# PAPERS AND SHORT REPORTS

# Guillain-Barré syndrome and Campylobacter jejuni: a serological study

J KALDOR, B R SPEED


## Abstract

**The association between Campylobacter jejuni infection and Guillain-Barré syndrome was investigated serologically in a retrospective study of 56 patients admitted to this hospital over four years. Evidence of preceding C jejuni infection was found in 21 (38%) of these patients, indicating that C jejuni was the most common single identifiable pathogen precipitating the disease. Among those patients who had presented with preceding diarrhoea the serum antibody response was similar to that in uncomplicated C jejuni enteritis. Patients with serological evidence of preceding C jejuni infection manifested a significantly more severe form of the disease. In cerebrospinal fluid the predominant specific antibody class was IgG, and this was closely related to the serum titres of specific IgG. IgA and IgM specific antibodies were found only in the cerebrospinal fluid of patients with recent C jejuni infection.**

**These findings support the possibility that humoral immune factors are responsible for the neural damage and demyelination seen in Guillain-Barré syndrome.**


## Introduction

Guillain-Barré syndrome (acute postinfectious polyneuritis) is the most common cause of acute neuromuscular paralysis. The aetiology and pathogenesis of the condition has yet to be established, but in roughly half the patients the paralysis has been noted to occur one to three weeks after an acute infection.[1] The preceding infection is often of a non-specific respiratory or gastrointestinal nature, but several specific pathogens or events have been described—namely, Epstein-Barr virus, herpes varicella-zoster and simplex viruses, cytomegalovirus, viral hepatitis, respiratory viruses, enteroviruses, typhoid, mycoplasma, vaccinations, and surgery.[1,2] More recently, two cases of Guillain-Barré syndrome occurring after *Campylobacter jejuni* enteritis have been reported,[3,4] and we also have observed this association.[5]

Since the original descriptions of *C jejuni* enteritis[6,7] this agent has become recognised world wide (and in our community) as one of the commonest causes of gastroenteritis. In view of the ubiquity of *C jejuni* its relation to the Guillain-Barré syndrome may occur more often than has been recognised. We therefore undertook a retrospective serological and clinical survey of a large group of patients with the syndrome to investigate this association.

## Patients and methods

From January 1979 to December 1982, 62 patients were admitted to this hospital suffering from the Guillain-Barré syndrome. A total of 216 serum specimens were available from 56 of the patients in addition to 22 specimens of cerebrospinal fluid from 20 of these patients. A further seven cerebrospinal fluid specimens were included from patients admitted before 1979 who also had Guillain-Barré syndrome. All specimens from patients with the syndrome were the remainder of those used for routine investigations and had been stored at −20°C.

Control samples comprised serum specimens from 27 patients suffering from neurological disorders other than the Guillain-Barré syndrome (encephalitis, myelitis, cerebrovascular accident, cerebral atrophy, alcoholic neuropathy); cerebrospinal fluid specimens from 29 patients suffering from central nervous system diseases (viral and bacterial meningitis, encephalitis, transverse myelitis); serum specimens from 73 patients suffering from culture proved *C jejuni* enteritis who had raised titres of specific antibody; and serum specimens from 30 healthy controls.

In every case the Guillain-Barré syndrome had been diagnosed by a specialist physician based on accepted clinical and laboratory findings.[8] Electrophysiological studies were not performed.

Class specific antibodies to *C jejuni* were measured in serum and cerebrospinal fluid using a solid phase enzyme linked immunosorbent assay.[9] With this method positive serum antibody titres are: IgA $>1/80$, IgM $>1/80$, IgG $>1/320$.

For this study a patient was regarded as having evidence of recent *C jejuni* infection if he satisfied the following criteria: (*a*) an increase

**Fairfield Infectious Diseases Hospital, Melbourne, Australia**
J KALDOR, MAPPSCI, senior biochemist
B R SPEED, MRCP, FRACP, specialist physician

Correspondence to: J Kaldor, Department of Biochemistry, Fairfield Hospital, Victoria, Australia 3078.

of two or more classes of antibody, and/or (*b*) serological conversion, and/or (*c*) a fourfold change in titre, provided that the peak titre was in the positive range. Cerebrospinal fluid antibody titres of $>1/4$ were taken as positive.

