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Serologic Evidence of Previous Campylobacter   Army Project Order
Jejuni Infection in Patients with the Guillain-Barre   90PP0820
Syndrome

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Serologic Evidence of Previous *Campylobacter jejuni* Infection in Patients with the Guillain-Barré Syndrome

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■ **Objective:** To determine if patients with the Guillain-Barré syndrome are likely to have had *Campylobacter jejuni* infection before onset of neurologic symptoms.

■ **Design:** A case-control study.

■ **Setting:** Several university medical centers.

■ **Patients:** Case patients met clinical criteria for the Guillain-Barré syndrome between 1983 and 1990 and had a serum sample collected and frozen within 3 weeks after onset of neurologic symptoms (*n* = 118). Disease controls were patients with other neurologic illnesses (*n* = 56); healthy controls were hospital employees or healthy family members of patients (*n* = 47).

■ **Measurements:** Serum IgA, IgG, and IgM antibodies to *C. jejuni* were determined by enzyme-linked immunosorbent assays. Assays were done in a blinded manner.

■ **Results:** Optical density ratios ≥2 in two or more immunoglobulin classes were seen in 43 (36%) of patients with the Guillain-Barré syndrome and in 10 (10%) of controls (odds ratio, 5.3; 95% CI, 2.4 to 12.5; *P* < 0.001). Increasing the optical density ratio or the number of immunoglobulin classes necessary to yield a positive result increased the strength of the association. The number of patients with the Guillain-Barré syndrome who had positive serologic responses was greatest from September to November (*P* = 0.02). Male patients were three times more likely to have serologic evidence of *C. jejuni* infection (*P* = 0.009); the proportion of patients with the syndrome who had a positive serologic response increased with age.

■ **Conclusions:** Patients with the Guillain-Barré syndrome are more likely than controls to have serologic evidence of *C. jejuni* infection in the weeks before onset of neurologic symptoms. *Campylobacter jejuni* may play a role in the initiation of the Guillain-Barré syndrome in many patients.

The Guillain-Barré syndrome, sometimes called "acute inflammatory polyneuropathy," is an inflammatory demyelinating disease of peripheral nerves characterized by various degrees of weakness, sensory abnormalities, and autonomic dysfunction (1-2). Since the marked decline in poliomyelitis, the Guillain-Barré syndrome has become the most common cause of acute neuromuscular paralysis in adults and children in the United States and has an annual incidence of 1.7 per 100 000 people (3, 4). Epidemiologic studies in all parts of the world have confirmed the association between the Guillain-Barré syndrome and previous acute infection, especially of the respiratory or gastrointestinal tracts (4-8). Most report that between 50% and 75% of patients have an infectious illness 1 to 3 weeks before onset of neurologic symptoms; previous diarrheal illness occurs in 10% to 30% of patients (4-6).

Bacteria of the genus *Campylobacter* are important human pathogens and are common causes of gastrointestinal illness in both developed and developing countries (9). Extraintestinal complications of *Campylobacter jejuni* infection such as reactive arthritis, pancreatitis, and carditis are well described (10), and in the last decade, clinical and epidemiologic evidence has suggested that infection with *C. jejuni* may be a precipitating factor for development of the Guillain-Barré syndrome. In small serologic studies of patients with the Guillain-Barré syndrome, as many as one third to one half of patients had increased levels of *C. jejuni* antibodies at the time of onset of neurologic symptoms (11-13). Routine stool cultures of eight patients with the Guillain-Barré syndrome in Japan yielded *C. jejuni* in seven (88%) patients (14). In an important U.S. investigation of 106 patients with the Guillain-Barré syndrome, *C. jejuni* was isolated from 4 (44%) of 9 patients who had antecedent clear-cut diarrheal illness; however, cultures had not been obtained from 97 other patients (15).

To assess the extent to which the Guillain-Barré syndrome is associated with recent *C. jejuni* infection, we did serologic screening with defined assays to determine the frequency of *C. jejuni* antibodies in a large group of patients with the syndrome and in two control groups.

