X-linked Adrenoleukodystrophy in Three Filipino Families

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ABSTRACT

We report five Filipino male children from three families with the childhood cerebral form of adrenoleukodystrophy. All patients demonstrated deterioration in mental and developmental skills with onset of symptoms ranging from 6 to 9 years of age. Diagnosis was confirmed by elevated very long chain fatty acid (VLCFA) levels and characteristic magnetic resonance imaging (MRI) findings. Carrier testing was carried out. Genetic counseling was performed on families, thereby aiding them to cope and accept the diagnosis.

Key Words: adrenoleukodystrophy, X-linked, adrenal function, carrier testing, genetic counseling

Introduction

Adrenoleukodystrophy (ALD) is an X-linked neurodegenerative disorder with an estimated incidence between 1:20,000 and 1:50,000 of the total population, and appears to be the same in most ethnic groups. The disease shows a wide range of phenotypic variation. Thirty five percent have the childhood cerebral form wherein affected boys suffer progressive neurologic deficit that leads to a vegetative state around 4-8 years of age. In another 35 to 40% of patients, the disorder presents in young adulthood as adrenomyeloneuropathy (AMN) with progressive paraparesis and sphincter disturbances that involves the long tracts in the spinal cord. Adrenal insufficiency can occur in 90% of the former type and in 60% of the latter type. Less common phenotypes include adrenal insufficiency without nervous system involvement, progressive cerebral dysfunction in adults and persons who are asymptomatic. Up to 50% of heterozygotes have mild neurologic disturbances that resemble AMN. Overt adrenal insufficiency is rare for them.[1]

ALD is the most frequent peroxisomal disorder characterized biochemically by the accumulation of very long chain fatty acids (VLCFA) in brain white matter, adrenal glands and in other tissues and body fluids. The defect results from the impaired capacity to degrade VLCFAs, a function that has been localized to the peroxisome. In this paper , we discuss X-linked ALD in 5 affected males from 3 different families who all presented with the childhood cerebral form of the disease.

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Clinical Reports

Family 1

Patient 1

The proband (III-F) was a 10 year old male who was born full term to a 27 year old primigravid after an uncomplicated pregnancy. Developmental milestones were achieved at appropriate chronological ages. He had an average school performance during the first 3 years of his primary education. Except for an ear trauma at 4 years old and an episode of acute glomerulonephritis which had a mild clinical course at 5 years old, his past medical history was not contributory.

He was apparently well until about 9 years of age when he had hearing difficulties followed by difficulty in reading, and later by deterioration in handwriting skills, difficulty in understanding speech and diminished school performance which led him to quit schooling. He also had frequent tantrums and became very irritable and sensitive. He eventually became dependent for his daily activities.

At the time of examination at 10 years old, his height was 122 cm (<5th percentile) and weight was 18.5 kg (<5th percentile). He was awake but restless, did not follow commands, was disoriented to time and place and did not respond to questions appropriately. Physical examination was essentially normal. There was no skin hyperpigmentation. Neurologic examination revealed briskly reactive pupils with visual acuity limited to counting fingers, normal funduscopy, full and equal extraocular muscles, no facial asymmetry and intact gag reflex. He was able to move all extremities against minimum resistance. There was bilateral Babinski and hyperactive deep tendon reflexes.

He was the eldest in a sibship of 6 born to non-consanguineous parents. The disease status of the entire family is shown in Figure 1. Inquiries into the health status of adult male relatives disclosed no symptoms nor signs pertaining to AMN nor Addison's disease. Likewise, female carriers in the family revealed essentially normal neurologic status.

