

Brief Report

PARANEOPLASTIC CEREBELLAR
ATAXIA DUE TO AUTOANTIBODIES
AGAINST A GLUTAMATE RECEPTOR

PETER SILLEVIS SMITT, M.D., PH.D.,
AYAE KINOSHITA, M.D., PH.D., BERTIE DE LEEUW, PH.D.,
WIEBE MOLL, M.D., PH.D., MICHEL COESMANS, M.Sc.,
DICK JAARMA, PH.D.,
SONJA HENZEN-LOGMANS, M.D., PH.D.,
CHARLES VECHT, M.D., PH.D., CHRIS DE ZEEUW, PH.D.,
NAOHIRO SEKIYAMA, PH.D.,
SHIGETADA NAKANISHI, M.D., PH.D.,
AND RYUICHI SHIGEMOTO, M.D., PH.D.

THERE are many types of cerebellar ataxia, including ataxia due to congenital or metabolic disorders and a paraneoplastic form in patients with gynecologic cancer, breast cancer, lung cancer, or Hodgkin's disease.¹ This paraneoplastic syndrome is the only type of cerebellar ataxia associated with autoantibodies against neuronal antigens. Often, the neuronal antigens are aberrantly expressed by the tumor cells.²⁻⁴ The antineuronal autoantibodies are believed to cause cerebellar ataxia, but this is unproved.^{5,6} In Hodgkin's disease, the lymphoma precedes the ataxia by months to years in 80 percent of patients, and ataxia often occurs during a prolonged complete remission.⁴ Among patients with this type of ataxia, 30 percent have anti-Purkinje-cell antibodies, some of which have the features of the neuronal antibody anti-Tr.^{4,7}

We identified a new autoantibody in two patients with severe cerebellar ataxia that developed while they were in remission from Hodgkin's disease. The antibody reacts specifically with the metabotropic glutamate receptor mGluR1 in mouse brain. Metabotropic glutamate receptors belong to a large family of cell-surface receptors that transmit signals into the cell by coupling to guanine nucleotide-binding proteins (G proteins) in the cytoplasm. Purified IgG from the serum of both patients blocked the glutamate-stimulated formation of inositol phosphates in Chinese-

hamster-ovary (CHO) cells that expressed mGluR1 α , and the injection of IgG from serum or cerebrospinal fluid into the cerebellar subarachnoid space of mice caused severe, reversible ataxia. These results indicate that antineuronal autoantibodies can cause disease of the central nervous system by blocking neuronal receptors.

CASE REPORTS

Patient 1

In 1995, when she was 19 years old, Patient 1 presented with subacute cerebellar ataxia. She had been treated with four cycles of mechlorethamine, vincristine, procarbazine, and prednisone plus doxorubicin, bleomycin, and vinblastine (MOPP-ABV) followed by subtotal nodal irradiation for stage IIA nodular sclerosing Hodgkin's disease. She had been in remission for two years when truncal ataxia, intention tremor, and gait ataxia developed. An examination of the brain with magnetic resonance imaging (MRI) was normal. The cerebrospinal fluid contained 28 mononuclear cells per cubic millimeter and had a protein concentration of 28 mg per deciliter. The IgG concentrations in cerebrospinal fluid and serum were 4.6 mg per deciliter (normal value, <8) and 0.89 g per deciliter, respectively. The albumin concentrations in cerebrospinal fluid and serum were 17 mg per deciliter and 4.8 g per deciliter, respectively. The calculated IgG index was 1.2 (an IgG index of more than 0.6 indicates intrathecal IgG synthesis).⁸ Cytologic examination showed no malignant cells. Serum and cerebrospinal fluid contained IgG antineuronal antibodies of unknown specificity.⁹

These findings strongly suggested a diagnosis of paraneoplastic cerebellar ataxia. The patient was treated with four plasma exchanges at intervals of two to three days, oral prednisone at a dose of 40 mg per day for six weeks, and two courses of intravenous immune globulin (total dose, 4 mg per kilogram of body weight). After the four plasma exchanges, the cerebrospinal fluid was acellular and the IgG concentration was less than 1 mg per deciliter. Over the following seven months, the ataxia slowly disappeared. An examination of serum for antineuronal antibodies when the patient was asymptomatic was negative. The Hodgkin's disease has remained in remission.

Patient 2

In 1996, at the age of 49 years, Patient 2 presented with severe cerebellar ataxia and short-term memory loss. She had been treated for stage II Hodgkin's disease (nodular sclerosing type) nine years earlier with carmustine, cyclophosphamide, vinblastine, procarbazine, and prednisone (BCVPP) and had been in remission since then. She also had polycystic renal disease and had required hemodialysis since 1991. On neurologic examination she was alert and oriented. She could repeat four words, but her recall after five minutes was limited to two words. Comprehension and naming were normal. She spoke with moderate cerebellar dysarthria. Severe appendicular and truncal ataxia with titubation of the head and trunk were present. She could walk only with support from another person. MRI examinations of the brain when ataxia was diagnosed and six months later were normal and did not show cerebellar atrophy. The serum contained IgG antineuronal antibodies that stained the cerebellum in a pattern identical to that of serum from Patient 1.

