Chapter 21

X-Linked Adrenoleukodystrophy

21.1 Clinical Features and Laboratory Investigations

X-linked adrenoleukodystrophy (XALD) is a genetically determined disorder that mainly involves the adrenal cortex and the CNS. Inheritance is X-linked recessive. The disease has a wide phenotypic variability. The rapidly progressive childhood cerebral form accounts for about one-third of the cases. Adrenomyeloneuropathy (AMN) has a later onset and slower progression and accounts for about 40–45% of the patients. The relative frequency of the Addison-only form varies with age and accounts for up to 50% of the cases in childhood. Less frequent variants include the adolescent and the adult cerebral forms. In addition, there are patients with unusual presentation and asymptomatic patients, a common observation below the age of 4 years and very rare above the age of 40. These different phenotypes may occur all within one affected family. Even identical twins may display different phenotypes. Females may also be affected, especially when they become older. The minimum frequency of males with the defect has been estimated to be 1:42,000 in the USA, whereas the minimum frequency of males with the defect and female carriers is estimated to be 1:16,800 (Bezman et al. 2001).

In the childhood cerebral form of XALD, the age at onset of neurological symptoms is usually between 5 and 9 years. Features of adrenal insufficiency may occur before overt neurological symptoms or may follow. In some cases a diagnosis of Addison disease is made 1–3 years before any neurological disorder is evident on the basis of increased pigmentation, fatigue, episodes of vomiting, or catastrophic reactions to intercurrent infections. In others, neurological deterioration may continue for some years without endocrine symptoms, and sophisticated investigations are needed to reveal evidence of adrenocortical dysfunction. Adrenocortical dysfunction is present in at least 80% of the patients.

The earliest neurological symptoms are often vague and frequently consist of behavioral changes. The changes vary from withdrawn to bizarre hyperactive and aggressive behavior and are often accompanied by poor school performance. Many boys receive psychiatric treatment until deteriorating learning capabilities and other neurological symptoms force recognition of the organic nature of the disease. Early neurological abnormalities are disturbances of gait, loss of vision, and impaired auditory discrimination. The course of the disease is relentlessly progressive and spastic tetraplegia and dementia become manifest in months. Decreased vision is caused by optic atrophy or bilateral occipital white matter lesions or, more often, a combination of the two. Initially, neurological findings are often asymmetrical with hemiparesis or hemianopia. Frequently noted subsequent problems are dysarthria, dysphagia, and hearing loss. Cerebellar ataxia or sensory disturbances may be present, but are not usually prominent. There are no clinical signs of peripheral nerve dysfunction. Progressive dementia occurs. Epileptic seizures occur and are often multifocal in origin. The pace of deterioration is variable. In the final stage a spastic quadriplegia is present and a variable degree of decorticate posturing. The affected boys are blind, deaf, and mute. A vegetative state or death is reached in 1–5 years. Most patients die within 2 or 3 years after the onset of neurological symptoms, but some live for many years in a vegetative state. However, some patients stabilize for years in an earlier stage of the disease.

The adolescent (onset at 10–21 years) and adult (onset after 21 years) cerebral forms resemble the childhood cerebral form, except for the later onset. Just like the childhood from, the adolescent and adult cerebral forms have a rapidly progressive course. The disease is often misdiagnosed. It may present as a psychosis or dementing illness, or as a single focal brain lesion that can be mistaken for a tumor.

The usual age at onset of AMN is within the third or fourth decade, but ranges from 14 to 60 years. Most patients with an adult onset form of XALD have AMN. Neurological deficits are primarily due to a myelopathy and to a lesser extent a neuropathy. Affected males have a slowly progressive paraparesis, disturbed vibration sense of the legs, voiding disturbances, and a variable degree of sexual dysfunction. Signs of a distal polyneuropathy may also be found. Approximately two-thirds of the patients have overt or biochemical signs of adrenocortical insufficiency, which may precede or follow neurological dysfunction. Twenty percent of the patients have signs of hypogonadism with infertility. Many patients have scanty scalp hair. The disease is slowly progressive over decades. About half of the patients with AMN develop some cerebral involvement, with usually mild cognitive dysfunction. Visual memory, spatial cogni-

tion, and psychomotor speed are affected most. Psychological dysfunction may also occur, with emotional disturbances and depression. About 20% of the patients with AMN develop a rapidly progressive cerebral white matter involvement as seen in the cerebral forms of XALD.

In the Addison-only form no neurological dysfunction is found. However, all patients are at risk of developing overt neurological symptoms sooner or later. Some patients develop Addison disease in childhood and neurological abnormalities only arise in adulthood.

The asymptomatic group includes males in whom the typical biochemical abnormalities of XALD are found and who are completely healthy. Most of the males included in this group are small boys who were investigated because of clinically evident XALD in an older brother. The group also includes adolescents and a few older males. The oldest asymptomatic male ever mentioned was 62 years old.

There are a few patients with unusual forms of XALD. Patients have been reported with predominantly cerebellar ataxia or a spinocerebellar syndrome. One patient, aged 57 years, has been reported who showed rapid neuropsychiatric deterioration and signs of cerebral demyelination at the site of a severe cerebral contusion suffered several months previously. Cerebral XALD presents rarely as an acute encephalopathy with seizures, status epilepticus, headache, vomiting, lowering of consciousness, or even coma. Papilledema may be seen. The presence of fever may suggest encephalitis. After this acute episode, the patient recovers with temporary encephalopathic signs, which resolve in the course of days and weeks.

Female XALD carriers may also develop clinical problems. Most women below the age of 30 years are uninvolved. The percentage of females with neurological problems increases with age. Approximately 50% of the female carriers of 40 years and older display signs of a mild myelopathy with increased tendon reflexes and distal sensory changes in the legs, but no or mild disability. About 15% of the females of 40 years or older have an AMN-like clinical phenotype, but with later onset and milder symptomatology. Cerebral involvement and adrenocortical insufficiency are very rare among female carriers at all ages. Cerebral involvement is exceedingly rare in childhood and then probably related to skewed, highly unfortunate X inactivation or a partial deletion of one X chromosome and a mutated gene on the other chromosome. Some older female carriers have mild intellectual impairment, which is only detected by detailed psychological tests. An adult female has been described with a lethal cerebral leukodystrophy.

Laboratory testing usually reveals signs of adrenocortical dysfunction. Urinary excretion of 17-hydroxycorticosteroids and 17-oxysteroids may be reduced. Further evidence for primary adrenocortical insufficiency is found in impaired cortisol responsiveness to adrenocorticotropic hormone (ACTH) in the presence of elevated baseline ACTH levels. Evidence of primary testicular insufficiency is provided by low testosterone levels and elevated luteinizing hormone (LH) or follicle stimulating hormone (FSH) levels. CSF protein is elevated in the majority of the symptomatic patients, sometimes combined with an elevation of γ-globulin level or moderate increase in lymphocytes.

Diagnosis of XALD depends upon the demonstration of abnormally high levels of saturated very-long-chain fatty acids (VLCFA) in plasma, cultured skin fibroblasts, or tissue. For routine purposes plasma is used. The concentration of C26:0 fatty acids is investigated as well as the ratios of C26:0/C22:0 and C24:0/C22:0 fatty acids. In over 90% of XALD patients all three parameters are abnormally elevated. In a minority of the patients only one or two of the three parameters are abnormal. VLCFA analysis and C26:0 fatty acid β-oxidation measurements in cultured skin fibroblasts are used for definite diagnosis. Patients with XALD already show the characteristic elevations in blood VLCFA during the first 2 weeks of life and even in cord blood.

Neurophysiological investigations are often used to establish the extent of disease in XALD. Nerve conduction studies usually show a normal conduction velocity in cerebral forms, but may also show a decreased velocity. In AMN the conduction velocity is decreased. In cerebral forms of ALD the EEG is as a rule abnormal, although nonspecifically, with diffuse slowing of the rhythm and a maximum usually in the posterior regions. Evoked potential studies may show abnormalities reflecting the central white matter involvement.