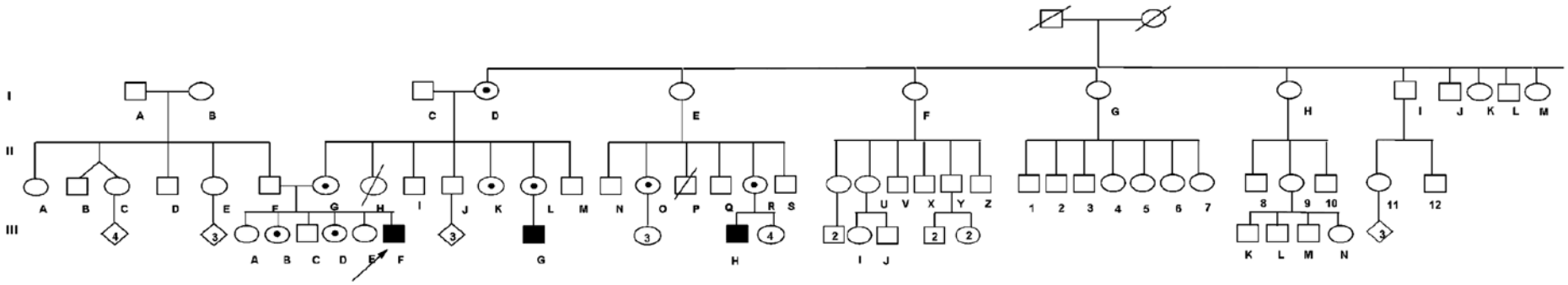

Figure 1. Pedigree of Family 1. Two female siblings of the proband (III-B) and (III-D) were carriers of the disease with high amounts of plasma C26:0 and likewise a high ratio of C24/22 and C26/22 than the normal. The plasma VLCFA of the maternal grandmother (I-D), 2 maternal aunts (II-K) and (II-L), 2 female first cousins of the mother (II-O) and (II-R) and a sister of the grandmother (I-H) were also consistent with being carriers of X-linked ALD. The proband's mother (II-G) had normal levels of C26:0 but had an increase in the C24/22 ratio. Although this provided an 80% assurance that she was not a carrier of X-linked ALD, she was an obligate heterozygote by pedigree analysis. One sister of his maternal grandmother (I-G) also had normal plasma VLCFA levels, however, two of his grandmother's sisters (I-E) and (I-F) were not available during the family consult. Two of the proband's cousins (III-G) and (III-H) were both affected with X-linked ALD with high plasma levels of C26:0 and high ratios of C24/22 and C26/22.

Laboratory results are summarized in Table 1.

Patient 2 (III-H)

He was a 7 year old boy who was born full term to a 21 y/o primigravid mother after an uncomplicated pregnancy. At 6 years old, he had blurring of vision that was followed months later by difficulty in ambulation. A cranial CT scan showed a well circumscribed symmetrical foci of hypodensities involving both the posterior corona radiata and centrum semi ovale. A nonspecific inflammatory process was considered. He was given oral steroids and was discharged after 4 days. However, no improvement was noted and he continued to have progressive deterioration in his motor, receptive and expressive language skills until he became non-ambulatory and aphasic.

He was the eldest in a sibship of 5 born to a non-consanguineous couple. All his siblings were females. At the time of examination (around 7 years old), he had notable spasticity of all extremities. He was quiet and had a blank stare. His head circumference was at the 25-50th percentile for age, while his length and weight were below the 5th percentiles. Physical examination was normal. Neurologic examination revealed briskly reactive pupils, with visual acuity limited to light perception. Deep tendon reflexes were hyperactive and there was bilateral Babinski. He died several months after evaluation due to pneumonia.

Patient 3 (III-G)

He was born full term to a 25 year old primigravid mother after an uncomplicated pregnancy. He was first seen at 6

Table 1. Summary of laboratory investigations

	Patient 1	Patient 2	Patient 3	Patient 4	Patient 5
Plasma VLCFA	an increased amount of hexacosanoic acid (C26:0) and a higher than normal ratio of tetracosanoic acid/ docosanoic acid (C24/22) and C26/22	Not done	Not done	an increase in the amount of C26:0 and an elevated ratio of C24/22 and C26/22	increased amounts of C26:0 and an elevated ratio of C24/22 and C26/22
Cranial findings	MRI showed symmetrical T2W hyperdensities in both the posterior temporoparietal and occipital lobes and smaller hyperdensities in the right and left pons and in the ventral medulla were consistent with ALD	CT Scan symmetrical foci of hypodensities involving both the posterior corona radiata and centrum semi ovale	CT scan initially revealed a non contrast enhancing signal abnormality involving the splenium of the corpus callosum with extension to the central occipital white matter; Follow up scan after a year revealed a radiographic evidence of progression of white matter disease with development of non enhancing signals involving the corticospinal tract bilaterally to the level of the cerebellar peduncles at midbrain level	CT scan showed white matter changes on both fronto-temporoparietal lobes Cranial MRI findings showed white matter changes on the temporoparieto-occipital lobes	cranial MRI was highly suggestive of ALD with white matter changes on the temporoparieto-occipital lobes
EEG	Normal	Not done	Not done	intermittent slowing of brain activity over the left hemisphere	Not done
Pure tone Audiometry (PTA)/ BAER	Mild hearing loss, bilateral (PTA)	Not done	<60dB left;<40dB right (BAER)	Abnormal (BAER)	Normal (BAER)
VER	Abnormal	Normal	Normal	Normal	Abnormal
Cortisol	Elevated	Not done	Normal	Normal	Normal
Serum Electrolytes	Normal	Not done	Normal	Normal	Normal

