2013•2014
FACULTY OF SCIENCES
Master of Statistics: Biostatistics

Masterproef

Assessing outbreak risk in highly vaccinated populations using spatial seroprevalence data on rubella

Promotor :
Prof. dr. Niel HENS
De heer Steven ABRAMS

Eleni Kourkouni

Master Thesis nominated to obtain the degree of Master of Statistics , specialization Biostatistics

Transnational University Limburg is a unique collaboration of two universities in two countries: the University of Hasselt and Maastricht University.

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Abstract

Background: The incidence of rubella has declined dramatically since the introduction of vaccines containing the rubella antigen, such as trivalent measles-mumps-rubella (MMR) vaccines. The objective of this report is to identify regions of high outbreak potential in Belgium based on regional estimates of the effective reproduction number R.

Methods: Susceptibility profiles in 2013 were estimated using a generalized additive model including age, gender and spatial location as covariates, and further informed by serological survey data on rubella from 2006 and available vaccination coverage information. Seroconversion and waning rates of the rubella antibodies play an important role in the estimation of susceptibility. Therefore, waning of vaccine-induced immunity is included in our analysis through the specification of an exponential decay model. Whether a vaccination program will achieve elimination of the disease at a particular time after the introduction of mass vaccination is determined by the effective reproduction number. For the estimation of the effective reproduction number, we rely upon a specific choice of the basic reproduction number R_0 equal to 8. A sensitivity analysis with regard to R_0 is presented as well.

Results: An extensive literature review yields an estimated combined seroconversion rate equal to 0.984. The overall exponential waning rates are equal to 0.015 and 0.016 after the first and second MMR dose, respectively. Based on the estimated age- gender- and location-specific susceptibility in 2013 and the assumed basic reproduction number, the estimated effective reproduction numbers for all Belgian provinces are well below the epidemic threshold of one.

Conclusion: Having well behaved estimates for the reproduction numbers does not mean that we should be reassured. Preventing rubella outbreaks, even when the risk is low, most likely requires various ingredients, in addition to a routine high-coverage two-dose vaccination program. Targeting specific age groups in specific localities where vaccination coverage is lower could be an efficient way of reducing the risk of new outbreaks. In the future outbreaks could occur as a result of a build-up of susceptibility in the population

Keywords: Effective reproduction number, susceptibility profiles, seroconversion, waning of vaccine-induced immunity

Contents

1. Introduction

Rubella, also known as German measles or three-day measles, is a disease caused by the rubella virus. This disease is usually mild and attacks often pass unnoticed. The disease can last from few to twelve days. Children attacked from rubella recover more quickly than adults. Rubella is a common childhood infection that can sometimes be fatal usually with minimal systemic upset although transientarthropathy may occur in adults. Serious complications such as deterioration of the skin are very rare. Apart from the effects of transplacental infection on the developing fetus, rubella is a relatively trivial infection (52). Similar diseases as rubella are measles and mumps that attack mostly children. All three diseases are vaccine preventable.

Vaccination is the most effective strategy to reduce morbidity and mortality from vaccine preventable diseases (1). For vaccination programs to be effective, it is essential to reach and maintain high vaccine coverage and rates of acceptance (2,3). The incidence of rubella has declined dramatically since the introduction of vaccines containing the rubella antigen, such as trivalent measles-mumps-rubella (MMR) vaccines. The first rubella vaccine—a live, attenuated vaccine—was licensed in 1969. The vaccine came available in most high income countries. In 1979, an improved live rubella vaccine the RA27/3 was introduced and had been used in Europe for years and offered superior protection against the disease. It also replaced the original rubella vaccine in the MMR combined shot, and is still used today (53). In Belgium, the combined measles, mumps, and rubella (MMR) vaccine was introduced in 1985 for children 12 to 15 months of age and the second dose of the MMR vaccine was implemented in 1995 for children 10 to 13 years of age. In 2003, the administration of the first dose of the MMR vaccine was modified to 12 months (4). Vaccination coverage is essential to understand age-specific immunity as well as the transmission dynamics. However, estimates of vaccination coverage remained unreliable during the first 15 years of the MMR program. Before 1985, MMR vaccine coverage was unlikely to have exceeded 50% (5), whereas it was estimated at around 66% in 1995 and 83% in 1999 in the Brussels and Flemish regions, respectively (6). In 2005, vaccination coverage of one dose of MMR vaccine was estimated at 94% for toddlers (18-24 months), 88% for 7-year-old schoolchildren and 84% for 14-year-old adolescents in the Flemish population (7). Although these estimates may seem high, they remain considerably below the elimination threshold (95%), even more since only 75% of the adolescents received both MMR

doses. It is also noticeable how vaccination coverage is showing variations throughout the Belgian regions. Hence, outbreaks of rubella are likely to occur in regions with low vaccination coverage.

Furthermore, an important factor for the occurrence of a rubella outbreak is the immunogenicity and effectiveness of the rubella component of the MMR vaccine. Low seroconversion rates and rapid decline of vaccine induced antibodies, increase the risk of occurrence of an epidemic. In most studies, the seroconversion rate for the rubella component reaches 95% or more after a single dose of the vaccine (54). There are no recent reports on rubella outbreaks in Belgium stating that rubella transmission is eradicated. In recent years, rubella outbreaks have been reported in highly vaccinated populations throughout Europe. On the one hand, Romania which had no vaccination program till 2002, experienced a large rubella outbreak in 2002-2003, with more than 115.000 reported cases nationwide (8). On the other hand, Finland was the first country documented in which indigenous rubella is eliminated as a result of a 12 year, 2-dose MMR vaccination program (9).

The objective of this report is to identify regions of high outbreak potential in Belgium based on regional estimates of the effective reproduction number R. Estimates of the effective reproduction number in the 593 different Belgian municipalities in 2013 are obtained using Belgian serological and vaccination coverage data. Firstly a meta-analysis was performed in order to estimate the seroconversion and waning rates of the rubella antibodies, which will be useful to predict the susceptibility profiles in 2013. Secondly a generalized additive model was considered to model the observed seroprevalence of rubella and later on to predict the proportion of susceptible individuals. Lastly, based on the susceptibility profiles, the effective reproduction numbers are calculated and graphically displayed on a spatial map of Belgium. The statistical software that was used for the analysis is R, due to the reason that this software is more flexible in programming such analysis.

2. Data Description

The data used in the statistical analysis were prospectively collected by diagnostic laboratories and blood transfusion centers in Belgium, from January 2006 to October 2007. Serum antibodies concentration against measles, mumps, and rubella were measured by enzyme-linked immunosorbent assay (ELISA). The cut-off points were determined by the ELISA manufacturer for each disease. Overall, immunoglobulin G (IgG) levels above the threshold are classified as seropositive, below as seronegative, and in between as inconclusive (equivocal). In this report, a cut-off point of 10 IU/mL was used for rubella (4). More specifically 3823 samples were tested for the presence of lgG antibodies against rubella. Equivocal and missing response cases were excluded from the statistical analysis. The serology is linked with the individual's age, gender and residence, if available. Otherwise, the spatial location of the test laboratories is used as a proxy for the residence of test subjects. From the available blood samples, 8% tested negative for the presence of antibodies against the rubella virus. Figure 1 shows the age-stratified cross-sectional serological profile of rubella anno 2006 in Belgium. The circles are proportional to the sample size. We expect the proportion immune to increase with age because of an increase in exposure time with age. We can see from the figure that rubella reaches high seropositive rates from early ages, which is as expected since rubella is a childhood infection.

