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Amyotrophic lateral sclerosis:
An exploration into the SOD1 protein and a representative case study

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Abstract

Amyotrophic lateral sclerosis (ALS) is a devastating neurodegenerative disease characterized by the continual deterioration of upper and lower motor neurons, which invariably leads to death. Like other complex diseases, no particular cause can be attributed to the great majority of ALS cases. Clear pathological mechanisms have not been elucidated, but current research is making headway into understanding superoxide dismutase 1 (SOD1), an important antioxidant that is heavily implicated in the pathogenesis of familial ALS. The goals of this thesis are to help understand the molecular basis of SOD1 in relation to ALS and to demonstrate the dire need for a greater understanding of this unrelenting disease.

This paper contains a general review of ALS disease, followed by a molecular exploration of the SOD1 gene. Finally, a representative case study is included at the end to highlight the disease from a clinical perspective.

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Introduction

Amyotrophic Lateral Sclerosis (ALS), also known as Lou Gehrig's disease, is a devastating neurodegenerative disease that impairs the function of lower and upper motor neurons. The demise of motor neurons causes paralysis and respiratory failure which invariably leads to death, usually within two to five years of the onset of the disease. Lower motor neurons implicated in ALS are located in the ventral horns of the spinal cord, and upper lower motor neurons are located the in the motor cortex and the brain stem [1]. Interestingly, unlike other neurodegenerative diseases, the effects of ALS are restricted solely to motor neurons; therefore, sensory, digestive, endocrinological, and cognitive functions are preserved in ALS, even in late stages of the disease.

ALS is a complex disorder. ALS is caused by genetic and environmental factors. Despite many years of ALS research, there is no clear, unifying mechanism that explains the etiology or molecular pathology of ALS; however, in the last few years, novel research approaches have allowed for a greater understanding about the effect of key molecules implicated in the onset and proliferation of ALS. This paper will first explore the clinical basics of ALS, current direction of molecular ALS research, discuss the structure and function superoxide dismutase gene, and end with a representative case study of ALS.

Clinical manifestations of ALS

ALS is late onset, fatal disease that leads to the degeneration of motor neurons. Spinal onset ALS, which comprises three fourths of ALS cases, leads to muscle weakness, atrophy, and stiffness, paralysis, cramps, and twitching, while bulbar onset

ALS leads to difficulty swallowing and articulating speech, spasms, and uncontrolled, over-responsive reflexes [2]. This impairment leads to patients displaying exaggerated or unarticulated expressions of emotional responses in muscles innervated by motor neurons. Patients also exhibit difficulty chewing, swallowing, making basic gestures, and difficulty breathing. In its initial stages, ALS is a difficult disease to diagnose because its symptoms can be typical of other diseases. Its diagnosis generally relies on the exclusion of other similar diseases and the confirmation of several ALS related symptoms [3].

In order to confirm diagnosis, physicians usually look for a few key characteristics [4]. Although ALS affects the peripheral nervous system, it does not affect sensory neurons—only motor neurons. With this in mind, patients that have fully functioning sensory function but impaired motor function. Doctors also look at the functions of the intestine and the bladder. Since the function of these organs depends on the autonomic nervous system, even in latest stages of the disease ALS patients retain proper function of these organs. Nervous system impairment in upper and lower motor neurons in at least three regions of the body is highly suggestive of ALS.

There is no definitive test of ALS. Electromyography, a technique that records electrical signals in skeletal muscles, can offer evidence to help physicians confirm an ALS diagnosis. Completely normal x-rays are seen in ALS patients. Researchers are trying to develop new methods to diagnose ALS.

Epidemiology

ALS occurs in about two cases for every 100,000 people. ALS mostly affects those over the age of forty, and incidence increases with each decade of life. Males are at a slightly higher risk of being afflicted with ALS than females (a 1.4:1 ratio) [5].

