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Effectiveness of Immunoprophylaxis in Suppressing Carriage of *Neisseria Meningitidis* in the Military Environment

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Abstract

Neisseria meningitidis, etiological factor of invasive meningococcal disease, is a human commensal that colonizes the nasopharynx. Colonization is usually asymptomatic, but it is a prerequisite for disease. Asymptomatic carriers are the major source of infection. In the present study, a survey of *N. meningitidis* carriage was conducted between January and March 2013 in a military unit in Poland. Single-time throat culture samples were collected from professional 559 soldiers (302 unvaccinated vs. 257 vaccinated individuals with the quadrivalent conjugate vaccine AC YW-135). Bacterial identification was performed with classic microbiological methods (culture, incubation, identification). Non-culture method (PCR) was used for confirmation of detected strains of *N. meningitidis* and determination of serogroups. We found 29 carriers in the group of unvaccinated soldiers (9.6 % of examined individuals) whereas among vaccinated soldiers only 3 persons were carriers of *N. meningitidis* (1.2 %). The most frequently identified serogroups among the carriers serving in the same military facility were serogroup B (28 %), followed by Y (25 %), and C (22 %). In conclusion, the initiation of mass vaccination with the quadrivalent conjugate vaccine ACYW-135 in the military environment seems an effective method of suppressing *N. meningitidis* carriage.

Keywords

Carriage • *Neisseria meningitidis* • Professional soldiers • Vaccination

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1 Introduction

Neisseria meningitidis are gram-negative, aerobic diplococci which cause severe infections in the form of bacterial meningitis and sepsis, defined as an invasive meningococcal disease (IMD). Bacteria colonize the nasopharynx and spread through inhalation of droplets of respiratory secretions or through a direct contact. The sources of infection are carriers and infected individuals. Humans are the only natural reservoir of *N. meningitidis* (Rosenstein et al. 2001). In most cases, the IMD is transmitted from asymptomatic nasopharyngeal carriers of *N. meningitidis* strains, who account for 5–10 % of the general population (Yazdankhah and Caugant 2004). In closed environments such as prisons, boarding institutions, orphanages, and military camps, meningococcal carriage may reach 40–80 % (Cartwright et al. 1991). Some studies have shown that meningococcal carriage prevalence differs across age groups, increasing during childhood and peaking in 15–24-year-olds (Soriano-Gabarro et al. 2011; Rosenstein et al. 1999). Another surveys assessed carriage across all ages, with increasing prevalence through childhood: from 4 to 5 % in infants to the peak of 23–27 % in 19-years-old persons and subsequently decreasing prevalence in adulthood to 7–8 % in 50-year-old persons (Christensen et al. 2010). Most of *N. meningitidis* strains in the population of asymptomatic carriers are not pathogenic. Only a minority of the nasopharyngeal isolates penetrate the human mucosa and gain access to the bloodstream, causing invasive disease (Rosenstein et al. 2001). Despite the opportunity to implement antibiotic therapy at an early stage of the disease and the development of intensive care facilities, the IMD remains one of the most severe contagious diseases in the world, with mortality reaching 10–13 %, and in case of a septic shock – as much as 50 % (Caugant et al. 1994). Invasive infections caused by *N. meningitidis* are a serious public health problem worldwide and have a heavy economic impact, not only in epidemic areas but also in regions where it occurs sporadically. It is estimated that approximately 500,000 cases and 50,000 deaths due to IMD occur worldwide every year (Wilder-Smith 2007). The incidence of invasive disease due to *Neisseria meningitidis* is highly variable according to geographical area and serogroup distribution (Panatto et al. 2011). The biochemical composition of the polysaccharide capsule determines the serogroups of meningococcal strains. There are usually 13 serogroups described, but the WHO reports 12 serogroups (Harrison et al. 2009). Of the 12 different polysaccharide capsular types, only six (A, B, C, W135, Y, and X) frequently cause disease globally. The major disease burden is in developing countries (e.g., 'the meningococcal belt' in Africa); in industrialized communities the IMD occurs sporadically. The European Surveillance System has revealed a considerable variability from one country to another in the incidence of meningococcal disease. The serogroups mostly associated with invasive cases are B and C, but serogroups W-135 and Y are also present, while serogroup A is only responsible for sporadic cases. The IMD due to serogroups Y and W-135, uncommon in most European countries, contributes to 10–23 % of all cases in Scandinavia and Slovenia (Trotter et al. 2007). In the US, serogroup Y is a major cause of meningococcal disease, accounting for more than one third of all cases (Bona and Guidi 2012). Serogroup X determines substantially invasive disease in sub-Saharan Africa, rarely in other parts of the world (Harrison et al. 2009). In recent years, especially in Europe, the incidence of invasive disease caused by serogroup C has declined owing to the introduction of vaccination programs with conjugated vaccine C in children and adolescents in some countries (Cohn et al. 2010). Immunoprophylaxis seems one of the most effective methods of inducing herd protection by preventing nasopharyngeal meningococcal acquisition. Mass vaccinations are particularly important in closed communities, where the risk of increasing meningococcal carriage and invasive disease is exceptionally high. An example of such a community are soldiers staying in military barracks, who living in crowded conditions, together with people from different