One year after the onset of ataxia, the patient received a diagnosis of probable paraneoplastic ataxia and was treated with 14 plasma exchanges, but there was no objective improvement of the truncal ataxia and she remained unable to walk without support. In 1998, after the 14 plasma exchanges, the cerebrospinal fluid was acellular and had an IgG concentration of 15 mg per deciliter. The serum IgG concentration was 0.84 g per deciliter. The IgG index was 0.62. High titers of antineuronal antibodies persisted in serum and cerebrospinal fluid. Hodgkin's disease remained in complete remission.

From the Departments of Neuro-Oncology (P.S.S., B.D.L., W.M., M.C., C.V.) and Pathology (S.H.-L.), Daniel den Hoed Cancer Center, University Hospital Rotterdam, the Netherlands; the Departments of Immunology (B.D.L.) and Anatomy (M.C., D.J., C.D.Z.), Erasmus University Rotterdam, the Netherlands; the Departments of Morphologic Brain Science (A.K., R.S.) and Biologic Sciences (N.S., S.N.), Faculty of Medicine, Kyoto University, Sakyo-ku, Kyoto, Japan; and the Laboratory of Cerebral Structure, National Institute for Physiologic Sciences, Myodaiji, Okazaki, Japan (A.K., R.S.). Address reprint requests to Dr. Sillevs Smitt at the Department of Neuro-Oncology, Daniel den Hoed Cancer Center, P.O. Box 5201, 3008 AE Rotterdam, the Netherlands, or at sillevs@neuh.azr.nl.

Drs. Sillevs Smitt and Kinoshita contributed equally to the article.
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METHODS

Samples

We analyzed samples of serum and cerebrospinal fluid from 3060 patients that had been sent to us for antineuronal-antibody testing. Of the 3060 patients, 26 had histologically proved Hodgkin's disease, including Patients 1 and 2. IgG was purified from specimens that were obtained from the first plasma exchange of Patients 1 and 2 and from normal serum with protein A Sepharose. The purified IgG was dialyzed against phosphate-buffered saline containing 10 mM lithium chloride or artificial cerebrospinal fluid and used in assays for inositol phosphates or for *in vivo* transfer experiments. Frozen tumor tissues were provided by the pathology department of the Daniel den Hoed Cancer Center. We studied 10 samples of Hodgkin's tissue, including a lymph node from Patient 1, and 5 samples of non-Hodgkin's lymphoma.

Immunohistochemical Analysis

Normal C57BL/6 mice and mGluR1-deficient C57BL/6 mice¹⁰ were deeply anesthetized with pentobarbital and perfused through the heart with a fixative solution containing 4 percent paraformaldehyde, 0.2 percent picric acid, and 0.05 percent glutaraldehyde. Parasagittal sections (40 μ m each) of brain from these mice were incubated overnight with a 1:1000 dilution of test serum or 0.5 μ g of rabbit anti-mGluR1 G18 antibody per milliliter¹¹ and were then incubated with biotinylated goat antihuman or antirabbit IgG (Vector). For confocal microscopy, sections underwent reaction with Texas red-avidin (Vector, Burlingame, Calif.) instead of avidin-biotin-peroxidase complex. To test for antineuronal antibodies, we incubated the 3060 samples of serum and cerebrospinal fluid with parasagittal frozen and acetone-fixed 6- μ m sections of rat cerebellum and then with the addition of fluorescein-isothiocyanate-labeled goat antihuman IgG (Dako, Glostrup, the Netherlands). Anti-Purkinje-cell antibodies were classified as anti-Yo when reactive with the paraneoplastic Yo62 antigen, as anti-Tr when an additional characteristic dotted staining pattern was present in the cerebellar molecular layer, or as of unknown specificity.⁹

Cell Labeling and Assay for Inositol Phosphates

Live CHO cells that expressed the mGluR1 isoform mGluR1 α or the closely related receptor subtype mGluR5a were incubated in culture medium¹² with 1:1000 dilution of each patient's serum for one hour. After being washed with phosphate-buffered saline, the cells were fixed with 4 percent paraformaldehyde for 10 minutes, and serum antibodies that bound to the cells were detected by staining with fluorescent-labeled antihuman IgG (Vector). For measurement of the formation of inositol phosphates, the receptor-expressing CHO cells were labeled with [³H]inositol (1 μ Ci per milliliter) for 24 hours as described previously.¹¹

The amino-terminal extracellular domain of mGluR1 is involved in glutamate binding.¹³ Antibodies raised against mGluR1 amino-terminal sequences inhibit the glutamate-stimulated formation of inositol phosphates in mGluR1 α -expressing cells.¹¹ With this system, we assessed the effects of purified IgG on the activation of mGluR1 by analyzing the glutamate-stimulated formation of inositol phosphates in CHO cells that expressed mGluR1 α . After we incubated the cells with phosphate-buffered saline for 20 minutes, we incubated them with phosphate-buffered saline that contained 10 mM lithium chloride in the absence or presence of the patient's purified IgG for 20 minutes at 37°C. Agonist stimulation was started by adding glutamate to a final concentration of 15 μ M; this caused an increase in the levels of inositol phosphates by a factor of two to three.¹¹ After incubation for 20 minutes at 37°C, the reaction was terminated by 5 percent trichloroacetic acid (wt/vol). [³H]inositol phosphates (IP1, IP2, and IP3) were separated by AG1-X8 chromatography (Bio-Rad, Hercules, Calif.), and the radioactivity was determined on a liquid scintillation spectrometer.