Investigation of the level of VLCFA is also a sensitive test in carrier detection: 85% of heterozygotes can be identified when the results of VLCFA assays in plasma and cultured fibroblasts are combined. Consequently, about 15% of the female carriers have false negative test results. Monoclonal antibodies have been raised against the XALD gene product (ALD protein, ALDP) and use of these antibodies in immunofluorescence studies of cultured skin fibroblasts or leukocytes may aid in the identification of heterozygous females, in particular when VLCFA concentrations in plasma and fibroblasts are normal. In 70–80% of the affected kindreds affected boys and men lack ALDP immunoreactivity; female carriers in these kindreds show a mixture of positive and negative immunoreactivity in their cells. Antenatal diagnosis can be established by measuring VLCFA in cultured amniocytes and chorionic villi, by measuring the activity of peroxisomal β-oxidation, and im-

munofluorescence analysis of ALPD. DNA techniques can also be used for carrier detection and prenatal diagnosis. A problem in genetic counseling and prenatal diagnosis is that the demonstration of the biochemical defect does not provide information about whether the patient will develop severe childhood XALD or the milder AMN.

21.2 Pathology

At autopsy the surface of the brain of a patient with cerebral XALD is either normal or shrunken, depending on the degree of tissue loss. The central white matter is grayish and indurated, sometimes cystic and cavitated. The thickness of the cortex is normal. The atrophic external appearance is, if present, secondary to loss of white matter. In cases of extensive loss of myelin the ventricular system is enlarged.

The pathological abnormalities are the same for childhood, adolescent, and adult cerebral XALD and consist of widespread demyelination of the white matter. The demyelinating lesion constitutes one large area extending across the corpus callosum and involving both hemispheres. In most cases of cerebral XALD the demyelinating process starts in the splenium of the corpus callosum and spreads bilaterally to the occipital region. Gradually the process spreads outwards and forwards as a confluent lesion until most of the cerebral white matter is involved. The frontal white matter is generally affected less severely and often asymmetrically. The subcortical U fibers are preserved until a far-advanced stage, and usually the U fibers in the occipital area are affected before those elsewhere. Other areas of the brain that are usually heavily involved are the fornix, the hippocampal commissure, the posterior limb of the internal capsule, the lateral two-thirds of the cerebral peduncles including the occipitoparietotemporopontine and pyramidal tracts, and the lateral lemniscus. In some patients the cerebellar white matter is involved, sometimes the cerebellar peduncles as well. In a smaller portion of the patients with cerebral XALD the demyelination starts bilaterally in the frontal area, also involving the anterior part of the corpus callosum. In these cases the anterior limb of the internal capsule and the medial third of the cerebral peduncles containing the frontopontine tracts are affected. In a few patients the demyelinating lesions are highly asymmetrical.

Within the white matter lesion three zones can be distinguished on histopathological examination. The outer zone shows evidence of active destruction of myelin with axonal sparing. Scattered PAS-positive and sudanophilic macrophages are present. The middle zone shows signs of active inflammation with marked perivascular mononuclear cell infiltration. This zone contains many large ballooned macrophages laden with lipids. There are many preserved demyelinated axons and little myelin remains. The large central area is destroyed and burnt out. There is no evidence of an active process. Axons, myelin sheaths, and oligodendroglia are absent. There are few lymphocytes and only occasional macrophages surrounding blood vessels. This area is filled with a dense mesh of glial fibrils and scattered astrocytes. Sometimes cavitation or calcium depositions are seen in this area.

In the brain stem and spinal cord degeneration of the tracts appears to proceed at the same rate throughout, with no evidence of a dying-back phenomenon. Here too the demyelinating process occurs in a continuous fashion. No small independent foci of demyelination are seen. Perivascular accumulations of inflammatory cells may well be noted.

The cytoarchitecture of the cerebral cortex is normal. Only in more advanced cases may neuronal loss and gliosis be seen, especially in the deeper cortical layers. The cerebral cortical damage is mainly found in the occipital region where the demyelinating lesion may not spare the U fibers and may be contiguous with the deep layers of the cortex.

Electron microscopic examinations show that many macrophages and microglia contain distinctive cytoplasmic inclusions, consisting of linear lamellae. An individual lamella has a trilaminar structure, consisting of paired electron-dense leaflets separated by an electron-lucent space. These trilamellar structures are often closely associated with lipid droplets. They are not found in oligodendroglia or astrocytes. In addition, macrophages contain myelin debris.

Microscopic examination of peripheral nerves reveals either no abnormalities or demyelination. On ultrastructural examination abnormal cytoplasmic inclusions may be seen in Schwann cells and in endoneurial macrophages. These inclusions have the characteristic linear, trilamellar appearance.

Pathological studies in AMN demonstrate bilateral, usually symmetrical, long tract degeneration in the spinal cord with most prominent involvement of the corticospinal and dorsal tracts. The distribution of the degeneration of these tracts conforms to a dying-back pattern in that the greatest loss of myelinated fibers is found in the cervical dorsal tracts and the lumbar corticospinal tracts. Axonal loss is equal to or greater than myelin loss. The above findings provide strong evidence for primary axonal degeneration. There are no inflammatory cells. Microglia are increased in numbers and appear the dominant responding cells. Dorsal ganglion cells are not decreased in numbers but are decreased in size. Peripheral nerves display both axonopathic and myelinopathic features. In Schwann cells and macrophages

the typical trilamellar inclusions are present. There are no inflammatory cells.

The cerebral involvement in AMN is highly variable. In most AMN patients, patchy, poorly defined, small areas of demyelination are scattered throughout the cerebral hemispheres, with relative or total axonal sparing, activation of microglia, presence of striated macrophages with trilamellar inclusions, and absence of inflammatory cells. In some patients, moderate diffuse demyelination is observed in the cerebral white matter and to a lesser extent the cerebellar white matter, with sparing of the U fibers and without inflammation. Still others display inflammatory demyelinating lesions with axonal loss, qualitatively similar to cerebral XALD but much more localized. Some AMN patients develop the cerebral form of XALD with rapidly progressive inflammatory demyelination. In addition to these inflammatory demyelinating lesions, AMN patients also demonstrate noninflammatory, bilateral, fairly symmetrical lesions with comparable loss of axons and myelin sheaths, involving most often the brain stem corticospinal and spinocerebellar tracts, medial and lateral lemnisci, cerebellar peduncles, posterior limb of the internal capsule, and the optic radiations.

The adrenal glands in XALD show gross atrophy of the cortex, the medulla being normal. The zona reticularis and zona fasciculata are particularly affected with ballooned cortical cells, in which a striated appearance of the cytoplasm may be seen. These striated cells are specific for XALD. Ultrastructurally the striations are shown to consist of linear, trilamellar accumulations within the adrenal cortical cell cytoplasm. Lymphocytic infiltrates are found in a minority of the patients. Light microscopic examination of the testis often reveals no abnormalities, although fully developed Leydig cells may be lacking. Interstitial cells, presumptive Leydig cell precursors, may contain the characteristic trilamellar profiles. In all tissues, the morphology of peroxisomes is normal.

21.3 Chemical Pathology

XALD is a lipidosis, in which accumulation of saturated VLCFA occurs in all tissues, especially in CNS white matter, peripheral nerve, adrenal cortex, and testis. Substantial quantities of these VLCFA are deposited as cholesterol esters, which appear as the characteristic lamellated cytoplasmic inclusions. These VLCFA vary in chain length from C23 to C32 with a peak at C25–C26. Several other lipids also contain an increased percentage of saturated VLCFA. These increases in VLCFA are found in cerebral XALD, AMN, and female carriers.

The changes in lipid composition of myelin and whole white matter have been investigated separately for regions with different stages of myelin breakdown in the cerebral form of XALD. In morphologically normal white matter, subtle changes in lipid composition are found. Phospholipids are increased, whereas galactolipids and cholesterol are slightly decreased. Only traces of cholesterol esters are found in histologically intact white matter. The fatty acid composition of cholesterol esters, cerebroside, and sulfatide in intact white matter is relatively normal, whereas phospholipids in the same area contain increased VLCFA, with the most striking increase in VLCFA in phosphatidylcholine. A moderate increase in VLCFA is seen in gangliosides. The area of active demyelination shows major changes in lipid composition. The water content is increased, the amount of total lipids is decreased, and there is a large increase in cholesterol esters, whereas unesterified cholesterol is severely decreased. Galactolipids are decreased, whereas phospholipids are stable as a proportion of total lipids. The fatty acid composition of cerebroside and sulfatide shows a slight elevation in VLCFA, whereas the VLCFA content of phospholipids, gangliosides, and cholesterol esters is greatly increased. The most striking rise in VLCFA among the phospholipids is seen in phosphatidylcholine and sphingomyelin. In the area of gliosis, the amount of remaining lipids is small, and the water content is high. The amount of galactolipids is relatively very low, whereas phospholipids and cholesterol are low in absolute content but constitute a relatively normal proportion of total lipids. Cholesterol esters are present in small but measurable amounts. Significant amounts of triglycerides and free fatty acids can also be measured. In gliotic tissue the VLCFA content of cerebroside, sulfatide, and phospholipids is barely elevated, whereas the VLCFA content of gangliosides and cholesterol esters is mildly to markedly elevated.

