, and knowledge about rubella.

Laboratory methods

A total of 1,242 anonymously serum specimens were collected from January 2005 to November 2006 and were stored at -20°C and tested for rubella IgG antibodies. Enzyme immunoassay method was used to detect and quantify human anti IgG antibodies for rubella virus in the serum according to the producer's instructions (Radim).

The sensitivity and specificity of the method used, as reported by the manufacturer, was 97.2% and 100% for IgG anti rubella respectively.

Samples with IgG values less than 15 IU/mL were considered non reactive for antirubella IgG antibodies (negative), samples with IgG values higher than 30 IU/mL were considered reactive for antirubella IgG antibodies (positive) and samples with IgG values between 15 and 30 IU/mL were considered equivocal and repeated for confirmation in accordance with the producer's instructions indicated in the rubella IgG Radim kit.

Women that were susceptible to rubella were offered vaccination at the relevant Local Health Unit.

Statistical analysis

Statistical analysis was carried out using the program EpiInfo 3.3. Data were analyzed through frequency distributions. The statistical comparison of data was performed using the Chi-square tests (χ^2) compared those with and without missing data on the relevant proportions for several
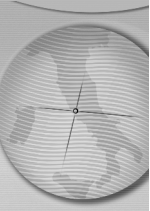
anagrafic date (age, educational level, ecc…) and a p-value <0.05 was considered statistically significant. Logistic regression was used to identify significant predictors of seropositivity. Results of the logistic analysis were expressed as Odds Ratio (OR) and 95% Confidence Intervals (95% CI). Finally, three different multivariate models, one for each outcome variable (seropositivity), were built using the stepwise approach (backward elimination procedure).

Results
Study population
    The mean age of the 1,242 females joining in the study, was 23.4 ±7.3 years (mode 16). The main characteristics of the study population are shown in Table 1. Most were 15-24 years old (66%), unmarried (71.2%), students (61%) and not having birth (87.9%).
Seroprevalence
    Rubella specific IgG antibody was positive in 77.9% of women, the distribution of age-stratified IgG antibody for rubella had a significantly higher seroprevalence in older women aged 40-45 years (90.3%) compared to women aged 15-19 years (68.8%) (p=0.0000). Higher IgG antibodies (>240 UI/ml) were detected in women aged 20-24 (35.0%) and 15-19 years (40.9%; p=0.0000) (tab.1).
Personal history of previous Rubella infections and vaccination
    Most of the women interviewed affirmed they did not remember having had rubella although more then half of them tested positive for rubella specific IgG antibodies (tab.2).
    Only 137 participants reported having received rubella vaccination although 9.5% of them were shown to be IgG negative; 287 women reported having had rubella infection and the 7.7% of them resulted IgG negative. Mostly females aged under 20 years (40%) and 21-25 years (27%) were vaccinated against rubella. However, we did not identify statistically significant differences among age groups.
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General knowledge
    The respondents' knowledge about rubella infection is summarized in Table 3.
    68.1% of the responders correctly knew the transmission pathway of the infection and with regards to transmission to the fetus during pregnancy, the risk was perceived by most women (78.1%) but only 20.3% knew that the infection could cause severe congenital malformations and only 1.7% knew that it could cause spontaneous miscarriage. Around half of the interviewees stated that they would not have known what to do if they were to contract a rubella infection during pregnancy, only 8.1% knew that newborns affected by CRS are contagious.
    36.2% of the women who had given birth declared that they were not aware of their own immunity status during pregnancy. Half of the women thought that their knowledge about rubella was insufficient and that they would specifically like more information from their family doctors (36.2%), gynecologists and paediatricians (25.7%). This result confirmed the percentage of women who were unaware of rubeotest.
    The bivariate analysis undertaken included correlating the data concerning the subjects' knowledge with their personal data, from which evidenced meaningful associations in relation to the age, the marital status and the title of study (tab. 4).
    Multivariate logistic regression analysis revealed that   the following variables would be independent predictors of susceptibility *age, marital status, having given birth, and rubella vaccination* (tab.5).

Discussion
    The success of the WHO Europe regional strategy in eliminating rubella depends mostly on the immunization coverage levels achieved with in EU countries. Suboptimal vaccination coverage, in fact, cause a reduction of incidence but also an increase of the average age with more cases being reported in teenagers and adults and a subsequent higher risk of CRS. Even though the
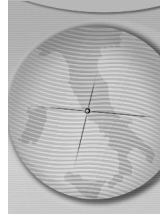
Table 1. Seroprevalence of rubella IgG antibodies subdivided according characteristics of participants.