Absorption Experiments

The patient's IgG was incubated with mGluR1 α - or mGluR5a-expressing CHO cells (1 \times 10⁸ cells per milligram of IgG) in phos-

phate-buffered saline for one hour. After centrifugation, IgG that remained in the supernatant was purified again with protein A and dialyzed against artificial cerebrospinal fluid. Successful absorption of the anti-mGluR1 antibodies with mGluR1 α -expressing CHO cells was confirmed by immunohistochemical assay, CHO-cell labeling, and assay for inositol phosphates with the use of the mGluR1 α -expressing CHO cells as described above. When the same assays were used, absorption with mGluR5a did not reduce the ability of the anti-mGluR1 antibodies to bind to mGluR1 α .

Transfer Experiments

A catheter was placed in the cisterna magna of C57BL/6 mice after they had received general anesthesia.¹⁴ At least 24 hours later, artificial cerebrospinal fluid containing IgG either from the patient or from normal serum (20 μ l [0.1 to 20 mg per milliliter]) was injected through the catheter over a period of 30 minutes. Footprints were made with ink applied to the hind paws of mice one hour after injection. For the rotarod test, animals were trained before injection. All animals managed to stay on a rod that was rolling at 20 rpm for longer than 60 seconds after several trials. Thirty minutes and every hour after the injection of IgG, each animal was tested in five trials, and the average time it remained on the rod was determined. The maximal time allowed was 60 seconds. In some experiments, aniline blue dye was injected concurrently to confirm the delivery of injected materials. For visualizing the penetration of human IgG into the cerebellum, the mice were perfused with the fixative solution 5 to 12 hours after injection, and transverse sections through the cerebellum were immunostained as described above with antihuman IgG primary antibody (Dako).

Expression of mGluR1 in Tumor Samples

Frozen sections (5 μ m each) from Hodgkin's and non-Hodgkin's lymphoma tissues were fixed in acetone and then incubated with biotinylated IgG from Patients 1 and 2 and normal human serum or with G18. We extracted RNA from the same tumors using the triple-extract reagent (Tri-Reagent, MRC, Cincinnati) followed by reverse transcription. For reverse transcriptase-polymerase chain reaction (RT-PCR), we used three primer sets specific to both intracellular and extracellular coding sequences of mGluR1: primer set 1: 5'TCTGGGGTGCATGTTCACTCC3' and 5'AGGCCGTCT-CATTGGTCTTCA3'; primer set 2: 5'CGAGAAAGTGCCCGA-GAG3' and 5'GTGGCTGAATAAGCGATCTG3'; and primer set 3: 5'TGAAGGCATAGTAGGTACAG3' and 5'GAGTGGAGCAACATCGAAT3'. The primers used for our positive control glyceraldehyde-3-phosphate dehydrogenase were 5'CCGAGCCACATC-TGCTCAGACAC3' and 5'GCCATCCACAGTCTTCTGGGT3'.

RESULTS

Samples of serum and cerebrospinal fluid from both patients had similar, specific immunohistochemical staining patterns on sections of mouse brain (Fig. 1A and 1E). Purkinje-cell bodies were strongly stained (Fig. 1C), and distinctive punctate staining, compatible with labeling of the Purkinje-cell spines, was observed in the molecular layer of the cerebellum (Fig. 1D). Strong staining of neurons and neuropil was also observed in the glomeruli of the olfactory bulb, the olfactory tubercle (including the islands of Calleja), the superficial layer of the cerebral cortex, the CA3 area of the hippocampus, the thalamus, the superior colliculus, and the spinal trigeminal nucleus. The immunohistochemical staining pattern appeared to be similar to the distribution of the metabotropic glutamate receptor mGluR1 (Fig. 1F).^{15,16} To test whether these antibodies were indeed directed against