geographic areas, have contact with diverse strains of *N. meningitidis* (Brundage et al. 2001). The international medical literature often reports on prevalence of meningococcal carriage and the use of immunoprophylaxis among new military recruits. However, there have only been a few reports on professional soldiers who, unlike recruits, are not living in military barracks, and rarely eat their meals in military canteens, which causes that the risk of meningococcal carriage, just like in the general population, is much lower among professionals than among recruits.

The aim of the study was to assess the prevalence of meningococcal carriage among professional soldiers, the effectiveness of immunoprophylaxis in suppressing the carriage of *N. meningitidis* in the military environment, and the serogroups of *N. meningitidis* on the basis of classic and non-culture microbiological methods.

2 Methods

2.1 Study Population

The study was approved by the Ethics Committee of the Military Institute of Medicine in Warsaw, Poland. The study of *Neisseria meningitidis* carriage was conducted in the military unit in Poland (25th Air Cavalry Brigade in Tomaszów Mazowiecki) between January and March 2013. The single-time throat culture samples were collected from 559 professional soldiers. They were on 8-h duties 5 days a week on the premises of the military unit, while the rest of time they stayed at homes, outside the military environment with the exception of 24-h duties assigned, on average, once a month. Among the 559 soldiers, 302 had never been vaccinated against meningococcal disease, while 257 had been vaccinated over the past 1–3 years with the quadrivalent conjugate vaccine ACYW-135. Apart from the data relating to vaccinations, socio-demographic and behavioral variables of soldiers, such as age, sex, place of residence, smoking of cigarettes were also assessed.

2.2 The Carrier Investigation

2.2.1 Classic Microbiological Laboratory Methods

Biological material was collected single-time from professional soldiers in a military unit (always by the same health care worker throughout the whole study period), on swabs from the back of the nasopharynx. Next, the samples were transported to a microbiological laboratory in the Military Institute of Medicine in Warsaw, Poland, where they were plated onto appropriate medium (Columbia agar with 5 % sheep blood, chocolate agar + PolyVitex VCAT3) in a laminar flow chamber. The plates were incubated in increased atmospheric CO_2 concentration at 37 °C for 48 h. After incubation, the colonies grown were macroscopically evaluated. ATCC (MicroBioLogics, St. Cloud, Minnesota, USA) living reference strains including *N. meningitidis* ATCC 13077 (serogroup A), ATCC 13090 (serogroup B), ATCC 13102 (serogroup C) were used for quality control. Colonies morphologically similar to the reference strains were isolated onto chocolate agar + PolyVitex (PVX) plates, and then incubated for 24–48 h (depending on the growth of microorganisms), in CO_2 atmosphere at 37 °C. The incubated pure colonies of bacteria (transparent, round, and slightly convex) were used to make gram-stained preparations, which were then observed under a light microscope (Gram-negative granuloma, most often in the shape of diplococci). Subsequently, catalase and cytochrome oxidase tests were performed (indicative tests ID Color Catalase, Oxidase Reagent; bioMerieux Polska, Warsaw, Poland). The identification was carried out by means of API NH biochemical sets and an automated system for identification of microorganisms using NH cards in compliance with the manufacturer's instructions (bioMerieux Polska, Warsaw, Poland). The strains identified as *N. meningitidis* were stored frozen in a temperature of −20 °C in Viabank, and then transported to the National Reference Center for Bacterial Meningitis in the National Medicines Institute in Warsaw, Poland, for re-identification and subsequent

characterization of isolates by means of molecular methods.

