Risk factors in multiple sclerosis: a population-based case-control study in Hautes-Pyrénées, France

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ABSTRACT - A population-based study of the prevalence and risk factors of multiple sclerosis (MS) was conducted in the Hautes-Pyrénées, the southwestern region of France. The prevalence rate per 100,000 was equal to 40. Data on the past medical history of 63 MS patients and matched controls were collected. The frequency and age at occurrence of common childhood infections were similar for both the MS cases and controls. There was no difference between the frequency of vaccination for MS patients and for controls. However, the age at which MS patients were immunized against poliomyelitis was significantly higher than the corresponding age for controls (15.8 years versus 8.9 years, P < 0.01). Antibody titers for various viruses were measured. The mumps antibody titer was significantly higher in the MS patients than in the controls. Also, MS patients tended to have higher titers for measles antibodies.

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The Hautes-Pyrénées department is situated in southwest France at a latitude of 42° to 43° north. It is a mainly rural area of 4500 square kilometers. At the time of the 1982 census, the population was 227,942 (Figure 1); 22% of which was concentrated in the main town of Tarbes. According to the census, approximately 84% of the resident population over age 20 had been born in the county.

Analysis of the geographical pattern of death rates from multiple sclerosis (MS) in France from 1968 to 1977 showed a risk gradient that increased from south to north (1). However, the Hautes-Pyrénées region recorded significantly higher mortality rates than the neighboring southern counties. We conducted an epidemiologic study in 1983 to investigate this phenomenon.

Methods

General practitioners and neurologists (n = 450) were asked to provide the names of individuals whose neurological disease suggested MS. Ninety-five percent of the physicians contacted were willing to cooperate. The criteria used to diagnose MS were those defined by Poser et al. (2). For each MS case, one unrelated control of the same age, gender, and parish of residence was selected. One investigator (J-B R) interviewed the patients and controls in their homes. A summary of questionnaire items is shown in Table 1.

Blood samples were taken from patients and controls. Antibody titers to 7 viruses (measles, rubella, varicella-zoster, mumps, herpes simplex...
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1, cytomegalovirus, coronavirus) were measured by ELISA. All specimens were tested blindly. Titers were compared by transforming the reciprocal of the dilutions to log 2.

Data were analyzed using statistical tests for matched-pair comparisons (3). A P value of 0.05 or less was considered significant.

Results

Patients

Ninety-one patients (60 women and 31 men) were living in the Hautes-Pyrénées county on prevalence day (1 January 1983). The prevalence rate per 100,000 was equal to 40; 4 patients died during 1983. Sixty-three patients with definite or probable MS for whom both questionnaire and blood samples had been obtained were available for matched-pair comparisons. Seventy-three percent of them were female (F:M ratio = 2.7). The mean age of MS onset was 30.8 years. The mean interval from onset to prevalence day was 13.1 years; the proportion of cases with onset greater than 15 years before prevalence day was 34% (22 of 63).

Thirty-six patients (57%) had a relapsing-remitting form of MS; 22 (35%) had a relapsing-remitting form leading to chronic progressive course; and 5 (8%) had a chronic progressive form with no remission. At the time of the examination, Kurtzke’s disability score (4) was greater than 5 in 16 (25%) of the 63 patients.

Medical history

The age of MS onset was noted for the patient in the patient-control pair, and medical events for each member of the pair were considered in relation to this age. Patients and controls were asked whether they had ever contracted the following infectious diseases: rubella, measles, varicella, mumps, viral hepatitis, herpes simplex, zoster, pertussis, chronic sinusitis, and upper airways infections (Table 2). On the whole, more patients than controls gave affirmative answers to these questions; but except for the incidence of zoster, the differences were not significant. Indeed, of the 11 patients who reported having zoster, 10 (17.5%) had been affected before the onset of MS compared with 3 (5.2%) of the controls. The interval between zoster and MS onset ranged from a few months to 17 years; in 5 patients, it was smaller than 5 years. Herpes zoster in varicella cases occurred 2.5 times as frequently in MS patients as in controls. Age at occurrence of common childhood infections was similar for patients and controls, although patients tended towards a higher mean age for rubella and varicella.

Past medical history, including tonsillectomy, operations under anaesthesia, and trauma were similar in patients and controls. Stressful life events before the age at MS onset were significantly more common in patients than in controls (67.2% versus 47.6%, P < 0.05). The mean age at menarche was higher in patients than in controls (13.5 years versus 12.7 years, P < 0.002). There was no relationship between age at menarche and age at MS onset in MS patients.

There was no difference in the frequency of vaccination against poliomyelitis and tuberculosis in patients and controls (Table 2). No data about other vaccinations had been recorded. There were 15 pairs for whom age at vaccination against poliomyelitis was known for both the MS patient and his or her matched control. Age at the time of immunization was significantly higher for patients than controls (15.8 years versus 8.9 years, P < 0.001).

Fig. 1. Distribution of MS cases in the Hautes-Pyrénées county. (Each point represents one case, except for the main town Tarbes, where the total number of cases is noted.)
A history of familial MS was reported by 4 patients but by none of the controls \((P < 0.05)\).