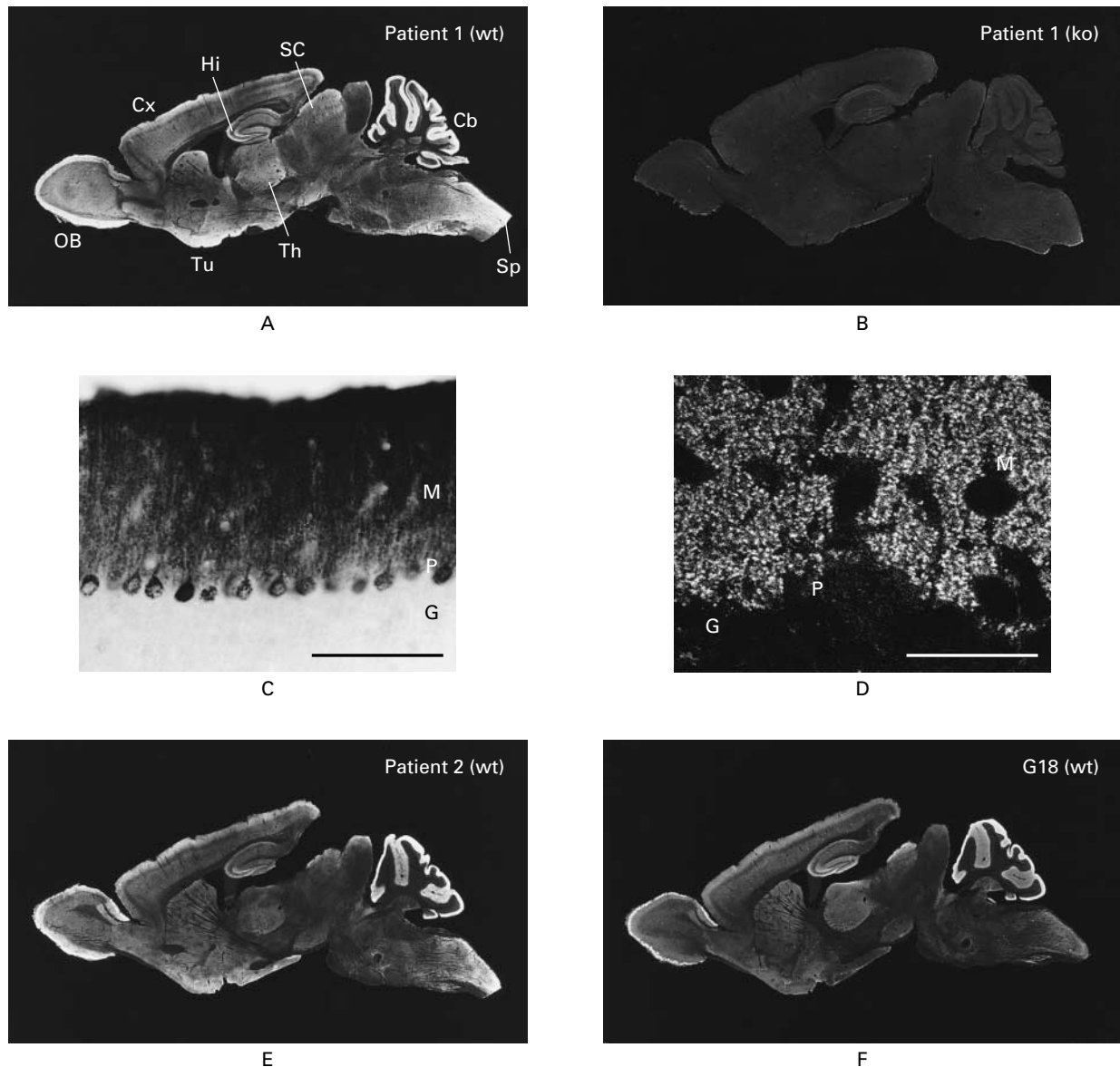


Figure 1. Immunohistochemical Analysis of Sections of Mouse Brain with Serum from Patients 1 and 2.

We performed an immunohistochemical analysis of parasagittal sections of mouse brain with the serum from Patient 1 (Panels A, B, C, and D) and Patient 2 (Panel E) and with a rabbit antibody (G18) to mGluR1 (Panel F). With brain tissue from mice (wild type [wt]), strong immunoreactivity was observed with both of the serum samples (Panels A, C, D, and E) in the Purkinje cells and the molecular layer (M) of the cerebellum (Cb), glomeruli of the olfactory bulb (OB), olfactory tubercle (Tu), CA3 area of the hippocampus (Hi), cerebral cortex (Cx), thalamus (Th), superior colliculus (SC), and spinal trigeminal nucleus (Sp). In the molecular layer, strong punctate labeling was observed with confocal microscopy (Panel D). These staining patterns resemble those found with a rabbit antibody against mGluR1 (Panel F) and were abolished (Panel B) in mGluR1-deficient mice (knockout [ko]). P denotes the layer of Purkinje cells, and G the layer of granule cells. The bar represents 100 μm for Panel C and 25 μm for Panel D.

mGluR1, we incubated the patients' serum and cerebrospinal fluid with sections obtained from mGluR1-knockout mice (Fig. 1B).¹⁰ These sections were not stained by either the serum or the cerebrospinal fluid (data not shown).

