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Hereditary Disorders and Human Mutations of Iron-Sulfur Assembly Genes

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Abstract

Multiple mitochondrial dysfunctions syndrome (MMDS) is a group of autosomal recessive mitochondrial disorders that is associated with deficiencies related to nuclear genes: *ISCA2*, *ISCA1*, *NFU1*, *IBA57*, and *BOLA3*. The syndromes are relatively new and recently discovered. Individuals with MMDS have reduced function of energy production stages in mitochondria. The dysfunctions are mostly related to iron-sulfur (Fe-S) clustering system (ISC) and its biogenesis. The signs and symptoms of the patients may begin early in life, and can be quite severe leading to death more or less during infancy. Affected individuals have various symptoms including brain dysfunction (encephalopathy), hypotonia, seizures, delayed developmental milestones, and cognition and psychomotor impairments. These individuals often have difficulty growing and gaining weight at the expected rate. Diagnosis of the disease can be challenging as in the case with most of the mitochondrial disorders. However, since the genetic causes of the MMDS are known, a laboratory test focusing on the causative genes will be helpful to determine the pathogenic mutations. This in turn would facilitate reducing the number of the diseases through carrier testing and genetic counseling and utilization of preimplantation genetic diagnosis in populations, especially those that display high rate of consanguinity, which are prone to have such autosomal recessive disorders.

Keywords: *BOLA3*, *IBA57*, *ISCA1*, *ISCA2*, *NFU1*, iron-sulfur (Fe-S) cluster (ISC), multiple mitochondrial dysfunction syndromes

1. Introduction

Mitochondria are double membrane-bound cellular organelles surrounded by outer and inner membranes [1, 2]. The organelle is considered cell's powerhouse generating adenosine

triphosphate (ATP) during cellular respiration; hence, facilitating energy conversion in eukaryotes. Uniquely, each mitochondrion has its own DNA and encodes mitochondrial genes; hence, contributing the cell's proteome independently. The inheritance of the mitochondrial genome differs from nuclear genome since the donor of mitochondrial DNA (mtDNA) is the egg rather than sperm whose mitochondria are marked for obliteration upon entering the egg [3]. Hence, the organelle's DNA is inherited through females known as "maternal inheritance." Since these organelles generate energy, most biochemical reactions in the eukaryotic cells occur in the mitochondria. These reactions include pyruvate oxidation, citric acid cycle, electron transport, and oxidative phosphorylation (OXPHOS) all needed for energy production. Mitochondria also have an important role in calcium signaling, regulation of cellular metabolism, heme synthesis, steroid synthesis, apoptosis, and the biosynthesis of iron-sulfur (IS) clusters (ISC). The high number of human diseases caused by the malfunction of the mitochondrial proteins—encoded by nuclear or mtDNA—drew attention to the importance of this organelle.

2. Mitochondria

Mitochondria are genetically controlled by both nuclear DNA and the mitochondrial genome [1, 4]. A wide range of molecular defects have been identified in the human mitochondrial genome [4–9]. Diseases due to mutations in the mitochondrial genome are clinically, genetically, and biochemically diverse [1, 2, 4, 6, 10]. Similarly, deficiencies in mitochondrial genes encoded by nuclear genome can also lead various mitochondrial disorders and a wide range of cellular perturbations such as undue reactive oxygen species and distracted apoptosis, aberrant calcium homeostasis, and deficient energy production. This in turn leads failure to meet the requirements of numerous organs, especially those with high energy needs. Hence, various pathological conditions appears due to impaired mitochondrial function in human body involving different cell types, tissues, and organs including heart and brain. Such multi-organ manifestations are all mitochondria related and these diseases varies from epilepsy to cardiac myopathies.

3. Mitochondria and genetics of mitochondria-related diseases

The mitochondrial genome is a multicopy, double-stranded circular DNA molecule, which is 16.6 kb in human [11]. This genome encodes 13 essential proteins for the OXPHOS system and 24 components of the RNA machinery: 2 rRNAs and 22 tRNAs [11]. It is intronless and the only noncoding region is the displacement region (D-Loop), a region of 1.1 kb. It contains both the replication origins and the transcriptional promoters. Although mitochondria are genetically controlled by both mitochondrial and nuclear genomes, mtDNA is only maternally inherited [3]. Mitochondrial genetics differ greatly from Mendelian genetics in size, number of encoded genes, number of DNA molecules per cell, lack of introns, gene density, replication, transcription, recombination, and mode of inheritance. The 13 proteins include 7 subunits of NADH

Dehydrogenase (complex I: ND1, ND2, ND3, ND4, ND4L, ND5 and ND6), Cytochrome b (subunit of complex III), 3 subunits of Cytochrome c oxidase or complex IV (COI, COII and COIII), and 2 subunits of F₀F₁ ATPase (ATPase 6 and ATPase 8). They are all encoded by mtDNA and synthesized in the organelle. While, complex II (Succinate Dehydrogenase) and the remaining subunits of complexes I, III, IV, and V are entirely encoded by the nuclear genome. These nuclear-encoded proteins are synthesized on cytosolic ribosomes and subsequently transported into the mitochondria.

