

Examination of the prevalence of vaccine-preventable pathogens and attitudes towards vaccinations

PhD thesis

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1. Introduction

1.1 Infectious diseases

The common history of infectious diseases and mankind goes back a long way, perhaps all the way to the beginning. Throughout history, many epidemics have shaken human communities. Although effective and safe vaccines are available against numerous diseases, this form of protection still encounter an obstacle, as vaccine hesitancy and, unfortunately, anti-vaccination also gained ground as never before.

Regarding infectious diseases, I focused on two important topics during my research. Firstly, with our research group we examined the prevalence of *Neisseria meningitidis* in Hungary, and the risk factors influencing carriage. Secondly, we wanted to understand better the attitudes towards varicella vaccination among parents and paediatric healthcare professionals before it was made mandatory in 2019 in Hungary. We also examined parental attitudes towards meningococcal vaccinations. Unfortunately, the increasing uncertainty concerning vaccinations in recent years is a barrier in their use. The availability of vaccines is unavailing if people are not willing to take them.

1.2 *Neisseria meningitidis*

Neisseria meningitidis is an obligate human pathogen. Encounter with the bacterium usually leads to asymptomatic carriage in the nasopharynx, which plays an important role in the spread of the pathogen. Disease develops only in a fraction of cases, when the bacteria enter the bloodstream, causing invasive meningococcal disease (IMD). Though IMD is rare, it can be associated with very high mortality. Based on the antigen structure of the capsular polysaccharide it can be divided into 12 serogroups, from which 6 causes most of the clinical cases (A, B, C, W-135, X and Y). There are several vaccines available for serogroup A, B, C, W-135 and Y, and there is one pentavalent vaccine that is also effective against serogroup X infection. Knowing the circulating strains in the

population is crucial to choose between the vaccine options, and for being prepared if a non-vaccine serogroup appears.

1.3 *Varicella-zoster virus*

Varicella-zoster virus (VZV) causes varicella in children and, later in life, may reactivate in the form of herpes zoster. Varicella is one of the most common childhood diseases, usually with mild, self-limiting symptoms, but rarely it can be associated with serious complications such as bacterial superinfection, pneumonia, and encephalitis in otherwise healthy children. In Hungary, varicella vaccine became available in 2003 on the private market and reached about 20% vaccination coverage. Awareness of the disease was raised by recommendations, professional and parental education, but still, vaccine coverage has reached a plateau. In 2019, the budget financed mandatory universal varicella vaccination was integrated into the NIP in Hungary.

1.4 Mandatory and non-compulsory vaccines in Hungary, vaccine hesitancy

In Hungary, vaccines to prevent several childhood diseases are involved in the national immunization program. Vaccination coverage with these vaccines is extremely high, the latest data from 2023 shows that 99,5-99,9% of the obliged population got vaccinated against tuberculosis, diphtheria, tetanus, pertussis, *Haemophilus influenzae b*, Poliovirus, pneumococcus, morbili, mumps, rubella and Hepatitis B virus. The incidence of these vaccine preventable diseases is very low in Hungary. Exact data on the occurrence of pathogens that are not included in the vaccination calendar and can be prevented by recommended, non-compulsory vaccinations are not available, these pathogens are probably more common. Vaccination with non-compulsory vaccines is typically low in Hungary. It is important to better understand the attitudes and the reasons for rejecting vaccinations, to increase childhood vaccination and reduce the number of serious diseases that can be prevented by vaccination.

2. Objectives

The objectives of our studies were the following:

- (1) To examine the knowledge, attitudes and factors influencing the support or rejection of varicella vaccination among parents and pediatric healthcare professionals in Hungary, before the introduction of varicella vaccination into the national immunization program. Our study sought to have an answer to the question of how to increase the trust and so the vaccination rate and safety of young children in relation to VZV vaccination and other vaccinations.
- (2) To assess knowledge about *Neisseria meningitidis* infection and the factors that determine refusal or support for vaccination among parents.
- (3) To assess the prevalence and risk factors of asymptomatic carriage of *Neisseria meningitidis* in high school and university students in Hungary, furthermore, to identify the currently circulating serogroups in these populations.