**Viral antibody titers**

Table 3 shows the distribution of viral antibody titers in MS patients and controls. Except for mumps, geometric mean titers were similar in both groups. Matched-pair analysis showed that there were 32 pairs in which the mumps antibody titer was higher in the patient than in the control, and 16 pairs in which the reverse was true (chi-square Mc Nemar = 5.3, \(P < 0.02\)). A similar trend was observed for antibodies against measles (35 pairs versus 21, \(P < 0.10\)).

In order to define the complete profile of viral antibody titers, we built an index ranging from 0 (where none of the 7 patients' titers was greater than the corresponding control's titer) to 7 (for the opposite case). The mean value of this index did not differ from the value predicted by the null hypothesis.

We did not find any consistent correlation between antibody levels to the various viruses either in patients, or in controls. High titers against both measles and mumps virus were observed in some patients, but this relationship was not statistically significant \((P = 0.13)\). There was no association between antibody levels and clinical features, including sex, age at onset of MS, and the course of the disease.

Agreement between the reporting of viral disease and the detection of corresponding antibodies was assessed by calculating the Kappa coefficient \((4)\). There was very poor agreement: all Kappa values for both patients and controls were smaller than 0.20.
Table 3
Viral antibody titers in MS patients and controls

| Virus       | Titer (<1/400) | Titer (1/2000) | Mean geometric titer |
|-------------|----------------|----------------|---------------------|
|             | MS(%)          | MS(%)          |                     |
| Measles     |                |                |                     |
| MS          | 7.1            | 45.6           | 7.4                 |
| Control     | 10.2           | 30.5           | NS                  |
| Rubella     |                |                |                     |
| MS          | 41.8           | 14.6           | 6.2                 |
| Control     | 32.8           | 8.6            | 6.3                 |
| Mumps       |                |                |                     |
| MS          | 33.3           | 10.5           | 6.3                 |
| Control     | 61.0           | 3.4            | 5.7                 |
| Varicella-zoster |        |                |                     |
| MS          | 36.8           | 17.5           | 6.4                 |
| Control     | 32.2           | 18.6           | 6.5                 |
| Herpes simplex |            |                |                     |
| MS          | 26.8           | 30.3           | 6.6                 |
| Control     | 25.4           | 27.1           | 6.6                 |
| Cytomegalovirus |            |                |                     |
| MS          | 61.4           | 15.8           | 5.7                 |
| Control     | 67.8           | 8.5            | 5.4                 |
| Coronavirus |                |                |                     |
| MS          | 22.8           | 28.1           | 6.8                 |
| Control     | 11.9           | 25.4           | 7.0                 |

(a) Proportion of subjects with titers between 1/400 and 1/2000 complemented the two proportions.

Discussion

With a prevalence of 40 per 100,000 population, the Hautes-Pyrénées can be considered a high-risk zone for MS. In the world-wide distribution of MS, the high frequency areas (including prevalence rates of 30 and above per 100,000) extend from 44° to 64° N latitude (5). The Hautes-Pyrénées region is located at the lower edge of this area.

Most MS studies have been of patients who had attended neurological clinics or who had been referred to neurologists. A possible effect of referral bias on MS research has recently been discussed (6). In contrast to these other studies, our patient sample was pooled from the general population. However, MS clinical features including sex ratio, age at onset, disease severity, and proportion of progressive forms are consistent with other studies (7, 8).

No evidence was found to incriminate demographic characteristics (9, 10), social class and occupation (9, 11-13), level of sanitation, diet (9, 14, 15), exposure to animals (16-18), or travel (19) as possible risk factors for MS. However, the paucity of statistically significant results may be partly due to the fact that controls, who shared an environment similar to that of the patients, were over-matched. It may also be related to study power.

On the whole, analysis of medical history revealed few consistent differences between MS patients and controls. In particular, we did not find associations suggested in previous studies relating MS with sinusitis (20), tonsillectomies, or operations under general anaesthesia (9, 21).

Age at menarche was significantly higher in patients than in controls. There is some evidence that disease course in females may be influenced by hormonal factors, particularly during pregnancy, post-partum, or menopause (22, 23). Also, the role of sex hormones on immunoregulation and autoimmune diseases is well established (24). The possible role of defects in the sex hormone balance in the etiology of MS in females has been recently postulated (25) and the present finding gives some support to this hypothesis.

Of particular interest is the fact that age at vaccination against poliomyelitis was significantly higher in patients than in controls. With respect to this result, it is important to note that in an investigation of MS in the Orkney and Shetland Islands, Poskanser (9) found that 3 of 88 MS patients (and no controls) reported having poliomyelitis.

The present study adds some findings to previous works on clinical viral infections, viral antibody titers, and MS. Unlike many authors (26-28) we did not find any relationship between measles infection and MS. Nevertheless, MS patients tended to have higher measles antibody titers than controls. An important observation is that the mumps antibody titer in MS patients is significantly higher than that of controls, even though there was no difference between the reported frequency of mumps infection in MS patients and controls.
Previous reports have involved mumps infection in the etiology of MS. Alter (29) and Compton (28) found that mumps infections were more frequent in patients than in controls. Several studies (29, 30) demonstrated a trend towards older age at infection by mumps in MS patients than in controls. Similarly, our findings support the hypothesis that mumps infection might be involved in the etiology of MS.

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