

A Case-control Study on Children with Guillain-barre Syndrome in North China¹

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Objective To explore the risk factors for Guillain-barre syndrome. **Methods** Case-control study design was used in 51 cases of Guillain-barre syndrome, and 51 matched controls. All of the 51 cases in this study had been examined by electrophysiology. Serum IgG antibodies specific for *C. jejuni* were determined in all the subjects by ELISA. Each case and control were interviewed using an ad hoc questionnaire, including his/her demographic information, onset of the illness, their personal hygiene and so on. **Results** The study showed that Guillain-barre syndrome was associated with a few factors, such as polio vaccine immunization before onset of illness ($OR=7.27$), no hand washing after defecation and before meals ($OR=6.15$). Infection of *C. jejuni* was strongly associated with the illness ($OR=9.5$, $P<0.001$). **Conclusion** It is suggested that occurrence of Guillain-barre syndrome may correlate to infection of *C. jejuni* and poor personal hygiene in children.

Key words: Guillain-Barre syndrome (GBS); Case-control study; *Campylobacter jejuni*

INTRODUCTION

Guillain-barre syndrome (GBS), characterized as acute flaccid paralysis (AFP), also called acute inflammatory demyelinating polyneuropathy and Landry's ascending paralysis, is an inflammatory disorder of the peripheral nerves. It is characterized by the rapid onset of weakness and often, paralysis of the legs, arms, breathing muscles and face. Abnormal sensations are often accompanied by general weakness. Although most people suffering from GBS take months to recover, some may have long term disabilities of various degrees, and less than 5% die. In China the prevalence of GBS is 16.2 per 100 thousand, and the occurrence of GBS in children under 14 years account for 42.6% of all the GBS cases. It is

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reported that GBS is prevalent in children living in North China^[1,2]. Based on its electrophysiological features, GBS can be classified into two subtypes^[3,4], one with typical acute inflammatory demyelinating polyneuropathy (AIDP) and the other with acute motor axonal neuropathy with extensive Wallerian-like degeneration of nerve fibers (AMAN). Each of both has its features in epidemiology, electrophysiology and pathology. Up to now, studies suggested that occurrence of GBS is correlated with a variety of risk factors equivocally, including infection of *C. jejuni*^[5]. Various diseases are described as antecedents of Guillain-barre syndrome (GBS), particularly gastroenteritis and respiratory infectious diseases, but epidemiological surveys are rare^[6]. To identify the risk factors for the disease using a case-control study is of importance for its prevention and control.

MATERIALS AND METHODS

Study Subjects

A case of GBS was defined based on the following characteristics^[7-9]:

(a) Acute onset, progressive motor weakness of more than one limb in less than four weeks, with areflexia, often with cranial nerve or respiratory paralysis, without fever at the onset of neurological symptoms.

(b) Normal sensory function, or mild sensory symptoms.

(c) Exclusion of poliomyelitis (with laboratory evidence), acute spinal shock, myasthenia gravis, polymyositis, etc.

(d) Autonomic dysfunction.

(e) Electrophysiological examinations during the first two weeks of the illness showed reduced amplitude of muscular compound action potential after stimulation, prolonged latency of distal motor nerve conduction, normal or slightly reduced velocity of nerve conduction, and normal or prolonged F-wave latency.

(f) Cerebral spinal fluid examination showed elevated protein content (>0.4 g/L) without pleocytosis (<10 cells/mm³).

(g) Precursor illness, such as upper respiratory infection, gastrointestinal infection, or vaccine immunization, surgical operation within four weeks before onset.

Diagnostic criteria for AIDP: Those with two or more of the following items in electrophysiological examinations at the first three weeks of onset could be diagnosed as AIDP.

(a) Amplitude of distal compound muscular action potential was greater than 50% of the lower limit, and nerve conduction velocity was less than 90% of the lower limit of the normal; or amplitude of distal compound muscular action potential was less than 50% of the lower limit, and nerve conduction velocity was less than 85% of the lower limit of the normal.

(b) Normal amplitude of distal compound muscular action potential, distal latency was 10% greater than the upper limit and amplitude of distal compound muscular action potential was 20% less than the lower limit of the normal.

(c) Amplitude of proximal compound muscular action potential or amplitude of distal compound muscular action potential was less than 0.7 with evidence of unequivocal temporal dispersed waves.

(d) F-wave latency was 20% greater than the upper limit of the normal.

Diagnostic criteria for AMAN:

- (a) No demyelinating manifestations mentioned above.
- (b) Amplitude of distal compound muscular action potential was less than 80% of the lower limit of the normal.
- (c) If more than two pieces of nerves showed loss of potential, no classification could be made.

