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Review

Sex and age as determinants of the seroprevalence of anti-measles IgG among European healthcare workers: A systematic review and meta-analysis

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ABSTRACT

Introduction: The international literature shows good evidence of a significant rate of measles susceptibility among healthcare workers (HCWs). As such, they are an important public health issue.
Methods: We conducted a systematic review and meta-analysis to estimate the prevalence of susceptible HCWs in EU/EEA countries and in the UK and to explore the characteristics (sex and age differences) and management of those found to be susceptible.
Results: Nineteen studies were included in the meta-analysis. The prevalence of measles-susceptible HCWs was 13.3% (95 %CI: 10.0–17.0%). In a comparison of serosusceptible female vs. male HCWs, the RR was 0.92 (95 %CI = 0.83–1.03), and in a comparison of age classes (born after vs. before 1980) the RR was 2.78 (95 %CI = 2.20–3.50). The most recent studies proposed the mandatory vaccination of HCWs.
Discussion: According to our meta-analysis, the prevalence of serosusceptible European HCWs is 13%; HCWs born in the post-vaccination era seem to be at higher risk. Healthcare professionals susceptible to measles are a serious epidemiological concern. Greater efforts should therefore be made to identify those who have yet to be vaccinated and actively encourage their vaccination.

Contents

Abbreviations: HCW, healthcare worker; MMR, measles, mumps, rubella; CDC, Center for Diseases Control and Prevention; ECDC, European Centre for Disease Prevention and Control; RR, risk ratio.

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1. Introduction

The international literature shows good evidence of a significant rate of measles susceptibility among healthcare workers (HCWs). In a 2015 review, 6% of HCWs in Europe were found to be seronegative for measles [1], and in a 2013 study from France the estimated prevalence was 8% [2]. However, a 2013 study from Spain found a susceptibility rate among HCWs of only 2% [3], and a recent meta-analysis from Italy [4] a rate of 9–17%, depending on the evaluation method (serological test vs. self-administered questionnaire).

Measles-susceptible HCWs represent a risk both to themselves and to the patients they care for and are thus an important public health issue [5]. A 2019 paper reported 12 nosocomial cases of measles in an Italian hospital, of which 5 involved unvaccinated HCWs, [6]. A study published in 2014, reviewing known cases of HCW-to-patient transmission of the most common vaccine-preventable infections in healthcare settings, concluded that vaccination is the primary method of protection from work-related infection risks for both HCWs and the patients in contact with them [7]. A 2020 study focused on the difficulty of HCWs in recognizing cases of these diseases, especially those with an atypical clinical picture, and concluded that these situations may contribute to nosocomial clusters [8].

The Center for Diseases Control and Prevention (CDC) recommends that HCWs have presumptive evidence of immunity to measles, defined as laboratory evidence of immunity, laboratory confirmation of disease, birth before 1957, or written documentation of vaccination with two doses of measles-containing vaccine [9]. Despite this recommendation, according to the European Centre for Disease Prevention and Control (ECDC) Measles Annual Epidemiological Report, there were 13,200 cases of measles in EU/EEA countries and the UK in 2019. While this was a decline compared to 2017 and 2018, the fact that measles cases continue to occur across the EU/EEA shows that vaccination coverage in many countries remains suboptimal, i.e., below the recommended objective of vaccination coverage of 95% of the population with two doses of measles-containing-vaccine [10]. Cases of measles among HCWs in Europe have been further documented in several other reports [6,11-14].

Strategies for the vaccination of HCWs in Europe differ from country to country. Maltezou HC et al. [15,16] found that, in 2019, only 14 European countries recommended measles immunization for all HCWs. Among the other countries, five recommended measles vaccine only for specific groups; in three, immunization against measles was mandatory for all HCWs; and in one country it was mandatory for specific groups only. Seven countries had no national measles immunization policy for HCWs (the vaccination strategy in Liechtenstein was not reported).

Against this background, we conducted a systematic review and meta-analysis to estimate the prevalence of measles-susceptible HCWs and medical school students (shown to be a potential source of measles outbreaks in hospitals [17]) in EU/EEA countries [18] and the UK. Our analysis made use of data on serological tests for circulating anti-measles IgG. We also evaluated the proposed options for the management of susceptible HCWs and the strategies aimed at improving immunization in the healthcare setting.

2. Methods

2.1. Search strategy and selection criteria

The Scopus, MEDLINE/PubMed, Google Scholar (pages 1–15) and the ISI web of knowledge databases were systematically searched. Research papers, including short reports, published between the January 1, 2015 and October 18, 2020 were included in the analysis. The following terms were used in the search strategy: (healthcare workers OR physician OR nurse OR resident OR student) AND (measles OR rubeola) AND (EU OR EEA OR United Kingdom OR Italy OR France OR Spain OR Portugal OR Austria OR Belgium OR Bulgaria OR Croatia OR Cyprus OR Czech Republic OR Denmark OR Estonia OR Finland OR Germany OR Greece OR Hungary OR Ireland OR Latvia OR Lithuania OR Luxembourg OR Malta OR Netherlands OR Poland OR Romania OR Slovakia OR Slovenia OR Sweden OR Iceland OR Norway OR Liechtenstein). Studies in all languages were included. Abstracts without full-text, letters to the editor, papers not reporting epidemiological data (editorials, commentaries, etc.), studies in which susceptibility was evaluated by surveys or those in which only vaccination coverage was reported, and all studies focused on issues not related to the aim of this review (vaccine hesitancy, vaccine knowledge, attitudes, etc.) were excluded. When necessary, the authors were contacted for further information. The list of documents was screened by title and/or abstract by two independent reviewers who applied the predefined inclusion/exclusion criteria. Discrepancies were recorded and resolved by consensus. The references of all articles were examined for additional studies. The extracted data included year, sample size, number of susceptible, professional category, country, management options for susceptible HCWs, and immunization strategies.

2.2. Quality assessment

The quality of the studies included in meta-analysis was assessed according to the STROBE checklist based on 22 methodological questions [19]. The only eligible short report described a cross-sectional study and its quality was therefore also evaluated using the STROBE checklist. The minimum and maximum scores of each study as evaluated by STROBE were 0 and 44, respectively. Depending on their score, the studies were classified as low quality (<15.5), moderate quality (15.5–29.5), and high quality (30–44). The risk of bias for each study was independently assessed by the two reviewers; discrepancies were recorded and resolved by consensus. The quality of papers not published in English was not assessed.