To confirm the specificity of the reactivity of the serum with native mGluR1 proteins, we incubated

living CHO cells that expressed rat mGluR1 α ¹² or mGluR5a¹⁷ with serum from both patients. The two serum samples strongly labeled CHO cells that expressed mGluR1 α but not cells that expressed mGluR5a (Fig. 2A, 2B, 2C, and 2D). The reactivity of the patients' IgG and cerebrospinal fluid with CHO cells that expressed human mGluR1 and with human

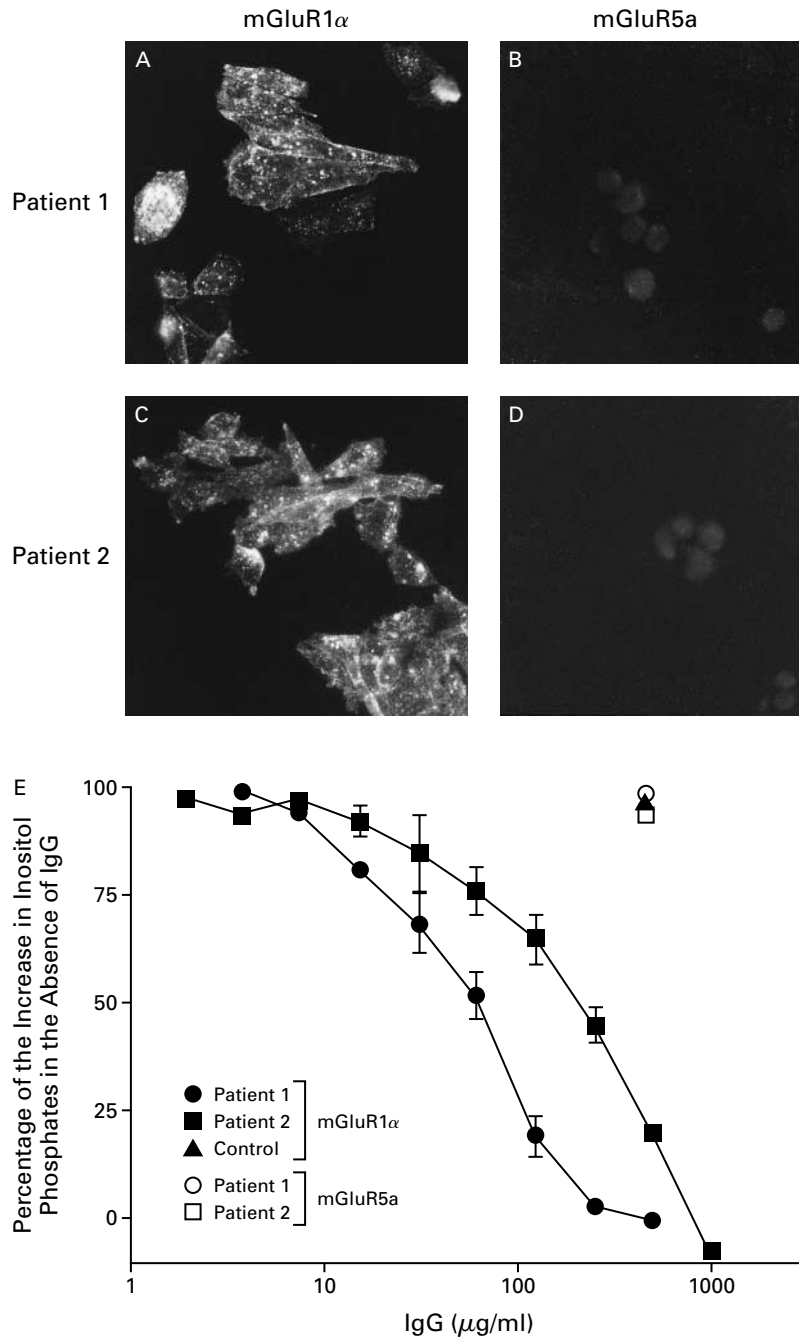


Figure 2. Functional Blocking of the mGluR1 Receptor by Autoantibodies.

Autoantibodies to mGluR1 blocked the glutamate-stimulated formation of inositol phosphates in mGluR1 α -expressing Chinese-hamster-ovary (CHO) cells. With the serum from Patient 1 (Panels A and B) and Patient 2 (Panels C and D), we incubated CHO cells that expressed either mGluR1 α (Panels A and C) or mGluR5a (Panels B and D). The serum samples from both patients reacted strongly with the native mGluR1 α protein on the cell surface but not with mGluR5a. Panel E shows the effects of purified IgG on the glutamate-stimulated formation of inositol phosphates in the CHO cells that expressed mGluR1 α (solid symbols) and mGluR5a (open symbols). The formation of inositol phosphates was stimulated with 15 μ M glutamate in the presence of IgG prepared from a control subject (triangles), Patient 1 (circles), and Patient 2 (squares). The values are the means (\pm SE) of three experiments performed in triplicate and are given as percentages of the increase in the formation of inositol phosphates stimulated with 15 μ M glutamate in the absence of IgG. The IgG of both patients inhibited the glutamate-induced production of inositol phosphates in a dose-dependent manner in CHO cells that expressed mGluR1 α but not mGluR5a. The mean (\pm SE) concentrations of IgG from Patients 1 and 2 that inhibited the activity of mGluR1 α by 50 percent were 58 \pm 9.4 and 194 \pm 36 μ g per milliliter, respectively. The IgG of the control subject had no effect at a concentration of 500 μ g per milliliter.