Prognosis

Patients will ALS on average live 3.5 years after the onset of their first symptom, although rarely some patients can live up to ten years or more. Younger patients are expected to live longer after the onset of ALS than older patients. Also, patients whose first ALS symptom began at a limb site tend to live longer with ALS than patients with bulbar onset ALS [6].

Unfortunately, upon diagnosis of the disease, patients can expect usually expect a devastating deterioration of their motor neurons and a death due to respiratory failure [4].

Treatment

Currently, the only treatment available for ALS is the drug Riluzole, and its exact mechanism is not very clear. Riluzole is a glutamate agonist. It is believed to extend the life of motor neurons by reducing glutamate excitoxicity [7], but this drug does not attack the largely unknown roots of ALS. Additionally, some studies have concluded that Riluzole only improves the survival time of patients by two to three months, and others have suggested that Riluzole fails to show any improvement in patient survival times [8].

At this time, there is no effective treatment for amyotrophic lateral sclerosis [9]. Physicians and caregivers should strive to keep patients comfortable and ensure that the patient has the most peaceful death possible.

Sporadic ALS

Sporadic ALS, ALS without a clear genetic or environmental cause, comprises the great majority of ALS cases, and the remaining cases are called familial because they exhibit clear modes of mendelian inheritance and high levels phenotypic expression; however, familial and epidemiological studies demonstrate that genetic factors contribute to sporadic ALS pathogenesis, although they do not contribute to its pathogenesis in a clear fashion [10].

Most gene associations to sporadic ALS are loose and/or limited to a small sample size. The high amount of relatively weak genetic associations in sporadic ALS might do more harm than good, since it seem to slow down progress and mislead research more than it produce practical information to understand the pathogenesis of ALS [11]. Very little progress has been achieved in understanding sporadic ALS. This is very unfortunate because the great majority of ALS patients suffer sporadic ALS rather than familial ALS.

Familial ALS

According to most sources, familial ALS accounts for about ten percent of ALS cases. In one family, ALS was inherited in an X-linked dominant fashion [12, 13], but in almost every other case, familial ALS is inherited in an autosomal dominant or recessive

fashion [12]. Mutations in the SOD1 are the most common cause for familial ALS. SOD1 mutations are responsible for twenty percent of familial ALS cases [14]. SOD1 is an enzyme that neutralizes superoxide radicals by converting them to elemental oxygen and hydrogen peroxide through a series of half reactions. Most SOD1 mutations are missense mutations, and, although they usually do not cause a change in the native state of the SOD1 enzyme, mutations in the SOD1 protein make it prone to denaturation which leads to deleterious effects in the affected organism [15].

In familial ALS, the type of ALS causing mutation determines how long a patient may be expected to live; for example, patients with the SOD1 Ala4Val mutation can expect to die rapidly, while patients with G37R or H46R mutations can expect to live at least ten years after the onset of ALS symptoms [12, 16, 17]. The SOD1 is the most well studied ALS gene. Despite much research, its exact mechanisms have just begun to be elucidated in the last few years. It is believed that misfolded SOD1 forms aggregates [15] and that these aggregates disrupt mitochondrial function, axonal transport, and cause stress in the endoplasmic reticulum [12, 15]. Most of the SOD1 related ALS mechanisms are studied using murine models that contain human SOD1 or mutant SOD1 genes.

FUS and TDP43, a nuclear genes involved in functions such as transcription, miRNA processing, local RNA transport and localization, and gene splicing [18], are commonly attributed to ALS [12]. Similar to SOD1, they form aggregates. FUS and TDP43 aggregates are present in both the nucleus and the cytoplasm of glial cells and neurons [19], cells implicated in ALS pathology. It remains unclear whether FUS and TDP43 related ALS pathogenesis is caused by a loss of function or a gain of toxicity. In a recent study [20], it was discovered that TDP43 mislocalizes in the cytoplasm of motor

neurons in SOD1 G39A mice that suffer from late stage ALS. This study offers hope that the SOD1 and TDP43 proteins might fit into a convergent mechanism that helps understand the molecular pathology of ALS.