Total protein concentrations in cerebrospinal fluid were measured by the method of Pesce and Strande.[10] Faecal culture for *C jejuni* was performed using the method of Skirrow.[7]

Throat swabs for viral culture were taken from 31 patients on admission, and specimens of urine and saliva for cytomegalovirus culture from 31 patients after admission. Various other serological tests were performed when indicated clinically (see below). Serum specimens were tested for hepatitis B surface antigen by radioimmunoassay,[11] and cytomegalovirus antibodies were detected using a complement fixation test.[12]

Statistical calculations were done using the $\chi^2$ test with Yates's correction.

## Results

### SERUM

Twenty one of the 56 patients with the Guillain-Barré syndrome (38%) were found to have evidence of recent *C jejuni* infection (CJ(+) GBS). Sixteen patients fulfilled two or more of the diagnostic criteria, and the other five had significant titres of two or more antibody classes. No serological evidence of recent infection was detected in either the 30 healthy controls, the 27 controls who had neurological disorders, or the other 35 patients with Guillain-Barré syndrome (CJ(−) GBS). The CJ(+) GBS patients were analysed with respect to the nature of their preceding illness, and table I outlines the results. There was a close association between recent *C jejuni* infection and preceding diarrhoeal illness. Eighty five per cent of these patients were CJ(+), whereas only 23% of those without diarrhoea were CJ(+) ($\chi^2=13{\cdot}5$; $p<0{\cdot}001$).

TABLE I—*Guillain-Barré syndrome and recent C jejuni infection: relation to preceding illness*

| Preceding illness | No (%) of patients studied | Proportion (No(%)) with recent *C jejuni* infection |
|---|---|---|
| Diarrhoea | 13 (23) | 11 (85) |
| Respiratory infection | 23 (41) | 4 (17) |
| Miscellaneous* | 4 (7) | 1 (25) |
| Nil | 16 (29) | 5 (31) |
| Total | 56 (100) | 21 (38) |

*Urinary tract infection, viral illness, hepatitis B, surgery.

The class specific antibody response in CJ(+) GBS patients was compared with that in the 73 controls with uncomplicated *C jejuni* enteritis (table II). Overall the proportion of patients with raised titres of IgA and IgM was similar in the two groups. A significantly larger proportion of the patients with Guillain-Barré syndrome, however, had raised IgG titres ($p<0{\cdot}05$). The magnitude of antibody response in the two groups was similar, except that IgA and IgM titres were somewhat lower in the CJ(+) GBS patients. This may be explained by loss of activity as a result of freezing the serum samples.

With regard to age and sex of the 56 patients with Guillain-Barré syndrome, 27 were men and the average age 46·6 years. There was no significant age or sex difference between those with and those without evidence of recent *C jejuni* infection. The time between onset of the preceding illness and onset of Guillain-Barré syndrome (latent period) was also analysed. In the 11 CJ(+) patients with preceding diarrhoea the latent period was 5-11 days (mean 8 days).

The severity of Guillain-Barré syndrome was assessed by measuring forced vital capacity, by the need for respiratory support, and by the duration of respiratory support. Table III shows the relation of *C jejuni* infection to these variables. A statistically significant correlation was apparent. In particular, 19 (90%) of the patients with CJ(+) disease required artificial ventilation.

TABLE II—*Class specific antibody response to C jejuni in seropositive patients: Guillain-Barré syndrome compared with uncomplicated enteritis*

| Antibody class | | Guillain-Barré syndrome (n = 21) | *C jejuni* enteritis (n = 73) | p* |
|---|---|---|---|---|
| IgG | No (%) positive | 17 (81) | 37 (51) | <0·05 |
| | Titre† | 1/1507 | 1/1573 | |
| IgA | No (%) positive | 18 (86) | 45 (62) | >0·05 |
| | Titre† | 1/285 | 1/422 | |
| IgM | No (%) positive | 16 (76) | 58 (79) | >0·05 |
| | Titre† | 1/226 | 1/416 | |

*$\chi^2$ with Yates's correction.
†Geometric mean of highest positive titres.