cerebellar sections was also demonstrated (data not shown). These results indicate that IgG from both patients reacted specifically with the amino-terminal extracellular domain of native mGluR1 α . Inhibition of the activation of mGluR1 α in a dose-dependent manner was also found with the IgG from each of the patients (Fig. 2E). The mean (\pm SE) concentrations of IgG that caused 50 percent inhibition of the activation of mGluR1 α (IC_{50}) for IgG from Patient 1 and Patient 2 were 58 ± 9 and 194 ± 36 μ g per milliliter, respectively. Normal human IgG had no effect. IgG from the two patients did not block the activation of mGluR5a, a finding that indicates its specificity.

We then examined the pathogenicity of the anti-mGluR1 autoantibodies by injecting purified IgG from the two patients (400 μ g; 20 μ l [20 mg per milliliter]) into the subarachnoid space of normal mice, near the cerebellum. Thirty minutes after the injection, the mice became increasingly ataxic, with a wide gait (Fig. 3A). They were unable to walk a straight line, and the distance between their steps was small and irregular, an indication of cerebellar dysfunction.¹⁸ At the peak of the symptoms, the most strongly affected mice could hardly walk or stand up because of severe truncal ataxia. As assessed by the rotarod test, the ataxic behavior peaked at about 2 to 4 hours after injection of IgG and subsided after 24 hours (Fig. 3B). Significant effects on the behavior of the mice ($P < 0.05$) could be detected with as little as 10 μ g of IgG (20 μ l [0.5 mg per milliliter]) from Patient 1, whereas no effects were detected with normal human IgG (20 μ l [20 mg per milliliter]).

To show that this *in vivo* effect of the IgG from the patients was caused by the anti-mGluR1 autoantibody, IgG was absorbed with CHO cells that expressed mGluR1 α or mGluR5a. The IgG that was absorbed with mGluR1 α completely lost its effect (Fig. 3B), but IgG that was absorbed with mGluR5a remained effective (the value on the rotarod test 2 hours after injection was 18 ± 12 seconds — not significantly different from the values for nonabsorbed IgG; $P > 0.3$). The injected IgG was restricted largely to the cerebellum (Fig. 3C and 3D); it had penetrated throughout various layers of the cerebellar cortex, as shown by immunohistochemical visualization of human IgG (Fig. 3D).

Furthermore, antibodies eluted from the cells that expressed mGluR1 α caused similar ataxic behavior in mice at a low concentration (20 μ l [about 0.15 mg per milliliter]) (Fig. 3B). These results clearly indicate that anti-mGluR1 autoantibodies from these patients caused cerebellar ataxia in mice by functional blocking of mGluR1 in the cerebellum.

Titers of the autoantibody in cerebrospinal fluid and serum samples from both patients were examined by end-point titration of immunohistochemical mGluR1 staining in sections of rat brain. Before Patient 1 received plasma exchanges, the titers in

her cerebrospinal fluid and serum were 512 and 3200, respectively. When these values were normalized according to concentrations of IgG, the anti-mGluR1 titer per unit of IgG was 31 times as high in cerebrospinal fluid as in serum, an indication of intrathecal synthesis. Using these values, we found that the anti-mGluR1 antibody content in the cerebrospinal fluid was 25 times as high as that in the IC_{50} of serum IgG (46 μ g per milliliter \times 31 \div 58 μ g per milliliter). In Patient 2, after she received plasma exchanges, the anti-mGluR1 titer in serum was 400 and the titer in cerebrospinal fluid was 256. Normalized according to IgG concentrations, the anti-mGluR1 IgG titer per unit of IgG was 36 times as high in cerebrospinal fluid as in serum. In Patient 2, the content of anti-mGluR1 antibody in cerebrospinal fluid was therefore 28 times that in the IC_{50} of serum IgG (150 μ g per milliliter \times 36 \div 194 μ g per milliliter).

To investigate whether the anti-mGluR1 autoantibodies were related to Hodgkin's disease, we examined a frozen biopsy specimen of a lymph node from Patient 1 and lymph nodes from nine other patients with Hodgkin's disease and five patients with non-Hodgkin's lymphoma by RT-PCR and an immunohistochemical assay, using biotinylated IgG from the patients. We could not detect mGluR1 RNA in any of these samples, but we did detect it in positive controls (rat and human cerebellum; data not shown). Also, an immunohistochemical assay with biotinylated IgG from the patients did not provide evidence for the expression of mGluR1 or a cross-reactive epitope in any of the tumor samples (data not shown).