4. Fe-S clusters (ISCs)

ISCs are evolutionarily ancient cofactors consisting of Fe (iron) and S (sulfur) associated to the cysteine sulfurs of proteins. The clusters are found in variety of organisms including archaea, protists, prokaryotes, and eukaryotes. In a eukaryotic cell, they can be found in the mitochondria, cytosol, and nucleus where they perform diverse functions [12]. ISCs play a critical role in many fundamental molecular processes and have roles in electron transfer, structural stabilization, gene regulation, enzymatic catalysis, metabolic regulation, and sensing environmental signals [13]. Almost 30 proteins in the mitochondria and the cytosol are involved in synthesizing and assembling these clusters. ISC have two most common forms [2Fe-2S] and [4Fe-4S] clusters. ISC-related proteins of the electron transport chain in the mitochondrion are mainly located in the inner membrane. Moreover, some of these proteins are also found in the mitochondrial matrix in the organelles. For the cluster assembly, two machineries are required, the mitochondrial ISC assembly machinery and the cytosolic IS protein assembly machinery [12].

Eukaryotic IS proteins are located in mitochondria, cytosol, and nucleus, where they perform diverse functions in cellular metabolism and regulation. The mitochondrial ISC assembly machinery matures all organellar IS proteins, and additionally contributes to the biogenesis of cytosolic and nuclear IS proteins by producing an unknown sulfur-containing compound (X-S) that is exported to the cytosol and used by the cytosolic IS protein assembly machinery. Hence, mitochondria are directly responsible for the essential functions (e.g., of nuclear IS proteins involved in DNA metabolism and genome maintenance).

Mitochondria forms iron-sulfur clusters of significant proteins such as DNA polymerase and DNA helicases, and, therefore, plays a significant role in survival. There are 17 different proteins forming iron-sulfur cluster machinery that places the clusters into the Apo proteins. The mechanism of formation of iron-sulfur clusters can be divided into three steps. First, it is synthesized on a scaffold protein. Second, it is bound to transfer protein after dislocation from scaffold protein. Third, the transfer protein, the cluster and the specific ISC targeting factor place the cluster into the Apo protein. The changes in the first two steps inhibit the maturation of extra mitochondrial Fe/S proteins and disturb the iron homeostasis [14]. Assembly of Fe-S cluster also takes place by NIF, SUF, and CIA machineries. Cysteine desulfurase is an enzyme that unites Fe-S assembly machineries. It is encoded by NFS 1 which functions to deliver sulfur to ISCU [15]. ISCU is an iron-sulfur cluster assembly enzyme; encodes component of

iron-sulfur scaffold protein. The changes in this gene result in severe myopathy and lactic acidosis (“ISCU Fe-S Cluster Assembly Enzyme [*Homo sapiens* (Human)] - Gene - NCBI”) Complexes 1, 2, and 3 contain Fe-S clusters. They function in electron transport by transfer of one electron in redox processes [16]. The assembly of the clusters is recently studied in Yeast. In photosynthetic organisms, the iron-sulfur clusters play role in chloroplast processes and are important for plastid functioning [17].

Yeast frataxin, *Isu1*, and *Nfs1* (cysteine desulfurase) take part in *de novo* synthesis of ISC. Many genes encode ISC assembly factors such as *BOLA3*, *NFU1*, *GLRX5*, *NUBPL*, *LYRM4*, *IBA57*, *ISCA1*, and *ISCA2*. These molecules have significant role in mitochondria. They are essential cofactors in the assembly of cluster. Deficiency of these genes leads to different diseases, for instance, *GLRX5* deficiency causes sideroblastic anemia, whereas *NUBPL* mutations lead to respiratory chain complex 1 deficiency. On the other hand, some of these deficiencies are classified under a unique category such as MMDS.