Methods

Study Population

Case Patients

Case patients included 126 persons with the Guillain-Barré syndrome admitted to the University of Maryland Medical Center, the University of Medicine and Dentistry of New Jersey, or to one of several hospitals in St. Louis between 1983 and 1990. A consecutive sample was used. All patients met the

National Institute of Neurologic and Communicative Disorders and Stroke (16) criteria for diagnosis of the Guillain-Barré syndrome; none had any exclusion criteria. Most of the patients had demyelinating polyneuropathy, although some had axonal damage. All had a monophasic illness that progressed during 1 to 4 weeks, plateaued, and almost all patients recovered to some extent; one patient died. Blood samples were collected within 3 weeks after the onset of neurologic symptoms from 118 of the patients; 8 patients had blood samples collected from 24 days to more than 3 months after onset of neurologic symptoms (median, 10 weeks). Because antibody response to C. jejuni infection is usually transient (17), specimens from these eight patients are not included in the analysis. Histories of symptoms of gastrointestinal or respiratory infections before onset of the Guillain-Barré syndrome were not available.

Controls

Two groups of controls were studied. One control group, a convenience sample, consisted of 56 patients with neurologic diseases other than the Guillain-Barré syndrome who were admitted to the neurology services at the University of Maryland, the University of Medicine and Dentistry of New Jersey, or one of the St. Louis hospitals. Diagnoses of these patients included optic neuritis, multiple sclerosis, amyotrophic lateral sclerosis, polyneuropathy associated with IgM paraproteinemia, chronic inflammatory demyelinating polyneuropathy, Charcot-Marie-Tooth disease, unexplained peripheral neuropathy, ischemic neuropathy, neuropathy associated with diabetes, and drug-induced neuropathy. The second control group, a voluntary sample, included 18 healthy employees of the neurology departments of the three universities and 29 healthy family members of case patients and of disease controls. Medical histories of healthy controls were not obtained.

Sera and Data Collection

Sera from case patients and controls was stored frozen at the collection sites until the serologic assays were done. Data collected included disease severity of case patients, sex, age, and date of sera collection. Information about race was available only from the patients at the University of Medicine and Dentistry of New Jersey.

Serologic Methods

Presence of serum IgA, IgG, and IgM antibodies specific for C. jejuni was determined by enzyme-linked immunosorbent assay (ELISA) as previously described (17). Antigens from three C. jejuni strains of Penner (O-) types 1, 2, and 3, which are commonly isolated from humans, were prepared as described by McCoy and colleagues (18); they contained a mixture of acid-extracted proteins common to C. jejuni strains (17, 19-20). The serum dilutions used were as follows: 1:50 for IgA, 1:200 for IgG, and 1:400 for IgM. The class-specific second antibody used (and the use dilution) was peroxidase-conjugated goat anti-human IgA (1:2000), IgG (1:2500), or IgM (1:1000) (Tago Inc; Burlingame, California). The plates were read at 414 nm on a Dynatech ELISA reader (Alexandria, Virginia), and results were expressed as an optical density value for each well. All sera had been coded and were assayed blindly three times on different days in duplicate wells for each run.

Calculation of Optical Density Ratios

The assays were standardized using sera from a patient with the Guillain-Barré syndrome and culture-proven C. jejuni infection (positive control) and sera from 38 healthy children in New York who had had no recent history of diarrheal illness (negative controls). Optical density values were determined serially for plates containing these sera, and the mean optical density of the positive control was plotted versus the mean optical density + 1 SD of the negative controls. For each immunoglobulin class, a regression line was calculated and then used each day the assay was done (using the same positive control serum) to generate a threshold value for determination of the optical density ratio. (Regression analysis for IgA assay, $r = 0.973$, $P < 0.001$; regression analysis for the IgG

assay, $r = 0.998$, $P < 0.001$; regression analysis for the IgM assay, $r = 0.981$, $P < 0.001$.) To control for day-to-day assay variation, the *optical density ratio* was defined as the ratio of the optical density of each unknown serum sample to the observed threshold on that day. A similar method has been used to standardize *Helicobacter* ELISA determination (21).