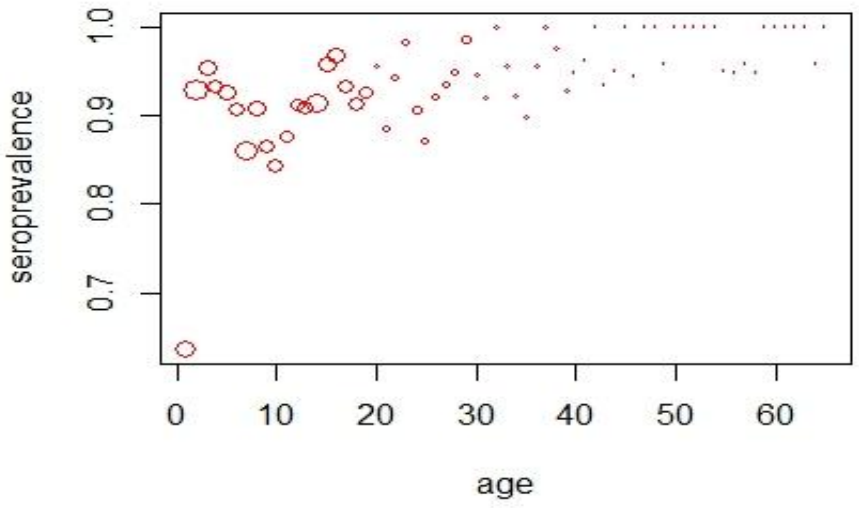

Figure 1: Serological rubella profile in Belgium anno 2006

3. Methods

3.1 Primary Vaccine Failure – Meta analysis

Primary vaccine failure occurs when an organism's immune system does not produce enough antibodies when first vaccinated. It is considered to be of very much importance in vaccination campaigns in order to explain the observed susceptibility levels. In the case of rubella, seroconversion rates are required to adjust available vaccination coverage information. In order to find an estimate for the seroconversion rate, we performed a literature review and combined available estimates using a meta-analysis model.

An extensive literature search in Pub Med and ISI Web of Knowledge resulted in 573 articles including duplicates (378 and 195 respectively). The keywords used in the search are “rubella" or “MMR" in combination with “immunogenicity", “seroconversion" or “primary vaccine failure". In total, 406 articles are considered for further investigation after removing duplicate studies. These articles were screened by title and abstract and selected if they investigated rubella seroconversion in healthy individuals. Currently, the trivalent MMR vaccine in Belgium contains, the Wistar RA 27/3 strain for the rubella virus, and therefore articles investigating properties of other strains were excluded. In total, we retain 23 eligible articles for the estimation of the seroconversion rate. In Figure 2, a flow chart with respect to the literature search on primary vaccine failure is presented. Table A.1 in Appendix A, shows details on the study designs for the different eligible articles.

For those 23 eligible articles, the seroconversion rates with 95% Clopper-Pearson confidence limits were estimated, and results are presented in the next session. The Clopper-Pearson interval is an early and very common method for calculating binomial confidence intervals. It is an exact interval since it is based directly on the binomial distribution rather than any approximation to the binomial distribution (10). These estimated rates are combined using a meta-analysis random effects model with the DerSimonian-Laird estimator for the between-study variability τ^2, the Freeman-Tukey double arcsine variance-stabilizing transformation and inverse-variance weighting. Random effects meta-analysis assumes that the effect differs from study to study and provides an estimate of the average effect. Interpretation of random effects meta-analysis is aided by a prediction interval, which provides a predicted range for the true effect in an individual study. This is simply the weighted average of the effect sizes of a group of studies. In addition, we

constructed confidence intervals for the combined effect in order to account for uncertainty with respect to τ^2 (11-13).

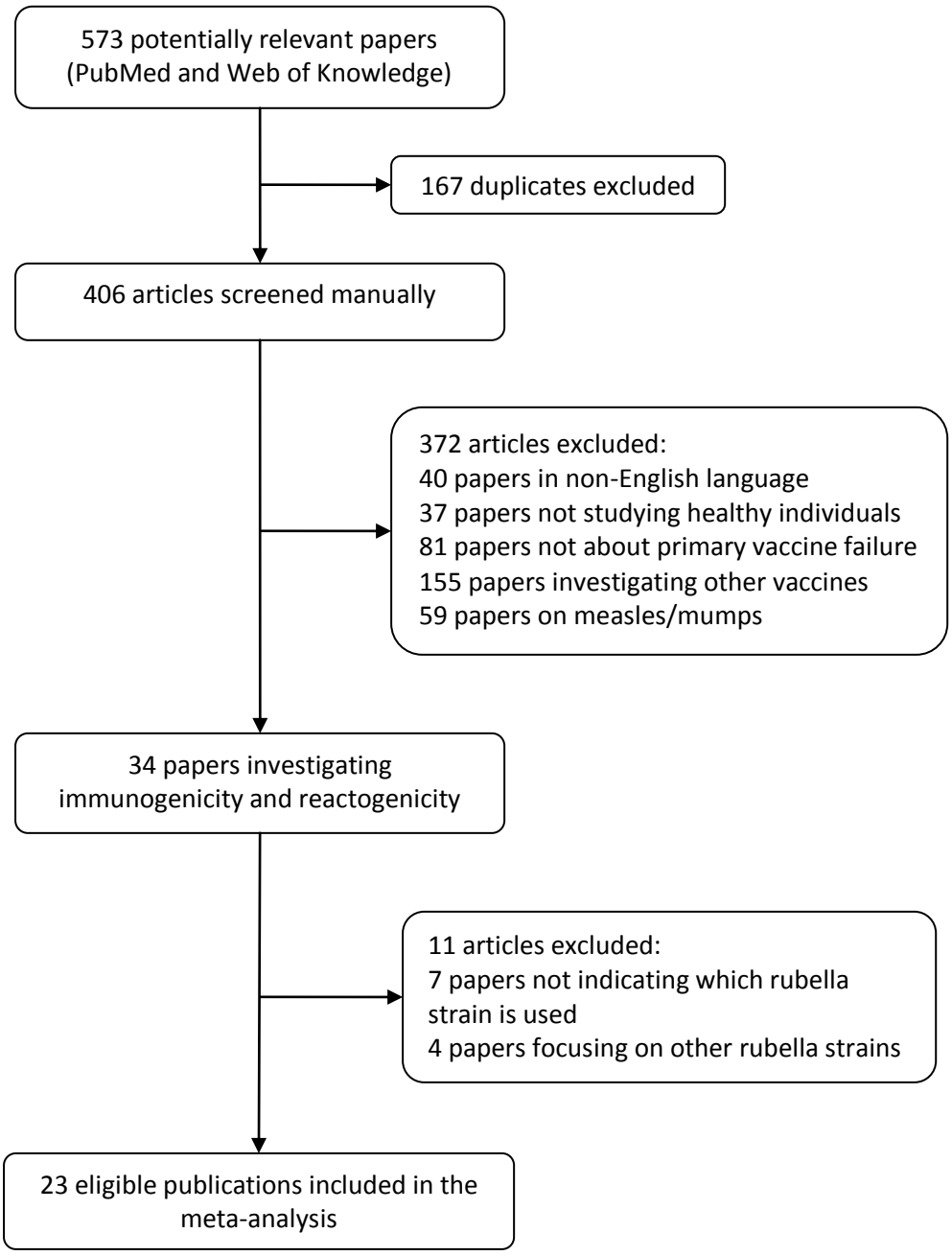

Figure 2: Flow diagram of the articles reviewed to estimate the seroconversion rate

3.2 Waning of rubella antibodies – Meta analysis

Secondary vaccine failure (waning) occurs when enough antibodies are produced immediately after the vaccination, but the levels fall with time since vaccination. While antibody levels always fall over time, this would be a more rapid loss of immunity than expected for each vaccine. Waning of antibodies has also an important effect on the observed rubella susceptibility in the population. In order to find an estimate for the waning of rubella-specific lgG antibodies after the vaccination a meta-analysis was performed.

An extensive literature search was performed in standard databases (Pub Med and ISI Web of Knowledge) for articles on rubella persistence after vaccination with the trivalent MMR vaccine. The keywords “rubella” and either “persistence”, “waning” or “immunization” were used. The search resulted in 225 articles including duplicates (116 and 109 respectively). Titles and abstracts were screened and articles were included for review whenever they met the following criteria. The vaccine administered to healthy human subjects is a trivalent MMR vaccine with the Wistar RA 27/3 strain. Furthermore, we implicitly retain papers for review in which the decay of the proportion of seropositive individuals with time since vaccination can be estimated based on a sufficient sample size ($n \geq 50$). After screening the articles base on the above criteria, 5 articles were eligible to be used in the estimation of the waning rate. In Figure 3, a flow chart with respect to the literature search on waning of vaccine-induced immunity is graphically depicted.