3. Methods

3.1 Examination of the attitudes towards varicella vaccinations

We recruited parents and pediatric healthcare providers via the internet to fill out a questionnaire about varicella vaccination (the original Hungarian questionnaires are attached to the thesis). The questionnaire was available between October 2018 and February 2019. 1146 parents and 194 healthcare professionals (189 pediatric health visitors and 5 pediatricians) completed the survey. The study included 1042 parents who responded to the primary outcome question about whether they had vaccinated at least one child against varicella. The primary outcome of the survey for professionals was whether they support universal varicella vaccination or not.

We employed logistic regression to explore the factors influencing vaccination decisions. The multivariate analysis for decision-making incorporated several key variables. We calculated odds ratios with corresponding 95% confidence intervals, excluding incomplete response sets. Statistical significance was determined by a p-value threshold of less than 0.05. To compare perspectives between professionals and parents, we used a two-sided Chi-square test. All statistical computations were made using MedCalc for Windows (version 19.0.4).

3.2 Examination of attitudes towards *Neisseria meningitidis* vaccination

Parents were invited to take part in an online survey assessing their knowledge of *Neisseria meningitidis* infection and their attitudes towards vaccination (the original Hungarian questionnaire is attached to the thesis). The online questionnaire was available between November 2020 and July 2021. Overall, 165 parents completed the survey, but only 159 of them responded to the primary outcome question about whether they had vaccinated at least one child against meningococcal infection.

To examine the factors influencing vaccination decisions among parents, logistic regression analysis was applied. We calculated odds ratios with 95% confidence intervals, excluding incomplete response sets. The determinants were considered significantly associated with the outcome if p-value was less than 0.05. All statistical calculations were made using MedCalc for Windows (version 23.2.0).

3.3 Assessing the frequency and risk factors of asymptomatic carriage of *Neisseria meningitidis*

Our study examining meningococcal carriage was conducted with the participation of 610 healthy adolescents and young adults between ages 15-31 from November 2017 through December 2018. The participants were nearly evenly divided between Budapest public high school students (307 individuals, 50.3%) and Semmelweis University third-year

students (303 individuals, 49.7%). Participants completed an anonymous questionnaire to assess potential meningococcal carriage risk factors.

We collected oropharyngeal swab samples and transported them to the laboratory in Transwab Charcoal medium at room temperature. DNA extraction was performed using QIAamp BiOstic Bacteremia DNA Kit. Final DNA concentration measurements were taken using a NanoDrop Lite spectrophotometer, then samples were stored at -80°C until analysis. The presence of *Neisseria meningitidis* was determined with real-time PCR detection of the species-specific *sodC* gene. For serogroup identification (A, B, C, X, Y, W) we used serogroup-specific gene detection following WHO and CDC protocol. Amplifications were conducted in 25µl volumes in 96-well plates, run in triplicate using LightCycler® 96 Real-Time PCR System and qTOWER3G thermal cycler, with high-sensitivity BioTaq polymerase.

For the statistical analysis we used univariate logistic regression to calculate unadjusted odds ratios, with 95% confidence intervals for colonization risk factors, with significance defined as $p < 0.05$. Comparison of variable prevalence between high school and university students two-sided Chi-squared tests were performed, with MedCalc for Windows version 22.014.

4. Results

4.1 Examination of the attitudes towards varicella vaccinations

Parent respondents were predominantly females, between 30-39 years old, university educated, and with good financial status. Participants represented various residential areas across Hungary. Healthcare professional respondents were almost entirely female health visitors working throughout different regions of Hungary.

Among parent participants, 53.3% (555) had vaccinated at least one child against varicella, with 46.3% (482) having vaccinated all their children,

7.0% (73) some of their children, and 46.7% (487) none of their children. Within the healthcare professional group, 76.3% (148) endorsed universal varicella vaccination, including all five pediatricians (100%) and 75.7% of health visitors.