2.3. Pooled analysis

Three different meta-analyses were performed. The first included all HCWs, the second compared susceptibility by sex (female vs. male), and the third by age class (born after vs. before 1980; this cut-off marked the pre- and post-vaccination eras and thus defined the naturally immunized, who probably came into contact with the wild virus several times, and those born and raised in the post-vaccination era, were presumably vaccinated, and whose contact with the wild virus was unlikely because mass vaccination reduced viral circulation). For the comparisons of sex and age class, the risk ratio (RR) and 95% confidence interval (95 %CI) were calculated. A sub analysis based on the sample size (<1,000 vs. 1,000+ HCWs) was performed for each meta-analysis. In addition, for each of the three meta-analyses, a separate analysis was carried out using only high-quality papers.

Two further sub-analysis were performed based on the country of the eligible studies and commercial immunoassay used to evaluate susceptible status of HCWs (the subsequent analysis per quality score was not performed due to the small number of papers for each subgroup).

The pooled proportions in the meta-analyses were calculated after Freeman-Tukey double arcsine transformation of the data, to stabilize the variance, and using the DerSimonian-Laird weights for the random effects model. The estimate of heterogeneity was taken from the inverse-variance fixed-effect model. The pooled prevalence with the relative 95% Wald CI and the forest plot were drawn. In addition, the I^2 statistic was calculated as a measure of

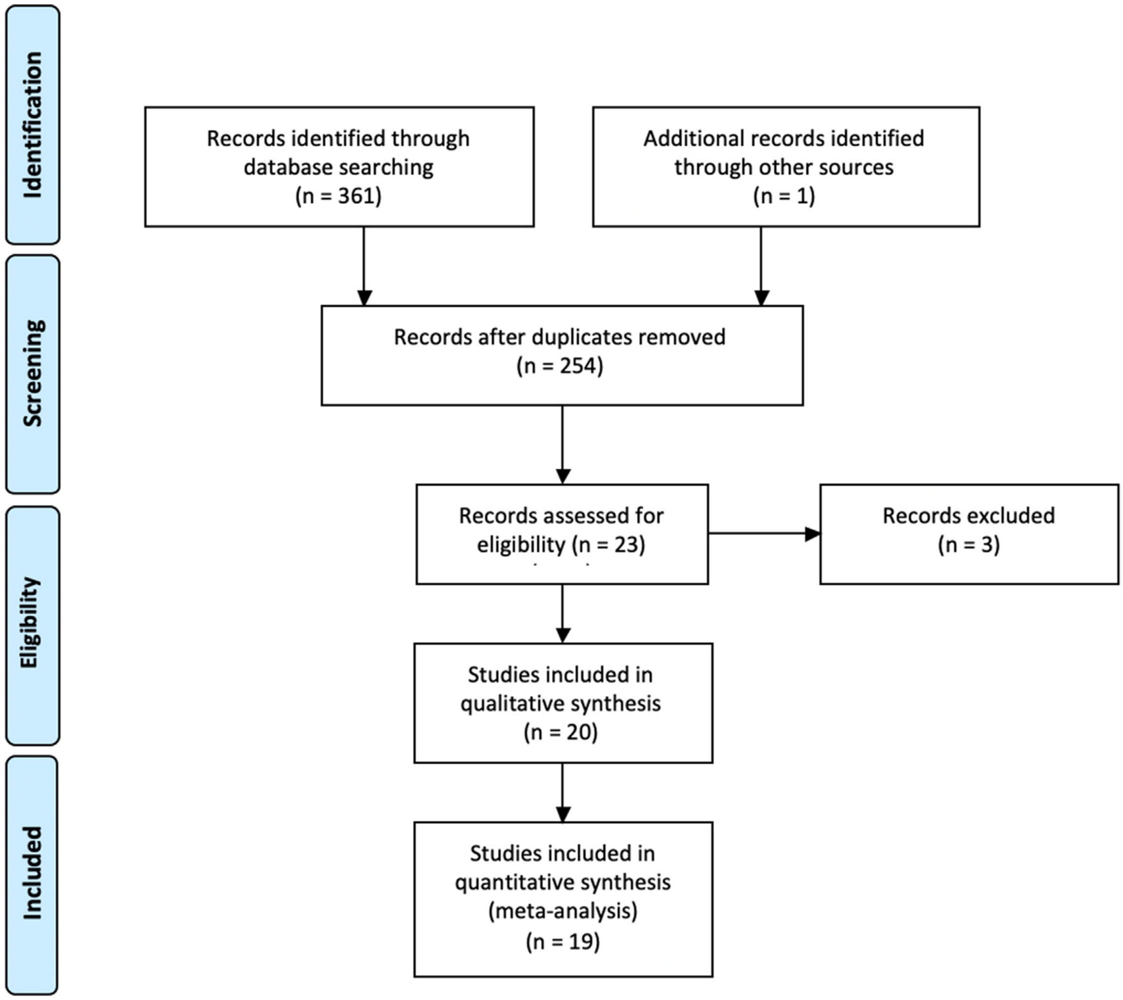

Fig. 1. Flow-chart of the bibliographic research.

the proportion of the overall variation attributable to between-study heterogeneity rather than to chance; the between-study heterogeneity of the different groups was also evaluated. For the heterogeneity, determinations, a p-value < 0.05 was considered statistically significant.

Funnel plots were used to assess publication bias. A study distribution with a symmetric funnel-shape indicated no significant bias, whereas an asymmetric funnel indicated publication bias. Egger's test for small-study effects was also performed.

A sensitivity analysis was conducted to evaluate stability, in which among the studies included in this systematic review one study at a time was excluded and the conclusion based on the others then re-evaluated for severe distortion.

The statistical analysis was conducted using STATA MP16 and Review Manager 5.4.1 software.

Strategies to promote vaccination among susceptible HCWs and the characteristics of serosusceptible HCWs were collected from all available studies and their respective findings compared, with particular attention paid to the evidence presented in several of the included papers.

3. Results

3.1. Identification of relevant studies

The article selection process, conducted according to the PRISMA indications [20], is summarized in the flow-chart in Fig. 1. Based on the aforementioned inclusion criteria, 9 articles were identified in Google Scholar, 16 in MEDLINE/PubMed, 11 in Scopus, and 8 in the ISI Web of Knowledge; one study was identified through other sources. After the exclusion of articles duplicated in more than one database, 23 studies were eligible for inclusion. Of these, two [41,42] were excluded because the data were not available and the authors did not provide them on request; another paper [43] was excluded because it evaluated the same phenomenon in a population studied in a more recent, more complete paper already included in the meta-analysis. Thus, overall, 20 studies were eligible [21-40] (Table 1), of which 19 were quantitative [21-39] and 1 was qualitative [40]; These studies were from Italy, Spain, Germany, Czech Republic, Finland, Hungary, and the Netherlands. The remaining 234 studies did not match the inclusion criteria [1,4-6,8,11,13,15,16,35,40,40-261].