Case Definitions and Statistical Methods

A *positive* serologic response was defined as an optical density ratio greater than or equal to a specified threshold in one or more immunoglobulin classes. Differences in the proportion of positive responses among case patients and controls were ascertained using an optical density ratio greater than or equal to 1, 2, or 3 as the threshold. Statistical analyses were done in a univariate manner using the Mantel Haenszel Chi-squared analysis or the Fisher exact test when indicated and by the calculation of odds ratios.

The sensitivity and specificity of the assays were determined as follows. Optical density ratios were determined as described above for 17 patients in the convalescent phase of culture-proven C. jejuni infection. These were U.S. soldiers who had become infected while on military exercise in Thailand in 1988; convalescent sera were obtained 4 weeks after onset of diarrhea. The comparison group comprised 19 healthy adults who were employees of or visitors to the Infectious Diseases Division of Vanderbilt University and included 20 healthy children in Nashville who had no recent history of diarrheal illness.

Results

Patients

Demographic characteristics of the 118 case patients and 103 controls are shown in Table 1. Age was known for 112 (95%) case patients and 93 (90%) controls; gender was known for 116 (98%) case patients and for 99 (96%) controls. Information about the month blood was drawn was available for 116 (98%) case patients and 80 (78%) controls. The groups were similar except that controls were somewhat younger (median age, 33 years compared with 41 years for case patients) and were more likely to have had their blood drawn during the summer months. Race of patients and controls (from the University of Medicine and Dentistry of New Jersey only) also was similar in the two groups. In both groups there were more male patients.

Serologic Assays in Persons with Campylobacter jejuni Infection and in Controls

Among 17 persons with culture-proven C. jejuni infections, in the IgA assay, 15 (88%) had an optical density ratio ≥1. Increasing the threshold for defining a positive serologic response to an optical density ratio ≥2 and ≥3 decreased the sensitivity of the test (Table 2). The specificity of the test was evaluated by the serologic responses in the control group. Defining a positive serologic response as an optical density ratio ≥1, the IgA assay was 85% specific (specificity determined by subtracting the percentage of controls exceeding threshold from 100%) (see Table 2). Increasing the threshold to an optical density ratio ≥2 or ≥3 improved the specificity of the test. The sensitivity and specificity of the IgG and IgM assays are also shown in Table 2. As expected, raising the stringency by increasing the optical density ratios or numbers of positive classes required to define a positive serologic response de-

Table 1. Demographic Characteristics of 118 Patients with the Guillain-Barré Syndrome and of 103 Controls, Including 56 Controls with Disease and 47 Healthy Controls

Characteristic	Patients with the Guillain-Barré Syndrome	Controls with Disease*	Healthy Controls	All Controls
		n(%)		
Male gender	116 (58)	53 (57)	46 (72)	99 (64)
White	57 (89)	11 (91)	17 (94)	28 (93)
Month blood drawn	116	46	34	80
Winter (Dec,Jan,Feb)	(24)	(35)	(26)	(31)
Spring (Mar,Apr,May)	(20)	(15)	(21)	(18)
Summer (Jun,Jul,Aug)	(22)	(26)	(47)	(35)
Fall (Sep,Oct,Nov)	(34)	(24)	(6)	(16)
Age, *y*	112	47	46	93
Range	(4 to 84)	(2 to 74)	(2 to 74)	(2 to 74)
Median	(41)	(36)	(31)	(33)

* Patients who had other neurologic diseases, including optic neuritis, multiple sclerosis, amyotrophic lateral sclerosis, drug-induced neuropathy, ischemic neuropathy, neuropathy associated with diabetes, Charcot-Marie-Tooth disease, chronic inflammatory demyelinating polyneuropathy, and polyneuropathy associated with IgM paraproteinemia.

creased the sensitivity but improved the specificity of the assays.