For those 5 eligible articles, the waning rates with 95% Clopper-Pearson confidence limits were estimated. These rates are combined using a meta-analysis fixed effects model, in order to avoid influential results from extreme values, as explained below in the results section. The fixed effect model provides a weighted average of a series of study estimates. The inverse of the estimates' variance is commonly used as study weight, such that larger studies tend to contribute more than smaller studies to the weighted average. In addition, we constructed confidence intervals for the combined effect in order to account for uncertainty in τ^2 (11-13).

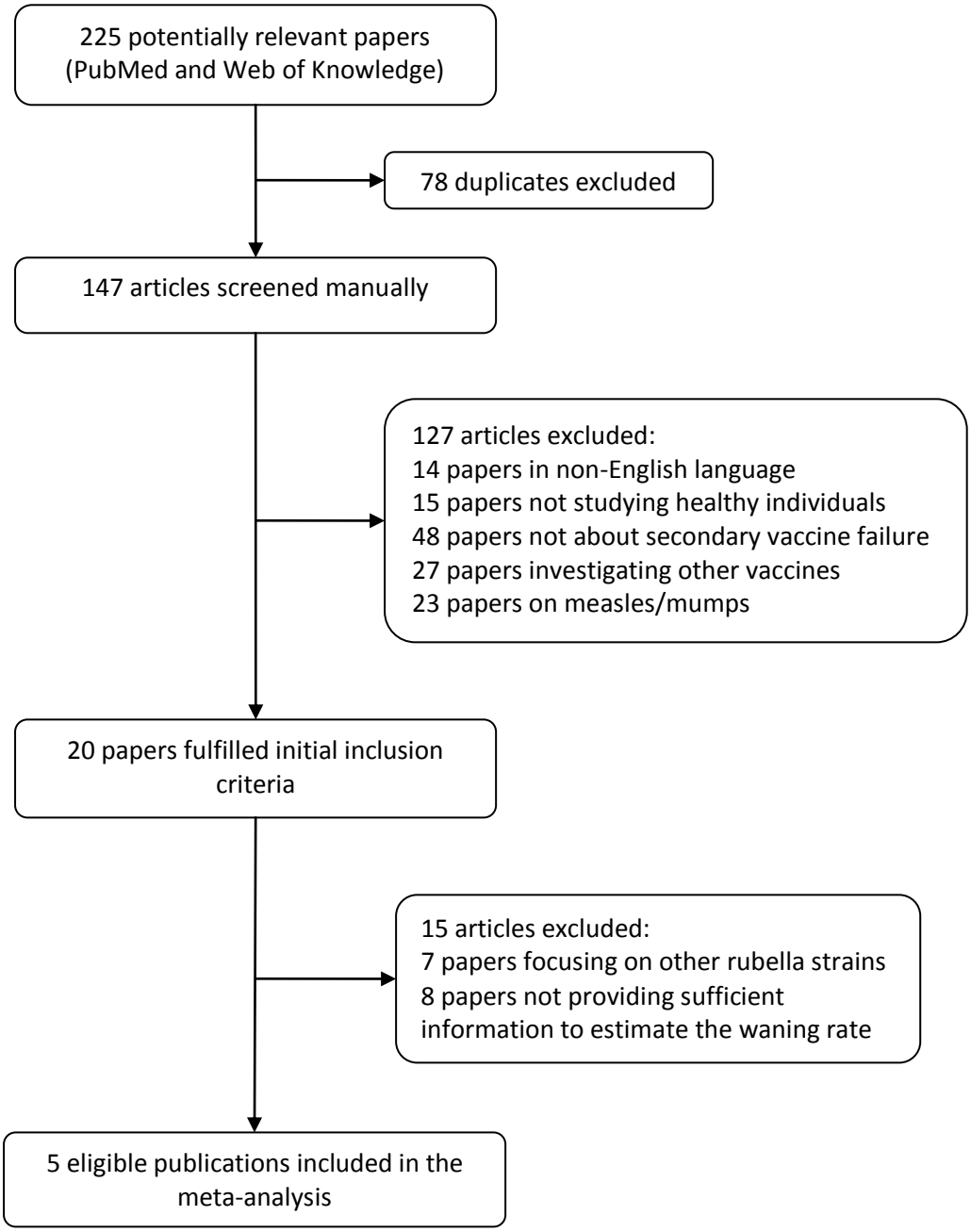

Figure 3: Flow diagram of the articles reviewed to estimate the exponential waning rate

3.3 Seroepidemiology of Rubella – Generalized Additive Model (GAM)

A generalized additive model (Hastie and Tibshirani, 1986, 1990) is a generalized linear model with a linear predictor involving a sum of smooth functions of covariates. The model allows for a rather flexible specification of the dependence of the response on the covariates, but by specifying the model only in terms of 'smooth functions', rather than detailed parametric relationships, it is possible to avoid some sort of cumbersome and unwieldy models. The linear predictor predicts some known smooth monotonic function of the expected value of the response, and the response may follow any exponential family distribution, or simply have a known mean variance relationship, permitting the use of a quasi-likelihood approach (14). In our analysis, in order to estimate the seroprevalence of rubella, a generalized additive model (GAM) with complementary log-log link is considered to model the observed seroprevalence as a function of the individual's age a, gender g and the spatial location (x, y) :

$$cloglog\big(\pi(a, g, x, y)\big) = f(a, g, x, y) \qquad (1)$$

where f is a smooth function of age a, gender g and spatial location (x, y)

and $\pi(a, g, x, y)$ is the proportion of seropositives of age a with spatial coordinates (x, y) and gender g

Submodels of this generalized additive model were fitted and the best fitting model was selected based on the Akaike Information Criterion (AIC,15).

Since the second dose of MMR vaccine is administered at the age of 12 years old, in our analysis, based on the serology from 2006, we take the restriction that the subjects are aged at least 13 years old in the year 2006. In this way we avoid influenced predictions by subjects for which the second dose of the MMR vaccine has not been administered at that time of the sample collection. Hence predictions of the proportion of susceptible individuals in 2013 are available only for those individuals aged 20 years and above. For younger age groups the susceptibility was deduced from available vaccination coverage information, i.e. approximately 90% to 95% coverage for the first dose and 70% to 85% for the second dose. Large differences in reported coverage estimates are observed between Flanders and Wallonia.

Since the generalized additive model yields estimates for the age-dependent proportion of susceptible individuals in 2006, we need to multiply these estimates with a factor in order to obtain susceptibility predictions for 2013. In our analysis, an exponential decay function $\exp(-\gamma * t)$, where γ is

the exponential decay rate and t represents the difference between the current calendar time and the time of data collection, i.e. 2006-2013.

3.4 Estimation of the Effective Reproduction Number R

Whether a vaccination program is achieving elimination at a particular time after the introduction of mass vaccination is determined by the effective reproduction number of the infection. The effective reproduction number denoted by R, is the average number of infectious individuals resulting from a single infectious individual introduced into a population not necessarily completely susceptible to infection, given the population mix of vaccine-acquired and naturally acquired immunity at that time (16). If $R < 1$, then, while infections still occur, for example by limited spread from imported cases, they cannot result in large epidemics. If the value of R is greater than 1, or below 1 but on the increase, additional control measures may be called for. Such calculations led to the measles and rubella mass vaccination campaign in 1994 in the UK (17). The effective reproduction number is closely related to the basic reproduction number R_0. Multiplying the equation (2) given below, with the proportion of susceptible individuals of age a at calendar time t, say $S(a,t)$ and taking the maximum eigenvalue of the resulting matrix yields the estimated effective reproduction number. Once we estimate the effective reproduction number R in each of the Belgian municipalities, we are able to express local information about the potential of epidemics to occur.