3.2. Quality assessment

The STROBE checklist score was calculated for all articles included in meta-analysis written in English; 70.6% of the eligible papers were determined to be of high quality (Table 1). The impact of study quality was assessed in a sub-analysis.

3.3. Pooled analysis

The meta-analysis of all HCWs showed that the prevalence of serosusceptibility to measles was 13.3% (95 %CI: 10.0–17.0%;

Table 1
Characteristics of the selected studies included in meta-analysis.

First author	Year	Quality	Susceptible HCWs (n)	Total sample	Study period	Country	Commercial immunoassay	Population
Quantitative study								
Anichini G [21]	2020	high	249	1,092	2018–2019	Italy	CLIA (LIAISON)	stu, res
Coppeta L [22]	2020	moderate	224	1,017	2019	Italy	CLIA (LIAISON)	HCWs (phy, nu, res, stu)
Coppeta L [23]	2020	high	80	358	2018	Italy	CLIA (LIAISON)	nurse
Camilloni B [24]	2020	moderate	51	461	n.r.	Italy	CLIA (LIAISON)	HCWs (phy, nu, oth)
Trevisan A [25]	2020	high	2,671	10,653	2004–2019	Italy	ELISA (Enzygnost)	Students
Malinova J [26]	2020	high	180	2,784	2018–2019	Czech Republic	ELISA (Immunolab)	HCWs (phy, nu, oth)
Rivas FV* [27]	2019	-**	152	2,614	2014–2019	Spain	n.r.	HCWs (n.r.)
Coppeta L [28]	2019	moderate	84	319	2018	Italy	CLIA (LIAISON)	students
Bianchi FP [29]	2019	high	29	447	2017–2019	Italy	CLIA (LIAISON)	HCWs (phy, nu, oth)
Coppeta L [30]	2019	high	450	2,940	2017	Italy	CLIA (LIAISON)	HCWs (phy, nu, oth, res, stu)
Ledda C [31]	2019	moderate	77	549	2017	Italy	CLIA (LIAISON)	HCWs (n.r.)
Galiàn Munoz I* [32]	2019	-**	15	138	2017	Spain	n.r.	HCWs (phy, nu, oth)
Bianchi FP [33]	2019	high	305	2,000	2014–2018	Italy	CLIA (LIAISON)	stu, res
Genovese C [34]	2019	moderate	7	140	2018	Italy	ELISA (Technogenetics)	HCWs (n.r.)
Hiller U [35]	2019	high	80	1,923	2017	Germany	ELISA (not specified)	HCWs (phy, nu, oth)
Lengyel G [36]	2018	high	204	2,167	2017	Hungary	ELISA (Serion)	HCWs (phy, nu, oth)
Koivisto K [37]	2017	high	29	157	2014	Finland	ELISA (Human-ELISA-IgG-Antibody-Test)	HCWs (phy, nu, oth)
Petersen S* [38]	2015	-**	1,420	9,993	2003–2014	Germany	ELISA (Enzygnost)	HCWs (phy, nu, oth)
Dorigo-Zetsma JW [39]	2015	high	18	154	2013	Netherlands	ELISA (Enzygnost)	HCWs (n.r.)

HCW = healthcare worker; phy = physician; nu = nurse; oth = other HCW; res = medical resident; stu = students n.r. = not reported; CLIA = chemiluminescence immunoassay; ELISA = enzyme-linked immunosorbent assay.
* not included in the systematic review.
** quality not assessed.

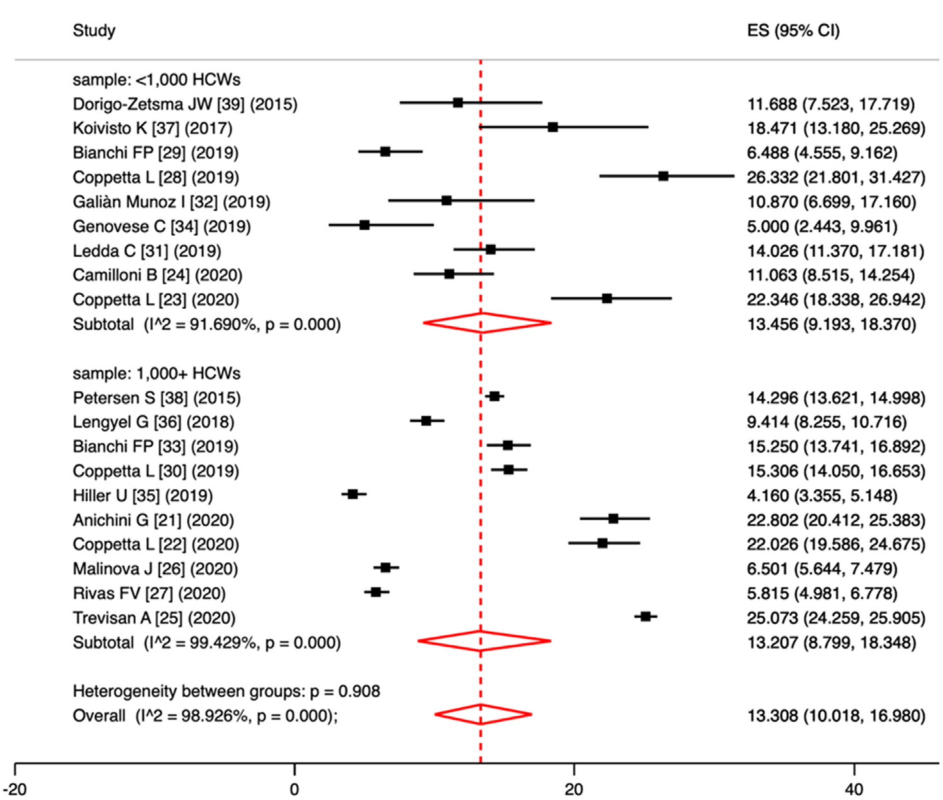

Fig. 2. Forest plot of the pooled prevalence of serosusceptibility to measles, per sample size (<1,000 vs. 1,000+).