Serologic Assays in Patients with the Guillain-Barré Syndrome and in Controls

Fifty-eight percent of the case patients (69 of 118) had an optical density ratio ≥1 in the IgA assay compared with 35% (36 of 103) of the controls (odds ratio, 2.6; $P < 0.001$) (Table 3). In the IgG assay, 56% of case patients and 41% of controls had an optical density ratio ≥1 (odds ratio, 1.9; $P = 0.02$). In the IgM assay, 61% of case patients and 42% of controls had optical density ratios ≥1 (odds ratio, 2.2; $P = 0.004$). When the threshold for defining a positive response was increased to an optical density ratio ≥2 or ≥3, the strength of the association between the Guillain-Barré syndrome and serologic evidence of previous *C. jejuni* infection also

increased. Seventy (59%) of case patients and 39 (38%) of controls had an optical density ratio ≥1 in two or more immunoglobulin classes (odds ratio, 2.3; $P = 0.001$). Increasing the required optical density ratio to ≥2 or ≥3 and increasing the number of immunoglobulin classes that must exceed the threshold for a serum sample to be classified as having a positive serologic response increased the strength of the association between the Guillain-Barré syndrome and serologic evidence of previous *C. jejuni* infection (see Table 3). Distribution of the optical density ratios was analyzed using the most specific definition of a serologic response that still provided more than 50% sensitivity, which was an optical density ratio ≥ 2 in two or more classes (specificity = 97%; see Table 2). Use of this criterion for seropositivity showed that patients with the Guillain-Barré syndrome were 5.3 times more likely to have

Table 2. *Campylobacter jejuni* Antibodies in Patients with Culture-proven *C. jejuni* Infections and in Controls*

Threshold	Percentage with Optical Density Ratio Exceeding Threshold		Odds Ratio	95% CI	*P* Value
	Infected Patients (*n* = 17)	Controls (*n* = 39)			
IgA					
≥1	88	15	41.2	5.1 to 175	<0.001
≥2	41	3	26.6	2.7 to 648	<0.001
≥3	18	3	8.1	0.6 to 222	<0.08
IgG					
≥1	82	18	21.3	4.2 to 130	<0.001
≥2	47	8	10.7	2.0 to 65.4	<0.002
≥3	24	0	ND†	NA‡	<0.007
IgM					
≥1	100	15	ND	NA	<0.001
≥2	94	5	296	8.9 to 2150	<0.001
≥3	94	5	296	8.9 to 2150	<0.001
Two classes					
≥1	100	10	ND	NA	<0.001
≥2	65	3	100	6.8 to 1740	<0.001
≥3	41	0	ND	NA	<0.001
Three classes					
≥1	71	8	33	2.0 to 200	<0.001
≥2	24	0	ND	NA	<0.07
≥3	6	0	ND	NA	>0.2

* The specificity may be determined by subtracting the percentage exceeding threshold from 100.
† ND = not defined because one cell had a value of zero.
‡ NA = not accurate.

Table 3. *Campylobacter jejuni* Antibodies in Patients with the Guillain-Barré Syndrome and in Controls