The adrenal and testicular content of cholesterol esters is abnormally high. The cholesterol esters contain an abnormally elevated amount of saturated VLCFA.

21.4 Pathogenetic Considerations

XALD is caused by mutations in the gene *ABCD1*, which is located on chromosome Xq28 and encodes ALDP, a peroxisomal ATP-binding cassette transmembrane transporter. A great number of different mutations have been identified. No consistent correlation between genotype and phenotype has been demonstrated.

The disease is biochemically characterized by elevated VLCFA due to reduced VLCFA β-oxidation. The first step in the β-oxidation of VLCFA is conversion of fatty acid to fatty acyl-CoA, catalyzed by the enzyme VLCFA-CoA synthase or ligase, present on the perox-

isomal membrane. The enzyme is more specifically called lignoceroyl- or hexacosanoyl-CoA synthase or ligase. The relation between ALDP and VLCFA-CoA ligase is still elusive. ALDP is not necessary for the import of this enzyme into peroxisomes or the import of VLCFA into peroxisomes. The reason for impaired VLCFA β-oxidation in XALD has yet to be found. A recent hypothesis assumes that ALDP facilitates the interaction between peroxisomes and mitochondria, and that ALDP deficiency leads to impaired β-oxidation in mitochondria. The repeated observation of structural mitochondrial abnormalities in XALD tissues, including lipid inclusions in mitochondria, is an argument in favor of this hypothesis.

The impaired degradation of VLCFA leads to enrichment of these fatty acids in various lipids at the expense of the normally degraded short-chain fatty acids. The accumulating fatty acids are saturated and have a chain length varying between C24 (lignoceroyl acid) and C32 with a peak at C26 (hexacosanoic acid). VLCFA are derived both from the diet and from endogenous synthesis by a microsomal system that elongates long-chain fatty acids. There is evidence that in XALD, not only β-oxidation of VLCFA is decreased, but fatty acid chain elongation activity is also elevated, contributing to the accumulation of VLCFA. It has been shown that the addition of monounsaturated fatty acids to culture medium has a dramatic effect in lowering the content of VLCFA in XALD fibroblasts. These monounsaturated fatty acids appear to inhibit the synthesis of VLCFA without having any effect on the degradation of VLCFA.

The exact mechanisms by which nervous tissue damage occurs are still unknown. There is a fundamental difference between the inflammatory demyelinative lesions seen in the white matter of patients with cerebral XALD and the axonopathic lesions seen in patients with AMN and AMN-like phenotypes. Both biochemical and immunological mechanisms may be important in the pathogenesis of the lesions. The greater length of the aliphatic chain causes VCLFA to be extremely insoluble. Abnormally high VLCFA levels alter membrane physiological properties and functions. Increasing levels of VLCFA in membranes may result in instability and breakdown of the membranes, leading to noninflammatory myelin loss and axonal degeneration, as seen in AMN. The destruction of adrenocortical cells, Leydig cells, and Schwann cells is also noninflammatory and may be related to the toxic effects of VLCFA. The rapidly progressive demyelination with a marked inflammatory reaction suggests additional immunological pathogenetic mechanisms in cerebral XALD. Further evidence for a role of immunological mechanisms is found in the occasional presence of signs of intrathecal immunoglobulin production, increased levels of IgA and IgG in XALD tissues, and high levels of myelin antibodies in serum of XALD patients. Tumor necrosis factor-alpha (TNF-α), a proinflammatory cytokine thought to be responsible for tissue damage in inflammatory brain disorders including multiple sclerosis, is expressed in astrocytes and macrophages at the active edge of the lesion. Reactive astrocytes, macrophages, and T lymphocytes are the most prevalent cellular elements. Most lymphocytes are CD8-positive cytotoxic T cells. CD1 molecules, which play major roles in lipid antigen presentation, have been found to be present, most conspicuously in the acute inflammatory lesions. It is hypothesized that the primary biochemical abnormality in XALD, i.e. the abnormal accumulation of VLCFA, leads to membrane instability and breakdown, leading to liberation of VLCFA-containing moieties, which may be antigenic and elicit an immune response. Not only peptide antigens, but also lipid antigens may play a key role in the pathogenesis of inflammatory demyelination in cerebral XALD. Lipids containing an abnormally high proportion of VLCFA may stimulate nearby astrocytes, microglia, and macrophages to initiate a cytokine cascade resulting in further myelin destruction by T cells, B cells, and complement. This two-stage hypothesis explains why the zone of active inflammation in cerebral XALD is found behind the zone of active demyelination. This location of the inflammation contrasts with the situation in multiple sclerosis, in which the inflammation is most intense at the edges of the lesion, with little or no inflammatory response in the inner zones of the lesion. Others hypothesize that accumulation of VLCFA in membrane domains associated with signal transduction pathways may trigger inflammatory processes through activation of microglia and astrocytes, resulting in loss of myelin.

Adrenal cortical cells, Leydig cells, and Schwann cells are also involved in the disease process. They accumulate VLCFA incorporated in cholesterol esters in the form of lamellar cytoplasmic inclusions. A cytotoxic pathogenesis has been proposed for the adrenal, testicular, and Schwann cell lesions. Furthermore, it has been shown that accumulation of VLCFA in adrenal cortical cell membranes leads to increased membrane microviscosity and can interfere with ACTH responsiveness. Impaired ACTH receptor function has been found, probably contributing to the adrenocortical insufficiency. It is noteworthy that the destruction of these cells is accompanied by little or no inflammation.

All phenotypic variants of XALD have the same basic defect and all variants may occur within the same family. No differences in fatty acid abnormality could be established in fibroblasts, erythrocyte membranes, or blood in repeated investigations. There is evidence for an autosomal modifier gene, which would explain the phenotypic variability of XALD within pedigrees. This modifier gene probably modu-

lates the immune response, considering the marked difference in immune reaction between cerebral XALD and AMN. It is likely that environmental factors may also influence the phenotype, as disparate phenotypes can be seen in sets of identical twins.

Many female heterozygotes suffer from a late-onset, slowly progressive neurological disorder. Inactivation of one of the two X chromosomes in female somatic cells is considered to be a random process. However, there is some evidence that, in XALD carrier females, selection mechanisms favor cells expressing the mutant allele rather than cells expressing the normal allele. This observation may explain the relatively frequent occurrence of symptoms in female carriers. There is also evidence that skewing of X inactivation is an important factor in determining the severity of disease manifestations in female XALD carriers.

21.5 Therapy

First and foremost, family counseling, carrier detection, and prenatal diagnosis are important in preventing the occurrence of further cases in known families.

Symptomatic care is essential. Hormonal substitution is necessary to correct the adrenocortical insufficiency, if present. Testosterone administration can be of help in AMN patients who have testicular insufficiency. Psychiatric care may be necessary, in particular in adult patients with cerebral XALD.

An important difference between cerebral XALD and the more benign AMN is in the presence or absence of an inflammatory response. Various modes of immunosuppression and immunomodulation have been tried in order to influence the inflammatory reaction, including the use of steroids, β-interferon, cyclophosphamide, pentoxifylline, thalidomide, cyclosporine, plasmapheresis, and high-dose intravenous immune globulin, and unfortunately so far none of these has been effective. They do not influence the course of the neurological disease. Incidentally, improvement under steroids has been reported, but this effect may be temporary.

Therapeutic strategies aiming to lower VLCFA have been the cornerstone of therapeutic strategies for a long time. Dietary restriction of saturated VLCFA is insufficient to lower blood VLCFA. Monounsaturated fatty acids compete with saturated fatty acids for the microsomal fatty acid elongation system. Diets enriched in monounsaturated fatty acids lead to decreased synthesis of VLCFA and are more effective in lowering blood VLCFA. The best results are obtained by combining the two. Diets restricted in saturated VLCFA combined with the use of a 4:1 mixture of glyceryl trioleate (GTO; oleic acid is a C18 monounsaturated fatty acid) and glyceryl trierucate (GTE; erucic acid is a C22 monounsaturated fatty acid), popularly referred to as Lorenzo's oil, can lead to complete normalization of blood levels of VLCFA. Moderate reduction in platelet count occurs as a side effect in 40% of the patients but does not lead to hemorrhages. This treatment does not alter the clinical disease course in patients in whom the neurological deterioration has already set in. However, there is increasing evidence that, depending on the degree of treatment compliance and decrease in VLCFA, the treatment has a protective effect when started in neurologically intact patients. Treated adequately, a smaller number of boys with the biochemical defect develop the aggressive cerebral form of the disease, and the onset of neurological symptoms is delayed. Unfortunately, dietary treatment is not an absolute preventive, and patients may still develop cerebral XALD despite perfect compliance and normalization of plasma VLCFA levels.