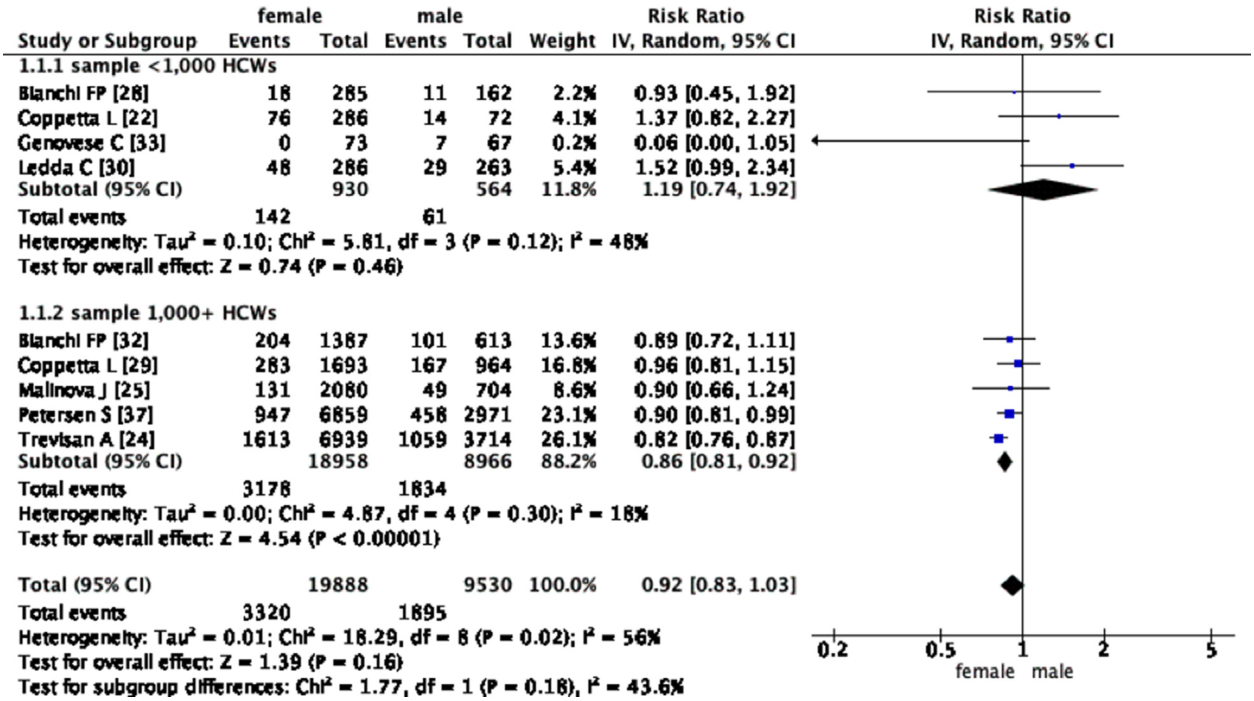

Fig. 3. Forest plot of the Risk Ratio in a comparison of serosusceptibility with respect to sex (female vs. male) and per sample size (<1,000 vs. 1,000+).

I^2 = 98.9%; p value for heterogeneity < 0.0001). Based on only the high-quality articles, the pooled prevalence among all HCWs was 13.5% (95 %CI = 8.6–19.3%; I^2 = 99.2%; p < 0.0001).

In a sub analysis of measles serosusceptibility according to sample size, the prevalence was 13.5% (95 %CI = 9.2–18.4%; I^2 = 91.7%; p < 0.0001) in HCWs for a sample size < 1,000 and 13.2% (95 %CI = 8.8–18.3%; I^2 = 99.4%; p < 0.0001) for a sample size of 1,000+, in accordance with a p value of 0.91 in the test of heterogeneity between sub-groups (Fig. 2). If only the high-quality articles were considered, the pooled prevalence among HCWs in a sample size < 1,000 was 14.1% (95 %CI = 6.7–23.6%; I^2 = 93.7%; p < 0.0001) and in a sample size of 1,000 + 13.2% (95 %CI = 8.6–19.3%; I^2 = 99.5%; p < 0.0001). The p value in the test of heterogeneity between sub-groups was 0.84.

In the comparison of measles serosusceptibility between female and male HCWs, the RR was 0.92 (95 %CI = 0.83–1.03; I^2 = 56.0%;

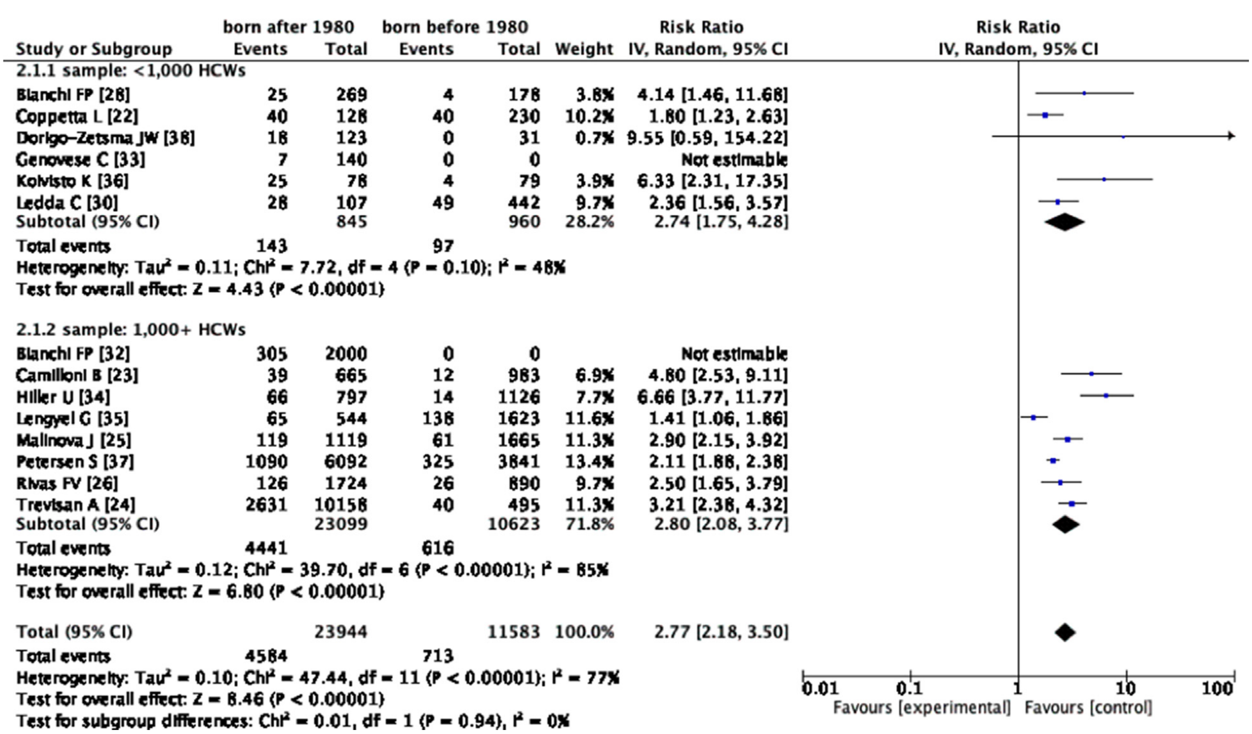

Fig. 4. Forest plot of the Risk Ratio in a comparison of serosusceptibility with respect to age class (born after vs. before 1980) and per sample size (<1,000 vs. 1,000+).