Basic Reproduction Number R_0

The basic reproduction number R_0 is the number of secondary cases produced by a single infectious person in a completely susceptible population. The basic reproduction number is defined as the leading eigenvalue of the next generation operator defined by the next generation matrix (18):

$$DN(a,t)\beta(a,a',t) \qquad (2)$$

where D is the mean duration of infectiousness

$N(a,t)$ denotes the number of individuals of age a in the population at calendar time t

and $\beta(a,a',t)$ represents the time heterogeneous transmission rates, i.e. the per capita rate at which an infectious individual of age a' makes an effective contact with a susceptible individual of age a

Estimates for the basic reproduction number for rubella are numerous in the literature. These estimates vary between countries and continents. Higher estimates are reported in Africa. In Europe R_0 varies from 6 to 12 (19). In our analysis we selected a basic reproduction number equal to 8 which is the average of the available estimates. Furthermore, a sensitivity analysis with respect to the R_0 is included.

Population Age Distribution $N(a,t)$

The population age distribution $N(a,t)$ can be estimated from demographic data using the following equation:

$$N(a,t) = \frac{N(t)}{L(t)} \exp\left(-\int_0^a \mu(u,t)du\right)$$

where $\mu(a,t)$ denotes the time-dependent natural mortality rates at age a,

$N(t)$ represents the total population size,

and $L(t)$ equals the life expectancy in the population at time t.

In the analyses, the population size equals N = 11,035,948 and the life expectancy L is estimated to be approximately 79 years. Data to calculate the population size and the life expectancy were available for the year 2012. We are considering those as proxies for those in 2013. The estimation of the mortality rates $\mu(a,t)$ relies on demographical data with respect to the number of deaths and the population size per age category from EUROSTAT. Furthermore, a generalized additive model with log link is applied to model the relationship between natural death and age:

$$\log(d_j) = \log(n_j) + s(a_j)$$

where j are age intervals of length one,

d is the number of deaths,

and n the population size for each interval.

In Figure B.1 in the Appendix B, the estimated survival function (solid line) and mortality rates (dashed line) are graphically depicted together with the observed death rate in Belgium anno 2010.

Transmission Rate $\beta(a,a',t)$

Using empirical social contact data has led to an improved estimation of the transmission rate. There are many authors using different approaches

to estimate these contact rates (20-22). In our case, the social contact hypothesis can be formulated in:

$$\beta(a, a', t) = q(a, a', t|c)c(a, a')$$

where $q(a, a', t|c)$ is an age- and time-dependent proportionality factor related to susceptibility and infectivity of individuals

and $c(a, a')$ are annual per capita contact rates between individuals of age a and a'

In our case q is restricted to be age and time-invariant. The social contact rates $c(a, a')$ are estimated from empirical data (22,23). Given the basic reproduction number R_0, the corresponding constant proportionality factor q in 2013 is estimated.

4. Results

4.1 Primary Vaccine Failure

Following the literature research for the primary vaccine failure regarding the rubella component of the trivalent MMR vaccine in healthy individuals, estimates for the seroconversion rates for each of the 23 eligible studies are presented in Table 1.

Source	$\hat{\rho}$	95% CI	
Samoilovich et al. (24)	0.959	0.932	0.978
Gatchalian et al. (25)	1.000	0.975	1.000
Bhargava et al. (26)	0.988	0.939	0.999
Mitchell et al. (27)	0.935	0.876	0.971
Forleo-Neto et al. (28)	0.990	0.947	0.999
Dos Santos et al. (29)	0.913	0.867	0.947
Lee et al. (30)	1.000	0.981	1.000
Khalil et al. (31)	1.000	0.915	1.000
Usonis et al. (32)	1.000	0.984	1.000
Vesikari et al. (33)	1.000	0.979	1.000
Robertson et al. (34)	0.990	0.966	0.998
Christenson et al. (35)	0.992	0.957	0.999
Lee et al. (36)	1.000	0.960	1.000
Lim et al. (37)	1.000	0.968	1.000
Nolan et al. (38)	1.000	0.950	1.000
Stuck et al. (39)	0.993	0.964	0.999
Klinge et al. (40)	0.974	0.927	0.994
Crovari et al. (41)	1.000	0.994	1.000
Tischer et al. (42)	0.981	0.963	0.991
Schwarzer et al. (43)	0.987	0.968	0.996
Rager-Zisman et al. (44)	0.933	0.851	0.978
Redd et al. (45)	0.943	0.927	0.957
Bottiger et al. (46)	0.997	0.987	0.999

Table 1. Estimated Seroconversion rates and Associated 95% Clopper-Pearson Confidence Intervals based on different studies with respect to the rubella component in a MMR vaccine.

These rates are combined using a meta-analysis random effects model with the DerSimonian-Laird estimator for the between-study variability τ^2. The estimated combined seroconversion rate equals to 0.984 (approximate 95% CI: 0.974, 0.9992). The seroconversion rate is assumed identical after the first and second MMR dose.

4.2 Waning of rubella antibodies

Results from the literature research for the secondary vaccine failure regarding the rubella component of the trivalent MMR vaccine in healthy individuals, are shown in Table 2. It is a list of the five studies retrieved by our search, showing the estimated exponential waning rates $\hat{\gamma}$, 95% confidence limits and corresponding vaccination information.

Source	$\hat{\gamma}$	95% CI		Number of doses
Davidkinn et al.(47)	0.000	0.000	0.000	2
Miller et al.(48)	0.001	0.000	0.003	1
Boulianne et al.(49)	0.006	0.002	0.010	1
Le Baron et al.(50)	0.016	0.013	0.020	2
	0.039	0.030	0.049	2
Poethko Muller et al.(51)	0.017	0.014	0.020	1
	0.008	0.006	0.010	2

Table 2. Estimated exponential waning rates based on different studies with respect to the rubella component in MMR vaccine.

A random effects meta-analysis model was first used to combine those estimates. The results lead to very wide confidence intervals due to influential extreme values such as the ones by Davidkin and LeBaron, which later on will a have big impact on the susceptibility profiles. Hence we decided to proceed with a fixed effects model for which the confidence intervals are smaller. The fixed effects model assumes that all included studies investigate the same population, use the same variable and outcome definitions, etc. This assumption is typically unrealistic as research is often prone to several sources of heterogeneity, but for the sake of our analysis we assume that all the studies are conducted in the same way. Based on a meta-analysis fixed effects model in which the rates are log transformed, the overall exponential waning rates after back-transformation are equal to 0.015 (approximate 95% CI: 0.012, 0.018) and 0.016 (approximate 95% CI: 0.013, 0.018) after the first and second MMR dose, respectively. Although evidence for waning of passively acquired immunity exists, rubella infections are generally accepted to induce lifelong immunity (52). Therefore we will assume that naturally acquired immunity is preserved for life, which is a conservative approach.

4.3 Susceptibility profiles for each municipality

Before estimating the local effective reproduction numbers, first we need to estimate the susceptibility profiles for each of the Belgian municipalities. These profiles will be constructed from available regional vaccination coverage data and information gained from the serology. As mentioned above in the methods section, we need a model that fits our data well in order to estimate the seroprevalence of rubella from the year 2006 accurately. Hence several submodels derived from equation (1) are fitted to the serological data from 2006. The model fit results are presented in Table 3.

Model	Linear Predictor	AIC
1	te(x,y,a)	932.8869
2	s(a) + te(x,y)	935.0413
3	s(x,y,a)	942.6044
4	te(x,y)	943.3454
5	te(x,y) + te(a)	935.0250
6	te(x,y,by=g) + te(x,y,by=1-g)	920.1460
7	**te(x,y,a,by=g) + te(x,y,a,by=1-g)**	**899.9828**
8	s(x,y,a,by=g) + s(x,y,a,by=1-g)	907.4935
9	te(a,by=g) + te(a,by=1-g)	930.2305

Table 3. Generalized Additive Models fitted to the Belgian seroprevalence data on Rubella infection with corresponding AIC-values.

Comparing the AIC values corresponding to the above models, model (7) has the lowest one in which age, gender and spatial location are found to be important in explaining the observed rubella seroprevalence. Henceforth model (7) is used it in our analysis and so the final generalized additive model is given by:

$$cloglog\big(\pi(a,x,y,g)\big) = \log\big(-\log\big(1-\pi(a,x,y,g)\big)\big)$$

$$= te(a,x,y,by=g) + te(a,x,y,by=1-g) \qquad (3)$$

where $\pi(a,x,y,g)$ is the proportion of seropositives of age α with spatial coordinates (x,y) and gender g,

and $te(.)$ represents tensor product thin-plate regression splines for which the basis is built up from tensor products of one-dimensional thin-plate regression splines.