Threshold	Patients with the Guillain-Barré Syndrome (n = 118)*	Disease Controls (n = 56)†	Healthy Controls (n = 47)	All Controls (n = 103)	Odds Ratio‡	95% CI	P Value
	←		%		→		
IgA							
≥1	58	34	36	35	2.6	1.5 to 4.8	<0.001
≥2	35	14	11	13	3.7	1.8 to 7.7	<0.001
≥3	17	7	2	5	4.0	1.4 to 12.5	0.005
IgG							
≥1	56	30	53	41	1.9	1.0 to 3.2	0.020
≥2	39	14	23	18	2.9	1.4 to 5.5	<0.001
≥3	24	7	9	8	3.7	1.5 to 9.1	0.001
IgM							
≥1	61	30	55	42	2.2	1.2 to 3.8	0.004
≥2	41	14	28	20	2.7	1.4 to 5.3	0.001
≥3	25	7	15	11	2.7	1.2 to 6.3	0.008
Two classes							
≥1	59	32	45	38	2.3	1.4 to 4.3	0.001
≥2	36	9	11	10	5.3	2.4 to 12.5	<0.001
≥3	15	2	4	3	5.9	1.6 to 25.0	0.002
Three classes							
≥1	33	9	23	16	2.7	1.3 to 5.6	0.003
≥2	16	2	2	2	10.0	2.1 to 50.0	<0.001
≥3	5	0	0	0	ND§	0.0 to 1.0	0.020

* Percentage with optical density ratio exceeding threshold.
† Patients who had other neurologic diseases. For complete list, see Table 1.
‡ Odds ratio for comparison of patients with the Guillain-Barré syndrome and all controls.
§ ND = not defined because one cell had a value of zero.

serologic evidence for *C. jejuni* infection than were the controls; we used this definition in all subsequent assays.

Epidemiologic Characteristics of Patients with the Guillain-Barré Syndrome Who Had *Campylobacter jejuni* Infection

Male patients with the Guillain-Barré syndrome were more likely to have had serologic evidence of previous *C. jejuni* infection. Of 67 male patients with the Guillain-Barré syndrome, 31 (46%) had positive *C. jejuni* serologic results compared with 11 (22%) of 49 female patients (odds ratio, 3.0; *P* = 0.009). In contrast, no differences were observed in the rate of seropositivity between male and female controls. The association between the Guillain-Barré syndrome and seropositive responses was observed in all age groups but was strongest in persons older than 60 years (Table 4). The rate of seropositive responses among patients with the Guillain-Barré syndrome increased from 15% in those younger than 20 years to 47% in those older than 60 years; among controls, the rate of seropositive responses did not increase with age (see Table 4). In a separate analysis, the mean age of patients with the Guillain-Barré syndrome and a positive seropositive response (42 years) was greater than that of patients with the syndrome who had a seronegative response (36 years) (*P* = 0.02).

An association between the Guillain-Barré syndrome and previous *C. jejuni* infection was observed in all seasons but reached statistical significance only in summer and autumn (Table 5). Because the peak incidence of *C. jejuni* infections in the United States occurs from August through October (22) and because in previous

anecdotal case reports the Guillain-Barré syndrome had occurred from 1 to 3 weeks after *C. jejuni* infection (23-25), we compared the proportion of serologically positive patients who had the syndrome occurring from September to November with those occurring during the rest of the year. Of 39 patients with the Guillain-Barré syndrome who had onset of symptoms from September to November, 20 (51%) had serologic evidence of previous *C. jejuni* infection, compared with 22 (29%) of 77 patients with onset of neurologic symptoms during other months (odds ratio, 2.6; 95% CI, 1.10 to 6.25; *P* = 0.02).

Discussion

Our study showed a strong association between the Guillain-Barré syndrome and serologic evidence of recent *C. jejuni* infection. Although the assay used was

Table 4. Rate of Seropositive Responses by Age Group in Patients with the Guillain-Barré Syndrome and in Controls

Age Group (y)	Patients	Controls	Odds Ratio	95% CI	P Value
	n (% positive)*				
0 to 19	20 (15)	14 (7)	2.3	0.2 to 50.0	>0.2
20 to 39	33 (36)	44 (9)	5.9	1.4 to 25.0	0.004
40 to 59	29 (38)	21 (14)	3.7	0.8 to 20.0	0.07
>60	30 (47)	14 (7)	11.1	1.2 to NA†	0.01
All	112 (36)	93 (11)	5.2	2.2 to 12.5	<0.001

* Seropositive response = optical density ratio ≥2 in two or more immunoglobulin classes.
† NA = not accurate.