5. Genetic factors of mitochondrial dysfunction syndromes

As the names imply, multiple mitochondrial dysfunction syndromes are disease conditions affecting mitochondria and usually lead to reduced function of more than one stages of energy production in the organelle [18]. The genetic factors causing these disorders are associated with the biogenesis of cellular ISC and currently these are the following genes: *ISCA2*, *NFU1*, *IBA57*, and *BOLA3*. More recently, *ISCA1* is also reported to lead a disease resembling MMDS and suggested to be a member of the group [19]. Interestingly, MMDS members appear to be inherited in autosomal recessive mode of inheritance (**Table 1**).

5.1. ISCA1

Iron-sulfur cluster assembly 1 (*ISCA1*) is one of the mitochondrial proteins required for the biogenesis and assembly of ISC [20]. This protein functions in the late stages of the ISC biogenesis and act as an iron binding molecule that may serve as a chaperone for biogenesis of Fe-S clusters [21]. It is believed that the molecule plays its pivotal role through its interaction with IOP1 (iron-only hydrogenase-like protein)/NARFL (nuclear prelamin A recognition factor-like). Knockdown of *Isca1* causes reduced activity of succinate dehydrogenase, mitochondrial aconitase, and cytosolic aconitase; hence, involving in both cytosolic and mitochondrial Fe-S protein biogenesis [22].

According to GenAtlas [23, 24], the gene has four exons and produces 14 kDa protein with 129 amino acids, which is known as mitochondrial Fe-S cluster assembly 1 homolog or otherwise HESB like domain containing 2. The gene is mapped to chromosome 9q21.33, and sits on genomic coordinates: 88.879.463–88.897.490. It is 2012 base pair long, generates four transcripts (splice variants) and highly expressed heart, esophagus, bladder, uterus, and cervix. Moreover, *ISCA1* is a member of consensus coding sequence (CCDS:35056.1) which are manually checked protein annotations on the reference mouse and human genomes that ensures

Gene	Cytoband	NCBI	Genomic location	MMDS-related phenotype	MIM PT#	IM	MIM LN
NFU1	2p13.3	27247	2:69,396,112-69,438,122	Multiple mitochondrial dysfunctions syndrome 1	605711	AR	608100
BOLA3	2p13.1	388962	2:74,135,400-74,147,911	Multiple mitochondrial dysfunctions syndrome 2 with hyperglycinemia	614299	AR	613183
IBA57	1q42.13	200205	1:228,165,807-228,182,256	Multiple mitochondrial dysfunctions syndrome 3	615330	AR	615316
ISCA2	14q24.3	122961	14:74,493,719-74,495,567	Multiple mitochondrial dysfunctions syndrome 4	616370	AR	615317
ISCA1	9q21.33	81689	9:86,264,545-86,282,574	Multiple mitochondrial dysfunctions syndrome 5	617613	AR	611006

AR: autosomal recessive; IM: inheritance mode; LN: locus number; MIM: Mendelian inheritance in man; MMDS: mitochondrial dysfunction syndromes; PT#: phenotype number.

Table 1. Genes and related mitochondrial dysfunction syndromes.

consistent representation of the tracks of NCBI, Ensembl, and UCSC Genome Browsers. The gene has several synonyms such as *hIsca*, *HBLD2*, and *ISA1*, and localizes to mitochondria as well as cytoplasm.

Effect of depletion of ISC-related proteins on the maturation of cytosolic 4Fe-4S proteins showed that some mitochondrial Fe/S proteins such as mitochondrial aconitase, SDH, several proteins of complex I, and Rieske Fe/S protein were decreased with the deficiency of *ISCA1*. On the other hand, cellular heme content and mitochondrial 2Fe-2S ferredoxin were unaffected by the depletion. This implies that *ISCA1* is crucial in the maturation of mitochondrial 4Fe-4S proteins [25]. In another study, *ISCA1* was found to be associated with multiple mitochondrial dysfunctions syndrome-5. A homozygous missense mutation at a conserved residue in the Fe-S biogenesis domain (c.259G>A, p.Glu87Lys) was identified in two unrelated Indian families. This mutation destabilizes the protein subsequently causing the syndrome [19].