Using the equation (3) we can predict the proportion of the susceptible individuals above 20 years of age. A parametric bootstrap approach is used to construct 1000 bootstrap samples, and the selected GAM model is fitted to

each of the generated samples. For individuals with age below 20 years, a similar procedure was performed only this time seroconversion rates and waning rates were added which were randomly sampled from a normal distribution with 95% confidence intervals equal to those reported before. Furthermore, for each bootstrap sample and municipality vaccination coverage estimates are randomly sampled from a normal distribution with 95% confidence intervals from available information. In order to end up with a smooth susceptibility curve for each municipality, an interpolating-spline model is fitted to each of the generated datasets. Note that our selected model includes a gender component as well. Hence susceptible profiles were calculated for males and females separately and these profiles were averaged to end up with the final age- and location-specific susceptible profile. Susceptibility curves for males and females are presented in Figure C.1 in Appendix C, where both curves follow the similar pattern with a slightly bigger susceptibility for males in some age groups. Since our concern is the averaged susceptibility details follow only the averaged curves.

In Figure 4 we illustrate the estimated susceptibility curves for three chosen localities, Hasselt, Liège and Brussels. The graphs show (1) estimated based the available vaccination coverage information and the seroconversion and waning rates of the vaccine induced antibodies, (2) estimated based on the serology and the waning of antibodies and (3) estimated based on the serology and the assumed lifelong natural immunity since there was no vaccination done. As expected the figure shows first an increase in susceptibility after the administration of the first dose of the vaccine caused by waning of vaccine-induced antibodies. At the time of the administration of the second dose of the vaccine, at the age of 12, we can clearly see a sharp decrease in the susceptibility, more noticeable in Hasselt and Liege. Waning of the antibodies from the second dose causes again an increase in susceptibility, to a lesser extent, and afterwards the profile remains low in the age group above 28 years of age since they are protected for life against rubella infection.

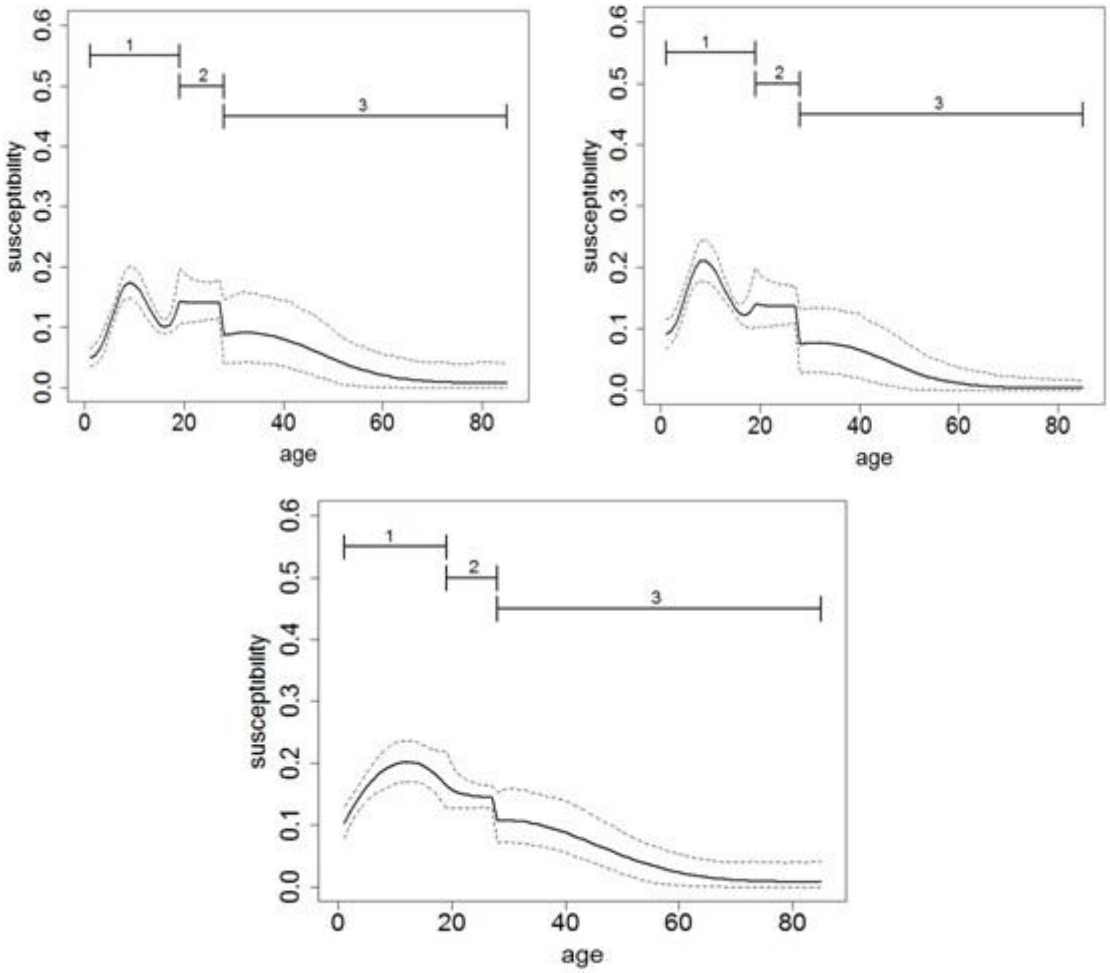

Figure 4. The estimated susceptibility in the urban areas of Hasselt (left upper panel), Liège (right upper panel) and Brussels (lower panel) in 2013 with 95% confidence limits in dashed lines.

4.4 Effective Reproduction Number R

As described before, once we have estimated the effective reproduction number R in each of the Belgian municipalities, we are able to express whether epidemics are likely to occur or not. Selecting the basic reproduction number R_0 to be equal to 8 and assuming based on literature (19) that the average duration of infectiousness for rubella is 7 days per year, one can estimate the corresponding constant proportionality factor q for 2013 to be $q = 0.0623$. Based on the estimated age- gender- and location-specific susceptibility in 2013, i.e. the proportion of susceptible individuals of age a at calendar time t, represented by $S(a,t)$ for each age class and the estimated constant proportionality factor q, one is able to estimate the effective

reproduction numbers R simply by taking the maximum eigenvalue of the resulting matrix.

Province	R (R_0=8)	CI	
Limburg	0.800	0.543	1.227
Antwerp	0.830	0.581	1.229
Flemish Brabant	0.849	0.643	1.135
West Flanders	0.696	0.511	1.042
East Flanders	0.781	0.582	1.074
Walloon Brabant	0.943	0.722	1.249
Hainaut	1.082	0.744	1.567
Liege	0.842	0.599	1.266
Namur	0.951	0.654	1.390
Luxembourg	0.928	0.592	1.514
Brussels	1.038	0.823	1.329

Table 4: Estimated effective reproduction numbers R for the provinces of Belgium for the year 2013

Results for the calculated effective reproduction numbers for each of the provinces of Belgium for the year 2013 are presented on Table 4. As mentioned before, if $R < 1$, then infections cannot result in large epidemics. If $R > 1$, additional control measures may be called for. In our case the effective reproduction numbers for all the provinces are well below the epidemic threshold of one. There are only 2 cases, namely the estimates for Hainaut and Brussels for which the estimated number are equal to 1 but that does not make any further concerns. This implies that further control measures are not required, at least for rubella. In general, the effective reproduction numbers tend to be larger in the Walloon region as compared to the other Belgian regions. That is mainly driven by vaccination coverage estimates that are substantially lower in Wallonia compared to those reported in Flanders. A graphical representation of the local effective reproduction number for each municipality on a spatial map and boxplots of the reproduction numbers in the regions of Belgium and are given in Figure 5 and 6, respectively. Both figures fit the description made previously. We clearly see a difference between the Walloon and Flemish region with respect to the estimated reproduction numbers.