years old and has remained neurologically asymptomatic up to present. His physical and neurologic examinations done at 6, 7 and 8 years old were within normal limits.

Laboratory results are summarized in Table 1.

He is currently being maintained on Coenzyme Q and cyclooxygenase 2 inhibitor.

Family 2
Patient 4

The proband (Figure 2) was seen at 9 years old. He was born full term to a 27 year old primigravid mother after an uncomplicated pregnancy. Except for congenital ptosis, his past medical history was unremarkable. His development was at par with his chronological age.

He was apparently well until at 8 years of age when tremors on both hands were noted along with a trembling gait. A cranial CT scan showed white matter changes on both frontotemporoparietal lobes. Acute disseminated encephalomyelitis was entertained and he was treated with IV steroids which did not improve the symptoms. His gait disturbance rapidly deteriorated into a state wherein he could not walk anymore without support. Three months after the onset of symptoms, he was brought to another physician where X-linked adrenoleukodystrophy was then considered.

On physical examination, he walked with assistance, had dysarthric speech but was receptive and able to follow some commands. He had a notable intermittent twisting motion of his head, trunk and extremities. Physical examination was normal. He had ptosis on the right eye and had pigmentation on both iris. Neurologic examination showed pupils that were equally reactive, had pale discs on funduscopy, motor strength of 5/5 on the upper extremities and 3/5 on both lower extremities, increased tone on the left upper extremity with resultant preferential movement of the right, hyperactive deep tendon reflexes (DTRs), bilateral clonus and babinski, and intention tremors on both hands.

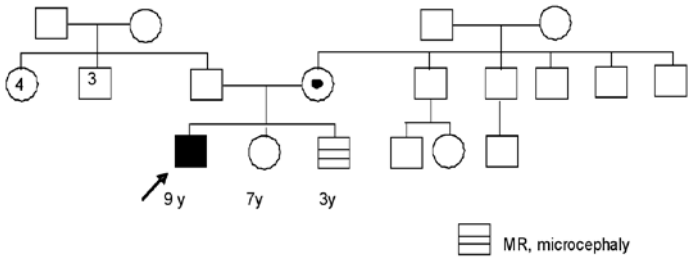

Figure 2. Pedigree of Family 2. The proband was the eldest in a sibship of 3 born to a non-consanguineous couple. A 3 year old male sibling has global developmental delay secondary to primary microcephaly. His mother was confirmed to be a carrier of the disease with high amounts of plasma C26:0 and high ratio of C24/22 and C26/22. No other family members had the similar illness or symptoms referrable to a myelopathic condition.

Family 3
Patient 5

The proband (Figure 3) was born full term to a 20 year old primigravid mother after a pregnancy complicated by urinary tract infection near term. He had no perinatal complications and had an uneventful neonatal course.

His development was observed to be at par with age and was an average grade 3 student until the symptoms set in. At 8 years old, he complained of blurring of vision, and was noted to have diplopia and hearing difficulties. Two months later, he started to have behavioral changes, deterioration of vision and handwriting. A cranial CT scan was done, which showed a suspicious low attenuation foci on the right temporal lobe.

He was seen at 8 year 10 months old and was noted to be restless, uncooperative and incoherent. Physical examination was normal. His visual acuity was limited to light perception but he was able to respond to some sounds. Muscle strength was normal in all 4 extremities. Deep tendon reflexes were hyperactive and there was bilateral babinski and clonus.

Laboratory results are summarized in Table 1.

