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Secretor status, smoking and carriage of *Neisseria meningitidis*

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SUMMARY

A survey of ABO blood groups, secretor status and smoking habits among 389 students and staff of a school in which there was an outbreak of meningococcal disease found no difference in the distribution of the ABO blood groups but a significantly higher proportion of non-secretors (37·6%) in the population examined compared with that reported for previous surveys of the neighbouring population in Glasgow (26·2%) ($P < 0·0005$). There was also a significantly higher proportion of non-secretors among carriers of meningococci (47%) compared with non-carriers (32%). Increased carriage of meningococci among non-secretors might contribute to the increased susceptibility of individuals with this genetic characteristic to meningococcal disease observed in previous studies. Although passive exposure to cigarette smoke has been associated with meningococcal disease, there was no association between passive smoking and carriage. There was, however, a significant association between active smoking and carriage.

INTRODUCTION

In December 1988 there was an outbreak of disease due to a serogroup B serotype 4 P1.15 sulphonamide-resistant strain of *Neisseria meningitidis* in Airdrie, Lanarkshire. There were five cases of the disease among students at the Airdrie Academy; one teacher developed the disease and died. Four of the cases occurred in the second year, three of these in one form. A child who attended another school was also infected.

During the week before the Christmas/New Year recess, teams from the Infectious Diseases Unit at Monklands District General Hospital and the Department of Community Medicine screened 1492 students and staff for carriage of meningococci, and issued each with a course of rifampicin (600 mg bd for 2 days). During the second week of January 1989 when the school had reopened, a selected group of students and staff was retested for carriage of meningococci and specimens of blood and saliva were also obtained. In addition, participants were interviewed about smoking habits or passive exposure to cigarette smoke that has been associated with meningococcal infections.

The genetically determined inability of an individual to secrete the glycoprotein form of his ABO blood group antigens (non-secretion) is associated with susceptibility to meningococcal infection [1]. Prolonged outbreaks of meningococcal disease have occurred in areas where there are higher proportions of non-secretors [2, 3]. In this study we determined if there is also a higher proportion of non-secretors in a population in which there was a single short outbreak of the disease. In the study in Stonehouse, Gloucestershire we found an increase in the proportion of non-secretors among carriers of meningococci within the community, but this was not significantly different from the proportion of non-secretors from whom no meningococci or *Neisseria lactamica* were isolated. In this study we determined if there is a higher proportion of non-secretors among carriers of meningococci within the school population.

Several environmental factors have been associated with susceptibility to meningococcal disease: passive exposure to cigarette smoke [4, 5]; overcrowded accommodation [6]; poverty and lower social class [7]; strenuous physical activity [5]; predisposing viral infections [8–10]. As the population affected was old enough to have enough smokers for statistical analysis, the effects of smoking or passive exposure to cigarette smoke on carriage of meningococci among students and staff was examined.

SUBJECTS AND METHODS

In December 1988, as part of an urgent public health measure to administer prophylactic antibiotics, 1492 staff and students at the Airdrie Academy were screened for carriage of meningococci by nasopharyngeal swabbing. In January 1989, 389 selected individuals at the school were screened for carriage of meningococci. From the second year class, 201 of the 226 pupils were examined as this was the year in which 4 of the 5 cases among the children occurred. From the other five classes, 85 carriers plus a group of 85 non-carriers from the same form matched as closely as possible for age and sex were examined: year one, 25 students; year three, 39 students; year four, 68 students; year five, 29 students; year six, 9 students. From the staff, there were ten members who were carriers matched as closely as possible for age and sex with eight non-carriers with similar duties in the school. Each participant supplied a saliva sample (5 ml), and 10 ml of blood was obtained from 371.

ABO and Lewis blood groups were determined by agglutination of blood specimens with monoclonal antibodies. Because the haemagglutination inhibition test for secretion with saliva can yield 'false' secretors if contaminated with blood, the Lewis test was used to determine secretor status: secretors express Lewis^b and non-secretors express Lewis^a. The nasopharyngeal swabs were cultured on GC selective medium (NYC/Thayer Martin medium) and *N. meningitidis* identified by Gram stain, oxidase test and the Gonochek II system (EY Laboratories). The serogroup, serotype and sensitivities to sulphonamide and rifampicin were determined at Ruchill Hospital, Glasgow.