2.2.2 Non-culture Laboratory Methods

DNA isolated from detected strains was used for polymerase chain reactions (PCR) to identify *N. meningitidis* to the species level by presence of *crg*A and *ctr*A genes and to capsular group level with primers specific for the genogroups A, B, C, W-135, and Y. The relatedness of isolates was determined by the restriction fragment length polymorphism (RFLP) analysis of genomic DNA in pulsed-field gel electrophoresis (PFGE), using *Spe*I restriction enzyme for DNA digestion. RFLP-PFGE patterns were analyzed using Bionumerics Software Package (Applied Maths NV, Sint-Martens-Latem, Belgium) and a dendrogram was constructed using the Dice coefficient of similarity and cluster analysis with the unweighted-pair group method with arithmetic averages (UPGMA). Both the position tolerance and the optimization were set up at 1 %.

2.3 Statistical Elaboration

The quantitative variables were characterized by arithmetic means ±SD or median, min/max (range) and 95 % confidence interval. The qualitative variables were presented in the absolute or percentage terms. Normality of distribution was checked with the Shapiro-Wilk test and homogeneity of variances with the Leven (Brown-Forsythe) test. Significance of differences between two unmatched groups was checked with an unpaired *t*-test (or Welch test in case of lack of homogeneity) or Mann-Withney *U* test. A paired *t*-test or Wilcoxon signed-rank test was used for matching groups, as required. For more than three unmatched groups, one-way ANOVA or Kruskal-Wallis tests were used, followed by *post hoc* Tukey's or Dunn's test, respectively. For matched groups, one-way ANOVA for repeated measurements or Friedman test was used, as required. Chi-squared tests for independence were used for qualitative variables, with Yates' correction for cell counts below 10 and a check of Cochrane's conditions for Fisher's exact test. To determine dependence, strength, and direction between variables, Pearson or Spearman's correlation coefficients were determined. Statistical significance was set at $p \leq 0.05$. The analysis was performed using a Statistica vr. 10.0 commercial package.

3 Results

The results revealed 29 carriers of *N. meningitidis* among the 302 unvaccinated (9.6 %) and 3 carriers (1.2 %) among the 257 vaccinated soldiers. There were no significant differences relating to age, military rank, or place of residence between carriers and non-carriers of *N. meningitidis*. Regression analysis indicated that females were 2.6-fold times more likely to become carriers of *N. meningitidis,* smoking cigarettes increased the risk of carriage, and vaccination reduced it. Among the carriers, there were significantly more females than males, more smokers than non-smokers, and more soldiers vaccinated against meningococcal infections than unvaccinated ones (Table 1).

In a group of 529 soldiers who were identified as non-carriers of *N. meningitidis*, the vaccinated soldiers were significantly older. There were notably more non-commissioned officers among the vaccinated. There were no other significant differences in the socio-demographic and behavioral variables between non-vaccinated and vaccinated individuals (Table 2), nor were there any such differences among 32 soldiers identified as carriers of *N. meningitides* (Table 3).

Testing at the National Reference Center for Bacterial Meningitis showed that all isolates belonged to *N. meningitidis* species and possessed *crg*A gene, but in two cases PCR product for *ctr*A gene was not detected. Genogrouping revealed that among the 32 carriage isolates, 9 belonged to genogroup B (28.1 %), 8 to genogroup Y (25.0 %), and 7 to genogroup C (21.9 %). Isolates belonged to rare genogroups were nongroupable in 8 cases (25.0 %), including 2 isolates with the lack of PCR product for *ctr*A gene. Among three vaccinated soldiers being

Table 1 Sociodemographic and behavioral variables in non-carriers and carriers of *Neisseria meningitidis*

Sociodemographic and behavioral variables	Non-carriers of *N. meningitidis* (*n* = 527)	Carriers of *N. meningitidis* (*n* = 32)	p
Age			
Mean ± SD	30.2 ± 4.8	29.3 ± 4.4	0.407
Range	21.0–52.0	22.0–43.0	
Median	29.0	29.0	
95 % CI	29.8–30.6	27.7–30.9	
Military rank			
Officer	30 (5.7 %)	3 (9.4 %)	0.093
Noncommissioned officer	137 (26.1 %)	3 (9.4 %)	
Private	360 (68.3 %)	26 (81.2 %)	
Sex			
Female	7 (1.3 %)	2 (6.3 %)	0.032
Male	520 (98.7 %)	30 (93.7 %)	
Place of residence			
City	341 (64.7 %)	21 (65.6 %)	0.916
Country	186 (35.3 %)	11 (34.4 %)	
Smoking of cigarettes			
Yes	174 (33.0 %)	22 (68.7 %)	0.001
No	353 (67.0 %)	9 (31.3 %)	
Vaccination			
Vaccinated	250 (47.4 %)	3 (9.4 %)	0.001
Non-vaccinated	277 (52.6 %)	29 (90.6 %)	

p values for the differences between non-carriers and carriers of *Neisseria meningitidis*