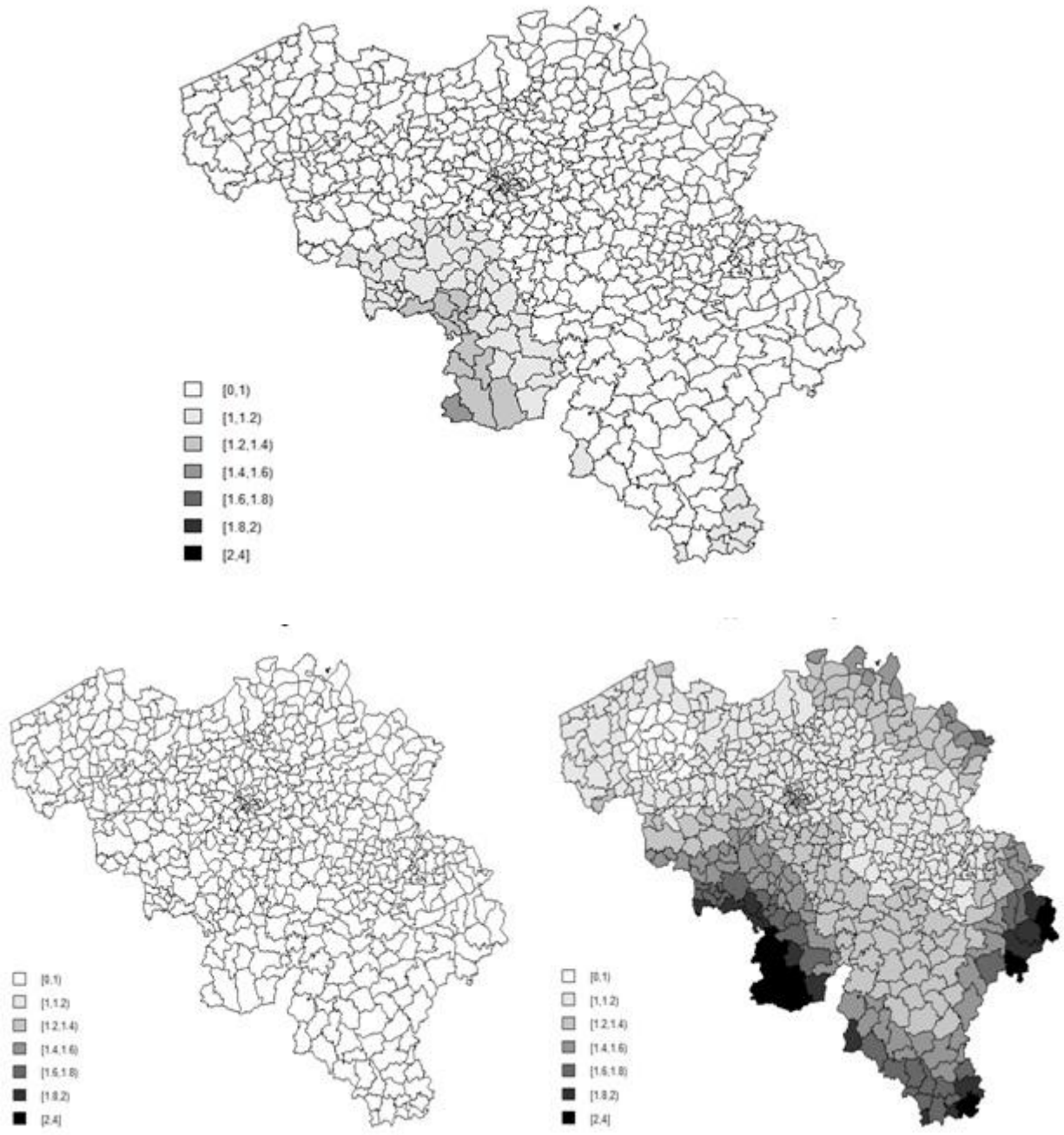

Figure 5. The estimated effective reproduction numbers in the Belgian municipalities on a spatial map (upper panel) with 95% confidence limits (bottom panels).

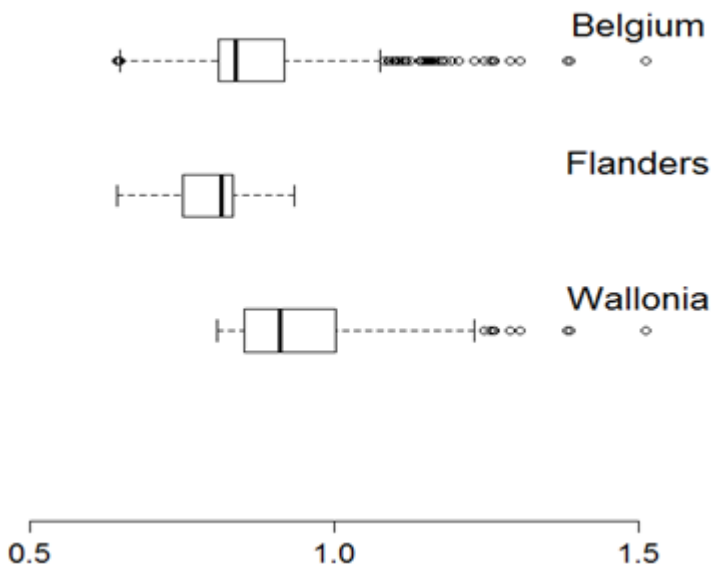

Figure 6: Boxplots of the estimated reproduction numbers for Belgium, Flanders and Wallonia for the year 2013.

4.5 Sensitivity Analysis with respect to R_0

Lastly a sensitivity analysis is conducted with respect to R_0. In our case we would like to determine how the estimates of the reproduction number differ when we change the basic reproduction number.

As mentioned previously, in the literature there are reported estimates for R_0 ranging from 6 to 12. Hence we conduct the same analysis as before using those two values for the basic reproduction number. The estimated effective reproduction numbers are compared to those reported in the previous section. The results are presented in Table 5. A graphical representation and boxplots are presented in Figure 7 and 8, respectively. Relying on $R_0 = 8$ the estimated effective reproduction numbers are not significantly larger than the threshold value one. As expected, using a more conservative approach of R_0 equal to 6, values of the effective reproduction number indicate a very small risk of future rubella epidemics. On the other hand, taking R_0 equal to 12 forces the estimated reproduction numbers above the epidemiological threshold. Almost all Belgian municipalities have an R value larger than one, implying that infections have the potential to spread and so intervention strategies should be conducted to reduce outbreak risk, for example vaccination programs. Of course this risk exists only when taking an extreme value of the basic reproduction number. Also we should keep in

mind that having effective reproduction numbers slightly above one, which is our case also at the extreme scenario, then potential outbreaks tend to extinct rapidly.

Province	R (R_0=6)	*R (R_0=8)*	*R (R_0=12)*
Limburg	0.600	*0.800*	1.201
Antwerp	0.623	*0.830*	1.245
Flemish Brabant	0.637	*0.849*	1.274
West Flanders	0.522	*0.696*	1.044
East Flanders	0.586	*0.781*	1.172
Walloon Brabant	0.707	*0.943*	1.415
Hainaut	0.811	*1.082*	1.622
Liege	0.631	*0.842*	1.263
Namur	0.713	*0.951*	1.427
Luxembourg	0.696	*0.928*	1.391
Brussels	0.778	*1.038*	1.557

Table 5: Estimated effective Reproduction Numbers R for the different values of the R_0 used.