Table 5. Rate of Seropositive Responses by Seasons in Patients with the Guillain-Barré Syndrome and in Controls

Month Blood Drawn	Patients	Controls	Odds Ratio	95% CI	P Value
	n (% positive)				
Winter (Dec, Jan, Feb)	28 (25)	25 (12)	2.4	0.5 to 14.3	0.20
Spring (Mar, Apr, May)	23 (30)	14 (7)	5.6	0.6 to 100.0	0.10
Summer (Jun, Jul, Aug)	26 (31)	28 (4)	12.5	1.3 to NA†	0.009
Autumn (Sep, Oct, Nov)	39 (51)	13 (7)	12.5	0.6 to 33.3	0.006

* Seropositive response = optical density ratio ≥2 in two or more immunoglobulin classes.
† NA = not accurate.

not highly sensitive or specific, increasing the specificity of the test by increasing the required optical density ratio for classification as a positive result increased the strength of the association. Furthermore, in our study the greatest proportion of patients with the Guillain-Barré syndrome who had *C. jejuni* antibodies occurred during the autumn months, which immediately follow the period of peak incidence of culture-proven *C. jejuni* infections (22). These observations suggest that the assays were detecting specific *C. jejuni* antibodies in truly infected persons.

Assay Validation

A number of features lend further credence to our results of this study. First, the assays were done using standardized techniques in a blinded manner, substantially reducing the possibility of investigator bias. Each serum sample was assayed several times in duplicate to assure reproducibility of results. Regression analyses provided more precise thresholds for determination of positive results than were used in previous investigations (11-13). The number of sera tested was large, and samples were obtained from three university medical centers making it unlikely that the results represented an institutional or regional phenomenon.

Description of Patients and Controls

The patient and control groups were similar for gender and race but were somewhat different for age (the control group was younger) and for the time of year blood was obtained (the control group was more likely to have blood drawn during the summer). However, because the incidence of *C. jejuni* infections is greater among younger adults and during the summer months (22), any bias introduced by these differences would have resulted only in an underestimation of the association between the Guillain-Barré syndrome and serologic evidence of *C. jejuni* infection. Furthermore, because *C. jejuni* infection may be present among household contacts, use of family members of patients with the Guillain-Barré syndrome as controls also may have resulted in an underestimation of the association between the syndrome and *C. jejuni* infection. Indeed, the optical density ratios among the healthy control group were greater than those observed in the disease controls and in the healthy control group used initially to determine the accuracy of the assay.

The distribution of seropositive responses among patients with the Guillain-Barré syndrome in our study

suggests that subgroups of patients may exist who are more likely to have had previous *C. jejuni* infection. The proportion of patients with the Guillain-Barré syndrome and *C. jejuni* antibodies was greatest in male patients, in persons infected in summer months. The proportion also increased with age. Although *C. jejuni* infections are known to peak during late summer (22), male gender and older age may represent additional risk factors for the development of the Guillain-Barré syndrome after infection with *C. jejuni*.

Previous Studies

Because the duration of convalescent excretion of *C. jejuni* is brief (mean, 16 days after onset of diarrhea) (22), and the Guillain-Barré syndrome associated with *C. jejuni* typically occurs 1 to 3 weeks after onset of diarrhea (24-27), reliance on isolation of *C. jejuni* from fecal cultures is an insensitive measure of previous infection. Other investigators have used serologic tests to determine the frequency of *C. jejuni* infection as an antecedent to the Guillain-Barré syndrome. However, the studies have been limited by use of nonstandardized methods, absence of appropriate controls, or failure to "blind" the investigators to the source of the sera studied (11-13). Using immunodot assays (28) to determine the frequency of *C. jejuni* antibodies, Gruenwald and colleagues (11) found that 3 (18%) of 17 patients with the Guillain-Barré syndrome had increased titers in two or more immunoglobulin classes. Similarly, using a complement fixation technique, Winer and colleagues (5) found that 14% of 99 patients with the Guillain-Barré syndrome had positive *C. jejuni* serologic test results. However, the absence of control groups in these investigations makes it difficult to interpret these results. Increased *C. jejuni* antibodies were found in 41% of 32 children diagnosed with the Chinese paralytic syndrome, but the control groups studied were from Thailand (29), and the relevance of this disease to the Guillain-Barré syndrome has not been established (30). Kaldor and Speed (12) used nonstandardized methods to study serum from 56 patients with the Guillain-Barré syndrome and from 57 controls; in their unblinded analysis, they found 38% of the patients and none of the controls met their criteria for positive serologic responses. Despite their limitations, each of these previous studies yielded results consistent with our findings.