There are several other gene mutations closely linked to ALS, but they each comprise a small minority of familial ALS cases and an almost negligible proportion (far less than one percent) of total ALS cases [12].

Directions of ALS

Like diabetes and cardiovascular disease, ALS is a complex disease. The great majority of ALS cannot be attributed to a single gene or environmental cause. Most ALS cases are believed to be attributed to the compound effect of genes and their environment. Currently, ALS patients must come to terms with the unfortunate diagnosis of a relatively rapid, untreatable, and devastating death. Unfortunately, there is no cure, but there is hope. One day, research into understanding the roles of mutations closely associated with ALS may unlock a unifying pathological mechanism of ALS or at least the mechanism of particular forms of ALS. The final purpose of understanding these mechanisms is to find a cure or at least an effective treatment for this unrelenting disease.

Structure and Function: SOD1 in Amyotrophic Lateral Sclerosis

Despite many efforts to understand the mechanism of amyotrophic lateral sclerosis, little conclusive progress has been made. Most types of ALS are sporadic. About ten percent of ALS cases are familial, and among the familial forms of ALS, SOD1 mutations are the most common culprits [14, 21]. SOD1 a superoxide dismutase converts reactive oxygen radicals to more stable products, hydrogen peroxide and diatomic oxygen, through a series of reactions. Even though the SOD1 gene has been one of the most well studied genes in familial ALS, little is known about the pathological mechanism of the disease.

Since the superoxide dismutase is so strongly associated with ALS, thorough explorations into its function in vitro and in vivo could help unlock knowledge about the pathological mechanisms of ALS. The oxidative damage hypothesis postulates that SOD1's normal role as a sequester of oxygen radicals places it at great risk to become damaged by the radicals it seeks to stabilize [22]. Subsequent support for the oxidative damage hypothesis from in vitro studies of metal catalyzed oxidation determined that the heavy exposure of free radicals could lead to changes in amino acid residues causing changes in structure that lead to aggregation [23,24].

The oxidative damage hypothesis is currently favored because it proposes an explanation to the aggregation of SOD1, demonstrates that SOD1 is highly prone to oxidative stress, and begins to explain the implication of SOD1 in disease pathology [15]. In this section, the effects of radical damage on the superoxide dismutase and SOD1 aggregation will be discussed.

The structure of the SOD1 protein

SOD1 is a homodimer with each subunit binding to a copper and a zinc atom [25]. A hydrophobic surface holds the two subunits together [26]. SOD1 is an extremely stable protein with a melting point between 85 and 95 degress Celsius [27]. Each monomer in the SOD1 dimer is composed of two beta barrels with eight strands, two large electrostatic loops, and a metal binding domain [26]. Bulges in the beta barrel accommodate metal binding [26], and a conserved disulfide bond helps greatly stabilize the dimer [28]. The copper helps catalyze the reaction that stabilizes oxygen radicals, and this copper is bound by four histadines [26]. The zinc atoms in SOD1 dimers play a structural role by helping distribute charge in the dimer, but they do not play a catalytic role in the conversion of radicals to more stable products [29]. SOD1 homodimers are connected by a disulfide bond that is critical to the structural integrity of the dimer because it serves as rigid bridge that connects each monomer to form the dimer [26].

Catalytic mechanism of SOD1

The function of the SOD1 enzyme is to convert the highly reactive superoxide radical $O_2^{\bullet-}$ to hydrogen peroxide, H_2O_2 and diatomic oxygen O_2. This process is made possible by the catalytic transformation between copper and copper (II) ion [30]. The following reaction sums up the process:

$2H^+ + 2\text{-}O_2 + Cu^+ \rightarrow H_2O_2 + Cu^{2+}$

$O_2^{\bullet-} + Cu^{2+} \rightarrow O_2 + Cu^+$

In the first reaction, copper (I) is oxidized to produce a hydrogen peroxide and an copper (II). In the second reaction, the superoxide radical is oxidized while the copper

(II) cation is reduced to produce diatomic oxygen and a copper (I) cation. The products of this reaction are far more stable than the reactants in the second step.