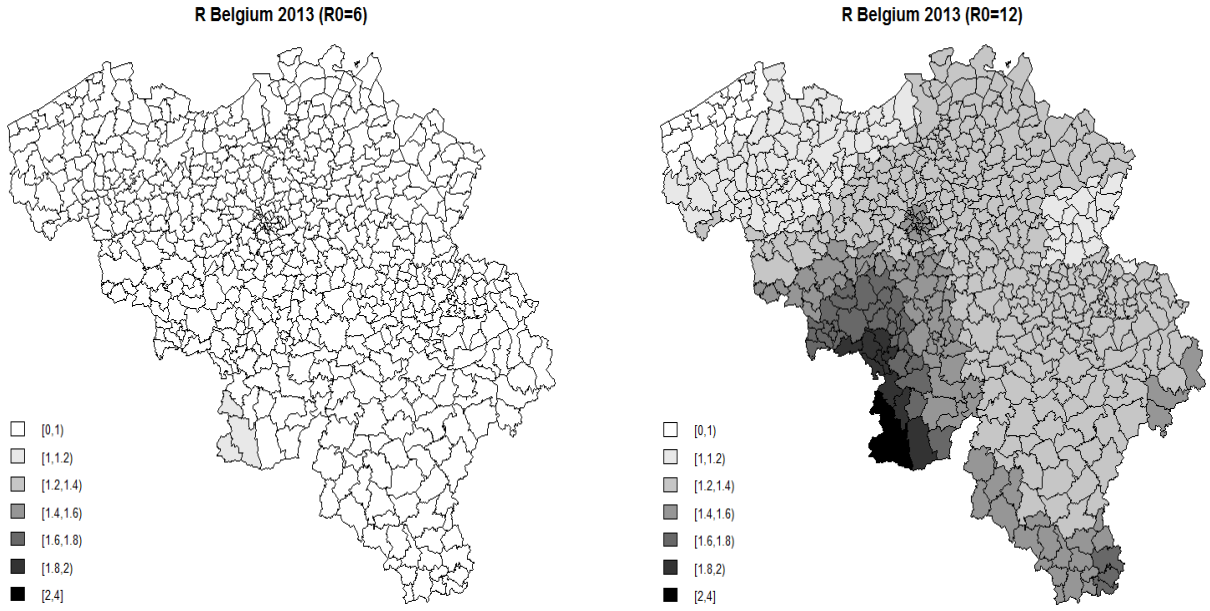

Figure 7. The estimated effective reproduction numbers in the Belgian municipalities on a spatial map for the different values of the basic reproduction number R_0(left panel R_0=6, right panel R_0=12).

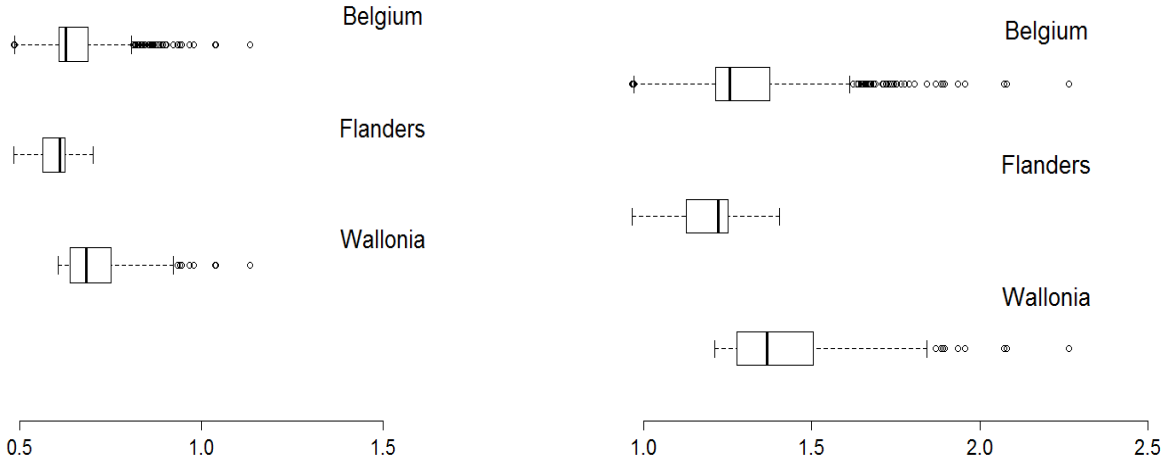

Figure 8. Boxplots for the estimated effective reproduction numbers in the Belgian municipalities for the different values of the basic reproduction number R_0 (left panel R_0=6, right panel R_0=12).

5. Conclusion and Recommendations

Measles, mumps and rubella related morbidities have declined considerably since the introduction of an effective and safe MMR vaccine. However, despite the fact that for almost three decades there have been vaccination programs employing live attenuated strains of measles, mumps and rubella viruses in trivalent MMR vaccines, periodic outbreaks of these diseases still occur. Sure thing is that for the rubella component outbreaks are fewer and more attenuated from the other two. This report presents a simple method to identify regions of high outbreak potential, informed by serological survey data and vaccination coverage information. Outbreak potential is quantified in terms of local estimates for the effective reproduction number R. In our case effective reproduction numbers for all the provinces are well below the epidemic threshold. It is noted in the report that naturally acquired immunity to rubella infection is believed to be lifelong. However, any deviation from this assumption leads to an even larger part of the population unprotected against new infections. This increases the estimated effective reproduction numbers and consequently enlarges the efforts required to prevent future outbreaks.

Susceptibility profiles in 2013, were estimated using a generalized additive model including age, gender and spatial location as covariates. Furthermore, BIC values were also calculated, apart from the AIC values. We do not report them and we depend our decision of the model choice only on the AIC values, because the BIC values indicate a model that does not include the spatial component. In our case we do need the spatial component because our coverage information is region-specific. We keep in mind though that the spatial heterogeneity shouldn't be over-interpreted.

Potential outbreaks are likely to result from a decline in protection with time since rubella vaccination. In addition to primary vaccine failure, waning of vaccine-induced immunity might be responsible for an increase in susceptibility. Seroconvertion and waning rates of the rubella antibodies play an important role in the estimation of susceptibility. Therefore, waning of vaccine-induced immunity is included in our analysis through the specification of an exponential decay model. The exponential waning rates after the first and second MMR dose are estimated from a literature review using a fixed-effects meta-analysis model. The susceptibility as it was expected, firstly shows an increase after the administration of the first dose of the vaccine caused by waning of vaccine-induced antibodies. At the time of the administration of the second dose of the vaccine there is a sharp decrease in

the susceptibility. Again waning of the antibodies from the second dose cause an increase in the susceptibility, and afterwards the profile remains low since they individuals are assumed to be protected for life against rubella infection. Information on vaccination coverage in different Belgian regions is disrupted, leading to uncertainty about the susceptibility in young age groups. Nevertheless, differences in susceptibility between regions are mainly driven by differences in vaccination coverages.

Estimation of the effective reproduction depends a lot on the choice of the basic reproduction number as seen in the sensitivity analysis, hence one should remain cautious about over-interpreting these results. Many publications give estimates for the basic reproduction number for rubella. Those estimates vary between countries and continents. In our analysis we consider the basic reproduction number to be equal to 8 which leads to a rather limited risk of future rubella outbreaks (19). However, having estimates for the reproduction numbers below one does not imply that we should be reassured. Currently the risk for rubella outbreaks is small. In the future outbreaks could occur as a result of a build-up of susceptibility in the population due to a decrease of individuals protected by natural acquired immunity, and an increase of individuals with waning immunity. Preventing rubella outbreaks, even when the risk is low, most likely requires various ingredients, in addition to a routine high-coverage two-dose vaccination program. Targeting specific age groups in specific localities where vaccination coverage is lower could be an efficient way of reducing the risk of new outbreaks. In order to be more sure about our predictions, a new analysis for Belgium would therefore require a new serological study. This means that surveillance should be a high priority for authorities. As it is proposed for the World Health Organization (WHO) in 2010, detection of specific antibodies (IgM) is not specific enough to confirm rubella infection. If a diagnosis is crucial, for example for a pregnant woman, an IgM+ result is inappropriate to confirm the diagnosis. A rubella infection must be confirmed by a molecular diagnosis test or by an avidity test (IgG). When elimination has been achieved, a laboratory diagnosis by means other than a serological test is required for all suspected cases.

Lastly it would be of a high interest to perform the analysis including more covariates such as immigrant rates, income, educational level, seasoning effects etc. and see how the susceptibility profiles differ and how prevention strategies should be redrawn.