Characteristics			Positive		Negative		Equivocal		*p*- value
	n	%	n	%	n	%	n	%	
Age group									
15-19	474	38.2	326	68.8	134	28.3	14	3.0	0.0000
20-24	346	27.9	281	81.2	60	17.3	5	1.4	
25-29	190	15.3	160	84.2	24	12.6	6	3.2	
30-34	101	8.1	86	85.1	8	7.9	7	6.9	
35-39	69	5.6	59	85.5	7	10.1	3	4.3	
40-45	62	5.0	56	90.3	3	4.8	3	4.8	
Total	**1242**	**100**	**968**	**77.9**	**236**	**19.0**	**38**	**3.1**	
Education level									
Secondary school	1064	85.7	824	85.7	208	88.1	31	81.4	0.0000
Graduated	66	5.3	54	5.6	10	4.2	2	5.3	
No answer	112	9.0	89	9.2	18	7.6	5	13.2	
Work activity									
Student	758	61	585	60.4	156	66.1	17	44.7	0.06
Employed	114	9.2	90	9.3	16	6.8	8	21.1	
Independent professional	32	2.6	28	2.9	3	1.3	1	2.6	
Housewife	40	3.2	34	3.5	4	1.7	2	5.3	
Unemployed	7	0.6	5	0.5	1	0.4	1	2.6	
Other	43	3.5	37	3.8	6	2.5	0	0	
No answer	258	20.0	189	19.5	50	21.2	9	23.7	
Marital status									
Unmarried	884	71.2	669	69.1	193	81.8	22	57.9	0.0014
Married	213	17.1	178	18.4	23	9.7	12	31.6	
Other	17	1.4	15	1.6	2	0.8	0	0	
No answer	128	10.3	106	11.0	18	7.6	4	10.5	
Havin given birth									
No	1092	87.9	838	76.6	224	20.5	30	2.7	0.004
Yes	150	12.1	130	86.7	12	8.0	8	5.3	
Pregnant women									
No	1085	95.4	924	95.3	228	96.6	33	86.8	0.0172
Yes	57	4.6	45	4.7	7	3.0	5	13.2	

incidence of CRS depends on the percentage of seronegative women in their procreative period, immunization of females of reproductive age is an effective but underutilized method of CRS prevention [20].

Nowadays in Italy, interventions targeting childbearing age women are not yet satisfactory and although rubella vaccine is free of charge for these individuals and preconceptional screening is available, these opportunities are substantially missed. Surveillance data show, in fact, that less than the 25% of women with suspected rubella during pregnancy had a rubeo-test before pregnancy and many others knowing their susceptibility are still not immunized [21,22].

Our investigation showed that women in each age group have an high proportion of susceptibility to rubella infection, far from the value of national sero-surveys in similar age groups (8%) [11].The overall susceptible rate was 28.3% in the 15-19 year age group and 17.3% in 20-24 year age group (tab.1) whereas to prevent

Table 2. Disease and vaccination history among Rubeotest.

Rubella infection	Diseases History (n)	Positives	Negatives	Equivocal	
Yes	287	89.9%	7.7%	2.4%	
No	275	68.4%	27.3%	4.4%	χ^2= 42.04
Unknow	343	78.7%	18.7%	2.6%	p-value < 0.001
Non responders	337	74.8%	22.3%	3.0%	
Rubella vaccine	**Vaccination History (n)**				
Yes	137	86.1%	9.5%	4.4%	
No	475	73.3%	23.8%	2.9%	χ^2= 19.74
Unknow	455	78.2%	19.2%	2.6%	p-value < 0.001
Non responders	175	83.4%	13.1%	3.4%	

Table 3. Number and percentage of women' general knowledge about rubella.

