

The diagnosis of neuromuscular disease increasingly is relying on the use of immunological laboratory tests. Conditions such as Guillain-Barré syndrome and myasthenia gravis can now be explained in terms of specific immunological profiles, as John Winer explains.

Antibodies and clinical neurological syndromes

Neurology has a reputation for being a largely clinical specialty and the diagnosis of many neurological diseases rests on the skill of the clinician in interpreting the neurological symptoms and clinical findings on examination. In recent years these clinical skills have been supported by an increasing number of immunological tests that permit much more precise diagnosis and the use of better therapy.

The measurement of antibodies targeted to antigens in the nervous system are the most reliable and well established of these tests and this article attempts to summarise the diseases and antibodies of the greatest clinical relevance to neurological practice. Table 1 lists the main clinical syndromes and the antibodies associated with them. Antibodies used in the diagnosis or monitoring of patients with paraneoplastic disease are not included as these are outside the scope of this paper.

Immune-mediated neuropathies

Guillain-Barré syndrome (GBS) is an acute, predominantly motor neuropathy that may be precipitated by infection and is usually monophasic in its course. Its name is derived from an original paper by Georges Guillain and colleagues in 1916 in which two patients seen by the authors during the First World War developed a rapidly progressive paralysis.

Guillain and colleagues examined the cerebrospinal fluid (CSF) of the two patients and found a high level of protein. However, unlike syphilis, the CSF cell count was normal – a finding they labelled “albuminocytological” dissociation.¹ The significance of the absent reflexes was noted and the correct conclusion drawn that this was a neuropathy and not a spinal cord disease, as had previously been considered the cause of such cases.

Many subsequent cases were described with a much more severe outcome and it was

CLINICAL SYNDROME	ANTIBODY PATTERN
Chronic sensorimotor demyelinating neuropathy	IgM against MAG
Chronic axonal sensory neuropathy	IgM sulphatide
Chronic sensory ataxic neuropathy	IgM NeuNacNeuNac
Multifocal motor neuropathy	IgM GM1
Miller Fisher syndrome	IgG GQ1b
GBS	GM1, GD1a, GT1
Myasthenia	AchR, MuSK

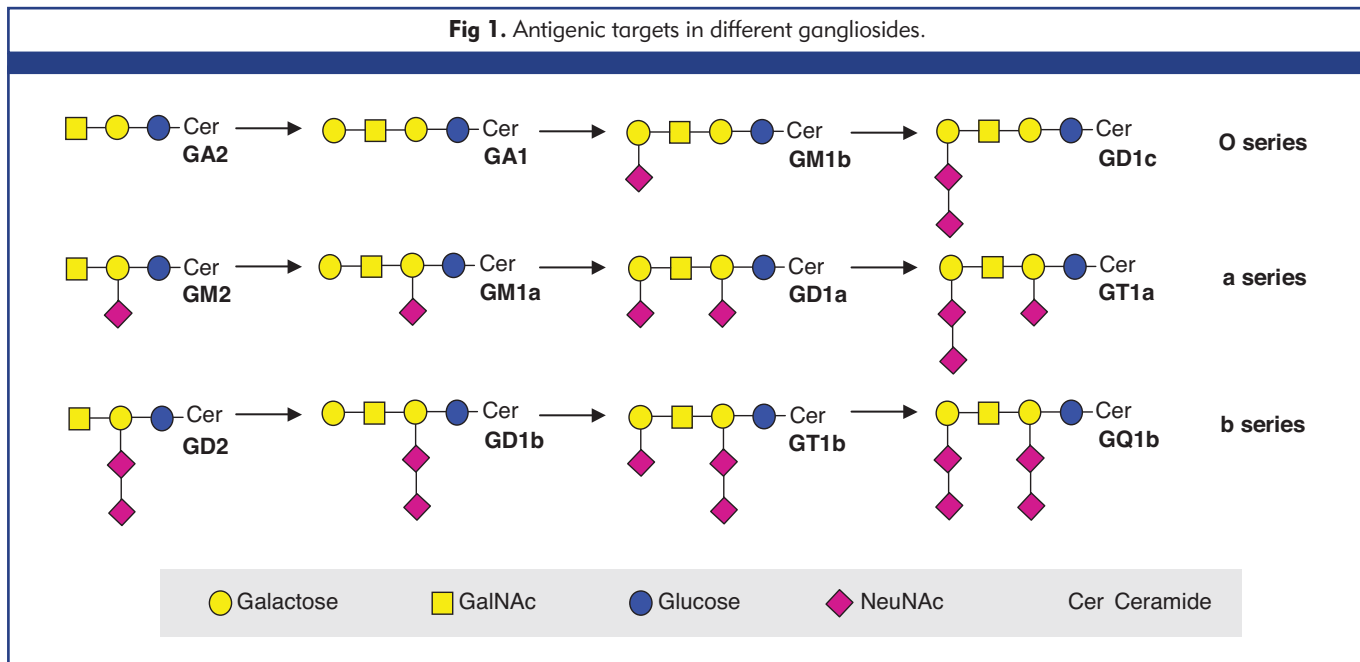
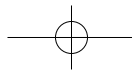
realised that respiratory muscles could be involved leading to death from hypoxia and retention of fluid due to a weak cough. Diagnostic criteria for GBS were formulated in 1978, mostly for the purposes of defining a uniform population for research studies.²

Neurophysiological studies suggest that patients with GBS can be divided into two main groups: those with a demyelinating neuropathy and those with a primarily axonal process. This either can be entirely motor (acute motor axonal neuropathy [AMAN]) or affect both motor and sensory nerves (acute motor and sensory neuropathy [AMSAN]). A striking variant of GBS involves the cranial nerves to the extraocular muscles and

produces ataxia and absent reflexes (Miller Fisher syndrome [MFS]). Clinical and pathological studies have better defined these variants of GBS,^{3,4} and it seems likely that the antigenic target varies between these subtypes of GBS (Table 2).