Fifty-one cases of GBS were admitted respectively to Beijing Children's Hospital, Beijing (29 cases), the First Hospital Affiliated to Norman Bethune University of Medical Sciences, Changchun (17 cases) and the First Hospital Affiliated to Inner Mongolia Medical College, Hohhot (5 cases), and they came from Beijing, Hebei, Jilin, Liaoning, Heilongjiang and Shanxi Provinces, as well as Inner Mongolia Autonomous Region.

Controls were selected from healthy children living in the same areas matched on age and sex.

Determination of Serum IgG Antibodies Specific for C. jejuni

Blood specimens were collected from the cases and controls and stored at -70°C. Sera were diluted 50-fold for detecting IgG antibodies specific for *C. jejuni* determined by ELISA using a preparation of surface antigen of *C. jejuni* C1 strain prevalent in North China isolated from the patients. The plates were read in duplicate at 450 nm with the results expressed as an optical density (OD) value for each well. A positive serological response was defined as an OD ratio (a ratio of the OD values for each serum sample of the case to the negative control) exceeding 2.0.

Poliomyelitis was excluded for each case by his/her negative serum IgM and IgG antibodies against poliomyelitis virus and negative fecal virus isolation.

Interview of Subjects

Each case and control were interviewed with his/her parents or guardians by a trained interviewer using an ad hoc questionnaire, including his/her demographic information, socioeconomic status, onset process of the illness, potential risk factors for the illness in environment and personal hygiene.

Data Analysis

All the data were input into a dataset using d-BASE III software in a microcomputer. Statistical analysis was performed with SAS release 6.04.

RESULTS

General Situation

Of the 51 cases, 20 were classified as AIDP, 28 AMAN and 3 unclassified; 26 were male (51.0%) and 25 female; and 6 living in urban areas and 45 in rural areas (88.2%). Their age at hospitalization averaged 6.1 years with a range of one to fourteen years, and 14 cases (27.4%) were one to four years, 19 (37.3%) were five to eight years, and 18 (35.3%) were nine to fourteen years.

Thirty-eight (74%) of 51 cases occurred the disease during June to October, which lasted 1 to 14 d, with an average of 6.3 d.

Thirteen of the 51 cases (25.5%) had previous medical history, 11 (21.6%) were hospitalized before and one was operated on.

Clinically, all the 51 cases were afebrile, 38 had initial symptom of weakness in both of their lower limbs which progressively ascended to upper limbs, 3 had weakness in upper limbs which progressively descended to lower limbs and 10 had weakness in four limbs, 25 had cranial nerve paralysis which was mainly involved in the IX and X cranial nerve, and 35 had abnormal CSF (protein content >0.4 g/L). Fever, diarrhea, common cold, weakness, or paresis were not found in their family members within four weeks before onset.

There was no significant difference in education level and occupation of their parents between cases and controls.

Annual household income averaged RMB1302.9 for the cases and 1678.6 for the controls, with no statistical significance.

Potential Risk Factors

Thirty-seven of the 51 cases had one or more precursor symptoms or illness four weeks before onset, 16 of them had fever (31.4%). 4 had diarrhea, 9 common cold, 13 cough, 3 sore throat, 8 myalgia, 2 arthralgia, and one had headache, neck pain, chickenpox, and mycoplasma infection.

Vaccine immunization history of the cases and controls is shown in Table 1.

TABLE 1

Comparison of Vaccine Immunization History Between Cases and Controls

Vaccine	Cases	Controls	OR	95% CI for OR	P
Polio	40	17	7.27	2.74-19.77	<0.01
Influenza	3	1	3.13	0.27-82.23	>0.05
Hepatitis B	17	7	3.14	1.06-9.61	<0.05
Japanese Encephalitis B	21	14	1.85	0.74-1.85	>0.05

Other factors relevant to the illness in the cases and controls are listed in Table 2.

TABLE 2

Comparison of Other Factors Contributing to GBS Between Cases and Controls

Risk Factors	Cases	Controls	OR	95% CI for OR	P
Have Been Rained on	5	0			
Excessive Fatigue	2	0			
Caught Cold	11	1	13.75	1.66-301.9	<0.01
Have Been to Forest	2	0			
Have Been to River/Lake Site	10	1	12.20	1.47-269.7	<0.01
Mountaineering	3	0			

Family Situation

There was no significant difference in family size, annual household income and housing area between the cases and controls (Table 3).