Another important cornerstone in the treatment of cerebral XALD is bone marrow transplantation. The outcome is highly dependent on the degree of cerebral involvement at the time of the transplantation. If applied early in the course of the disease, bone marrow transplants may prevent further deterioration and may even lead to improvement of cerebral lesions on MRI. In the brain, the donor-derived cells are microglia, and it is assumed that the beneficial effect of bone marrow transplantation is that donor microglia metabolize VLCFA, thus reducing their levels in the brain. The observation that improvement appears to commence only 6 months after transplantation is compatible with the slow turnover of microglia. The therapy is not recommended for patients with rapidly advancing or severe cerebral involvement, because these patients do not tolerate the temporary worsening associated with the procedure or the delay in onset of beneficial effect. Generally, an IQ of 80 (performance IQ appears to be especially important) is considered the limit because outcome in patients with a performance IQ below 80 is poor. In these patients transplantation may even lead to rapid worsening and sometimes an early death. Delayed stabilization in a poor neurological condition or vegetative state may also occur. Bone marrow transplantation is also not recommended for asymptomatic patients, because of the over 50% chance that they will not develop the serious cerebral form of the disease, and the high mortality associated with bone marrow transplantation. Transplantation is also not recommended in stable patients, because the duration of the stable period is unclear and may be many years. Unfortunately, the prognosis of XALD cannot be predicted by VLCFA measurement, mutation analysis, or the expression of the disease in relatives. It is presently not possible to predict whether a young asymptomatic boy is destined for a severe phenotype, for which

bone marrow transplantation should be performed, or a mild phenotype. For this reason, monitoring of the disease on MRI and using neuropsychological tests from the presymptomatic stage onwards is extremely important. As soon as lesions appear, and appear to be progressive, bone marrow transplantation should be attempted. Extensive screening of family members should be performed as soon as the diagnosis of XALD or AMN has been established in one patient and HLA typing should initiated in all persons who are potential future candidates for bone marrow transplantation in order to search for a suitable donor and be prepared for transplantation. Bone marrow transplantation is presently not performed in adult patients with AMN. This is because the morbidity and mortality of bone marrow transplantation are higher in adults than in children, whereas AMN is a relatively slow disease that may take decades before death ensues. At present, it is not clear whether bone marrow transplantation at a young age for incipient cerebral XALD will prevent the development of AMN in adulthood.

Promising alternative therapies have been proposed, but no definitive data on their clinical effects are as yet available. 4-Phenylbutyrate up-regulates the expression of a protein referred to as ALDR, which is a protein with a high homology with ALDP and may be able to substitute at least in part for its function. The gene encoding this protein is called *ABCD2*. There is evidence that the drug may lead to lowering of plasma and tissue VLCFA. Lovastatin also induces the increased expression of ALDR. It improves the capacity of XALD fibroblasts to metabolize VLCFA and normalizes VLCFA in plasma of XALD patients. Lovastatin may also inhibit inducible nitric oxide synthase and proinflammatory cytokines. However, simvastatin, an analogue of lovastatin with similar pharmacokinetics and effects on plasma VLCFA, failed to decrease VLCFA tissue content.

Ultimately, gene therapy may hold the greatest hope for the treatment of XALD patients. One possible strategy would be bone marrow ablation followed by autologous transplantation with genetically corrected hematopoietic cells of the patient's own bone marrow. Transplantation and oligodendrocytes or pluripotent stem cells to repair damaged tissue are other options.

21.6 Magnetic Resonance Imaging

In imaging the brain of patients with XALD, different patterns can be detected. The most commonly occurring pattern in cerebral XALD consists of predominantly parieto-occipital white matter abnormalities (Fig. 21.1). The lesion starts in the splenium of the corpus callosum and spreads out into the parieto-occipital white matter. The arcuate fibers are relatively or completely spared, which is most easily seen on T_1-weighted images. In most patients two zones can be distinguished, the anterior zone where demyelination advances being less severely affected than the posterior zone. After administration of contrast, a rim of enhancement can be seen surrounding the most severely affected area, separating this area from the less severely affected and normal white matter (Fig. 21.1). These three zones are in accordance with the three zones recognized histologically. The outer and advancing zone is in the process of active demyelination without inflammation. The middle zone shows signs of prominent inflammation. The innermost and most posterior region is completely demyelinated and burnt out. In this latter area, cavitation and calcification may be present. The calcium deposits are best seen on CT (Fig. 21.2). The lesions are essentially symmetrical, but are often not so in detail. Demyelination advances in the frontal direction. Structures affected relatively early on are the lateral and medial geniculate bodies, in some cases the lateral-inferior part of the thalamus, the posterior limb of the internal capsule, and the external capsule. The frontal lobe is affected last and more variably. In the end, almost all cerebral white matter is affected. In the end-stage, serious white matter atrophy is seen. The cerebellum is not usually involved early in the course of disease, in contrast to the brain stem. Typically, the involved tracts are the occipitoparietotemporopontine and pyramidal tracts, the brachium of the inferior colliculus, the brachium of the superior colliculus, and the lateral lemniscus. The frontopontine tracts are preserved. This MRI pattern is present in about 80% of patients with childhood cerebral XALD and has a lower percentage in older patients. The overall fre-

Fig. 21.1. An 8-year-old boy with cerebral XALD. The T_2-weighted axial images show the characteristic pattern of symmetrical lesions in the parieto-occipital white matter. The images show that the lesion contains three zones. The inner and more posterior zone has a slightly higher signal than the outer and more anterior zone; there is a rim of low signal in between. The contrast-enhanced T_1-weighted images confirm the two zones, the inner and more posterior zone having the lowest signal, whereas the outer and more anterior zone has a less low signal. The rim in between shows prominent contrast enhancement. The three zones of the disorder can be visualized this way: the outer and more anterior zone being inactive and completely demyelinated, zone 2 showing marked inflammation and advanced demyelination, and zone 3 representing the start of demyelination. Zones 1 and 3 can usually be distinguished on T_2-weighted images. The splenium of the corpus callosum is affected and the lateral part of the midbrain as well as the corticospinal tracts in the pons