TABLE III—*Relation of C jejuni infection to severity of Guillain-Barré syndrome. Figures are numbers of patients*

| | *C jejuni* positive | *C jejuni* negative | p* |
|---|---|---|---|
| Respiratory support | 19/21 | 14/35 | 0·001 |
| Forced vital capacity <2·5 l | 19/21 | 21/35 | 0·05 |
| Respiratory support ≥30 days | 17/21 | 7/35 | 0·001 |
| Significant residual disability | 5/14 | 4/29 | 0·05 |

*$\chi^2$ with Yates's correction.

The outcome of Guillain-Barré syndrome was also evaluated: a total of four patients died (7%), three of whom were in the CJ(+) group. Of the 43 survivors assessable for final recovery, 9 (21%) were found to have significant residual disability (requiring walking aids). A larger proportion of the CJ(+) patients had residual disability, but the difference was not statistically significant (table III).

### CEREBROSPINAL FLUID

Cerebrospinal fluid from three groups of patients was examined for class specific antibody to *C jejuni* (table IV). Of 11 specimens tested from CJ(+) GBS patients, 10 were positive, whereas significantly smaller proportions from the other two groups were positive ($\chi^2$ test: $p<0{\cdot}001$) and their mean antibody titre was lower. IgG was the predominant antibody class found in cerebrospinal fluid; IgA and IgM were found only in the cerebrospinal fluid of CJ(+) GBS patients. There was a close correlation between serum IgG titres and cerebrospinal fluid IgG titres in CJ(+) GBS patients ($r=0{\cdot}88$; $p<0{\cdot}001$) but not in CJ(−) patients ($r=0{\cdot}21$; $p>0{\cdot}05$). In the CJ(−) GBS patients with raised cerebrospinal fluid IgG titres, however, there was a correlation between serum and cerebrospinal fluid levels when the cerebrospinal fluid total protein was allowed for (cerebrospinal fluid IgG $\propto$ serum IgG / cerebrospinal fluid protein); $r=0{\cdot}82$; $p<0{\cdot}02$). Among CJ(−) GBS patients, those with increased titres of specific cerebrospinal fluid IgG had a higher mean concentration of cerebrospinal fluid total protein than those without increased titres (1394 and 1116 mg/l respectively).

TABLE IV—*Cerebrospinal fluid findings in patients with Guillain-Barré syndrome and recent C jejuni infection compared with findings in seronegative cases and controls*

| | Guillain-Barré with recent *C jejuni* infection | Guillain-Barré without recent *C jejuni* infection | Other neurological diseases |
|---|---|---|---|
| No positive for IgG (titre*) | 10/11 (1/45) | 8/18 (1/13) | 2/29 (1/11) |
| No positive for IgA | 3/11 | 0 | 0 |
| No positive for IgM | 3/11 | 0 | 0 |
| Mean (1 SD) total protein (mg/l) | 1317 (1085) | 1239 (647) | 638 (426) |

*Geometric mean of positive titres. Normal cerebrospinal fluid antibody titre <1/4.

### OTHER LABORATORY INVESTIGATIONS

*Bacteriology*—Faecal culture was performed in only four patients; *C jejuni* was isolated from one and was the only pathogen.

*Virus isolation*—Only one positive isolate (herpes simplex virus)

was recovered from 31 throat swabs taken on admission of patients suffering from Guillain-Barré syndrome, and the patient concerned was CJ(+). No virus was isolated from cerebrospinal fluid (19 patients) or faeces (eight). Urine and saliva specimens collected specifically for cytomegalovirus isolation in 31 patients yielded cytomegalovirus in nine and herpes simplex virus in seven.

*Serology*—Testing for several other agents was performed in varied combinations according to clinical features. Testing for hepatitis B surface antigen was the only test performed routinely on all patients, and one gave a positive result. That patient had acute hepatitis B associated with Guillain-Barré syndrome and was CJ(−). Testing of paired sera for cytomegalovirus antibody was performed in 31 patients, and only two CJ(+) GBS patients had evidence of recent infection with this agent. Testing of paired sera for antibodies to the following pathogens was performed and gave negative results: Epstein-Barr virus (six patients), hepatitis A virus (seven), influenza (five), mycoplasma (four), psittacosis (two), adenovirus (one). A Paul-Bunnell test was performed in six patients and all gave negative results.