Table 2 Sociodemographic and behavioral variables between non-vaccinated and vaccinated with conjugate meningococcal vaccine A, C, Y, and W-135 non-carriers of *N. meningitidis*

Sociodemographic and behavioral variables	Non-vaccinated non-carriers (*n* = 277)	Vaccinated non-carriers (*n* = 250)	p
Age			
Mean ± SD	29.1 ± 4.4	31.4 ± 5.0	0.001
Range	21.0–46.0	22.0–52.0	
Median	28.0	30.0	
95 % CI	28.6–29.6	30.8–32.0	
Military rank			
Officer	14 (5.1 %)	16 (6.4 %)	0.001
Noncommissioned officer	54 (19.5 %)	83 (33.2 %)	
Private	209 (75.4 %)	151 (60.4 %)	
Sex			
Female	5 (1.8 %)	2 (0.8 %)	0.314
Male	272 (98.2 %)	248 (99.2 %)	
Place of residence			
City	174 (62.8 %)	167 (66.8 %)	0.339
Country	103 (37.2 %)	83 (33.2 %)	
Smoking of cigarettes			
Yes	89 (32.1 %)	85 (34.0 %)	0.649
No	188 (67.9 %)	165 (66.0 %)	

p values for the differences between non-vaccinated vs. vaccinated individuals

Table 3 Sociodemographic and behavioral variables between non-vaccinated and vaccinated carriers of *N. meningitidis*

Sociodemographic and behavioral variables	Non-vaccinated carriers of *N. meningitidis* ($n = 29$)	Vaccinated carriers of *N. meningitidis* ($n = 3$)	p
Age			
Mean ± SD	29.2 ± 4.6	30.0 ± 1.0	0.516
Range	22.0–43.0	29.0–31.0	
Median	29.0	30.0	
95 % CI	27.5–31.0	27.5–32.5	
Military rank			
Officer	3 (10.3 %)	0	0.683
Noncommissioned officer	3 (10.3 %)	0	
Private	23 (79.4 %)	3 (100 %)	
Sex			
Female	2 (6.9 %)	0	0.639
Male	27 (93.1 %)	3 (100 %)	
Place of residence			
City	20 (69.0 %)	1 (33.3 %)	0.216
Country	9 (31.0 %)	2 (66.7 %)	
Smoking of cigarettes			
Yes	19 (65.5 %)	3 (100 %)	0.220
No	10 (34.5 %)	0	
Serogroup of *N. meningitidis*			
B	7 (24.1 %)	2 (66.7 %)	0.555
Y	8 (27.6 %)	0	
C	7 (24.1 %)	0	
A, B, C, W, Y (−)[a]	7 (24.1 %)	1 (33.3 %)	

p values for the differences between non-vaccinated vs. vaccinated individuals
[a]Serogroups A, B, C, W-135 or Y have not been identified

meningococcal carriers, two carried isolates of genogroup B and one meningococci not belonging to any of the above outlined genogroups.

Patterns generated by PFGE of *Spe*I-digested DNA of 32 meningococcal isolates were classified into 25 PFGE types. Among them, two types were subdivided into two PFGE subtypes. Isolates of genogroups B and C were heterogenous because except for one pair of meningococci in each genogroup, they possessed dissimilar PFGE patterns. Eight isolates of genogroup Y were divided into five PFGE types, including three patterns (1, 2, and 3) showing more than 80 % similarity. Three Y meningococci had the same PFGE pattern (type 4A) and were also closely related to one isolate not belonging to the genogroups A, B, C, W-135, and Y, with PFGE type 4B (Fig. 1).