Statement	Correct Number	Percentage	Incorrect Number	Percentage	Unknown Number	Percentage	Non responders Number	Percentage
What is rubella?	821	66.1	127	10.2			294	23.7
Is rubella contagious?	1001	80.6	21	1.7	82	6.6	138	11.1
How is rubella transmissitted?	846	68.1	16	1.3	192	15.5	188	15.1
Is foetus at risk?	970	78.1	9	0.7	106	8.5	157	12.6
What to do during pregnancy?	344	27.7	132	10.6	582	46.9	184	14.8
Is rubella virus excreted by infants with CRS?	108	8.1	222	17.9	747	60.1	173	13.9
Is't a vaccine?	1020	82.1	9	0.7	66	5.3	147	11.8
What precautions are needed after vaccination?	232	18.7	126	10.1	701	56.4	183	14.7
What is rubeo test?	625	50.3	11	0.9	387	31.2	219	17.6

CRS it has been estimated that the proportion of susceptible females of reproductive age should not exceed 5% [8]. The over represented number of younger females allowed us to further investigate rubella susceptibility in this more susceptible age group, this was important considering that the young women of the province of Frosinone may be exposed to an increased risk of rubella infection related to the history of local vaccination campaigns and outbreaks in the surrounding area, for example in 2002 reported cases of rubella primarily concerned those aged between 15 and 19 as well as those aged between 20-34 years [14].

It should be highlighted that most of the women interviewed did not have evidence of their serological or immunization status. Mostly females aged under 20 years (40%) and 21-25 years (27%) were vaccinated against rubella, 137 participants reported having received rubella vaccination but 7.7% of them tested IgG negative.

There is a lack of basic information about rubella so education and screening programmes should be put in place to disseminate and promote screening and immunisation in school girl and susceptible women of childbearing age.

As recently pointed out, CRS prevention requires strong integration and coordination between medical services in order to reach the objective to immunize 95% of susceptible women - those who are post-partum or post abortion.

There should be an emphasis on the need to disseminate counselling whenever a childbearing woman interacts with general and hospital practitioners, gynaecologists and obstetricians. These practitioners should recommend rubella virus antibody screening and as well as provide recommendations for immunisation screening [5,19] .

The results of this study need to be interpreted with caution. Several limitations are recognised first of all that this research covered a single Italian province in Central Southern Italy, therefore, it is not possible to extrapolated the results for the whole country. The sample was recruited on a voluntary basis and the over representation of younger females could bias the results.

This study shows that younger women may be exposed to an increased risk of rubella infection but these results might not be generalized to other females populations and replication studies
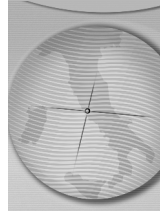
Table 4. Bivariate analysis to correlate knowledge about rubella and personal data.

Statement	Correct knowledge	p-value
Definition of rubella		
Age group		
15-19	46.5%	0.0000
Marital status		
Unmarried	72.5%	0.0000
Married		
Education status		
Secondary School	45.1%	0.0000
Modality of transmission		
Age group		
15-19	35.3%	0.01
Marital status		
Unmarried	74.5%	0.0000
Married		
Education status		
Secondary School	57.7%	0.0000

Statement	Lack knowledge	p-value		
Judgment on the own personal knowledge				
Age group				
15-19	43.2%	0.0000		
Marital status				
Unmarried	78%	0.0000		
Married				
Education status				
Secondary School	53.4%	0.0000		

| Statement			Yes	p-value
It's a prevention?				
Age group				
15-19			41%	0.0002
Marital status				
Unmarried			76.2%	0.02
Married				
Education status				
Secondary School				
Graduate			54%	0.0000
It's a vaccine?				
Age group				
15-19			33.7%	0.01
Marital status				
Unmarried			63%	0.0000
Married				
Education status				
Secondary School			44.4%	0.0000
Graduate				
Risk to the foetus				
Age group				
15-19			41%	0.0001
Marital status				
Unmarried			76.8%	0.0000
Married				
Education status				
Secondary School			52.2%	0.0000
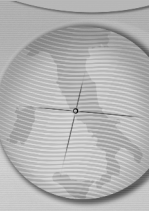
Table 5. Predictors and associated odds ratio of rubella susceptible rate.

Characteristics	OR	(95% CI)	p- value
Age group			
15-19	2.0	1.1-2.4	0.001
20-24	1.4	1.2-1.6	0.0000
>=25	1.0		
Marital status			
Unmarried	2.3	1.4-3.6	0.0003
Married	1.0		
Having given birth			
Yes	3.4	2.1-4.6	0.0005
No	1.0		
History of rubella			
Yes	0.6	0.4-0.9	0.01
No	1.0		
Rubella vaccination			
Yes	1.0		
No	2.5	1.4-4.3	0.001
Unknown	1.9	1.2-3.06	0.004

of different sizes need to be conducted in different geographic area.

Acknowledgements

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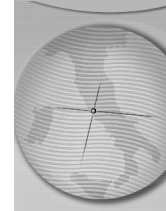
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