p = 0.02); when only the high-quality studies were considered, the RR value was 0.89 (95 %CI = 0.80–0.99; I^2 = 31.0%; p = 0.20). the male vs. female sub analysis of sample sizes < 1,000 and 1,000+, the RRs were 1.19 (95 %CI = 0.73–1.93; I^2 = 48.0%; p = 0.12) and 0.86 (95 %CI = 0.81–0.92; I^2 = 18%; p = 0.30), respectively, and the p value in the test for heterogeneity between sub-groups was 0.18 (Fig. 3). In the analysis based only on the high-quality studies, the RR for a sample of < 1,000 was 1.20 (95 %CI = 0.79–1.83; I^2 = 0.0%; p = 0.39) and that for a sample of 1,000 + 0.86 (95 %CI = 0.79–0.93; I^2 = 19.0%; p = 0.29); the p value in the test for heterogeneity between sub-groups was 0.12.

In the comparison based on age class (born after vs. before 1980) the RR was 2.77 (95 %CI = 2.18–3.50; I^2 = 77.0%; p < 0.0001); for the subs-et of high-quality articles, the RR was 3.05 (95 %CI = 2.05–4.53; I^2 = 82.0%; p=<0.0001). The age class (born after vs. before 1980) sub analysis according to a sample size < 1,000 and 1,000+, the RRs were 2.74 (95 %CI = 1.75–4.28; I^2 = 48.0%; p = 0.10) and 2.80 (95 %CI = 2.08–3.77; I^2 = 85.0%; p < 0.0001), respectively; the p-value in the test for heterogeneity between sub-groups was 0.94 (Fig. 4). In the analysis of only high-quality studies, the RRs for sample sizes of < 1,000 and 1,000 + were 3.48 (95 %CI = 1.57–7.69; I^2 = 61.0%; p = 0.05) and 2.94 (95 %CI = 1. 71–-5.07; I^2 = 90.0%; p < 0.0001), respectively; the p value in the test for heterogeneity between sub-groups was 0.74.

In a sub analysis of measles serosusceptibility according to country, the prevalence of serosusceptible HCWs was 15.8% (95 % CI = 11.9–20.1%; I^2 = 97.7%; p < 0.0001) in Italy (7 studies), 5.9% (95 %CI = 5.0–6.8%; I^2=-; p = -) in Spain (2 studies), 12.3% (95 %C I = 11.7–12.9%; I^2=-; p = -) in Germany (2 studies), 34.2% (31.2–37.0%) in France (1 study), 9.4% (95 %CI = 8.3–10.7%) in Hungary (1 study), 18.5% (95 %CI = 13.2–25.3%) in Finland (1 study), 6.5% (95 %CI = 5.6–7.4%) in Czech Republic (1 study) and 11.7% (95 %C I = 7.5–17.7%) in the Netherlands (1 study).

In a sub analysis of measles serosusceptibility according to commercial immunoassay, the prevalence of serosusceptible HCWs was 16.8% (95 %CI = 13.5–20.3%; I^2 = 94.2%; p < 0.0001) using chemiluminescence immunoassay (CLIA) LIAISON (9 studies), 16.9% (95 %CI = 9.4–26.1%; I^2=-; p = -) using enzyme-linked immunosorbent assay (ELISA) kit Enzygnost (3 studies), 5.0% (95 %CI = 2.4–10.0%) using ELISA kit Technogenetics (1 study), 18.5% (13.2–25.3%) using ELISA kit Human-ELISA-IgG-Antibody-Test (1 study), 6.5% (95 %CI = 5.6–7.5%) using ELISA kit Immunolab (1 study), 9.4% (95 %CI = 8.3–10.7%) using ELISA kit Serion (1 study), 4.1% (95 %CI = 3.4–5.1%) using a not specified ELISA kit (1 study) and 5.9% (95 %CI = 5.0–6.8%; I^2=-; p = –) using a not reported test (2 studies).

The sensitivity analysis did not show a severe distortion by a specific study. In the publication bias analysis, there was no obvious asymmetry in the funnel plots and no strong evidence of publication bias, especially for those studies with a large sample size (Fig. 5). The p-value in the Egger's test was 0.37 for the sub-analysis based on sex and 0.06 for that based on age class.

3.4. Suggestions and procedures to manage measles susceptibility in HCWs

All studies concluded that the screening of HCWs is essential to prevent nosocomial clusters and that the promotion of an adequate immunization program should be a priority of Occupational Medicine services. The four studies [20,34,35,39] that focused on the cost-effectiveness of such strategies consistently found that that an immunization strategy with pre-vaccination screening was