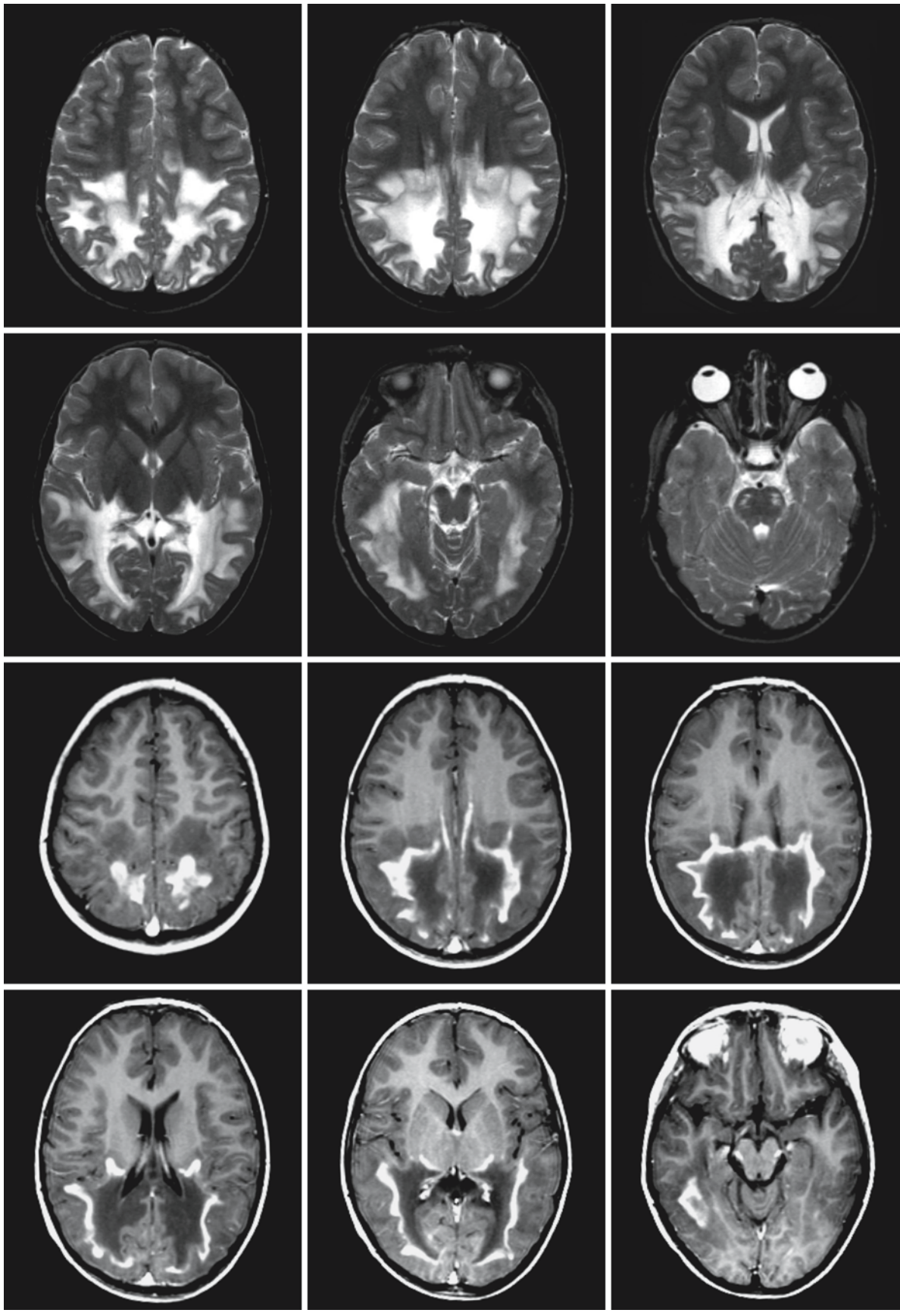

Fig. 21.1.

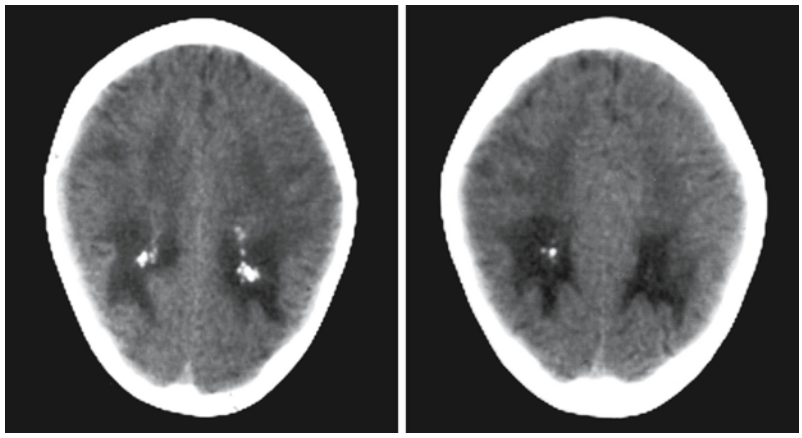

Fig. 21.2. CT scan of an 8-year-old boy with cerebral XALD shows calcium deposits in the lesion

quency of this pattern among patients with cerebral XALD is about 65–70%.

The pattern of cerebral XALD may be reversed, starting bilaterally in the frontal area with concomitant involvement of rostrum and genu of the corpus callosum (Fig. 21.3). In such cases the anterior limb of the internal capsule is involved instead of the posterior limb, and the frontopontine brain stem tracts instead of the occipitoparietotemporopontine tracts. Again, there are three distinguishable zones with contrast enhancement of the middle zone. This pattern is most commonly seen in teenagers and adolescents with cerebral XALD. The overall frequency among patients with cerebral XALD is about 15%.

In a few patients, there is concomitant but independent frontal and parieto-occipital white matter involvement. This pattern is rare and occurs mainly in children.

In some patients the frontopontine or corticospinal projection fibers are involved without involvement of the periventricular or deep white matter. In this pattern lesions are initially seen in the internal capsule and brain stem tracts. With progression of disease abnormalities most often develop in cerebellar peduncles, the splenium of the corpus callosum, and optic radiation. Contrast enhancement may be present. This pattern is seen in 10–15% of all cerebral XALD patients. It is mainly seen in adult XALD patients with clinical signs of AMN (Fig. 21.4), but has also been reported in children with mainly pyramidal signs.

Another pattern consists of symmetrical involvement of cerebellar white matter, middle cerebellar peduncles, and brain stem tracts. This pattern is rare and is mainly seen in adolescents.

In exceptional cases patients have unilateral disease initially and bilateral but markedly asymmetrical disease in later stages (Fig. 21.5). The presence of two zones in the lesion on unenhanced images and contrast enhancement of the rim in between these zones are helpful diagnostic clues. One asymptomatic adult patient suffered a severe cerebral contusion in the left temporal area after which the course was downhill. Demyelination started in the area of the contusion, and proceeded to affect both hemispheres, but asymmetry remained. Marked contrast enhancement was seen and extensive inflammation on brain biopsy.

In general, patients with the parieto-occipital pattern and those with the frontal pattern have rapid disease progression if the abnormalities are present at an early age and if contrast enhancement is present. Patients with both frontal and parieto-occipital abnormalities have the most rapid disease progression. Patients with predominant involvement of frontopontine or corticospinal projection fibers or predominant involvement of the cerebellar white matter generally have much slower disease progression.

The primary finding in AMN consists of spinal cord atrophy. In half of the symptomatic AMN patients, abnormalities are noted on brain MRI. This percentage depends on age: it is lower among the younger AMN patients and higher among the older AMN patients. Involvement of corticopontine and corticospinal projection fibers, described above, is most commonly seen. However, AMN patients may also develop much more extensive white matter abnormalities, involving mainly the parieto-occipital white matter and splenium of the corpus callosum, the frontal white matter and anterior part of the corpus callosum, or the cerebellar white matter.

Only a few female heterozygotes show abnormalities on MRI of the brain and spinal cord, even when they display an AMN-like clinical phenotype. The pattern of full-blown cerebral demyelination is very rare.

Apart form its role in diagnostics, MRI has a major role in monitoring of the disease (Figs. 21.6 and 21.7). Monitoring is an essential part of treatment, first of all to determine which patients are eligible for a particular treatment and secondly to monitor treatment effects. At present, there are two cornerstones in the