4 Discussion

Studies into the prevalence of meningococcal carriage in the military environment are widely available in international medical literature. However, they are limited to one type of community only, i.e., young recruits who had just been drafted into the military. Compulsory military service was abolished in some European countries, including Poland in 2009, which led to professionalization of the national armed forces. The changes caused that the age of soldiers has risen (19–20 year old recruits have been replaced by 25–30 year and older professional privates). Recruits used to serve in a given military facility 24 h a day, 7 days a week, and they were provided with full board in military canteens. Professional soldiers,

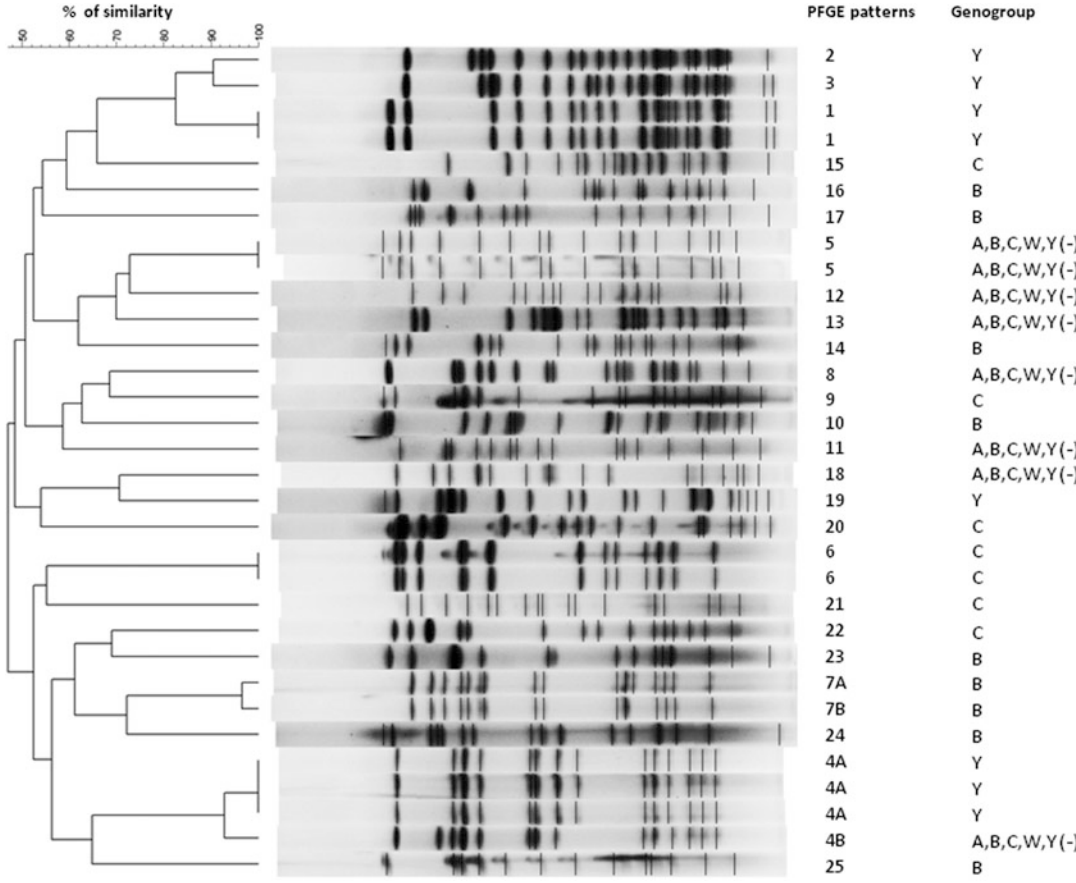

Fig. 1 Dendrogram of similarity obtained with Dice coefficients and arithmetic averages (UPGMA) clustering method, using BioNumerics software, showing the relatedness of meningococcal isolates by PFGE (n = 32)

on the other hand, work for 8 h a day and they are not provided with board and accommodation on the premises of a military unit, with the exception of infrequent 24-h duties and military exercise or military operation. In fact, professional military service has changed from 24 h/7 days service in crowded and close contact conditions to a typical regular job.