The superoxide radical is highly reactive due to its strong tendency to act as a powerful reducing agent that could add electrons to reactants and thereby produce undesired products in the cell. This can cause unwanted effects in living cells but not necessarily the development of ALS.

Effects of SOD1 mutants on misfolding

There are many SOD1 mutations that cause ALS and still retain the proper enzymatic function, although sometimes impaired to different degrees, of the SOD1 enzyme [31]. Interestingly, in studies where mice completely lacked the SOD1 gene, no ALS appeared [33], though other deleterious effects were observed, but they were probably due to largely unrestrained free radical damage to the cells of those mice. The fact that no ALS symptoms appeared in SOD1 knockout mice confirms that a lack of function in SOD1 does not cause ALS. On the other hand, transgenic mice that express a human variant of SOD1 exhibit ALS symptoms [32]. It is therefore believed that SOD1 creates a gain of toxic properties rather than a loss of function. Mutants have a higher propensity to demonstrate toxic properties than the wild type or knockout SOD1.

At their native state, SOD1 mutants are nearly identical in structure and function to wild type SOD1. According to the oxidative damage hypothesis, the SOD1 enzyme, whether it is in mutant or wild type form, will seek out superoxide radicals that it intends to stabilize. After long term exposure to these radicals, both wild type and mutant SOD1 proteins will begin losing their structure; however, mutant SOD1 enzymes denature to

radical damage much more easily than the wild type SOD1 [15]. In their native state, wild type and mutant SOD1 proteins exhibit no denaturation, similar functionality, and identical structure. It seems that SOD1, either wild type or mutant, becomes toxic once SOD1 proteins begin losing their native structure.

Currently, the mechanisms that lead to the pathogenesis of ALS are not yet known. A few recent studies have suggested that the amount of SOD1 protein misfolding, which differs by mutant, seems be related to the severity of ALS symptoms [36.37].

SOD1 aggregation

There has been strong evidence suggesting that changes in the structure of SOD1 lead to the deleterious aggregation of SOD1 proteins [15, 34, 35, 36] which may be the cause of ALS. SOD1 is ubiquitously expressed in cells, but in some tissues it is found in higher concentrations than others [38]. It is clear that SOD1 mutants become unstable over time and then form aggregates, suggesting that there is an underlying mechanism that causes their denaturation and their subsequent aggregation [38]. In familial ALS where SOD1 mutants are involved, there is a clear consensus that SOD1 aggregation is one of the first signs of ALS pathogenesis, and it is therefore critical to understand the SOD1 molecular and pathogenic mechanism of SOD1 aggregation [39].

One study showed that upon thermal or chemical denaturation, SOD1 mutants tended to show high increases in the amount of surface hydrophobicity and that mutants deficient in metal content and stability tend to also demonstrate an increase in hydrophobic surface exposure [39]. This last finding suggests that the metal domains of SOD1 proteins may not only serve to provide their already known functional roles to but

also to provide overall structure stabilization in the enzyme. Also, little could be determined from the amino acid sequence and its effect on denaturation and surface hydrophobicity [39]. Most of these effects were only seen upon examining the denaturation of the overall structure of mutants in conditions of chemical or thermal stress [15, 39]. The exposure of hydrophobic surfaces precedes the aggregation of SOD1 [39]. Diverse treatments that induce the exposures of hydrophobic surfaces drive SOD1 proteins to form amyloid like aggregates, which are most likely driven by hydrophobic interactions. In addition, the wild type SOD1 was much less likely to demonstrate an increase in hydrophobic regions after various treatments in comparison to the mutants [39].