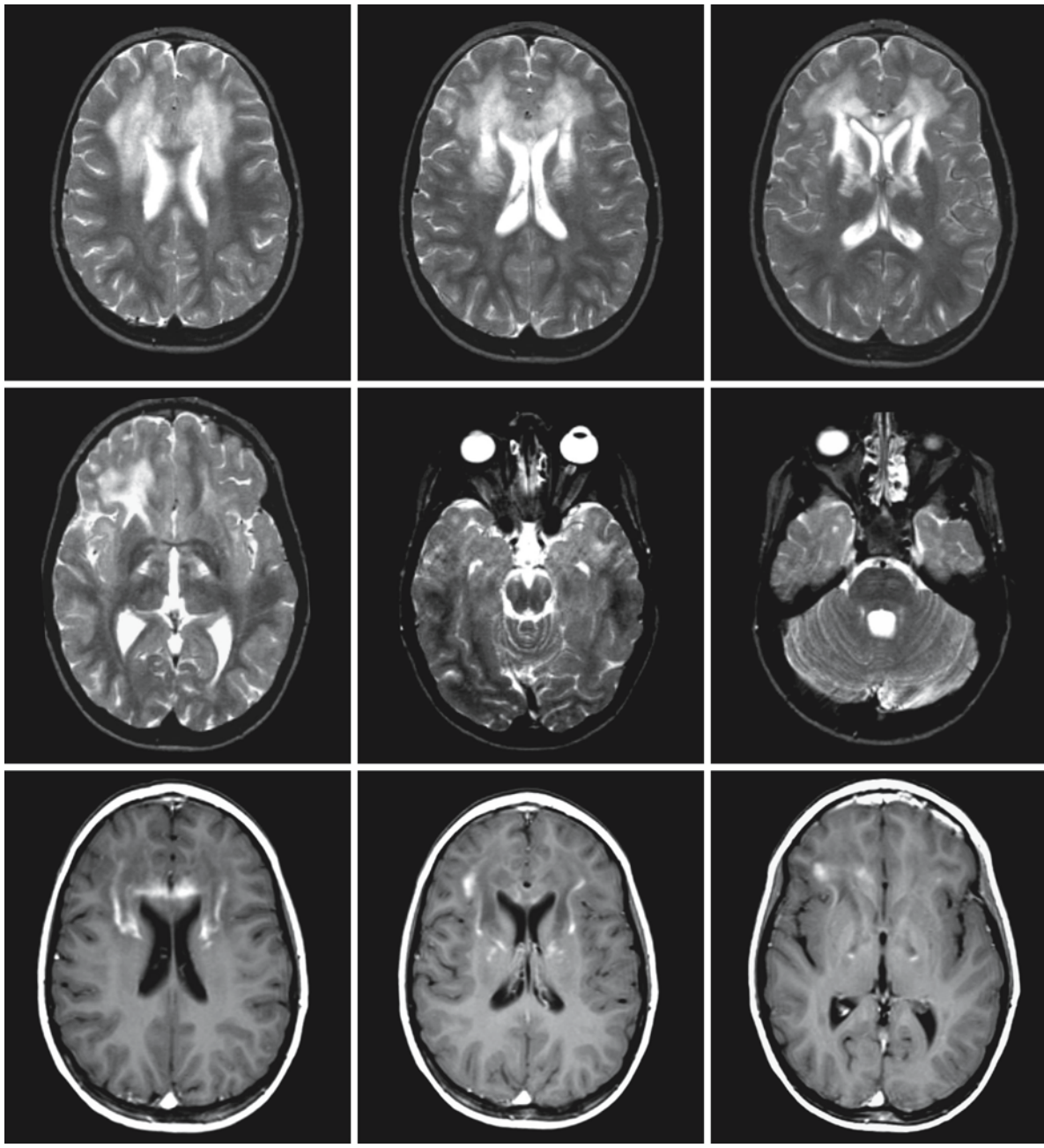

Fig. 21.3. A boy with cerebral XALD. The *first three rows* show images at the age of 9 years. Note the frontal predominance of the lesions with involvement of the genu of the corpus callosum and the anterior limb of the internal capsule. The U fibers are relatively spared. Also note the involvement of the fronto-pontine tracts at the level of the midbrain. The three zones are visible, although less clearly than in Fig. 21.1, with enhancement of the middle zone on the T_1-weighted images. (Continue see next page)

treatment of XALD: dietary treatment and bone marrow transplantation. Dietary treatment should be applied in all asymptomatic patients in an attempt to delay and, hopefully, prevent the onset of clinical symptoms. Unfortunately, dietary treatment is not an absolute preventive. Bone marrow transplantation is indicated for patients in whom cerebral demyelination is starting, especially young patients without AMN, but not for patients with a normal MRI or patients with entirely stable MRI abnormalities. Loes et al. (1994 and 2003) developed a 34-point scoring method for the grading of MRI abnormalities. It has

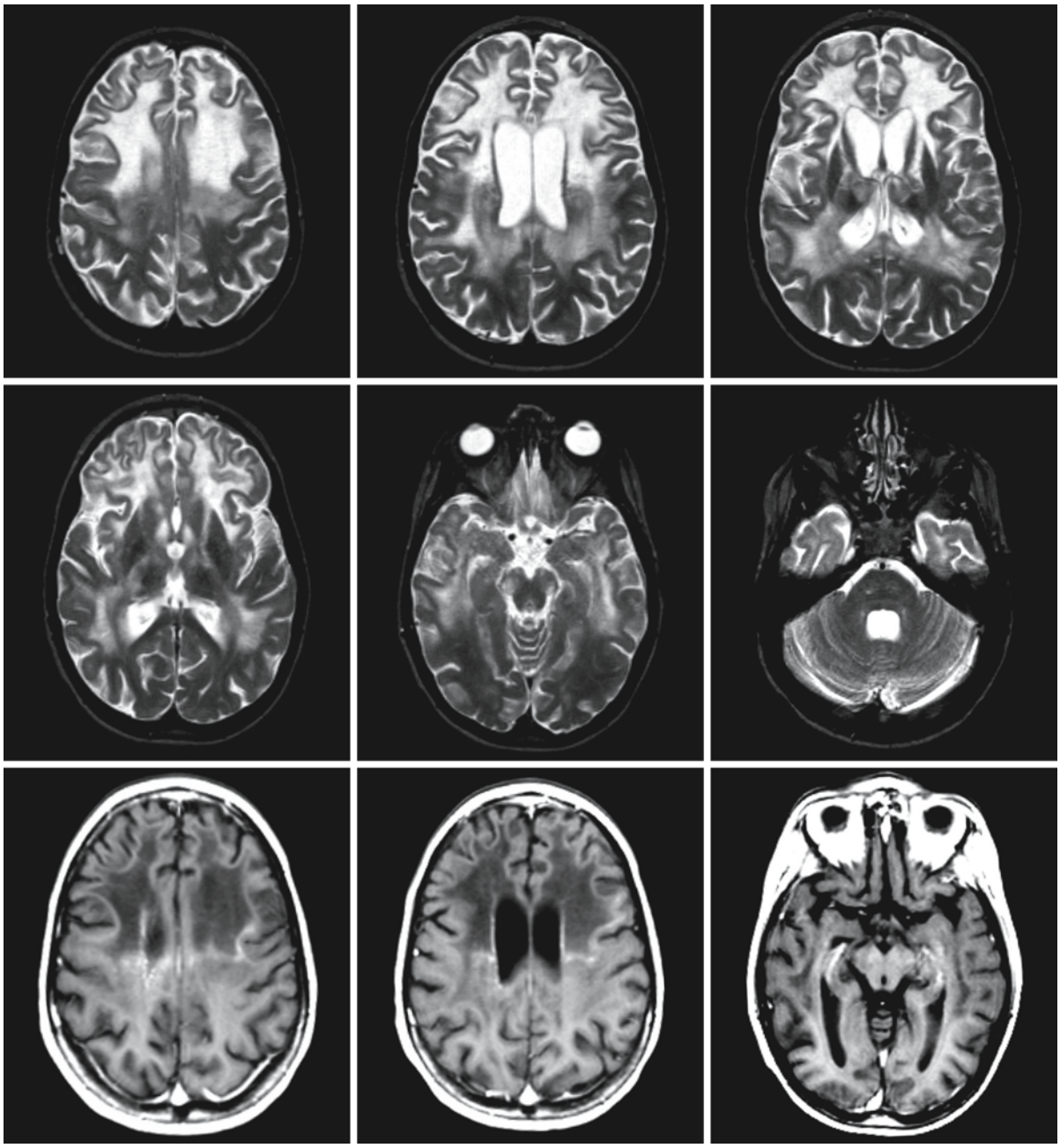

Fig. 21.3. (*continued*). A boy with cerebral XALD. The *second three rows* show the follow-up MRI, obtained 1.5 years later. The images demonstrate serious progression of the disease with spread over almost the entire brain. Only the parieto-occipital region is partially spared. The cerebellar white matter remains intact

been found that the Loes score has predictive value. With a score higher than 3, almost all patients worsen, irrespective of age; only 10% remain neurologically stable. With a score of 1–3, 60% of patients worsen, irrespective of age, but survival is longer. If the Loes score is below 1, prognosis depends on the age of the patient: among patients aged between 3 and 7 years, 30% will develop rapidly progressive cerebral demyelination; in the age group between 7 and 10 years, 10% will develop rapidly progressive cerebral demyelination; while in the age group above 10 years, rapidly progressive cerebral demyelination is rare and AMN is more likely. Contrast enhancement also has predictive value: absence of enhancement is associated with stable disease in 80–85% of the patients, whereas clear contrast enhancement is an indication