Data on the rate of *Neisseria meningitidis* carriage among recruits serving in European armed forces show a high carriage prevalence, regardless of the size of a given study group or country of origin. Recruits constitute a high risk group for meningococcal carriage and invasive disease, with a reported incidence of four to ten times greater than that of the general population (Biselli et al. 1993). Two longitudinal surveys of *N. meningitidis* carriage were conducted in Polish military recruits in non-outbreak settings in 1998 (n = 151 and n = 168). Overall, carriage prevalence in these studies was 36 and 61 %, respectively (Tyski et al. 2001). Andersen et al. (1998) have described that the carriage rate among 3 cohorts of 1,069 Danish recruits was constant over time and season at the level of 39–47 %. The meningococcal carriage surveillance study was conducted between November 1999 and March 2000 among 1,179 German recruits in 6 military camps in Bavaria. Three hundred and eighty-four soldiers (32.6 %) were carriers of *N. meningitidis* (Claus et al. 2005). In Norway, meningococcal carriage study in 126 military recruits was performed between February and July 2003. A total of 78 carriers

(61.9 %) was identified (Caugant et al. 2007). In the present study, carriage of *N. meningitidis* among 559 Polish professional soldiers amounted to 32 individuals, which was 5.7 % of the study population; the rate comparable with that in the general population. Due to the progressive professionalization of the Polish Armed Forces, it is highly probable that meningococcal carriage in the military environment will considerably decrease. The examination of carriers revealed a substantial disproportion between non-vaccinated (9.6 %) and soldiers vaccinated with the quadrivalent conjugate vaccine ACYW-135 (1.2 % persons), which may confirm the effectiveness of immunoprophylaxis in suppressing carriage of *Neisseria meningitidis* and invasive meningococcal disease. Mass vaccinations carried out in some European armed forces also confirm this hypothesis. In the 1980s, high rates of meningococcal disease, mainly caused by serogroup C, were observed among Italian recruits. A mass immunization campaign against meningococcal infections has been carried out in the Italian army since 1987. Thanks to the program the disease prevalence has been largely reduced (Stroffolini 1990). In 1985, 52 cases of meningococcal disease were diagnosed in the Italian military (48 cases with serogroup C) and in 1991 only 1 case (serogroup B). The authors demonstrated that vaccination is highly effective as to seroconversion (18 days after immunization 84 and 91 % had seroconversion against serogroup A and C, respectively). The protective efficacy of the vaccine A + C was 91.2 % (12 cases of serogroup C and A from 150,000 unvaccinated and 1 case of serogroup C from 150,000 vaccinated Italian soldiers in 1987) (Biselli et al. 1993). In connection with a sharp increase in meningococcal disease prevalence among Israeli soldiers, the Department of Epidemiology of the Army Health Branch in Israel adopted an immunization policy with the quadrivalent vaccine for all recruits. As a result of the mass vaccination program, there has been a dramatic drop in the incidence of vaccine-preventable meningococcal disease (from 1.3 cases per 100,000 person-years in 1983–1994 in the period preceding the start of immunization to 0 cases in 1995–2007). From 1983 to 2007, 42 cases of laboratory-confirmed meningococcal disease were reported, all caused by serogroup B after the onset of the vaccination program (Mimouni et al. 2010). According to Panatto et al. (2011), since the introduction of vaccination programs with conjugated vaccine C in children and adolescents, most cases of invasive meningococcal disease in reported countries have been caused by meningococcus B. It is important to underline that invasive meningococcal disease will not be controlled, until safe and effective vaccines for meningococcal B are available and widely used. Scientific publications issued in 2012 confirm the immunogenicity and safety of the 4CMenB (Bexsero, Novartis) vaccine against meningococcal disease caused by serogroup B. The European Medicine Agency registered this vaccine in the territory of European Union in January 2013. The Polish Armed Forces are considering the purchase of the vaccine for their troops. Serogroup B is still a leading cause of meningococcal carriage in the military environment. It was the most frequently identified serogroup (46 %) among 151 carriers of *N. meningitidis* diagnosed in French soldiers serving in the same military facility in 1991 (Chapalain et al. 1992). In 1998, 156 carriers were identified in the population of Polish recruits, 54 % of isolates were nongroupable, among the remaining strains serogroup B was predominant (32 % of all carrier strains) (Tyski et al. 2001). The meningococcal carriage surveillance studies which were conducted between 1999 and 2000 among German soldiers in military facilities revealed that serogroup B was also the most common (42 %) (Claus et al. 2005). In our present study, serogroup B was the most frequently identified as well (28 %). According to most surveys, roughly 50 % of the strains isolated from meningococcal carriers are not serogroupable (Yazdankhah and Caugant 2004; Yazdankhah et al. 2004). In this study, we were able to establish the genogroup for 75 % of isolates tested. Interestingly, we found a high percentage of genogroup Y vs. C carriage in comparison with the general population tested in the Czech Republic, Greece, and Norway:

25.0 % vs. 10.2 % and 21.9 % vs. 4.8 %, respectively (Yazdankhah et al. 2004). An increase in carriage frequency of serogroup Y was recently indirectly reported in young adult population in the UK. The authors concluded that the carriage rise may help explain the recent growth in the incidence of serogroup Y disease in the country, as it was the case during the late 1990s in the US, where a similar increase in serogroup Y carriage was linked to a concomitant increase in serogroup Y disease (Ala'aldeen et al. 2011). Increasing prevalence of meningococcal carriage of serogroup Y demonstrated in this study among non-vaccinated soldiers as well as in other surveys seems to confirm the validity of implementing the quadrivalent conjugate vaccine ACYW-135, which may induce immunological response in humans and facilitate the flow of antibodies to the nasopharyngeal mucosa and eventually affect the colonization of *Neisseria meningitidis*. Additional carriage studies, including extensive molecular strain characterization, should be performed before and after vaccination in countries where mass vaccination programs have been implemented (Caugant et al. 2007). Apart from the analysis of serogroups, it is also important to assess the risk factors affecting the rise in *N. meningitidis* carriage. Smoking, active as well as passive, is one of the strongest risk factors for becoming a meningococcal carrier (Stuart et al. 1989). This has been confirmed by our studies carried out in Polish soldiers. Some studies have also demonstrated a coincidence between meningococcal carriage and symptoms of upper respiratory tract infections. There are also slightly more carriers in males than females and a low socioeconomic status appears to increase the risk factors for carriage (Caugant et al. 2007). Our research revealed that respiratory tract infections occurred sporadically among carriers of *N. meningitidis*, while analyzing gender of soldiers we demonstrated that females exhibit a higher risk for meningococcal carriage than males. Given that the meningococcal carrier state may be chronic, intermittent, or transient (Broome 1986), it is necessary to continue the research into the prevalence of *Neisseria meningitidis* carriage in the military environment taking into consideration its dynamics, risk factors, and serogroup characteristics.

5 Conclusions

The initiation of mass vaccination with the quadrivalent conjugate vaccine ACYW-135 in the military seems to be an effective tool in suppressing *N. meningitidis* carriage. Vaccination contributes to the elimination of carriage, but does eliminate serogroup B. Therefore, it is recommended that the 4CMenB vaccine should be introduced into the vaccination schedule. Surprisingly, a high prevalence of carried serogroup Y observed in the soldiers of this study can indicate the presence of this serogroup also in the general Polish population.

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Conflicts of Interest The authors declare no conflicts of interest in relation to this article.

References

Ala'aldeen DA, Oldfield NJ, Bidmos FA, Abouseada NM, Ahmed NW, Turner DP, Neal KR, Bayliss CD (2011) Carriage of meningococci by university students, United Kingdom. Emerg Infect Dis 17:1762–1763

Andersen J, Barthelsen L, Bech Jensen B, Lind I (1998) Dynamics of the meningococcal carrier state and characteristics of the carrier strains: a longitudinal study within three cohorts of military recruits. Epidemiol Infect 121:85–94

Biselli R, Fattorossi A, Matricardi PM, Nisini R, Stroffolini T, Amelio RD (1993) Dramatic reduction of meningococcal meningitis among military recruits in Italy after introduction of specific vaccination. Vaccine 11(5):578–581

Bona G, Guidi C (2012) Meningococcal vaccine evolution. J Prev Med Hyg 53:131–135

Broome CV (1986) The carrier state: Neisseria meningitidis. J Antimicrob Chemother 18(Suppl A):25–34

Brundage JF, Ryan MAK, Feighner BH, Erdtmann FJ (2001) Meningococcal disease among United States military service members in relation to routine uses of vaccines with different serogroup-specific components, 1964–1998. Clin Infect Dis 35:1376–1381

Cartwright KA, Stuart JM, Robinson PM (1991) Meningococcal carriage in close contact of cases. Epidemiol Infect 106:133–141

Caugant DA, Hoiby EA, Magnus P, Scheel O, Hoel T, Bjune G, Wedege E, Eng J, Froholm LO (1994) Asymptomatic carriage of *Neisseria meningitidis* in a randomly sampled population. J Clin Microbiol 32:323–330