Some SOD1 mutants become more charged than others. It was found that more charged versions of SOD1 tend to aggregated less than neutral SOD1. This is probably due to charged SOD1 proteins repelling each other and neutral ones being driven by attraction of the hydrophobic portions of neutral SOD1 proteins to other neutral SOD1 proteins [36.37].

The study “Early Steps in Oxidation-Induced SOD1 Misfolding: Implications for Non-Amyloid Protein Aggregationin Familial ALS” [15] provides novel evidence of the oxidative damage and its subsequent effects on aggregate structure, dimer alterations, and the metal ligands. Most protein aggregates form an amyloid, a particular type of aggregate conformation, but SOD1 mutants form a non-amyloidal formation. It is believed that oxidative damage leads to the changes in SOD1 structure that cause aggregation [15]. Research was conducted in order to understand the cause of the non-amyloid, amorphous shape of SOD1 aggregates Researchers discovered that basal levels

of oxidative stress can lead to SOD1 misfolding and aggregation, which are strongly linked to ALS onset and progression. A key finding in this research study is that radical species affected the structure of both wild type and mutant forms, but mutants were more affected by oxidative stress. The hydrophobic areas in the dimer increased significantly, and mutants showed a greater increase in the amount of hydrophobic surfaces exposed in the oxidatively stressed mutant SOD1 proteins than the wild type. This research suggests that increase in hydrophobicity of SOD1 proteins in conditions of oxidative stress may be the cause for the atypical aggregation patterns seen in SOD1 proteins. Again, the exposure of hydrophobic surfaces in oxidatively stressed SOD1 homodimers supports the idea that the SOD1 aggregation is driven by hydrophobic effect.

Perhaps the most interesting finding in this study is that SOD1 protein misfolding occurs over a series of steps and intermediates over time. This study found that there is one intermediate that the SOD1 protein must go through in order to cause pathogenic aggregation. This intermediate is key to downstream misfolding of the SOD1 protein [15].This intermediate demonstrates a small but measureable increase in hydrophobic surface area relative to the native state. Since this intermediate is critical in downstream misfolding and kinetically stable, the researchers in this study postulate that this globular intermediate may serve as a target for drugs.

Pathogenic role of SOD1 localized in mitochondrial intermembrane space

Currently, we know that SOD1 mutations are strongly associated with the onset of ALS and that SOD1 mutations tend to lead to SOD1 aggregates, but still little is known about the pathological mechanism that causes ALS. Attempts have been made to understand the effect of SOD1 mutations and their aggregates in relation to the clinical onset of the disease.

The intermembrane space of mitochondria (IMS) a common place for the localization of SOD1 [22]. SOD1 in the IMS serves to protect the mitochondria from oxidative damage. In vitro studies have demonstrated that, in cells where mutant SOD1 accumulates in the IMS, abnormalities are seen in mitochondrial function, axonal transport, and structural differences [41, 42].

A recent study analyzed the effects of SOD1 in vivo cells [40]. Transgenic mice were created to produce human wild type and mutant G93A SOD1 to target mitochondrial intermembrane space of muscle cells and neurons. Researchers fused part of mitofilin, a protein that targets into with human SOD1 in order to create a chimeric mito-SOD1 protein that localizes in the intermembrane space. Mutant and wild type chimeric proteins were created. This mito-SOD1 protein expressed in high concentrations in the brain, spinal cord, and skeletal muscle—cells relevant in ALS. Interestingly, mito-WT SOD1 and mito-G93ASOD1 mice were expressed at similar levels in the mitochondria, but only mito-G93ASOD1 mutant mice demonstrated the ALS disease phenotype. Previous studies, suggested that the deleterious effects of SOD1 were possibly due to an increase in SOD1 proteins, but this finding suggests that the concentration of the wild type SOD1, even when comparable to the concentrations of

deleterious mutant SOD1, does not contribute to the expression of the SOD1 phenotype; however, the presence of the mutant SOD1 in the mitochondria in high levels induces the ALS phenotype. Additionally, the levels of chimeric mitochondria bound mito-G93ASOD1 and the untarget mutant G93A SOD1 were present in about the same levels.