We also examined serum samples from more than 3060 patients for the presence of paraneoplastic antineuronal antibodies. These samples included serum samples from 26 patients with Hodgkin's disease. Only the serum from Patients 1 and 2 showed anti-mGluR1 immunoreactivity. Of the 24 serum samples from other patients with Hodgkin's disease, 5 reacted with Purkinje cells; the pattern was anti-Tr in 3 of these. These anti-Tr serum samples did not bind to CHO cells that expressed mGluR1 and showed the same immunolabeling pattern of Purkinje cells in wild-type and mGluR1-knockout mice (data not shown).

DISCUSSION

IgG from serum and cerebrospinal fluid from two patients with cerebellar ataxia bound to mGluR1 receptors in the brain and caused ataxia in mice. In Patient 1, the most striking symptom at presentation was gait ataxia; she was unable to walk with a tandem gait. At that time, the titers of the anti-mGluR1 autoantibodies in serum and cerebrospinal fluid were 3200 and 512, respectively. After 25 days of treatment with prednisone and four plasma exchanges, her gait improved and the serum and cerebrospinal

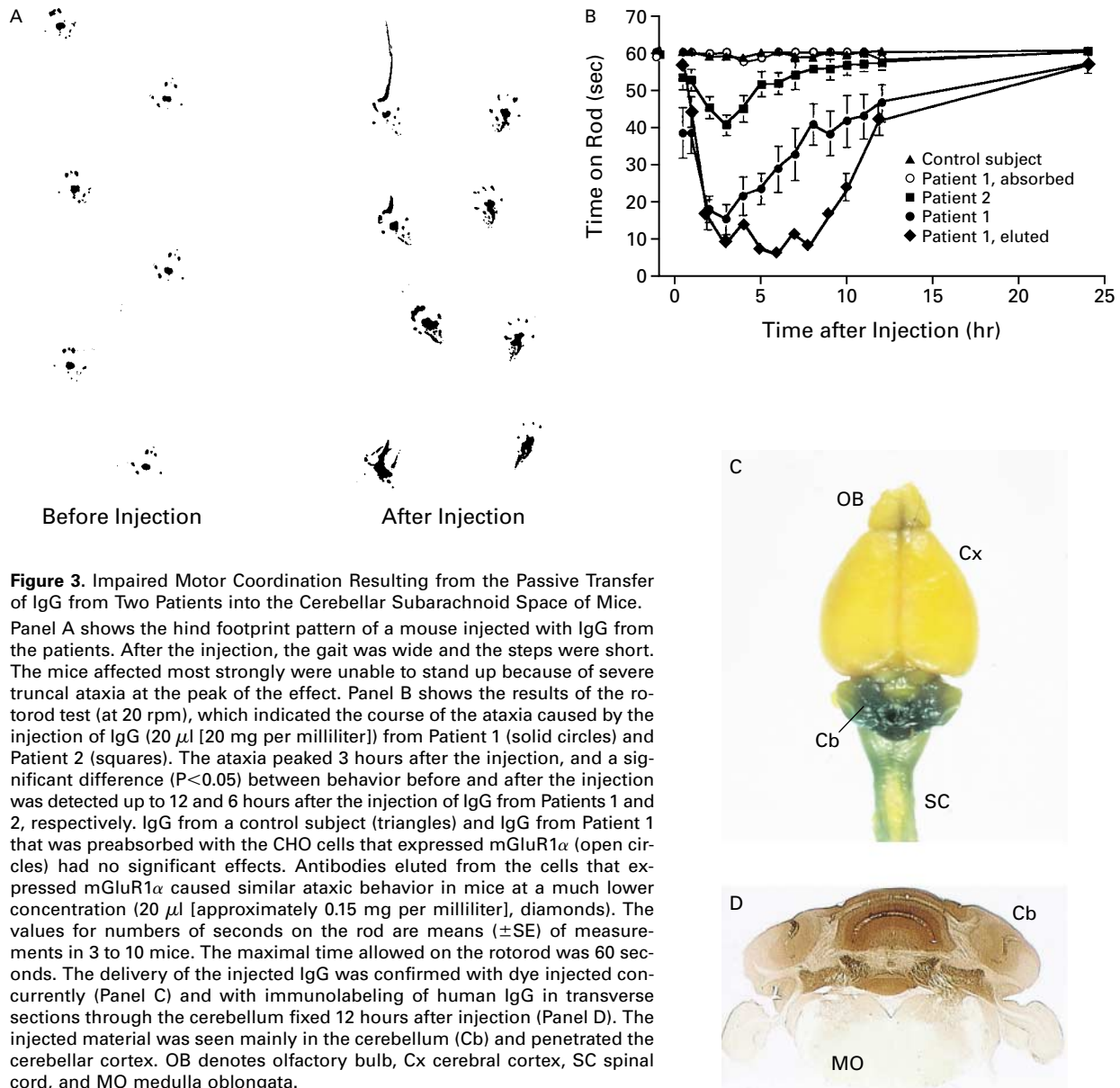


Figure 3. Impaired Motor Coordination Resulting from the Passive Transfer of IgG from Two Patients into the Cerebellar Subarachnoid Space of Mice.