Caugant DA, Tzanakaki G, Kriz P (2007) Lessons from meningococcal carriage studies. FEMS Microbiol Rev 31:52–63

Chapalain JC, Guibourdenche M, Perrier-Gros-Claude JD, Bartoli M, Riou JY (1992) The chemoprophylaxis of cerebrospinal meningitis using rifampin in a military population. Pathol Biol 40:230–233

Christensen H, May M, Bowen L, Hickman M, Trotter C (2010) Meningococcal carriage by age: a systematic review and meta-analysis. Lancet Infect Dis 10:853–861

Claus H, Maiden MC, Wilson DJ, McCarthy ND, Jolley KA, Urwin R, Hessler F, Frosch M, Vogel U (2005) Genetic analysis of meningococci carried by children and young adults. J Infect Dis 191:1263–1271

Cohn AC, MacNeil JR, Harrison LH, Hatcher C, Theodore J, Schmidt M, Pondo T, Arnold KE, Baubach J, Bennett N, Craig AS, Farley M, Gershman K, Petit S, Lynfield R, Reingold A, Schaffner W, Shutt KA, Zell ER, Mayer LW, Clark T, Stephens D, Messonnier NE (2010) Changes in *Neisseria meningitidis* disease epidemiology in the United States, 1998–2007: implications for prevention of meningococcal disease. Clin Infect Dis 50(2):184–191

European Medicines Agency. Bexsero. Available at: http://www.ema.europa.eu/ema/index.jsp?curl=pages/medicines/human/medicines/002333/human_med_001614.jsp&mid=WC0b01ac058001d124. Accessed 28 Jan 2013

Harrison LH, Trotter CL, Ramsay ME (2009) Global epidemiology of meningococcal disease. Vaccine 27 (Suppl 2):B51–B63

Mimouni D, Bar-Zeev Y, Huerta M, Balicer RD, Grotto I, Ankol O (2010) Preventive effect of meningococcal vaccination in Israeli military recruits. Am J Infect Control 38:56–58

Panatto D, Amicizia D, Lai PL, Gasparini R (2011) Neisseria meningitidis B vaccines. Expert Rev Vaccines 10:1337–1351

Rosenstein NE, Perkins BA, Stephens DS, Lefkowitz L, Cartter ML, Danila R, Cieslak P, Shutt KA, Popovic T, Schuchat A, Harrison LH, Reingold AL (1999) The changing epidemiology of meningococcal disease in the United States, 1992–1996. J Infect Dis 180:1894–1901

Rosenstein NE, Perkins BA, Stephens DS, Popovic T, Hughes JM (2001) Meningococcal disease. N Engl J Med 344:1378–1388

Soriano-Gabarro M, Wolter J, Hogea C, Vyse A (2011) Carriage of *Neisseria meningitis* in Europe: a review of studies undertaken in the region. Expert Rev Anti Infect Ther 9:761–774

Stroffolini T (1990) Vaccination campaign against meningococcal disease in army recruits in Italy. Epidemiol Infect 105:579–583

Stuart JM, Cartwright KA, Robinson PM, Noah ND (1989) Effect of smoking on meningococcal carriage. Lancet 2(8665):723–725

Trotter CL, Chandra M, Cano R, Larrauri A, Ramsay ME, Brehony C, Jolley KA, Maiden MC, Heuberger S, Frosch M (2007) A surveillance network for meningococcal disease in Europe. FEMS Microbiol Rev 31:27–36

Tyski S, Grzybowska W, Dulny G, Berthelen L, Lind I (2001) Phenotypical and genotypical characterization of Neisseria meningitidis carrier strains isolated from Polish recruits in 1998. Eur J Clin Microbiol Infect Dis 20:350–353

Wilder-Smith A (2007) Meningococcal vaccine in travelers. Curr Opin Infect Dis 20(5):454–460

Yazdankhah SP, Caugant DA (2004) *Neisseria meningitidis*: an overview of the carriage state. J Med Microbiol 53:821–832

Yazdankhah SP, Kriz P, Tzanakaki G, Kremastinou J, Kalmusova J, Musilek M, Alvestad T, Jolley KA, Wilson DJ, McCarthy ND, Caugant DA, Maiden MC (2004) Distribution of serogroups and genotypes among disease-associated and carried isolates of *Neisseria meningitidis* from the Czech Republic, Greece, and Norway. J Clin Microbiol 42:5146–5153