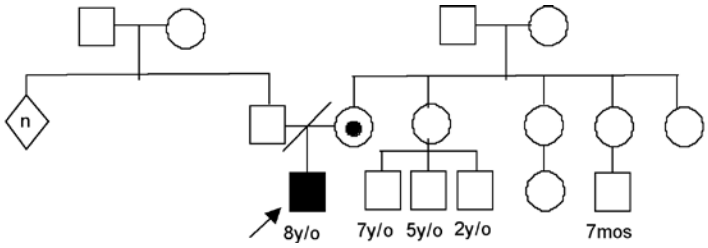

Figure 3. Pedigree of Family 3. The proband was the only child of a non-consanguineous couple. The plasma VLCFA of the mother was consistent with a carrier state with an increased amount of C26 and higher than normal ratios of C24/22 and C26/22. His mother had 4 other female siblings with male children who were asymptomatic. No carrier testing has been done on them at this time.

Discussion

All of the affected males in the 3 families just described had the childhood cerebral form of X-linked ALD. Patient 1, 2 and 5 initially presented at less than 10 years of age with vision and auditory disturbances followed by progressive behavioral and cognitive defects which eventually led to a vegetative state. Patient 4 primarily presented with a movement disorder that was dystonic in form. This movement disorder, to our knowledge, is an uncommon neurologic manifestation of X-linked ALD and has not been reported in literature. It may be indicative of basal ganglia involvement that could be secondary to the active demyelination and inflammation in the temporoparieto-occipital areas. Patient 3, on the other hand was neurologically asymptomatic at 8 years of age although serial brain magnetic resonance

imaging revealed a progressive course of white matter abnormalities.

Based upon historical analysis of 388 X-linked ALD patients, 35.87% had developed neurologic symptoms at 10 years of age defined as childhood cerebral ALD, while 64.13% of them remained free of neurologic symptoms at this age. It has been estimated therefore, that between 31% and 39% of untreated ALD patients develop neurological symptoms prior to age 15 years and conversely that 61-69% escape this fate.[2]

An abnormal MRI generally has serious prognostic implications irrespective of age. A normal MRI is a favorable sign in patients who are more than 7 years old while in patients below 7 years of age, MRI does not provide a similar reassurance.[3]

Loes scoring method for brain magnetic resonance (MR) observations in X-linked ALD patients has been developed to help define better the natural history of ALD and monitor response to developing therapies. Based on MR severity scores done serially in the MR positive group, 52% showed progressive disease, 18% had progressive disease with subsequent stabilization, 24% had stable disease and 6% had minimal improvement.[4] Thus, it is not possible at this time to predict whether patient 3 is destined for a severe or a milder phenotype of the disease based on MRI findings alone. His neurologic and neuropsychologic functions depend greatly on his MRI scores.

X-linked ALD presents a challenge because of the striking variability of clinical course even among members of the same family. For Family 1, the 3 affected members all have the childhood cerebral form of ALD but had obvious phenotypic variability. Patient 2 had an earlier onset of neurologic symptoms at 6 years old while Patient 1 had symptoms at 9 years of age. Patient 3 is neurologically normal at 8 years of age despite evident white matter changes on MRI. The cause of this phenotypic variability is unknown. It is not attributable to the nature of the mutation nor to the severity of the abnormality of VLCFA levels nor to the expression of the disease in relatives.[5]

Segregation analysis suggests the action of an autosomal modifier gene as the possible etiologic mechanism for the varied phenotype expression.[6,7] Environmental factors may also influence the severity of the patient's phenotype.[8]

The gene for X-ALD has been mapped to Xq28. It codes for the peroxisomal membrane protein ALDP with homology to the ATP binding cassette (ABC) transporter superfamily of proteins. Proteins of this family are involved in transport across cellular and subcellular membranes of a wide variety of ligands such as ions, fatty acids and proteins.[5,9] Alterations in the ALD gene have been identified in more than 300 different ALD families.[1] Most mutations are 'private' and 68% of the identified mutations are non recurrent. Of all the disease causing mutations, 53% are missense mutations, 24% are frameshift, 5% are in-frame deletions or insertions and 2.5% are splicing defects. Mutation analysis has confirmed that X-linked ALD phenotype cannot be predicted based on X-linked ALD genotype. The mechanism by which the ALD gene defect leads to VLCFA accumulation have not yet been determined. Several hypotheses suggested the following: that ALDP might be required for association of VLCS with the peroxisomal membrane, or that ALDP might translocate VLCS into the peroxisomes and that ALDP could be required for import of a substrate necessary for the activation and or stabilization of VLCS. The pathological effects of VLCFA accumulation include alteration of membrane structure and function in adrenocortical and nerve cell membrane, increased proportion of fatty acid precursors of cholesterol esters and impaired function of the ACTH receptor in adrenal dysfunction, and cell death that initiates the inflammatory demyelinating processes in the brain.