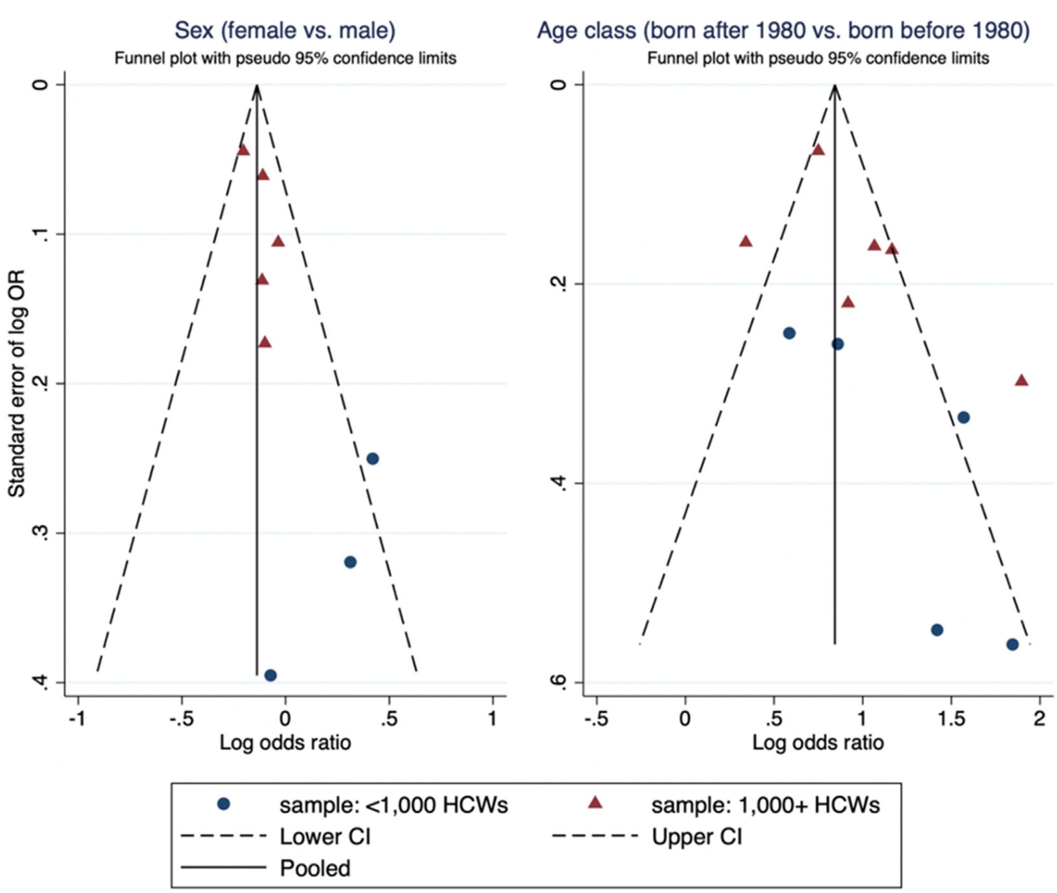

Fig. 5. Funnel plot with pseudo 95% confidence limits.

more cost-effective than a hypothetical vaccination strategy without screening. A 2017 Dutch study [40] of hospital culture and organization suggested that the guidance offered to hospitals regarding outbreaks of vaccine-preventable disease should take into account the need for a distinct step-wise approach to policy implementation and a clear chain of responsibilities.

In most studies, the immunization status of the person prior to serological testing is known and includes several non-seroprotected individuals who remained unvaccinated; however, in many cases, among the twice vaccinated are those who are still serosusceptible [26,28,30,33]. Bianchi FP et al. [33] conducted a serosurvey of 2,000 fully vaccinated individuals and determined that 15.3% were still susceptible to measles. In the study of Anichini G et al. [21], 161 (23.6%) of the 682 participants who had received two doses of vaccine remained seronegative. Malinova J et al. [27] found a seronegativity rate of 6.3% in the cohorts fully immunized only by vaccination (participants 19–43 years of age); the proportions of fully vaccinated among serosusceptible HCWs ranged from 13% to 25% [29,31].

Many of the included studies [21-26,28,31,35-37,39] reported a higher proportion of serosusceptible HCWs among those born in the post-vaccination than in the pre-vaccination era and thus naturally immunized. Serosusceptibility among the former can be traced to the fact that IgG antibody titers induced by the measles-mumps-rubella (MMR) vaccine decline by 5–7% per year even after a second dose of the vaccine. In this context, two studies [25,33] determined that the interval since the last dose of MMR vaccine seemed to influence the persistence of circulating antibodies. By contrast, according to Malinova J et al. [26] the persistence of seropositivity is not dependent on the time since the acquisition of immunity, but on how immunity was acquired, i.e., by vaccination or by childhood measles. However, all authors agreed that, while vaccine-induced humoral immunity persists for 15 years or longer, naturally acquired immunity is always longer and probably life-long.

Only a few studies compared the proportion of non-seroprotected male vs. female HCWs. Two studies [23,31] determined a higher proportion of serosusceptible female than male nurses. Coppeta L et al. [23] suggested a higher vaccination rate among females via MMR vaccination programs whereas male nurses were more often not vaccinated but were more likely to have natural and thus longer-lasting immunity, resulting in their paradoxically higher seroprotection. The other studies [21,22,24,25,33,36] did not find any significant difference in the seroprotection of male and female HCWs.

The management of serosusceptible HCWs was not addressed by nine studies [21-23,25,26,34,36,37,39]; two studies [24,31] reported that the MMR vaccine was offered to serosusceptible HCWs but neither compliance nor the seroconversion rate was reported. Coppeta L et al. [28,30] reported that almost 50% serosusceptible HCWs received the MMR vaccine but provided no data on the seroconversion rate among those who were vaccinated. Hiller U et al. [35] vaccinated > 95% of the non-seroprotected HCWs in their hospital. Of the eight HCWs who years before the serological test were vaccinated with two doses of MMR, seven were re-vaccinated and showed a sufficient increase in measles-IgG after 2 weeks. Bianchi FP [29,33] described the management of serosusceptible HCWs, medical students and medical residents at Bari Policlinico General University Hospital (Italy), regardless of vaccination status (none or two doses of vaccine) and recall of having had the disease. For the never immunized group, the measles vaccination protocol consisted of two doses of MMR vaccine administered 28 days apart and followed by a blood test. For the fully vaccinated group, a booster dose of MMR vaccine was provided, followed 20–25 days later by a second blood test to retest IgG titers. If the value determined in the re-evaluation exceeded the cut-off, the HCW was classified as seroconverted; if the titer was still negative, another vaccine dose (28 days after the first booster) was administered and again after 20–25 days IgG levels were measured. For medical students and residents who after this protocol remained seronegative, a re-evaluation for measles infection was recommended in all cases of exposure, with the possible administration of immunoglobulin. Screening was voluntary and vaccination was not mandatory, with its rejection without consequences in terms of work suitability [33]. Thus, at the end of screening the Occupational Health physician listed the placement options for each potential HCW according to his/her susceptibility/immunity status and an evaluation of risk. For susceptible HCWs who refused one or more vaccines, exclusion from occupational settings that included patients at high infectious risk (e.g., immunocompromised patients) was recommended [29]. The authors reported high vaccination compliance among susceptible HCWs and medical students/residents and a seroconversion rate > 90% after a booster dose(s). The latter were not followed by any serious adverse events.

The need for one or more MMR vaccine doses in serosusceptible HCWs was discussed in many of the studies. Malinova J et al. [26] concluded that adult, fully vaccinated or naturally immunized HCWs do not need to be systematically revaccinated with one or more vaccine doses. Dorigo-Zetsma JW and Lengyel G [36,39]. reported that vaccinating non-seroprotected HCWs may be an opportunity to reduce the risk of nosocomial clusters. Bianchi FP et al. [29,33] presented the protocol described above. Finally, Anichini G et al. [21] suggested monitoring of the population 10–15 years after vaccination, in order to revaccinate individuals who were seronegative.