This study demonstrates the G93A SOD1 leads to mitochondrial damage that then causes the loss of motor neurons in the spinal cord, abnormalities in muscle and brain cells and bioenergetics dysfunction [40]. Despite the fact that mitochondrial accumulation leads to the onset of SOD1 type mutants, this study suggests that there are other factors, outside the accumulation of mutant SOD1 in the mitochondria, that lead to disease pathogenesis; this is supported by the slow degeneration progress and the lack of denervation of mito-G93ASOD1 in comparison to the typical, untargeted G93A mutants which are not strictly bound to the mitochondria [40]. Although we now know that SOD1 mutants may accumulate in mitochondrial spaces and then exhibit many of the symptoms of ALS, additional efforts are clearly needed to paint a fuller picture of the pathogenesis of SOD1 induced ALS [40].

Conclusion

Despite many extensive studies, we still do not have strong experimental evidence that allows us to see a cure for sporadic and familial ALS. Recently, many studies have been published that help us to understand the role of SOD1 in disease pathophysiology. The ultimate goals are to explain ALS pathophysiology with clear mechanisms that help us thoroughly understand the role of this critical protein.

The conceptual and experimental analysis outlined above captures some of the most novel and relevant roles of SOD1 related pathogenesis: the oxidatative stress hypothesis, idea that aggregation contributes to ALS pathology, and the effect of mutants in intermembrane space on ALS pathogenesis. The newly acquired understanding of these three ideas may help develop effective therapies in the future. The oxidatative stress hypothesis and the idea that aggregation is responsible for disease pathogenesis and proliferation should probably be treated as a coupled idea. Treatments in the future can search for ways to stabilize SOD1 proteins in order to avoid the deleterious denaturation and the subsequent aggregation of SOD1.

Since recent studies have implicated high SOD1 concentrations in the mitochondria as pathogenic in the development of ALS, a possible therapy strategy would likely focus on removing SOD1from the mitochondria or inhibiting SOD1 entrance into the mitochondria.

Though these last few ideas seem to hold great promise for the understanding of at least one form of ALS, familial SOD1 ALS, even more research is needed to understand sporadic and familial ALS not related directly to SOD1. Even though SOD1 research mainly focuses on one particular type of ALS, the hope is that understanding the SOD1 mechanisms may open up pathways of research that allow us to understand and then eventually treat other additional forms of ALS.

Much work remains, but current ALS research seems to be making headway into the mechanistic foundations of this crippling disease. Hopefully, the recent advances into understanding this disease are only the beginning of a new, productive era of ALS research.

Clinical case study

Chief complaint

James came to clinic because, lately, he laughs and gestures uncontrollably for no apparent reason.

History of present illness [2-6, 8-9, 43-46]

James is an active thirty-eight year old male. He enjoys spending his free time playing sports, especially basketball. A few months ago, he was elbowed during a game of basketball. Although he did not lose consciousness, he said the blow was so hard that he was on the brink of passing out. He was considering going to the hospital but assumed his symptoms would go away. Weeks after the incident, James felt dizzy at times. He also became forgetful and says he did not feel like he could concentrate on tasks as well as before. He also began feeling a bit clumsier than usual.

Despite all of his complications following the blow, he did not seek out medical attention because he assumed that his symptoms would simply leave on their own. Although his dizziness subsided and although he can concentrate better now, he feels uncoordinated, especially with his hands. He has been dropping many objects, such as cups and toothbrushes, from his hands; he sometimes stumbles when he walks; and for about six months, he has been making strange facial gestures without intention.