In view of its severity and the striking clinical picture of severe, rapidly progressive weakness, often in young patients, the pathogenesis of the disease has been the subject of many studies. The association of the syndrome with preceding infection with *Campylobacter jejuni*,⁵ Epstein Barr virus and cytomegalovirus⁶ is well documented, although the risk of developing GBS after infection is very low.

ANTIBODIES	
AMAN	GD1a, GM1, GM1b, Ga1NAC
AMSAN	–
MFS	GQ1b
GBS/MFS	GQ1b
Pharyngeocervicobrachial	GT1a, GQ1b
AIDP minority	GM1, GM2, gal c
AIDP majority	Unknown



Complement fixing antibodies against peripheral nerves was first noted in some patient with GBS in the early 1960s.⁷ Extensive searches for antibodies against myelin proteins proved unrewarding, but a much higher frequency of antilipid antibodies was detected. These are largely directed against gangliosides, which share important epitopes so that antiganglioside antibodies frequently will react with a variety of different gangliosides, giving a characteristic pattern (Fig 1).

Patients with AMAN usually have antibodies against GM1 gangliosides, while patients with MFS have antibodies against GQ1b gangliosides. The association of GQ1b and MFS is very tight, with more than 90% of patients having positive antibody serology in the acute phase of the disease.⁸ There is evidence that these antibodies interfere with neuromuscular junction transmission, at least in *in vitro* animal preparations,⁹ and the detection of these antibodies is of considerable diagnostic use.

A variable proportion of patients with more typical GBS have antibodies against other gangliosides (eg GD1a and GT1) but these are probably in a minority. The value of these

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antibodies’

antibodies in diagnosis is much less than is the case with GQ1b, but their presence in patients with an undiagnosed neuropathy increases the chance of a post-infective or immune-mediated pathogenesis. While these antibodies might simply mark the presence of another immunological process, there is evidence that GM1 antibodies might be more fundamental in the disease process. Yuki has shown that rabbits injected with anti-GM1 antibodies develop an illness that resembles AMAN in humans in both its clinical picture and histological findings.¹⁰

Chronic neuropathies

The clinical picture of a relatively slowly progressive chronic sensory ataxic neuropathy may be associated with an IgM kappa paraprotein. Several studies have demonstrated that these patients usually have circulating antibodies to the protein myelin-associated glycoprotein (IgM anti-MAG),¹¹ which is a minor component of myelin.

While IgM anti-MAG is very helpful diagnostically, there is some debate about whether or not it has an important causal role in these neuropathies. Frequently, immunosuppression is ineffective in changing the course of these patients, although there have been a few reports of response to treatment with drugs such as rituximab.¹²

Another sensory ataxic neuropathy is associated with IgM anti-disialosyl antibodies against an epitope consisting of NeuAc(α 2-8)NeuAc(α 2-3)Gal, which is present in many gangliosides including GD1b, GD3, GT1b and GQ1b (Fig 1). These IgM antibodies may also be cold agglutinins.

The clinical picture is of a progressive sensory ataxia with relatively preserved motor function and areflexia. A high proportion of cases have motor weakness of the oculomotor or bulbar muscles, which may fluctuate. These features are similar to MFS but show a much more chronic course.

‘A wider range of
antibody tests for
neuromuscular
disease is likely to
become more
generally available
over the next
few years’

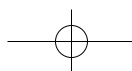
Interestingly, acute-phase disialylated ganglioside antibodies are a feature of MFS. The acronym CANOMAD (Chronic Ataxic Neuropathy, Ophthalmoplegia, IgM paraprotein, cold Agglutinins, Disialosyl antibodies)¹³ has been applied to these patients.

Myasthenia

Myasthenia gravis is an uncommon disease with an incidence of about four cases per 100,000 population. The classical features of myasthenia are weakness and fatigue, which frequently affects the extraocular muscles. While some cases of myasthenia are congenital and reflect genetic disorders of the proteins involved in signalling at the neuromuscular junction, most cases are immune-mediated.

Approximately 80% of patients with myasthenia have antibodies against the acetylcholine receptor (anti-AChR), which is a transmembrane protein containing four subunits arranged in a pentamer (α 2, γ δ). In myasthenia the majority of the antibodies are directed against the α -subunit.

In those patients without anti-AChR antibodies there is evidence of another



serum-derived factor that could interfere with neuromuscular transmission *in vitro*.¹⁴ Recently, this has been shown to be largely an antibody to muscle specific kinase (MuSK).¹⁵ Antibodies against both AChR and MuSK have become very useful diagnostic tools.

A view of the future

Immunological tests for antibodies play an important part in the diagnosis and management of patients with a wide variety of neuromuscular diseases. New antibodies are being described with increasing frequency. While some of these antibody tests (eg anti-AChR antibodies) have become established in many laboratories, others (eg anti-G1bQ ganglioside) currently are only available in a small number of laboratories. However, they are likely to become more generally available over the next few years. .

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