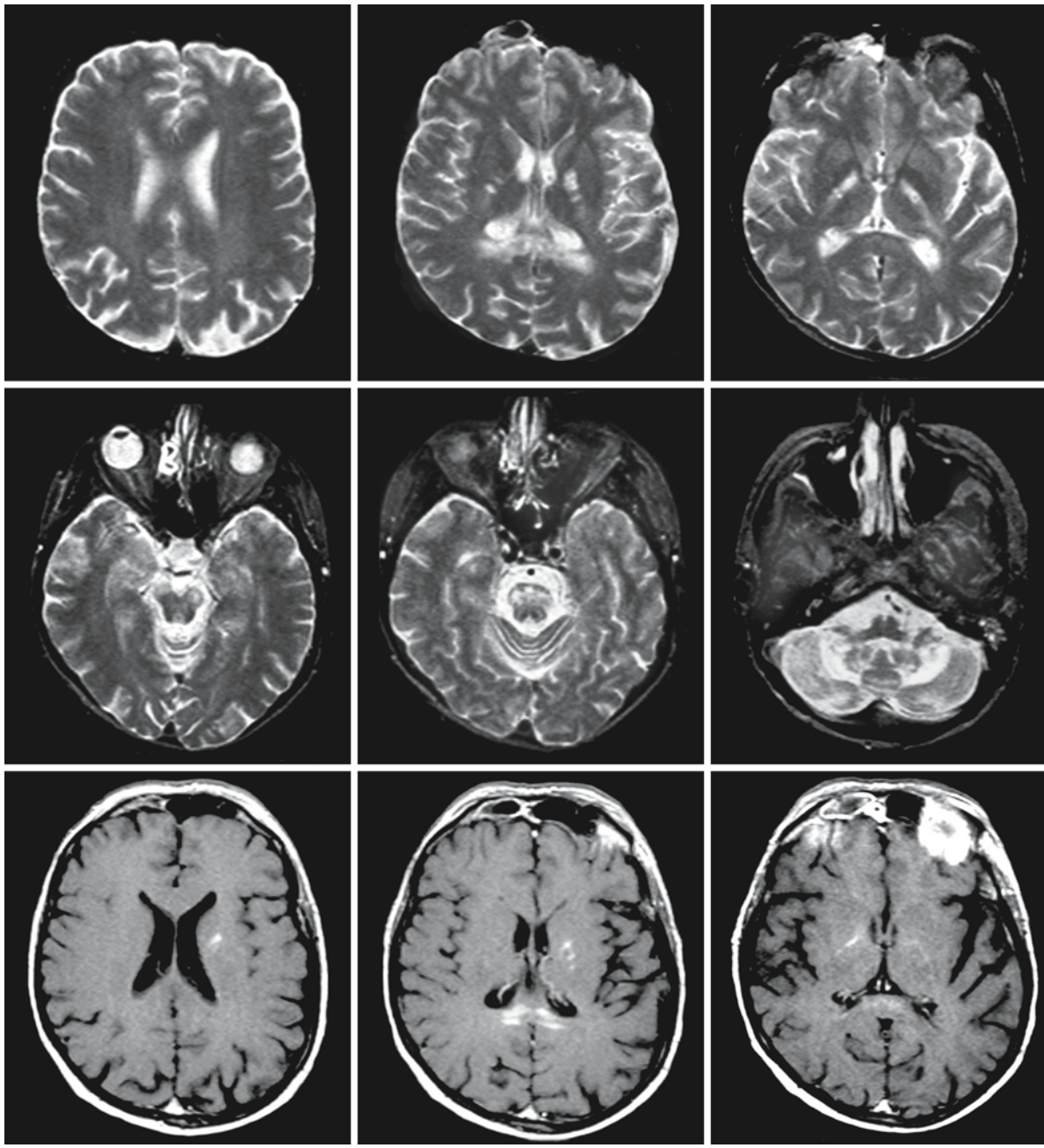

Fig. 21.4. A 45-year-old man with an AMN clinical phenotype. The T_2-weighted images (*upper two rows*) show the involvement of the cerebellar white matter, the brain stem tracts, the splenium of the corpus callosum, and the posterior limb of the internal capsule. Some enhancement occurs after injection of contrast (*third row*)

of active inflammatory demyelination and is associated with progression in 85–90 % of the patients. Diffusion tensor imaging may be more sensitive than conventional MRI and show abnormalities in areas that are not (yet) abnormal on conventional images. The potential role of magnetization contrast imaging in the monitoring of XALD patients is presently not clear. The decrease in magnetization transfer ratios reflects the three zones of white matter involvement, the lower ratios being present in the central burnt-out region. Normal-appearing white matter has a normal magnetization transfer ratio. The findings at MRS of the brain also have predictive value, in particular the findings just outside the visible lesion. If the *N*-acetylaspartate:choline ratio is decreased, this is evidence for impending demyelination in the area and disease

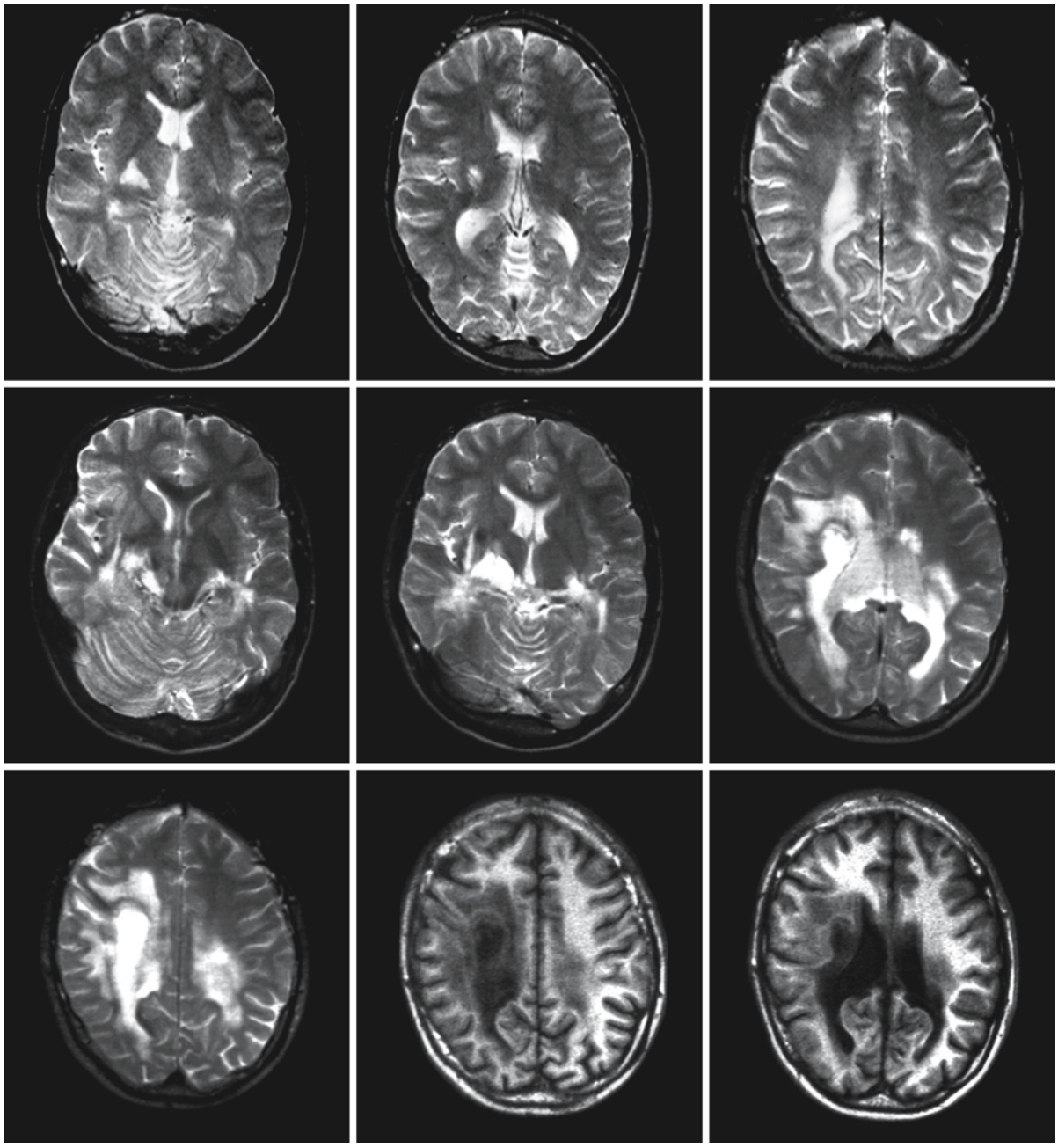

Fig. 21.5. Adult male patient with cerebral XALD. The MRI of the *first row* was obtained when he was 29 years old; the MRI of the *second* and *third row*s when he was 31 years old. The process started unilaterally and progressed to a bilateral but highly asymmetrical disease with predominant involvement of the right side of the brain. Still the MR characteristics suggest cerebral XALD with a zoned lesion, the middle zone having a low signal on the T_2-weighted images. The zone are particularly prominent on the T_1-weighted images

progression. A normal *N*-acetylaspartate:choline ratio provides evidence for a stable situation. For these reasons, combined MRI and MRS studies every 6 months are recommended for asymptomatic XALD boys up to the age of 10 years. If abnormalities are seen on MRI, contrast-enhanced images are added to the protocol. As soon as abnormalities are found, a repeat MRI is obtained after 2–3 months to document progression. If progression of abnormalities is demonstrated and the Loes score becomes 3 or higher, bone marrow transplantation is performed as soon as possible. Between the ages of 10 and 20 years, the