James notices that he is not very coordinated when he plays sports anymore. He shrugged off his decrease in sports finesse as an effect of old age. He became concerned after his loss of coordination began to affect him in his everyday life. He was also troubled but a subtle but definite loss in grip strength, causing him to drop objects quite often. Expanding on his clumsiness, James says that nearly a year ago he noticed that he

felt weaker and clumsier than usual when he tried to sprint. He felt that his right leg was weaker than usual, and then weeks later his left leg felt weaker too. Initially, his weakness was very mild, so he did not think much of it. Now he is at a point where cannot run nearly as fast as he did a year ago. He attributed his decrease in speed to his old age and lack of proper conditioning, but now he is worried because he has difficulty jogging and sometimes walking too.

Late last night, his wife became very worried because she says that he laughed, smiled, frowned, and raised his eyebrows incessantly for a few minutes. Thinking that he was joking, she told him to stop doing that, but James expressed his lack of control over these gestures. His wife says that this has been occurring for a few months, but the frequency and length of James's episodes has steadily increased. He admitted that coworkers have also mentioned that he frowns and smiles when there is no reason to do either. He is quite embarrassed that others around him have noticed that he gestures when he probably should not. James says that this was never a problem until a few months ago. When his wife stepped out of the room, he admitted that he has been stressing a lot over his recent clumsiness and that he is extremely worried about his uncontrollable gestures.

He experiences no pain. Apart from engaging in physical activities (although he is not as active as he used to be), he enjoys reading books and spending time with his family. He works as an accountant at a firm, and he says that he has been stressed out for the last few months at his job. Apart from the busy days at his job, he generally enjoys his life.

Past Medical History

As a young child, James suffered from chicken pox. The chicken pox cleared away. About ten years ago, James has hospitalized after he broke his tibia and fibula during a soccer match. He was hospitalized, and he recovered with no complications. He has suffered minor concussions from playing basketball and soccer, but he has never sought medical care for them. His recent blow to the head was the most severe hit to the head that he has received while playing a sport.

He takes one multivitamin a day. He does not take any other medications. He does not have any allergies. All of his immunizations are up to date. He has never had any blood transfusions.

Family History

His mother and father suffer from high blood pressure. His mother has diabetes. Family members on his mother's side have suffered from Huntingdon's disease. His mother has no symptoms. His immediate family members have no other medical problems.

Social/Sexual History

James leads a healthy social life. He spends his free time with his wife, his friends, or exercising. He enjoys a monogamous relationship with his wife. He does not smoke cigarettes. He does not use drugs for recreational purposes. Occasionally, he drinks alcohol with his friends, but he rarely exceeds three or four drinks.

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ROS/Physical Exam [43-44]

General: The patient seems well nourished and exhibits no deformities.

Head, ears, eyes, nose, and throat: No obvious deformities to the head. Normal tympanic membrane. Pupils react to light normally. Mucous membranes are dry. No palpable lymph nodes.

Head, neck, and eyes: His head, scalp, neck, and thyroid were inspected and palpated. The carotids were auscultated. The pupils were checked to respond to light. No remarkable findings.

Ears, nose, and throat: His ears, nostrils, and mouth were inspected. No remarkable findings.

Pulmonary: The patient's lung expansion, percussion, and lung sound were checked. No remarkable findings.

Heart and blood vessels. His precordium, wrist arterial pulses, dorsalis pedis pulses were inspected and palpated. Capillary refill in fingers was also assessed. His heart was auscultated and blood pressure measured (128/84). No remarkable findings.

Abdomen: His bowed sounds were checked. His abdomen was inspected for rigidity, tenderness, and distension. No remarkable findings.

Musculoskeletal: Bilateral muscle weakness was seen in the patient's hands. Muscle weakness was detected in the patient's hands, a 4+ on the standard scale of 0-5. Both of the patient's feet exhibited difficulty with dorsiflexion. The patient was asked to walk. He demonstrated a subtle but noticeable steppage gait.