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Appendix A

Source	n	Age	Vaccine	Time	Test	Sero+
Samoilovich et al. (24)	324	12-24m	Trimovax	2-2,5m	ELISA	>=1:100
Gatchalian et al. (25)	149	12-24m	Priorix/MMR-II	40-63d	ELISA	4 IU/ml
Bhargava et al. (26)	89	18-24m	MMR(RA 27/3)	4w	ELISA	_
Mitchell et al. (27)	124	12-24m	MMR-II	1m	ELISA	>=1.00
Forleo-Neto et al. (28)	103	9m	Trimovax	6w	HIG	>=1/10
Dos Santos et al. (29)	219	6-12y	MMR-II	21-30d	ELISA	_
Lee et al. (30)	202	12-18m	Priorix/MMR-II	40-63d	ELISA	4 IU/ml
Khalil et al. (31)	42	12m	MMR-II	8w	ELISA	>7 IU/ml
Usonis et al. (32)	228	12-24m	Priorix/MMR-II	60d	ELISA	4 IU/ml
Vesikari et al. (33)	174	14-24m	MMR(RA 27/3)	_	_	_
Robertson et al. (34)	212	13m	MMR(RA 27/3)	42d	ELISA/HA	_
Christenson et al. (35)	129	18m	MMR-II	2m	HIG	_
Lee et al. (36)	91	12-23m	Priorix/MMR-II	5-8w	ELISA	4 IU/ml
Lim et al. (37)	115	12-18m	Priorix	42d	ELISA	4 IU/ml
Nolan et al. (38)	72	12m	Priorix	60d	ELISA	4 IU/ml
Stuck et al. (39)	154	12-24m	Priorix/MMR-II/Trivarent	60-70d	ELISA	2 IU/ml
Klinge et al. (40)	118	9-17m	MMR-Vax	4-6w	ELISA	>= 10 U/ml
Crovari et al. (41)	677	12-27m	Priorix/Trivalent	60d	ELISA	4 IU/ml
Tischer et al. (42)	427	15m/2y/8y/12y/20y	MMR-Vax/Pluserix/Trivivarent	49d/4-6w	ELISA	factor of >= 2
Schwarzer et al. (43)	320	14-24m	Priorix/MMR-II	6w	HI/ELISA	>= 8
Rager-Zisman et al. (44)	75	12m	MMR-II	30d	ELISA	_
Redd et al. (45)	957	9/12/15m	MMR-II	24m	ELISA	_
Bottiger et al. (46)	441	18m/12y	MMR-II	_	HIG	_

Table A.1. *Overview of different studies, included in the meta-analysis, with respect to primary vaccine failure of the rubella component in the trivalent MMR vaccine*

Appendix B

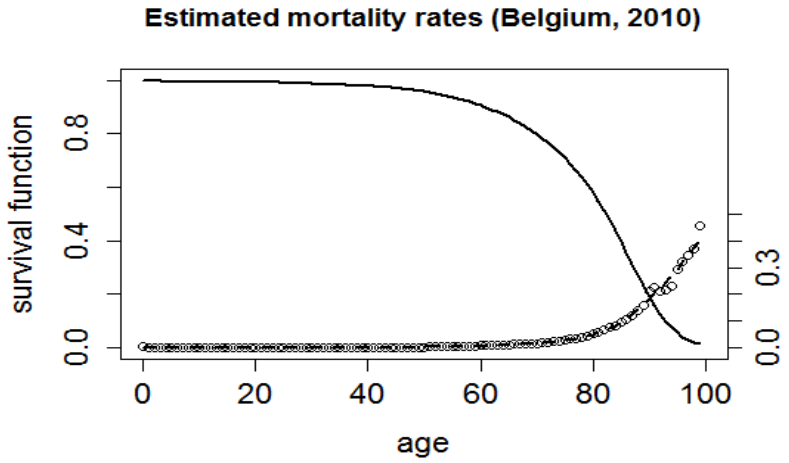

Figure B.1*: The estimated survival function (solid line) and mortality rates (dashed line) with observed death rate (dots) for Belgium year 2010.*

Appendix C

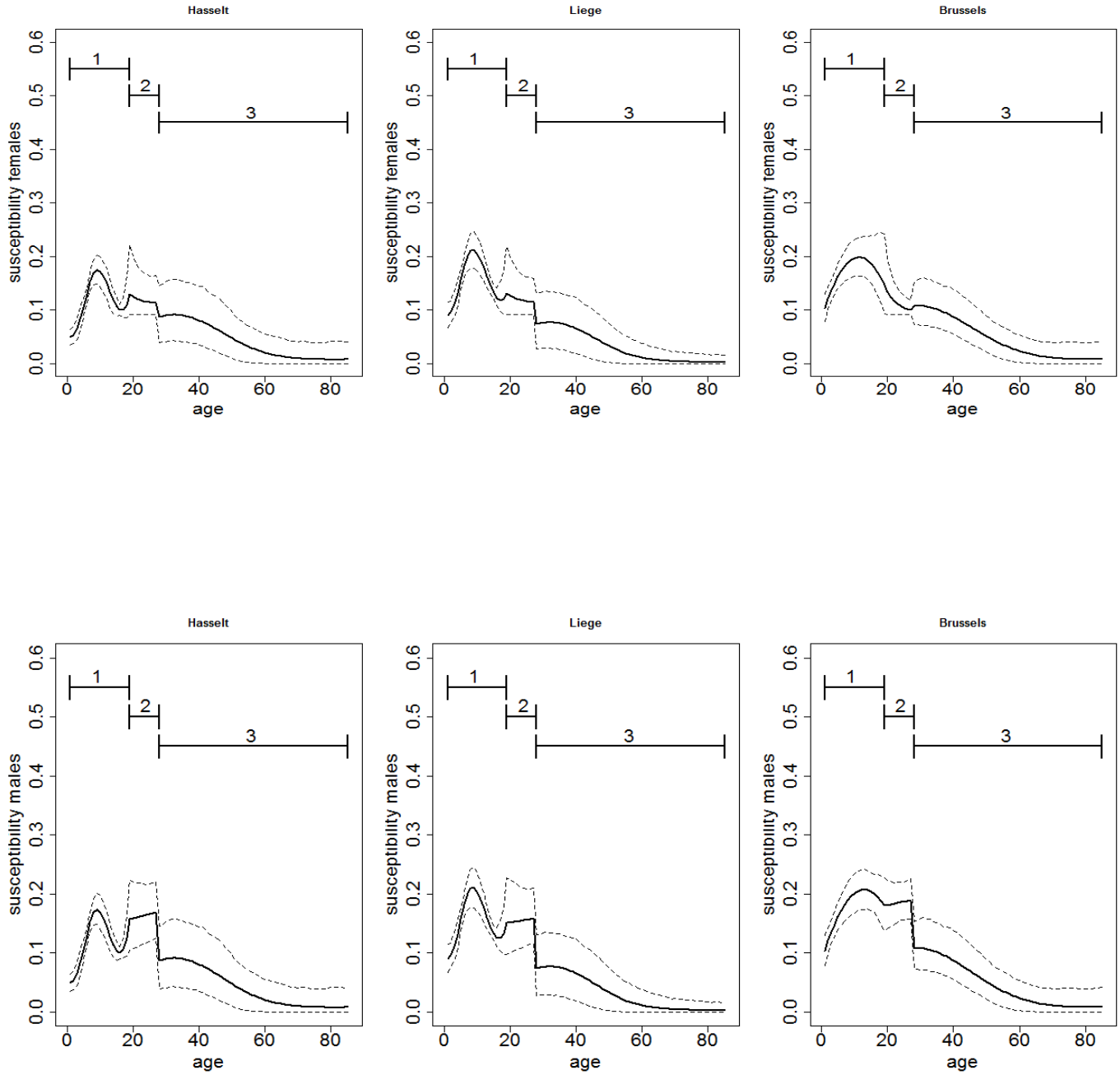

Figure C.1. The estimated susceptibility for females (upper curves) and males (lower curves) in the urban areas of Hasselt, Liege and Brussels in 2013 with 95% confidence limits in dashed lines.

Auteursrechtelijke overeenkomst

Ik/wij verlenen het wereldwijde auteursrecht voor de ingediende eindverhandeling:
Assessing outbreak risk in highly vaccinated populations using spatial seroprevalence data on rubella

Richting: **Master of Statistics-Biostatistics**
Jaar: **2014**

in alle mogelijke mediaformaten, - bestaande en in de toekomst te ontwikkelen - , aan de Universiteit Hasselt.

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Voor akkoord,

Kourkouni, Eleni

Datum: **5/02/2014**