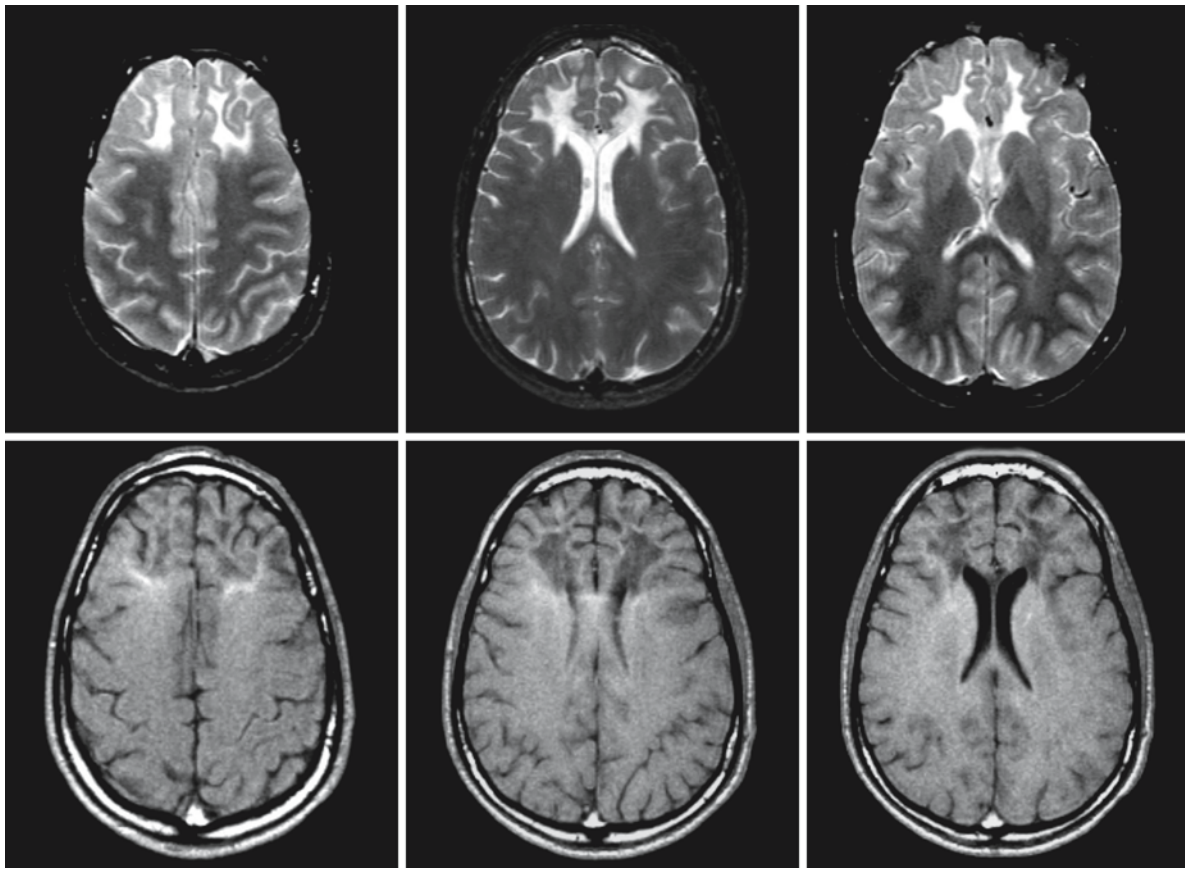

Fig. 21.6. This 18-year-old boy had learning and behavioral problems from early on. At the age of 15 years he experienced an encephalitis-like episode from which he fully recovered. Since then he has been clinically stable with a normal neurological examination. Between the ages of 15 and 18 years MRI findings have remained unchanged with frontal white matter abnormalities and white matter atrophy. The genu of the corpus callosum is involved. The entire cerebral white matter appears slightly elevated in signal, with a loss of contrast between white and gray matter. There is some contrast enhancement at the rim of the lesion. In view of the static nature of the disease, hematopoietic stem transplantation has not been considered

frequency of MRI and MRS is decreased to once a year. After bone marrow transplantation, various outcomes are seen. In some patients MRI abnormalities regress or disappear; in others the abnormalities become stable. Unfortunately, bone marrow transplantation is not a great success in all patients and further deterioration may also occur.

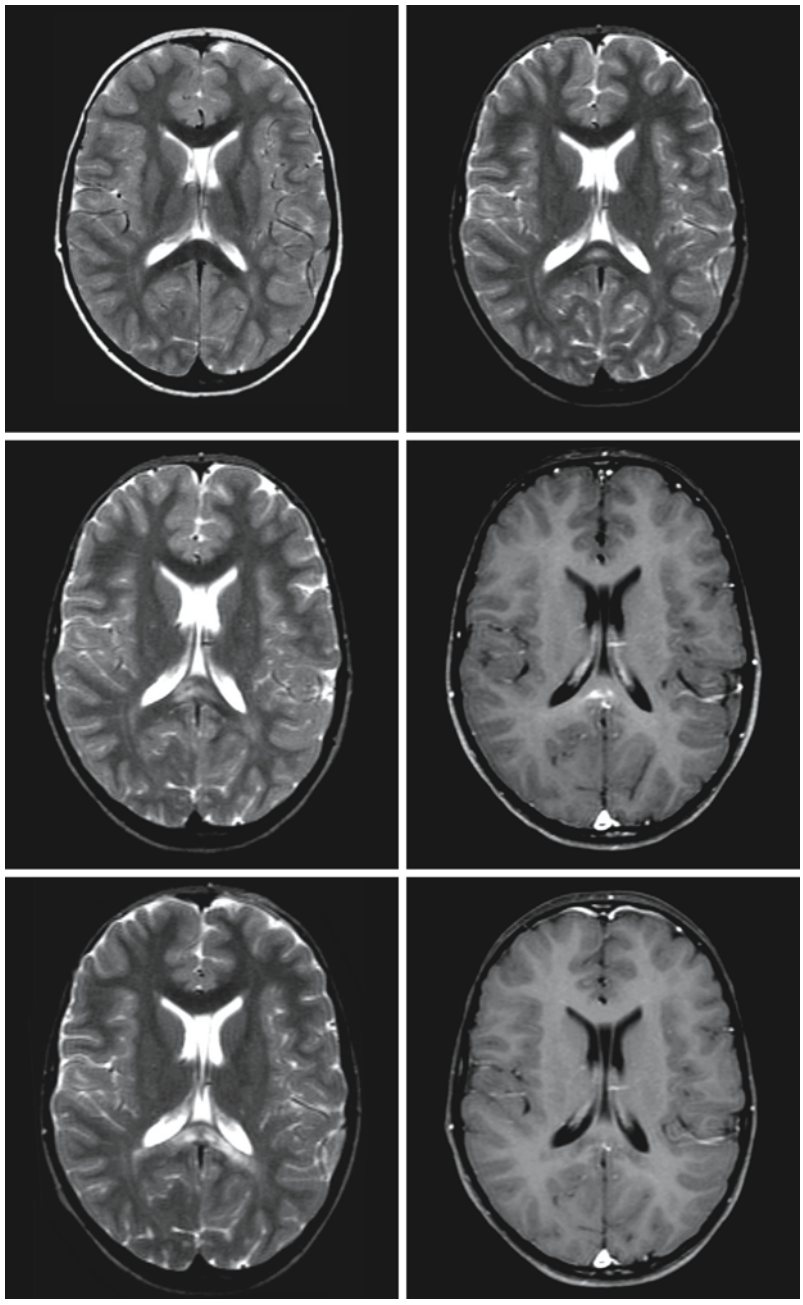

Fig. 21.7. This boy is the younger brother of the patient shown in Fig. 21.1. He was diagnosed with the biochemical defect of XALD after his brother was diagnosed with the disease. He was monitored by MRI and MRS every 6 months and normal results were found (*upper row*, *left*). When he was 6 years old a lesion was found in the splenium of the corpus callosum (*upper row*, *right*), which had increased in size after 3 months (*middle row*, *left*). At that time contrast enhancement of the lesion was found (*middle row*, *right*). Bone marrow transplantation was performed soon after without major complications. The first MRI after the procedure showed some further progression of the lesion, but the lesion has been stable since and he is now 9 years old (*third row*, *left*). There is at most minimal contrast enhancement remaining. He is clinically intact