5.2. ISCA2

ISCA2 stands for iron-sulfur cluster assembly 2 protein and the gene encodes for A-type iron-sulfur cluster protein. Fe-S clusters are inorganic cofactors, mostly found in metalloproteins. The gene is located on chromosome 14 and expressed from the plus strand. According to Ensembl, this gene generates 4 different transcripts and has 96 orthologues. *ISCA2* is a regulatory protein found in mitochondria as well as extra mitochondrial sites such as cytosol and nucleus. The protein takes part in assembly of Fe-S clusters in mitochondria which further take part in oxidation reduction (especially in complex 1 and 2), substrate activation, iron/sulfur storage, regulation of gene expression, and enzyme activity. Alternative name for *ISCA2* is "HESB-like-domain-containing protein 1" for humans. First human mutation of *ISCA2*

(c.229G>A; p.Glu77Ser) identified in the patients from five consanguineous families was a homozygous ancestral founder mutation that led to neurodegeneration, developmental failure to thrive, quadriplegia, truncal hypotonia, optic atrophy, and leukoencephalopathy [26]. Later, additional 10 cases with the same founder mutation were also described [27]. Recently, two other patients with the same mutation were also studied with detailed functional experiments revealing complex 2 and 4 deficiencies [28]. Interestingly these patients were all Arab descent. Most recently, a second mutation, a compound heterozygous variant (a single basepair deletion causing frameshift with a premature stop codon: mutation: c.295delT; p.Phe99Leufs*18 and a missense mutation c.334A>G; p.Ser112Gly) in *ISCA2* was reported in a 2-month-old girl from Italy [29]. These mutations cause disorder of energy metabolism which results in respiratory failure, severe hypotonia, nystagmus, lactic acidosis, poor neurologic development, hyperglycemia, leukodystrophy of the brainstem with longitudinally extensive spinal cord involvement, and mtDNA deficiency ultimately leading to death [26].

5.3. NFU1

NFU1 is one of the human mitochondrial components that is involved in the assembly of the Fe-S protein cluster. It helps in the transfer of [4Fe-4S] clusters to specific protein targets and facilitates their maturation [30]. *NFU1* is mapped on the 2p13-p15 chromosomal region and codes for the NFU1 protein. During Fe-S assembly, two NFU1 monomers are needed to assemble one 4Fe-4S. Complex I, II, and III of oxidative phosphorylation have multiple Fe-S clusters. Therefore, any deficiency in these clusters causes dysfunctions of respiratory chain complexes [31]. Previous studies showed that the function of the NFU1 has been associated by the fatal mitochondrial disease, multiple mitochondrial dysfunctions syndrome 1 (MMDS1) [30]. Patients with NFU1 mutations usually manifest feeding difficulty, weakness, lethargy, and decreasing responsiveness within a few days after birth and a few had epileptic seizures [31]. It has been shown that the patients with mutations in the NFU1 gene have similar biochemical features to that seen in patients with lipoic acid defects. Thus, NFU1 mutation appears to have some effect on Fe-S enzyme lipoic acid synthase (LAS). In conclusion, NFU1 is an ISC assembly protein, and there is strong evidence that LAS deficiency is important in NFU1 mutation-related disease [31].

5.4. IBA57

IBA57 is a member of the Fe-S cluster assembly group. It is known as putative transferase CAF17 and Fe-S cluster assembly factor homolog. *IBA57* is located on 1q42.13 and codes for the IBA57 protein that is located in the mitochondrion. This protein functions in the late stages of the biosynthesis of mitochondrial 4Fe-4S proteins. Any deficiency in *IBA57* can cause an autosomal recessive spastic paraplegia-74 or multiple mitochondrial dysfunctions syndrome 3. In a previous study, it was found that the depletion of IBA57 in cell culture caused striking alterations in mitochondrial morphology, including a vast enlargement of the organelles and a loss of cristae membranes. It is also found that the function of IBA57 protein is conserved from bacteria to human, according to a study that provides an evidence for the requirement of bacterial and yeast relatives of human IBA57 for efficient maturation

of [4Fe-4S] proteins. Moreover, potential diseases caused by mutations in these genes are expected to cause defects in mitochondrial respiration and in lipoic acid-dependent proteins [25]. Another study reported two siblings from consanguineous parents died with a condition characterized by generalized hypotonia, respiratory insufficiency, arthrogryposis, microcephaly, congenital brain malformations, and hyperglycinemia. Catalytic activities of the mitochondrial respiratory complexes I and II were deficient in skeletal muscle, a finding suggestive of an inborn error in mitochondrial biogenesis. Homozygosity mapping identified IBA57 located in the largest homozygous region on chromosome 1 as a culprit candidate gene. Their analysis of IBA57 revealed the homozygous mutation c.941A>C, p.Gln314Pro in those two patients [15].