## Discussion

This study shows that *C jejuni* is the most common single pathogen yet reported in association with Guillain-Barré syndrome. Twenty one out of 56 patients with the syndrome (38%) had serological evidence of recent infection with *C jejuni*. By comparison, cytomegalovirus has been reported in 15% of cases and Epstein-Barr virus in 8%.[2]

Although isolation of *C jejuni* from the faeces is the method of choice for diagnosing acute infection, this was not possible in most of our cases. Firstly, by the time a patient came under medical attention for Guillain-Barré syndrome the faecal culture would often be negative, as the median duration of excretion is two to three weeks.[13] Secondly, in order to evaluate a large number of patients our study was retrospective. Hence detection of specific antibody was the means of diagnosis in nearly all our cases.

Our serological findings are supported by the following: (*a*) previous reports have linked culture positive *C jejuni* enteritis with Guillain-Barré syndrome[3 4]; (*b*) one patient in our study had bacteriologically confirmed *C jejuni* enteritis preceding the syndrome and a diagnostic rise in all three classes of specific antibody; (*c*) the antibody response in CJ(+) GBS patients was similar to that seen in uncomplicated *C jejuni* enteritis; (*d*) 11 of our patients suffering from the syndrome (20%) had a preceding diarrhoeal illness and were CJ(+); (*e*) diarrhoea was the preceding illness most closely associated with serological evidence of recent *C jejuni* infection; (*f*) normal controls and those with other neurological disorders had no evidence of recent *C jejuni* infection.

We found that 10 patients who had not suffered from preceding diarrhoea were CJ(+), which might suggest cross reaction with other agents. Asymptomatic infection with *C jejuni* has been described,[13] however, and we have recorded four asymptomatic patients in whom there was a typical acute antibody response. Two of the CJ(+) patients with no preceding illness also had acute cytomegalovirus infection, but it was not possible to elucidate the relative roles of the two agents in these cases. Herpes simplex virus was the only other agent isolated and occurred in four CJ(+) cases. In three of these the virus was isolated only from specimens taken 20 or more days after admission, and probably these represented reactivation of latent virus; no other specific micro-organisms were detected in our series of patients.

There was a clear association between recent *C jejuni* infection and severity of Guillain-Barré syndrome (table III). Nineteen of the 21 (90%) CJ(+) patients in our series developed a forced vital capacity of less than 2·5 l, and all of these required artificial respiration. There was no obvious explanation, but the finding strongly suggests that Guillain-Barré syndrome after *C jejuni* infection behaves differently from that occurring after other infections. A possible explanation is that different infections produce neural damage by different immunological mechanisms.

We confirmed our previous finding of specific *C jejuni* antibody in the cerebrospinal fluid of patients with Guillain-Barré syndrome[5] (table IV). The origin of cerebrospinal fluid IgG in CJ(+) patients appears to be the result of passive diffusion across the altered blood-brain barrier. Other reports have shown that the increase in total cerebrospinal fluid IgG regularly seen in Guillain-Barré syndrome also occurs in this way.[14] The presence of low titres of IgG in the cerebrospinal fluid of eight (CJ(−) GBS patients and in the cerebrospinal fluid of two controls is less easy to explain; but when the increase in total cerebrospinal fluid protein was allowed for there appeared to be a relation. Four of the CJ(+) GBS patients possessed IgA or IgM or both in their cerebrospinal fluid. As these are much larger molecules, their presence suggests local production, which in turn suggests the presence of specific antigen in the nervous system.

Several mechanisms for the pathogenesis of Guillain-Barré syndrome have been postulated, and most have in common an immunological basis. Both cellular[15 16] and humoral[17–19] mechanisms have been suggested, and there is evidence in favour of each. The timing of the onset of the syndrome one to two weeks after an antecedent illness corresponds to the interval required to mount a humoral immune response. In the CJ(+) GBS patients who had suffered from diarrhoea, the mean time to onset of the disease was eight days, by which time the immune response to *C jejuni* is well established.[9] The role of *C jejuni* antibodies in production of Guillain-Barré syndrome is uncertain but there are three broad possibilities.