Parents with one child showed the highest vaccination rates (53.3%), which decreased significantly in families with three or more children (46.2%). Significantly higher vaccination rate was associated with parents aged 30-39 (58.0%, vs. only 48.8% for parents under 30 years of age and 44.3% over the age of 40), those holding university degrees (60% compared to 42.9% for parents who finished high school and 42.0% for parents who finished elementary school), and urban residents (65.8% in the capital city versus 38.5% in villages). Financial circumstances, respondent gender, and personal history of varicella did not significantly impact vaccination decisions, though male participants were underrepresented. Vaccination likelihood increased significantly among parents who considered varicella severe (77.7% vaccination rate) and those who had witnessed disease complications. Healthcare professional recommendation emerged as the strongest predictor of vaccination, with 77.8% of parents vaccinating after receiving such advice.

Of the 487 non-vaccinating parents, 365 provided explanations for their decision. The primary reasons included: perception that varicella was not serious enough to warrant vaccination (33.7%), concerns about vaccine side effects (31.0%), doubts about vaccine efficacy (19.7%), insufficient knowledge about the vaccine (15.6%), negative recommendations from healthcare providers (11.8%), financial constraints (8.8%).

Among the 228 parents who initially declined but later decided to vaccinate subsequent children, the major factors changing their minds were unexpectedly severe disease in their unvaccinated child and improved vaccine availability. Conversely, 76 parents who vaccinated their first child but declined it for later children typically cited

breakthrough infection despite vaccination or adverse reactions to the vaccine as their reason.

Support for universal varicella vaccination was higher among healthcare professionals who were younger (under the age of 30), worked in the capital, and had no children. The perception of varicella severity strongly predicted support, with 100% of those viewing it as severe disease supporting vaccination. Direct experience with disease complications significantly increased support rates, which rose with the frequency of witnessed complications.

Among the 46 health visitors (23.7%) who opposed universal vaccination, key objections included viewing varicella as not severe enough to justify vaccination, skepticism about vaccine effectiveness, and concerns about adverse effects.

4.2 Examination of attitudes towards *Neisseria meningitidis* vaccination

Among the 159 participating parents, who responded for the main outcome of the study, 78.6% (125) had vaccinated at least one child against meningococcal disease. The majority of parents, 71.7% (114), had vaccinated all their children, 6.9% (11) chose to vaccinate some of their children, while 21.4% (34) decided not to vaccinate any of their children.

The number of children had no influence on parental decision regarding meningococcal vaccination, as parents with 1, 2, and 3 or more children showed 79.2%, 80.0% and 79.3% vaccination rates. Age did not have a significant effect on vaccination rates, but the pattern showed that younger adults are more likely to vaccinate their children, as vaccination rate moderately decreased with increasing age (87.5% under the age of 30 years, 81.9% between 30-39, and 70.0% over the age of 40). Though male participants were underrepresented in our study, we did not detect significant association between gender and vaccination rates.

Significantly higher vaccination rate was associated with those holding higher education/university degrees (84.7% compared to 63.8% for parents who finished high school or lower education), and urban residents (89.7% in the capital city versus 69.2% in villages). We found that 92.9% of participants with very good financial status and 79.5% of parents with good financial status vaccinated their children, whereas only 63.6% of those with low financial status decided to vaccinate, though this reduction was not statistically significant. Interestingly, personal history of IMD did not impact vaccination decisions, moreover, those parents who did not know someone with meningococcal disease showed slightly higher vaccination rate. Vaccination rate increased significantly among parents who considered meningococcal disease severe (88.3% compared to 51.9% for those finding meningococcal disease moderate, and 36.4% for those considering it mild or do not know the severity). Healthcare professional recommendation was the strongest predictor of vaccination, as 94.7% of parents decided to vaccinate after receiving such advice. Only 37.8% of parents vaccinated their children against meningococcal disease, when they did not get healthcare professional recommendation. We also found that parents with health-related studies are significantly more likely to vaccinate their children against meningococcal disease (89.9% versus 67.9%).