Each participant in the January survey was interviewed to obtain information regarding smoking habits and exposure to cigarette smoke. The information was coded and analyzed with the laboratory results by a Database II programme. Because all the participants could not be matched for sex, the results were compared by the χ^2 test rather than McNemar's test.

RESULTS

Among the 1492 students and staff tested in December 1988 *N. meningitidis* was isolated from 126 (8·4%). There were 11 isolates which were serogroup B or non-groupable with increased resistance to sulphonamide and with the serotype and/or subtype of the outbreak strain; 6 of these were isolated from students in the form (2A) where 3 of the 5 cases occurred. In December none of the isolates was resistant to rifampicin; however, in January 3 of the 13 isolates, including 2 with characteristics of the outbreak strain, were resistant to rifampicin.

ABO blood group distribution and secretor status of the school population

Blood specimens were obtained from 371 students and staff. The distribution of the ABO groups is shown in Table 1; this does not appear to differ markedly from that observed in earlier studies for either Edinburgh [11] or Glasgow [12]. The distribution of the Lewis^a/non-secretors and the Lewis^b/secretors in the population tested does differ significantly from the figures reported earlier for Edinburgh [11] ($\chi^2 = 9·03$, $P < 0·005$) and Glasgow [13] ($\chi^2 = 12·14$, $P < 0·0005$) (Table 2). Among the 185 students in the second year class whose red cells expressed Lewis antigen, 65 (35%) were Lewis^a/non-secretors, a significantly higher proportion compared with the figure for nearby Glasgow ($\chi^2 = 4·81$, $P < 0·05$). In form 2A where 3 of the 5 cases among the students occurred, 13 of the 26 (50%) children tested were non-secretors; but this was not significantly increased compared with the other forms in year 2. Only 2 of the 5 cases were non-secretors.

Specimens were obtained from 109 individuals who had been carriers in December. There were 51 non-secretors (47%) among the carriers compared with 83 non-secretors among the 262 non-carriers (32%) ($\chi^2 = 4·94$, $P < 0·05$). The majority of these strains did not have the characteristics of the outbreak strain. Specimens were obtained from 10 of the 11 students who were carriers of isolates with characteristics of the outbreak strain; 6 of the 10 carriers were non-secretors.

Meningococci were isolated from 13 of the 389 tested in January and 5 of the carriers were non-secretors (38·5%). The outbreak strain was isolated from 2 students, one secretor and one non-secretor, both of whom had received rifampicin.

Smoking and carriage of meningococci

There was no association between passive exposure to cigarette smoke (father and/or mother smokers) and carriage; but there was an association between students' smoking habits and carriage. There were 17 carriers among the 37 students who were smokers (46%) compared with 87 carriers among 349 students who were non-smokers (25%) ($\chi^2 = 6·48$, $P < 0·025$) (Table 3). Four (31%) of the 13 students who were carriers in January were smokers. In the December survey, 6 of the 10 members of staff who were carriers were smokers, but none of the eight

Table 1. *Comparison of the distribution of ABO blood groups for the Airdrie Academy population with those of Glasgow and Edinburgh*

	Blood group							
	A		B		O		AB	
	No.	(%)	No.	(%)	No.	(%)	No.	(%)
Airdrie	118	(32)	47	(13)	198	(53)	8	(2)
Glasgow	1906	(32)	637	(11)	3177	(54)	178	(3)
Edinburgh	104	(31)	42	(13)	173	(52)	15	(4)

Table 2. *Comparison of secretor status of the Airdrie population with those of Glasgow and Edinburgh*

	Lewis[b]/secretor		Lewis[a]/non-secretor			
	No.	(%)	No.	(%)	χ^2	P
Airdrie	222	(62·4)	134	(37·6)	—	—
Glasgow	371	(73·8)	132	(26·2)	12·14	<0·0005
Edinburgh	245	(73·4)	89	(26·6)	9·03	<0·005

Table 3. *Carriage of meningococci among smokers and non-smokers*

	Carrier		Non-carrier	
	No.	(%)	No.	(%)
Smoker (37)	17	(46)	20	(54)
Non-smokers (349)	87	(25)	262	(75)
Total	104		282	

non-carrier controls from the staff were smokers. The proportion of girls who smoked 27/208 (13%) was significantly higher than that of boys who smoked 10/171 (6%) ($\chi^2 = 4{\cdot}64$, $P < 0{\cdot}05$). The proportion of carriers was, however, similar for both boys and girls, 47/171 (28%) and 57/208 (27%) respectively.