(1) *Immunological cross reaction between C jejuni and neural tissue*—Antibodies to myelin and other neural proteins have been described in Guillain-Barré syndrome,[17–19] and *C jejuni* may possibly stimulate their production. A search for evidence of this cross reactivity is in progress. Antibodies to *C jejuni* which penetrate into the cerebrospinal fluid may be responsible for the neural damage. Antineural IgG has been detected in several studies,[17–19] and significantly more of our CJ(+) GBS patients than controls with uncomplicated gastroenteritis had raised serum titres of specific IgG. In addition, CJ(+) GBS patients were more likely to have raised cerebrospinal fluid IgG titres than the CJ(−) GBS patients, and the levels of the titres were higher. Antigenic similarities between neural glycopeptides and bacterial capsules have been described,[20] which may be the possible immunological link in the case of *Campylobacter*.

(2) *Toxic neural damage—C jejuni* has recently been shown to produce a cholera-like enterotoxin,[21] and cholera toxin binds avidly to gangliosides.[22] This suggests another possible mechanism based on the role of a toxin produced by *C jejuni* that binds to neural tissue.

(3) *Cell mediated immune damage*—As yet there are no reports describing the cell mediated response in *C jejuni* infection, but because such mechanisms have been described in Guillain-Barré syndrome[15 16] they must remain a possibility in the case of *C jejuni*.

Despite the high incidence of *C jejuni* enteritis in the community, Guillain-Barré syndrome remains a rare disease. This suggests that it occurs after infection only with one particular serotype or that patients who develop the disease differ in their response to the infection.

It is clear that *C jejuni* infection often precedes Guillain-Barré syndrome, but the nature of this association and the mechanism of neural damage remain uncertain. This study supports the possibility that immunological mechanisms play a part, and the eventual elucidation of these processes may well be relevant to other demyelinating diseases.

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# Circumstances of death from asthma

A J JOHNSON, A J NUNN, A R SOMNER, D E STABLEFORTH, C J STEWART


## Abstract

**Mortality from asthma in England and Wales has remained unchanged for at least 20 years, even in the age group 15-44. Yet in those 20 years "modern" drugs have been introduced for the treatment of asthma, such as $\beta_2$ agonist bronchodilators and corticosteroids. Why do patients still die?**

**Detailed review of the circumstances of 90 deaths from asthma showed that a few were inevitable but that in the remainder four main sets of circumstances in the fatal attack contributed to the death. These were, firstly, the patient's failure to recognise the severity of the asthma; secondly, very rapid progress in the severity of the attack; thirdly, misjudgment in the management of the attack; and, fourthly, delay from many causes.**

**Patients admitted to hospital with severe acute asthma usually survive. Those at risk of a life threatening attack should be identified and taught to monitor the severity and progress of their asthma objectively. Their direct admission to hospital should be facilitated.**



**Kent and Canterbury Hospital, Canterbury CT1 3NG**
A J JOHNSON, MB, MRCP, consultant chest physician

**MRC Tuberculosis and Chest Diseases Unit, Brompton Hospital, London SW3 6HP**
A J NUNN, MSC, statistician

**Preston Hospital, North Shields, Tyne and Wear NE29 0LR**
A R SOMNER, MD, FRCP, consultant chest physician

**East Birmingham Hospital, Birmingham B9 5ST**
D E STABLEFORTH, MB, MRCP, consultant in thoracic medicine

**Ipswich Hospitals, Suffolk**
C J STEWART, MD, lately consultant chest physician

Correspondence to: Dr C J Stewart, 159 Henley Road, Ipswich, Suffolk IP1 4NU.


## Introduction

In 1982 the Research Committee of the British Thoracic Association (Society) reported on the death from asthma of 90 patients aged 15-64 years living in the West Midlands and Mersey regions in 1979.[1] Like other reports of death from asthma in recent years,[2-5] that report considered principally the deficiencies of management and the modern drug treatment preceding the fatal attack. In this paper we review in more detail than was possible in the original report the circumstances of the fatal attack of asthma.

## Methods

The procedure and methods were fully described in the original report.[1] Of particular value in that retrospective inquiry was the independent panel of three physicians who reviewed and assessed each case. Altogether 153 patients died, of whom they agreed that 90 had died of asthma. They then assessed the management and treatment of the asthma in the last year and the last month of life and the events of the fatal attack. Our report is largely based on those assessments (see tables I and II). The 12 case histories set out below represent the main categories of death.