Out of the 34 parents who decided not to vaccinate their children, 31 provided explanations for their decision. The most common reason was insufficient knowledge about the vaccine (45.2%), followed by the lack of recommendation from their pediatrician (29.0%) and finding it unnecessary (16.1%). Concerns about vaccine side effects, doubts about vaccine efficacy and financial constraints were mentioned only in 9.7% of answers.

4.3 Assessing the prevalence and risk factors of asymptomatic carriage of *Neisseria meningitidis*

Most participants were non-smokers (82.8%), lived in households without passive smoke exposure (66.2%), had not used antibiotics in the previous two months (81.3%), reported no recent upper respiratory infections (58.7%), and did not regularly attend crowded social gatherings (63.0%). While most characteristics were comparable between high school and university students, significant differences emerged in two areas: university students reported higher rates of party attendance (37.0% compared to 22.8% for high school students, $p=0.0001$), while high school students showed higher meningococcal vaccination coverage (17.9% versus 10.2% for university students, $p=0.0063$).

Regarding vaccination history, most participants (50.7%) were uncertain about their meningococcal vaccination status. Among the 86 participants (14.4%) with confirmed vaccination, 41.9% had received monovalent vaccines targeting serogroup C (Meningitec, Menjugate, NeisVac-C), 24.4% had received tetravalent vaccines against ACWY serogroups (Mencevax, Menveo, Nimenrix), and 9.3% had been vaccinated against serogroup B (Bexsero). Six participants reported receiving multiple vaccine types, while 25 could not identify their specific vaccine.

The study identified 212 carriers of *Neisseria meningitidis* among the 610 tested students, representing 34.8% carriage rate (95% CI: 31.0-38.5%). Gender differences were significant, with males showing higher carriage rates than females (42.4% versus 33.1%, $p=0.0279$). High school students demonstrated significantly higher colonization rates than university students (48.9% versus 20.5%, $p < 0.0001$), accounting for 70.8% of all positive cases. Age emerged as a strong predictor of meningococcal carriage, with the 17-19 age group showing the highest rate at 49.8% (147/295).

Several factors were associated with lower carriage rates, though without statistical significance: smoking (OR 0.73), household passive smoke exposure (OR 0.96), and recent antibiotic use (OR 0.72). No association

was found between carriage status and recent upper respiratory infections. Regular attendance at social gatherings showed a slight, non-significant negative association with colonization (OR 0.80). Vaccination status did not significantly affect carriage rates. Among high school students, number of siblings became relevant only with larger families; those with four or more siblings showed particularly high carriage rates (74.2%, OR 1.27).

The majority, 87.3% of carriage isolates (n=185) were non-typable by real-time PCR, representing a 30.3% NT carriage rate in the overall study population. Serogroup B was identified in 9.0% (n=19) of all isolates, with significantly higher prevalence among high school students compared to university students (11.8% versus 3.2%, $p=0.0282$). Serogroup C accounted for 2.4% of all carriage isolates, with no significant difference between university and high school students (3.2% versus 2.0%, $p=0.6007$). Serogroups A, X, and W were not detected in the study population. From the vaccinated carriers 5 were colonized with vaccine-type meningococci: four students vaccinated against serogroup C carried serogroup B, and one student with menACWY vaccination carried serogroup C meningococcus.

5. Conclusions

5.1 Attitudes towards varicella vaccination

Our research about varicella vaccination attitudes revealed correlations between several factors that can influence vaccination decisions, such as socioeconomic background, healthcare professional recommendation, and parents' information about the disease, its complications and about vaccines. Despite the vaccine not being provided for free in Hungary at the time of our survey, we found a relatively high vaccination rate among participating parents.

Parents with university degree and those living in the capital demonstrated higher vaccination rates, showing that educational level

and the type of settlement significantly influence vaccine uptake, probably due to better access to health and vaccination related information. Financial status of the family is another important factor, we found higher vaccination rates between parents with good financial status, suggesting that the cost of the vaccine can strongly influence parental decisions. Healthcare professional recommendation emerged as the strongest positive predictor of vaccination, as vaccination rate decreased significantly when parents did not get recommendation from them. This result shows that healthcare professionals play a crucial role in parental vaccination decisions.