Neurological exam [44-46]

Patient is oriented and mentally alert. No test was conducted for CN 1 and the parasympathetic stimulation of CN 10. In the CN 7 test, the patient exhibited slightly asymmetric nasolabial folding. A comparison with his two year old driver's license picture shows no pronounced change. Otherwise CN 2-12 were unremarkable. He demonstrated a positive Babinski sign. During the examination, the patient exhibited pseudobulbar affect when he laughed and gestured without provocation for a moment. Additionally, his sensory function, bowel, and bladder control were completely normal. The patient has a solid grasp of time and place. He was given a Mini Mental Status Exam. He exhibited no deficits in cognition or memory. He also seemed very coherent during the entire examination.

Lab Results

*** indicates abnormal value

Complete blood count

- WBC—7.5K cells/mcL
- RBC—5.2M cells/mcL
- Platelets—303 billion/L
- Hemoglobin— 14.3 grams/dL
- Hematocrit—43%

Complete metabolic panel

- Albumin—4.5 grams/dL
- Alkaline Phosphatase—103 units/L
- Alanine amino transferase—27 units/L
- Aspartate amino transferase—22 units/L
- Bilirubin—0.9 mg/dL
- Calcium—9.8 mg/dL
- Carbon dioxide—25 mmol/L
- Chloride—104 mmol/L
- Creatinine—1.05 mg/dL
- Glucose—0.88 mg/dL
- Potassium—4.4 mmol/L
- Protein, total—7.5 g/dL
- Sodium—142 mmol/L
- Urea nitrogen—14 mg/dL

Additional blood tests

- ***Creatine kinase—504 units/L
- Thyroid stimulating hormone—2.5 mU/mL

Radiology

- Conventional MRI of brain, cervical spine, and thoracic spine: Normal

Differential diagnosis and assessment [43-44]

From the patient's history and exam, a differential diagnosis containing the following diseases can be made: Kennedy's disease, cervical radiculomyelopathy, progressive bulbar palsy, primary lateral sclerosis, hyperthyroidism, and amyotrophic lateral sclerosis. Kennedy's disease impairs both the spinal and bulbar lower motor neurons, however, upper motor neurons are spared; the patient's upper motor neurons are also affected; Kennedy's disease must be discarded. Cervical radiculomyelopathy can impair function of both the upper and lower motor neurons, but it also typically creates sensory abnormalities, sphincter dysfunction, and leads to an abnormal cervical MRI; the last three characteristics are inconsistent with the patient. Progressive bulbar palsy and primary lateral sclerosis only affect the lower and upper motor neurons, respectively; both must be discarded since the patient's upper and lower motor neurons are affected. Hyperthyroidism may cause signs that appear to be upper motor neuron damage and peripheral neuropathy, which may create symptoms that mimic our patient's symptoms; the TSH test can rule this out. The evidence obtained from our patient's history, in clinic exam, and lab results overwhelmingly suggests amyotrophic lateral sclerosis.

The patient demonstrates pseudobulbar affect, a positive Babinski reflex, hand weakness, footdrop leading to steppage gait, and possible asymmetric nasolabial folding. Pseudobulbar affect and a positive Babinski reflex are typical of upper motor neuron damage, and they affect two distinct regions of the body—the cranial and lumbosacral regions respectively. Hand weakness, footdrop leading to steppage gait, and nasolabial folding are typical of lower motor neuron damage, and they affect three distinct regions of the body—the cervical, lumbosacral, and cranial regions, respectively.

For definitive ALS diagnosis, upper and lower motor neuron damage must appear in at least three regions. The patient fits all the criteria and can, therefore, be diagnosed with amyotrophic lateral sclerosis.

Treatment plan:

James is to be prescribed Riluzole, a glutamate agonist [7], in hope of increasing the his survival time. James is asked to come every three months to check up on the progression of his ALS symptoms. James and his family are advised to seek counseling to help cope with the psychological stress experienced with the disease diagnosis and progression. Although nothing can be done to stop the progression of the disease, we hope that monitoring James closely can allow for timely treatments to ameliorate some of his symptoms [8].

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