TABLE 3

Comparison of Family Situation Between Cases and Controls ($\bar{x} \pm s$)

Family Features	Cases	Controls	F	P
No. Members	3.7±1.0	3.7±0.8	0.08	>0.05
Annual Income (yuan)	1661.2±1417.2	2038.3±1851.2	1.06	>0.05
Total Housing Area (m ²)	89.2±185.1	104.3±274.5	0.15	>0.05
Housing Area Per Capita (m ²)	22.1±36.4	28.0±68.4	0.00	>0.05

Kinds of crops planted in the families of the cases and controls are shown in Table 4.

TABLE 4

Comparison of Crops Planted In the Families of Cases and Controls

Crops Planted	Cases	Controls	OR	95% CI for OR	P
Wheat	15	16	0.73	0.28-1.90	>0.05
Barley	1	1	0.86	0.02-33.2	>0.05
Corn	25	22	0.96	0.39-2.4	>0.05
Millet	1	1	0.86	0.02-33.2	>0.05
Rice	2	2	0.86	0.08-9.1	>0.05
Sorghum	1	0	----		
Soy Bean	4	5	1.16	0.20-7.14	>0.05
Peanut	0	0	----		
Cotton	1	2	0.42	0.01-6.32	>0.05

There was no significant difference in exposure to pesticide, domestic animal raising, exposure to animals and insects, sources of drinking water, distances between water source and toilets, between chicken pens and water source, and between pigsties and water source in the cases and controls.

Personal Hygiene

Hand washing habit before meals and after defecation in cases and controls is listed in Table 5.

TABLE 5

Hand-washing Before Meals and After Defecation in Cases and Controls

Hand-washing	Cases	Controls	OR	χ^2	P
Every Time	13	16	1.00		
Often	7	21	0.41	2.46	>0.05
Occasionally	20	4	6.15	8.29	<0.01
Unknown	11	10	----		

Serum IgG Antibodies Specific for *C. jejuni*

Positive serum IgG antibody response for *C. jejuni* in 19 of the 51 cases was compared with that in 3 of the 51 controls, with an OR of 9.5 (95% CI of 2.37-44.22).

OD value for serum IgG antibodies against *C. jejuni* averaged 0.171 ± 0.145 in cases and 0.063 ± 0.0073 in controls, with statistically significant difference (Kruskal-Wallis $H=14.67$, $P<0.01$).

DISCUSSION

GBS has been named for about eighty years^[8]. It is prevalent in children living in North China^[1,2]. But many cases were diagnosed only based on clinical symptoms and signs without any electrophysiological or pathological examination. All of the 51 cases in this study had been examined by electrophysiology, moreover, and pathological changes were identified in 8 cases mainly as axonal denaturalization or demyelination through the sural nerve biopsy. All the diagnoses were reliable.

In the past, GBS was always known as only one subtype-AIDP, but now we find that there is still another subtype-AMAN existing in North China. So GBS can be classified into

two subtypes-AMAN and AIDP in North China^(1,2). Some researches have shown that infection of *C. jejuni* is associated with GBS^(5,10). But most of the researches were case reports or series of case reports, and only few was designed using a case-control study. In this case-control study we found that among the 51 cases in this study, 19 cases were positive on IgG antibodies specific for *C. jejuni*. (11 cases were diagnosed as AMAN and 8 cases as AIDP), while there were only 3 whose IgG antibodies specific for *C. jejuni* were positive in the controls, in which implicated infection of *C. jejuni* was a potential risk factor, which was strongly associated with the illness ($OR=9.5$, $P<0.001$), and also contributed to explain the pathogenesis of GBS and its variety of clinical manifestations. In this study we could not analyze AMAN and AIDP because the number of the cases was too limited. Evidence of recent or ongoing *C. jejuni* infection has been found in about one out of every four cases of Guillain-Barre syndrome (GBS)⁽¹¹⁾. A case-control study reported that fifteen percent of cases versus 5% of hospital controls had *C. jejuni* infection ($P<0.003$, $OR=3.96$, 95% CI: 1.08-17.85)⁽¹²⁾. It is increasingly accepted that *C. jejuni* infection is an important causal factor for GBS.

It was reported that the outbreak of GBS was associated with water pollution⁽¹³⁾. *C. jejuni* was named in 1963. The most obvious symptom of *C. jejuni* infection is diarrhea. The main hosts of *C. jejuni* are poultry⁽¹⁴⁾, which could partly explain why this disease is prevalent in rural areas.

Results of the case-control study show that GBS is also associated with a few factors, such as polio and hepatitis B immunization, getting cold and going to river or lake site before onset of the illness. Thirty-five of the 51 cases had precursor symptoms before onset (68.6%). This is coincident with other studies, suggesting that most cases have respiratory infection and vaccine immunization history⁽⁶⁾. Poor personal hygiene is also a potential risk factor for the illness.

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