Among vaccine-hesitant parents, besides finding the vaccine unnecessary, and being afraid of side effects, insufficient information, questioning vaccine efficacy, and not finding varicella severe were common reasons for not to vaccinate. These suggest that reliable information could lower vaccine hesitancy, and that public education is needed about this topic.

Healthcare providers generally supported universal varicella vaccination, especially those who recognized varicella as a serious disease and those who saw complications of varicella. However, concerns about vaccine necessity, efficacy, and potential side effects remained in some of them, suggesting that not only the general public, but professionals also need targeted education about the risks of natural infection, and about vaccine safety and efficacy.

5.2 Attitudes towards meningococcal vaccination

We found that vaccination rates were relatively high despite that the vaccine is not available for free, moreover, MenB vaccines have quite high price.

As key determinants leading to higher vaccination rates, we identified higher education/university degree and living in the capital, both probably providing better access to health-related information. Most

significant positive determinant for vaccination was healthcare professional recommendation, confirming the same finding in our previous study. Additionally, parents with health-related educational backgrounds showed significantly higher vaccination rates, likely due to greater knowledge about IMD and vaccines in general.

Primary reasons from parents for not vaccinating their child were insufficient information, lack of pediatrician recommendation, and finding the vaccine unnecessary. Affordability, fear of side effects, and questioning vaccine efficacy were less frequent reasons. Parents' view on the severity of meningococcal disease also strongly influenced vaccination decisions. All these highlight the importance of public health education about IMD severity and about vaccines to prevent it.

Enhancing healthcare professional education about meningococcal vaccination and communication is essential, as their recommendations strongly influence parental decisions. Targeted public educational campaigns addressing disease severity and vaccine efficacy could result in improved parental vaccine acceptance. To further increase vaccination rates consideration should be given to financial assistance programs or to implementation of meningococcal vaccines into the national immunization program.

5.3 Asymptomatic meningococcal carriage in high school and university students

Our study is the first Hungarian meningococcal carriage report. We enrolled numerous participants of at-risk age groups across different educational levels. We found considerably high prevalence of *Neisseria meningitidis* carriage among Hungarian adolescents and young adults, compared to previous international studies. Our study indicated significant difference in carriage rate between high school students and university students, which shows the complexity of meningococcal colonization in young adults. Age emerged as an independent risk factor, with peak colonization occurring in the 17–19-year-olds, confirming

previous findings on the epidemiology of *N. meningitidis* carriage. We found that gender is also a significant determinant, with males showing higher carriage rates, which is in accordance with other international studies. Other factors frequently associated with meningococcal carriage, such as smoking, passive smoking, recent respiratory infections, recent antibiotic use, party attendance, and vaccination status did not show significant association in our study.

The predominance of non-groupable meningococci in carriers aligns with other studies describing carriage. Among groupable strains, the distribution reflected the Hungarian IMD epidemiology, with serogroup B and serogroup C dominating, followed by serogroup Y.

There are various findings in literature regarding the relationship between meningococcal vaccination and meningococcal carriage. In our study we had limited information about vaccination status, so we cannot make definitive conclusions, however, we found that vaccination does not necessarily influence carriage, as half of the MenC vaccinated students remained carriers, although MenACWY vaccines showed some protection. Combined vaccines need further investigation, as we found that there were no carriers between those who received two different types of meningococcal vaccinations.

These findings provide important information to the understanding of meningococcal carriage dynamics in Hungary, and in Europe, highlighting the importance of region-specific epidemiological monitoring. The high carriage rates found in our study support recommending broad-spectrum meningococcal vaccination for Hungarian adolescents and young adults. As this research predates COVID-19 social distancing measures, it provides valuable baseline data for future investigations.

6. Bibliography of publications

6.1 Publications related to the topic of the thesis

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6.2 Publications not related to the topic of the thesis

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6.3 Conference presentation

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6.4 Conference poster

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