DISCUSSION

The proportion of non-secretors in most western European populations is 20–25% [14]. In areas where there have been prolonged outbreaks of meningococcal disease such as Iceland, Nigeria and Stonehouse in Gloucestershire, we have found increased proportions of non-secretors [2, 3]. The present study provided an opportunity to determine if the same pattern would be found in short localized outbreaks. Although the distribution of the ABO blood groups was similar to that previously reported for Glasgow and Edinburgh (Table 1), the proportion of Lewis[a] non-secretors was 37·6% among the staff and students tested, significantly higher than those previously reported for Glasgow (26·2%) [13] or Edinburgh (26·6%) [11] (Table 2). This increase in the proportion of Lewis[a]/non-secretors is similar to that observed among the residents of Stonehouse, Gloucestershire (32·7%) compared with blood donors in the south west region (23·4%) [3].

In the survey reported here, there was a significant increase in the proportion of non-secretors who were carriers of meningococci (47%) compared with that among the non-carriers (32%). In the Stonehouse study, the proportion of non-secretors among carriers did not differ significantly from that of the participants from whom no neisseria were isolated [3].

The main reason for the difference in the two studies is probably an underestimate of the proportion of non-secretors among the carriers in the Stonehouse survey [15]. The secretor status of each of the participants in the Stonehouse survey was determined from the saliva specimen collected. Blood contamination of saliva specimens can result in false 'secretors', so the Lewis blood group was determined for a proportion of the blood specimens as a control for secretor status. Although the ABO blood group was determined for each specimen at the time of collection, the large number of individuals examined in the Stonehouse survey (>6000) limited the determination of the Lewis blood group to every eighth specimen.

For the specimens obtained in the Stonehouse study, the results of the saliva and the Lewis blood group agreed for more than 92% of the pairs of specimens tested. In the report on the Stonehouse data, it was emphasized that agreement between the two tests was closest in the areas of the town where socioeconomic conditions were better [3]. Differences between the blood and saliva tests indicating 'false' secretors occurred significantly among the specimens obtained from residents of Park Estate and Verney Fields where (*a*) 14 of the 15 cases of meningococcal disease occurred [15], (*b*) the carrier rates for both outbreak and non-outbreak strains were higher [16] and (*c*) the socioeconomic indicators were lower [15]. Blood in saliva might be due to poor oral hygiene, periodontal disease or smoking; each can affect the integrity of the oral mucosa and each is associated with poorer socioeconomic conditions.

The results of this study provide evidence for our hypothesis that one of the factors contributing to the susceptibility of non-secretors to meningococcal disease is increased carriage of these bacteria among such individuals. This parallels the reports of increased proportions of non-secretors among patients with rheumatic fever and among carriers of *Streptococcus pyogenes* [17]. A significant increase in the proportion of non-secretors has also been found among healthy carriers of *Candida albicans* [18], and we have found a similar pattern among patients with non-insulin dependent diabetes mellitus [19].

Passive smoking is associated with meningococcal disease [4, 5], but we found no association with passive exposure to cigarette smoke (mother and/or father who were smokers) and carriage of meningococci. There was however, a significfant increase in the proportion of carriers among smokers (46%) compared with that among non-smokers (25%) ($P < 0{\cdot}025$). The higher proportion of girls who were smokers (13%) compared with boys who were smokers (6%) agrees with the recently reported trends of increased smoking among teenage girls [20].

A slightly higher proportion of males to females has been noted among carriers of meningococci; in the study of Cartwright and co-workers [16] the ratio was 3:2, males:females. If smoking has a significant effect on carriage, in the population examined in the present study, the higher proportion of girls who smoked might contribute to the similar proportions of boys (28%) and girls (27%) who were carriers. Analysis of smoking and secretor status among carriers and non-carriers indicate that these are two independent factors affecting carriage.