4. Conclusion

Our meta-analysis showed an overall prevalence of susceptible European HCWs of 13.3% (95 %CI = 10.0–17.0%), more than double the rate reported in a 2015 review (6%) (a meta-analysis was not performed) [1] and higher than the rate reported in a 2019 meta-analysis that investigated Italian HCWs (9%) (that included not only serosurveys but also papers based on self-administered questionnaire) [4].

To our knowledge, this is the first study to find that female HCWs were less likely than their male counterparts to have circulating anti-measles IgG (RR = 0.92; 95 %CI = 0.83–1.03). While the statistical significance of this conclusion was low, it was supported by the sub analysis based on study quality and larger sample size, both of which had a RR < 1 and less heterogeneity among studies. Sex differences in the response to vaccination or infection has been examined in several studies [262-266], but our analysis is the first to demonstrate sex-based differences for measles infection/vaccination. Females generally have more effective immune responses following immunization and against infection, with immunological, hormonal, genetic, microbiotic, and environmental factors likely contributing to the difference between males and females with respect to measles. Furthermore, as women have been the primary target for the elimination goal of rubella and congenital rubella, they are likely to have received the rubella or MMR vaccine during their youthful age; therefore the administration of the MMR formula could explain the different serosusceptibility to measles compared to males.

Our study also showed a higher risk of a loss of seroprotection in HCWs born in the post-vaccination era (RR = 2.78; 95 %CI = 2.20–3.50) and thus unlikely to be exposed to the wild virus, the circulation of which has decreased since the introduction of immunization. A 2020 Italian study [267] evaluated the proportion of individuals with detectable anti-measles IgG in two groups, those vaccinated with two doses of anti-MMR vaccine and those with a self-reported history of measles infection. Among the 611 students and residents who were tested, 94 (15%) had no detectable protective anti-measles IgG. This proportion was higher among vaccinated individuals (20%) than among those with a self-reported history of measles (6%; p < 0.0001); the seroconversion rate after two doses of MMR vaccine in the disease group was 100% (95 %CI = 59–100%), while in the vaccinated group it was 86% (95 %CI = 73–94%), concluding that the difference in the response to the booster dose(s) may have reflected the greater persistence of immunological memory in naturally immunized individuals. Although the immune responses induced by the vaccine are qualitatively similar to those induced by infection, antibody levels are lower after vaccination. Vaccination at a young age enhances the quality and quantity of the antibody response but has a minor effect on T cell responses. Over time, the levels of virus-specific antibodies and vaccine-induced CD4 + T cells decrease, accounting for a secondary vaccine failure rate of 5% 10–15 years after immunization [268]. Several studies have investigated the duration of humoral immunity and the role played by circulating IgG antibodies both in patients who have overcome infection and in the vaccinated population. Although the results showed a stronger antibody response (titer) induced by natural disease than by vaccination, a 1994 study [269] found that, for MMR immunity, serological memory after vaccination is similar to that after natural infection. However, the second dose of the MMR vaccine is essential, as the antibody titer slowly declines during the first 10 years after the first vaccination of the basal protocol [269]. The levels of neutralizing antibodies 10 years after the second dose of vaccine were shown to remain above the level considered protective and to confer long-lasting immunity, although they fall in the years thereafter [270]. Several studies reported that circulating anti-measles IgG antibodies decrease ∼15 years after the second dose of MMR vaccine administered according to the basal protocol [22-26,28,31,35-37,39]. However, the authors cautioned against questioning the role of measles vaccination, as the complications of measles are more frequent and more serious than any vaccine-related adverse reaction [271-273]. For example, in a recent study published in *Science* [274], Mina et al. described the long-term damage to immune memory caused by measles infection. They found that measles infection can greatly diminish previously acquired immune memory, potentially leaving individuals at risk of infection by other pathogens. In light of these evidences, we can conclude that the MMR vaccine remains the most effective, safe, and cost-effective tool for preventing measles.

Regarding the sub-analysis per country, the higher prevalence of serosusceptible was found in France (34%) and the lowest in Spain (6%); it must be underlined that this analysis is strongly biased due to the small number of selected papers per subgroup. No particular differences among serosusceptibility can be evidenced considering the vaccination policy of each country; indeed, only in Czech Republic the measles vaccine is mandatory to get hired for specific GCWs groups or settings, in Hungary and the Netherlands it is not mandatory nor recommended, while in the other countries it is recommended [15]. Considering the distribution of measles cases and notification rates per 1,000,000 population by country (from 2014 to 2018) the picture was very heterogeneous; the higher rates were found in Italy (89.1 in 2017), followed by France (43.6 in 2018), Germany (30.4 in 2015), Czech Republic (21.1 in 2014), the Netherlands (8.6 in 2014), Spain (4.8 in 2018), Hungary (3.7 in 2017) and Finland (2.7 in 2018) [275]. These values seem to be difficult to correlate with the susceptibility prevalence highlighted in our paper; an ad hoc study should be set up to correlate the circulation of the virus to the seroprevalence values in the HCWs in the selected countries. No data were found regarding the overall vaccine coverage in HCWs for each country.

Considering the commercial immunoassay, similar values of prevalence were evidenced considering CLIA LIAISON (16.8%) and ELISA kit Enzygnost (16.9%); the other commercial kits were used by too few studies to draw solid considerations. On the other hand, the type of commercial kit used should not be a cause for concern; a 2015 study, in fact, compare CLIA LIAISION with Enzygnost ELISA, with final classification of discrepancies by indirect immunofluorescence, concluding that the sensitivity and specificity of CLIA against ELISA were 95.5% (95 %CI = 89.5–98.3%) and 100% (95 %CI = 91.8–100%) respectively [276]. Latner DR et al. [277] in a 2020 paper compared two ELISA tests whit three manufacturer-specific automated equipment tests; the results demonstrate differences in the sensitivity and specificity of individual IgG tests, even if all the commercial platforms demonstrated good agreement of qualitative results. Up to 11% of samples gave discordant results in comparisons of the most-sensitive versus the least sensitive platforms; the discrepant results were in the low-positive, equivocal, and high-negative ranges for all platforms.