VLCFA levels are increased in approximately 85% of women who are heterozygotes for X-linked ALD, but a significant 15% of them have false negative results on VLCFA testing.[10] Thus, DNA based mutation analysis can be done after the disease causing mutation has been identified in the family. It specifically provides the most definitive technique in obligate heterozygotes like the mother of patient 1 who had false negative results on plasma VLCFA assays.

Cerebral ALD and its other phenotypic variants such as AMN are frequently associated with Addison's disease.[11] Adrenal insufficiency may precede, co-exist or develop after neurologic dysfunction. It was reported to be present before onset of neurological symptoms in 92% of the youngest children with clinical onset before the age of 6 years. In comparison, 80% of older patients with clinical onset after the age of 20 years had neurological symptoms preceding the endocrinologic symptoms. The interval between the onset of neurologic and endocrinologic symptoms may be longer than 10 years regardless of whether neurological or endocrinological symptoms occur first. No correlation was found among ALD gene mutation, VLCFA levels and endocrinologic dysfunction. Adrenocortical dysfunction is defined by clinical symptoms of adrenal insufficiency or an impaired cortisol response in the conventional ACTH stimulation test, the standard for evaluating the adequacy of adrenal cortical function.

Our patients had no signs and symptoms such as hyperpigmentation, exercise intolerance, vomiting or addisonian crisis. Patient 1 had increased serum cortisol level while patients 3,4 and 5 had normal serum cortisol levels. Patient 2 was not available for serum cortisol determination. Isolated measurements of plasma cortisol levels are insufficient and may lead to the false conclusion that adrenal insufficiency has been ruled out.[1] Unfortunately, ACTH stimulation test was not done in all patients, so that the following assumptions have been made regarding the interpretations of the serum cortisol results of our patients. An increased cortisol response under stress may indirectly imply an intact adrenal function for patient 1. However, for patients 3, 4 and 5, the normal cortisol levels could either signify a reduced cortical response to stress or may truly

denote a normal cortisol response.

Management issues for ALD patients include supportive therapy and counseling of families. Although of no proven benefit yet, they were started on Coenzyme Q10 and cyclooxygenase 2 (COX 2) inhibitor. The rationale for the administration of CoQ10 is based upon recent results that there is mitochondrial dysfunction in X-linked ALD in addition to the well known peroxisomal defect. Cyclooxygenase 2 is present in the nervous system and may contribute to the brain inflammatory response that leads to the rapid progression of the cerebral form of the disease, thus the utilization of COX 2 inhibitors.[12]

Lorenzo's oil, a 4:1 mixture of glyceryl trioleate (GTO) and glyceryl trierucate (GTE) presumably inhibits the endogenous fatty acid elongation system.[1,13] But the diet failed to halt the neurologic progression and did not improve the endocrine dysfunction in symptomatic patients with childhood cerebral ALD. Trials regarding oral administration of clofibrate and carnitine, dietary restriction of VLCFAs and administration of other immunosuppressants such as cyclophosphamide likewise failed to bring about convincing clinical improvement.

Bone marrow transplantation (BMT) is currently the most effective therapy for the early cerebral stages of the disease but it carries a significant risk. Considerations include neuropsychological deterioration, normalization of VLCFA prior to and during transplantation and lack of neurologic defects of severe degree. The proposed mechanism of beneficial effect is mediated by brain microglia which at least in part is bone marrow derived.[14] Long term beneficial effects of BMT for childhood onset cerebral X-linked ALD when the procedure is done at an early stage of the disease include stabilization of demyelination, normal or improved motor function and verbal intelligence, stability or improvement of performance of non verbal abilities and reduction to almost 55% in plasma VLCFA concentration.[15] MR images also indicate an abatement of the process compared to untransplanted patients.[16]

Patient 3 can be a good candidate for BMT, however, factors such as assessment of neuropsychologic function, refinement of brain imaging technique and the mortality risk associated with the procedure should be carefully evaluated.

Genetic counselling is an important part of management and was done on all families. ALD being an X-linked condition carries a 50% recurrence risk for males and a 50% risk of being carriers for females.

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