Incidence and Pathologic Mechanism

The incidence of the Guillain-Barré syndrome is slightly higher in men (4), and in our study, male pa-

tients outnumbered female patients, 3 to 2. The incidence of *Campylobacter* infection is also slightly higher in men (22). Male patients with the Guillain-Barré syndrome were three times as likely as female patients to have serologic evidence of previous *C. jejuni* infection. Host factors, including immunologic response to *C. jejuni* infection that is genetically linked (31), may predispose patients to the development of the Guillain-Barré syndrome after *C. jejuni* infection. Of 6 patients with the Guillain-Barré syndrome in Japan who had antecedent *C. jejuni* infection, all had the HLA-B35 antigen, compared with only 14% of 3090 healthy controls (32). Previous studies of patients with the Guillain-Barré syndrome had not shown an association with a particular HLA type (33), but the data had not been stratified for presence of *C. jejuni* infection.

The hallmark of the Guillain-Barré syndrome is segmental demyelination of peripheral nerves accompanied by mononuclear infiltrates and edema. However, the pathogenic mechanism involved in the Guillain-Barré syndrome after *C. jejuni* infection is not known. Some investigators have suggested that relatively few *C. jejuni* strains are capable of triggering an immunologic response that is associated with myelin destruction. The findings of two Japanese groups indicated that a particular serotype of *C. jejuni*, Penner type O19, was associated with the initiation of the Guillain-Barré syndrome (34, 35). Whether the O19 strains represent a particularly virulent clone, or whether the O19 polysaccharide is itself a virulence determinant or a marker for other virulence factors is not known. The neurologic target for immunologic injury also is unknown. Fujimoto and Amako (36) showed that serum from a patient who acquired the Guillain-Barré syndrome shortly after a *C. jejuni* infection reacted strongly with P0, a peripheral nerve myelin-specific protein. They suggested that *C. jejuni* has antigenic characteristics that stimulate production of antibodies that react with peripheral nerve myelin and cause the Guillain-Barré syndrome. Similarly, other investigators have proposed that gangliosides found in myelin may be the ultimate target of antibodies formed in response to *C. jejuni* infection. Thus, anecdotal reports exist of the Guillain-Barré syndrome occurring after administration of parenteral gangliosides (37, 38).

Conclusion

It is difficult to estimate the exact number of patients with the Guillain-Barré syndrome who had previous *C. jejuni* infection. Previous epidemiologic studies have found that 10% to 30% of patients with the Guillain-Barré syndrome have antecedent acute gastrointestinal illnesses. Our results suggest that the specificity of the test is close to 100% (the number of controls exceeding threshold is close to zero) when the definition of a positive response is an optical density ratio ≥ 3 in two or more immunoglobulin classes or optical density ratio ≥ 2 in all three classes. Using this most stringent definition of "positive," about 15% to 20% of patients with the Guillain-Barré syndrome are classified as positive; this provides a conservative estimate of the number of patients with the syndrome who also had

C. jejuni infection. Although the biological significance of this association cannot be established by this study, our results provide further evidence that *C. jejuni* infection is a common initiator of the Guillain-Barré syndrome.

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