Few studies described the management of susceptible HCWs but the protocol developed by Bianchi FP et al. [29,33] was shown to have high efficacy and safety. However, the management of HCWs vaccinated with two doses but still without circulating antibodies remains problematic. Should they receive one or more MMR booster doses? The literature includes reports of measles in fully vaccinated HCWs during an outbreak. For example, in the 2018 study by Machado RS et al., 67 of 96 HCWs (age 18–39 years) with confirmed nosocomial measles had previously been vaccinated with two doses of measles or MMR vaccine [11]. In a 2016 Dutch study, six of eight HCWs confirmed to have measles had been vaccinated twice, such that among 106 potentially exposed HCWs the estimated effectiveness of two doses of measles vaccine was 52% [12]. In that study, two of the HCWs had pre-exposure neutralizing antibodies, evidenced in samples collected 4 months and 8 years before illness, respectively. Among 99 cases in Greece, six (6.1%) of the affected HCWs had been vaccinated twice [13]. In an outbreak in England in 2013 that involved 110 individuals, 30 had been fully immunized with two doses of MMR vaccine [14]. Toner et al. [44] identified two fully vaccinated HCWs among 52 who developed measles in Catalonia. The authors concluded that the assessment of immunization status and the implementation of a two-dose vaccination protocol in those lacking evidence of immunity are needed to eliminate the risk of acquiring and spreading measles in healthcare settings.

Furthermore, this systematic review and meta-analysis determined a consistent proportion of non-seroprotected HCWs, especially younger ones, among those vaccinated with two doses. The role of cell-mediated immunity and circulating antibodies in the long-term response to the vaccine/disease (and consequent protection against measles) is discussed controversially in the scientific literature. Amanna et al., in a study reported in 2017 [278], conducted a prospective observational analysis of antibody titer changes in 45 individuals over a period of more than 26 years. Antigen-specific memory B cells were also measured and their levels compared with those of the corresponding antibodies. The authors determined an association between the levels of memory B-cell and the concentration of antibodies against measles, based on the assumption that serum antibodies and memory B cell levels are equally stable but independently maintained. However, a direct cause-and-effect relationship could not be established [278]. A 1975 study highlighted the role of cellular immunity and postulated that the cell-associated immune system is the main host defense against measles. The findings were based on the observed responses to measles in agammaglobulinemic children and the death of these children but not those with a thymus deficiency who also contracted measles [270]. However, a 2016 study found a much smaller contribution of T cells to protection than of neutralizing antibodies [279]. Thus, whether HCWs born in the post-vaccination era represent a potential risk for nosocomial outbreak remains to be determined.

The main limitation of this meta-analysis was the high heterogeneity across studies, as indicated by the I^2 values; but our use of a random effect analysis minimized this bias. Also, the lack of information from most European countries may have distorted the general picture and introduced bias. Thus far, Italy is the country with the highest number of studies, followed by Spain, Germany, Czech Republic, Finland, Hungary, and the Netherlands; indeed, the results of our study can be generalized only to the above described counties and not to the entire Europe. This difference could be due to the thousands of measles cases that have occurred across the Italian peninsula since 2017, including several cases among HCWs [46] that may have piqued interest in scientific research into the immune status of health personnel and the management of those found to be susceptible. Nonetheless, measles outbreaks involving HCWs have also been reported in the rest of the EU / EEA and in the UK [10]. Another limitation of this study was the difficulty in obtaining data not easily deducible from the included papers; in fact, many authors were unwilling to provide the data. Differences in the techniques used to analyze blood samples also complicated comparisons between studies. The chemiluminescence-based method of the LIAISON® Measles IgG system [280] was used in nine studies, and other techniques with different cut-offs defining immunity in the other ten studies; as reported above, this does not seem to be a critical issue. It was also not possible to stratify susceptible HCWs on the basis of their vaccination status or previous illness. However, a strength of our review and meta-analysis was the large sample size that resulted from collating the selected papers, which improved the statistical analysis and provided a better view of measles immunity among European HCWs. Furthermore, since most studies investigated a recent cohort of HCWs (mostly since 2017) this view is up-to-date and reliable. Finally, the sub analysis by age class and sex provided information, including RR values, not previously reported in the literature.

The elimination of measles is a 20-year objective of national and international public health institutions [281], but the many elements highlighted by this study and reported in the recent scientific literature highlight the challenges in achieving this goal.

Firstly, epidemic outbreaks affect both unvaccinated and vaccinated individuals [46]; secondly, in addition to the D8 genotype, the worldwide re-emergence of measles has been attributed to an emerging clone, the B3 strain [46]; however, Fatemi Nasab et al. [282] found that neutralizing antibody titers were lower for the B3 genotype than for the H1, D4, and A genotypes. Thirdly, vaccinated individuals lose antibodies over time such that between now and the next 10–20 years, the measles susceptibility of the vaccinated population may increase. The impact of these different observations on the goal of eradication, as proposed by Fenner in 1998, are unknown [283].

It is therefore the task of national and international public health institutions to support the development of innovative strategies aimed at addressing the measles risk, especially in the nosocomial setting. Attempts to educate HCWs and medical school/nursing students [47-49], as the efforts thus far have proved insufficient to bridge the immunization gap [50]. The solution proposed in most of the recent scientific literature is to mandate the vaccination of HCWs [16,51,52] in order to reduce the risk of nosocomial transmission by patients and the HCWs themselves. In Italy, three regions approved a specific law that made vaccinations for HCWs semi-mandatory, based on work suitability as assessed by occupational health physicians [34], similar to the protocol described by Bianchi FP [29]. The impact of this law on the immunization status of HCWs has yet to be reported, but are expected to be encouraging.

Moreover, considering the ease of travel, cultural exchanges, and working in other countries, increasingly common in the modern era, it is essential that mutual strategies are put in place in all European countries, as also advocated by Maltezou HC et al. [16]. Yet, the path to a common mandatory strategy in the EU/EEA and UK countries is likely to be an uphill one, including for reasons of informed and medico-legal consent [284,285]. Thus, from the perspective of the current epidemiological framework, hospital administrations must work to overcome vaccine hesitancy among HCWs [286]. Nonetheless, the need for registry for vaccinations of HCWs to follow vaccination rates in real-time and a reminding systems remains critical.

In conclusion, even in the era of the COVID-19 pandemic, diseases such as measles (and other vaccine-preventable diseases, such as influenza, hepatitis B, mumps, rubella, pertussis) still represent a threat in hospital and community settings that cannot and must not be forgotten by policy makers. Indeed, it is highly probable that in the two years pandemic the anti-measles vaccine coverages in general population may be dropped and so the risk of measles outbreaks in the following years is possible. In the emergency state in which the world currently finds itself, quick and firm decisions must be taken. The reduction of susceptible HCWs would reduce the risk of a measles outbreak and therefore measles-related morbidity and mortality, absenteeism, and their direct and indirect costs [35,53-55,287]. Other, perhaps more difficult to solve problems for international public health institutions are the management of HCWs vaccinated with at least two doses of vaccine but who remain sero-susceptible and the decrease in circulating antibody over time among the vaccinated.

5. Footnote page

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Declaration of Competing Interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

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