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REFERENCES

1. Blackwell CC, Jonsdottir K, Hanson M, et al. Non-secretion of ABO antigens predisposing to infection by *Neisseria meningitidis* and *Streptococcus pneumoniae*. Lancet 1986; ii: 284–5.
2. Blackwell CC, Jonsdottir K, Mohammed I, Weir DM. Non-secretion of blood group antigens – a genetic factor predisposing to infection by *Neisseria meningitidis*. In Poolman JT ed., et al., Gonococci and meningococci. Dordrecht: Kluwer Academic Publishers, 1988.
3. Blackwell CC, Weir DM, James VS, Cartwright KAV, Stuart JM, Jones DM. The Stonehouse study: secretor status and carriage of *Neisseria* species. Epidemiol Infect 1989; **102**: 1–10.
4. Stuart JM, Cartwright KAV, Dawson AJ, Rickard J, Noah ND. Risk factors for meningococcal disease: a case control study in Southwest England. Community Med 1988; **10**: 139–46.
5. Haneberg B, Tonjum T, Rodahl K, Gedde-Dahl T. Factors preceding the onset of meningcococcal disease with special emphasis on passive smoking, stressful events, physical fitness and general symptoms of ill health. National Inst Public Health Annals 1983; **6**: 169–74.
6. Kaiser AB, Hennekens CH, Saslaw MS, Hayes PS, Bennett JV. Sero-epidemiology and chemoprophylaxis of disease due to sulphonamide resistant *Neisseria meningitidis* in a civilian population. J Infect. Dis 1974; **130**: 217–24.
7. Farries JS, Dickson W, Greenwood E, Malhotra TR, Abbot D, Jones DM. Meningococcal infection in Bolton 1971–1975. Lancet 1975; ii: 18–21.
8. Nichol KP, Cherry JD. Bacterial-viral interrelations in respiratory infections of children. N Engl J Med 1967; **277**: 667–72.
9. Eichenwald K, Kotsevalov O, Fasso LA. Some effects of viral infection on ariel dissemination of staphylococci and on susceptibility to bacterial colonization. Bacteriol Rev 1961; **25**: 274–81.
10. Krakinski K, Nilson JD, Butler S, Laby JP, Kusmiesz H. Possible association of mycoplasma and viral respiratory infections with bacterial meningitis. J Epidemiol 1987; **125**: 499–508.
11. Kinane DF, Blackwell CC, Brettle RP, Weir DM, Winstanley FP, Elton RA. Blood group secretor state and susceptibility to recurrent urinary tract infections in women. Br Med J 1982; **285**: 7–9.
12. MacSween MP, Syme UA. ABO blood groups and skin diseases. Brit J Dermatol 1965; **77**: 30–4.
13. Cameron JM. Blood groups in tumors of salivary tissue. Lancet 1958; i: 239–40.
14. Mourant AE, Kopec A, Domaniewska-Sobszak K. The distribution of the human blood groups and other polymorphisms, 2nd ed. London: Oxford University Press, 1976.
15. Stuart JM, Cartwright KAV, Jones DM, et al. An outbreak of meningococcal disease in Stonehouse: planning and execution of a large-scale survey. Epidemiol Infect 1987; **99**: 579–89.
16. Cartwright KAV, Stuart JM, Jones DM, Noah ND. The Stonehouse survey, nasopharyngeal carriage of meningococci and *Neisseria lactamica*. Epidemiol Infect 1987; **99**: 591–601.

17. Haverkorn MJ, Goslings WRO. Streptococci, ABO blood groups and secretor status. Amer J Human Genet 1969; **21**: 360–75.
18. Burford-Mason AP, Weber JCP, Willoughby JMT. Oral carriage of *Candida albicans*, ABO blood group and secretor status in healthy individuals. J Med Vet Mycol 1988; **26**: 49–56.
19. Blackwell CC, Aly FZM, Weir DM, et al. Blood group, secretor status and oral carriage of yeasts among patients with diabetes mellitus. Diabetes Res. In press.
20. Wald N, Kiryluk S, Darby S, Doll R, Pike M, Peto R. UK smoking statistics. Oxford: Oxford University Press, 1988.