Panel A shows the hind footprint pattern of a mouse injected with IgG from the patients. After the injection, the gait was wide and the steps were short. The mice affected most strongly were unable to stand up because of severe truncal ataxia at the peak of the effect. Panel B shows the results of the rotarod test (at 20 rpm), which indicated the course of the ataxia caused by the injection of IgG (20 μ l [20 mg per milliliter]) from Patient 1 (solid circles) and Patient 2 (squares). The ataxia peaked 3 hours after the injection, and a significant difference ($P < 0.05$) between behavior before and after the injection was detected up to 12 and 6 hours after the injection of IgG from Patients 1 and 2, respectively. IgG from a control subject (triangles) and IgG from Patient 1 that was preabsorbed with the CHO cells that expressed mGluR1 α (open circles) had no significant effects. Antibodies eluted from the cells that expressed mGluR1 α caused similar ataxic behavior in mice at a much lower concentration (20 μ l [approximately 0.15 mg per milliliter], diamonds). The values for numbers of seconds on the rod are means (\pm SE) of measurements in 3 to 10 mice. The maximal time allowed on the rotarod was 60 seconds. The delivery of the injected IgG was confirmed with dye injected concurrently (Panel C) and with immunolabeling of human IgG in transverse sections through the cerebellum fixed 12 hours after injection (Panel D). The injected material was seen mainly in the cerebellum (Cb) and penetrated the cerebellar cortex. OB denotes olfactory bulb, Cx cerebral cortex, SC spinal cord, and MO medulla oblongata.

fluid titers of the autoantibodies had dropped to 200 and 64, respectively. Later, when she was asymptomatic, we could not detect anti-mGluR1 autoantibodies in her serum.

The serum of Patient 2 was first tested when she had had severe ataxia for almost one year. The titer of anti-mGluR1 autoantibodies in her serum was 3200 at that time. After 14 plasma exchanges, she continued to have severe ataxia and was unable to walk without support. Although the plasma anti-mGluR1 titer had dropped to 400 after the plasma exchanges, the cerebrospinal fluid titer remained high, an indication of ongoing intrathecal synthesis of anti-mGluR1 autoantibodies.

Several molecules have been identified as autoantigens associated with nervous system diseases. These include the acetylcholine receptor in myasthenia gravis,¹⁹ voltage-gated calcium channels in the Lambert-Eaton syndrome,²⁰ presynaptic potassium channels in Isaacs' syndrome (neuromyotonia),²¹ GluR3 in Rasmussen's encephalitis,²² and Hu antigens in paraneoplastic encephalomyelitis.²³ So far, functional effects of such autoantibodies have been found only in disorders of the peripheral nervous system, such as myasthenia gravis (blocking of acetylcholine receptors)¹⁹ and the Lambert-Eaton syndrome (blocking of presynaptic voltage-gated calcium channels at the neuromuscular junction).²⁴ Our results indicate

that autoantibodies may also affect the central nervous system by blocking neuronal receptors.

We detected anti-mGluR1 autoantibodies in the serum of only two of 3060 patients with a variety of disorders. These two patients had Hodgkin's disease, but we were unable to show conclusively that the ataxia and the Hodgkin's disease were linked in a paraneoplastic syndrome. We did not detect expression of mGluR1 in the tumor-containing lymph node of Patient 1, a finding that would have tied the two disorders together. Nevertheless, in about 50 percent of cases, the cerebellar syndrome of nonhereditary subacute ataxia in adults is paraneoplastic and can occur when Hodgkin's disease is in remission.^{1,4}

The mGluR1 receptors are located at the perisynaptic site of the Purkinje-cell dendritic spines, which form excitatory synapses with parallel fibers or climbing fibers.²⁵ The activation of mGluR1 receptors is necessary for the induction of cerebellar long-term depression, which is probably the mechanism of cerebellar motor learning.^{10,11,26,27} Mice that lack the mGluR1 gene have ataxic gait and intention tremor and impaired cerebellar long-term depression and motor learning.¹⁹ The ability of the anti-mGluR1 autoantibodies to cause ataxic behavior in mice by blocking mGluR1 in the cerebellum indicates that the activation of mGluR1 is necessary for normal cerebellar coordination.

Impaired cerebellar long-term depression and motor learning, which result from the blocking of mGluR1,^{10,11,26} are unlikely to be the cause of ataxia in our study, because the effects of the injected antibodies were evident in the short term. The activation of mGluR1 induces slow inward-outward currents as well as a depression of parallel fiber-mediated excitatory postsynaptic currents in Purkinje's cells.²⁶ The blocking of these mGluR1-mediated effects in the parallel fiber synapses may have a role in the ataxic behavior. Finally, we should consider the possibility that the short-term ataxic effect results partly from impaired mGluR1 activation at other locations, such as climbing fiber synapses — activation that is necessary for normal motor coordination.²⁸

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