5.5. BOLA3

BOLA3 is another essential protein in the Fe-S clusters production and involves in the normal maturation of lipoate-containing 2-oxoacid dehydrogenases. Another critical role of the molecule is to facilitate the assembly of the respiratory chain complexes. *BOLA3* was identified in the year 2008 during a search for similar sequences for bacterial BolA and cloned together with *BOL1* and *BOL2* [32]. According to Ensembl, the gene has five different transcripts and two isoforms. The main isoform is longer and localizes to mitochondria while the shorter isoform lacking exon 2 is restrained in the cytoplasm [18, 33]. The main transcript (ENST00000327428.9) has four exons comprising 68 variations [33]. The mRNA is nearly ubiquitously expressed in human tissues. The protein has seven domains including two low complexity segments. The main BolA domain consists of a helix-turn-helix structure close to its C terminus. The gene has three published mutations (c.123dupA; p.Glu42Argfs; c.200T>A, p.Ile67Asn; c.136C>T, p.Arg46Ter) [18, 34–36] in addition to a 5 bp deletion [37] and a single basepair insertion [18]. These mutations were identified in ethnically different families. The first patients were initially described in 2001 in a mapping study [38] that included a singleton from a consanguineous family as well as three siblings from a nonconsanguineous family. Since all the patients from two different families had similar metabolic abnormalities, a mapping strategy was employed to identify the genetic interval for the causative gene. This approach located the gene on chromosome 2. Further positional cloning studies on the subjects yielded a single significant interval on p arm extending ~5 centiMorgan region and excluding the region positioned on the q arm. Interestingly, both of these families were utilized in a follow-up study that yielded deficiencies of two ISC-related genes in each family. While the larger family with three siblings were identified to harbor splice site mutation in *NFU1* (c.545G>A) [18], the singleton had a single nucleotide duplication leading to a frame shift and eventually a premature stop codon (p.Glu42Argfs*13) in *BOL3* [18]. Later on, a few more follow-up studies revealed additional mutations in the gene. The first follow-up study focused on two patients (male and female) with quite similar clinical course appeared with hypotonia, severe neonatal lactic acidosis, and intractable cardiomyopathy [35]. A missense mutation (c.200T>A, p.Ile67Asn) was identified in the patients' DNA using exome sequencing. The other studies [34, 37] provided two additional missense mutations in *BOLA3*. Interestingly, while c.287A>G (p.His96Arg) causes a lethal infantile mitochondrial disorder [37], c.136C>T (p.Arg46*), a severe truncation mutation, leads to nonketotic hyperglycinemia [34] in the affected individuals.

Table 2 consists of previously published mutations in some Fe-S cluster genes.

Gene	Mutation type	Mutation	Disease and phenotype	References
<i>BOLA3</i>	Missense	c.200T>A; p.Ile67Asn	Multiple mitochondrial dysfunctions syndrome	Haack et al. [35]
<i>BOLA3</i>	Missense	c.287A>G p.His96Arg	Lethal infantile mitochondrial disorder	Kohda et al. [37]
<i>BOLA3</i>	Nonsense	c.136C>T; p.Arg46*	Nonketotic hyperglycinemia,	Baker et al. [34]
<i>BOLA3</i>	Microdeletion	c.225_229delGAGAA; p. Lys75*	Lethal infantile mitochondrial disorder	Kohda et al. [37]
<i>BOLA3</i>	Microduplication	c.123dupA	Combined respiratory chain and 2-oxoacid dehydrogenase deficiency	Cameron et al. [18]
<i>IBA57</i>	Missense	c.313C>T; p.Arg105Trp	Leukodystrophy with acute psychomotor regression	Torraco et al. [39]
<i>IBA57</i>	Missense	c.316A>G; p.Thr106Ala	Leukodystrophy with acute psychomotor regression	Torraco et al. [39]
<i>IBA57</i>	Missense	c.436C>T; p.Arg146Thr	Leukodystrophy, fatal infantile	Debray et al. [40]
<i>IBA57</i>	Missense	c.586T>G; p.Trp196Gly	Leukodystrophy, developmental delay, feeding problems and recurrent vomiting	Torraco et al. [39]
<i>IBA57</i>	Missense	c.686C>T; p.Pro229Leu	Leukodystrophy, developmental delay, feeding problems and recurrent vomiting	Torraco et al. [39]
<i>IBA57</i>	Missense	c.706C>T; p.Pro236Ser	Leukodystrophy with acute psychomotor regression and feeding difficulties	Torraco et al. [39]
<i>IBA57</i>	Missense	c.757G>C; p.Val253Leu	Leukodystrophy with acute psychomotor regression	Torraco et al. [39]
<i>IBA57</i>	Missense	c.941A>C; p.Gln314Pro	Myopathy and encephalopathy	Ajit Bolar et al. [15]
<i>IBA57</i>	Splice	IVS2 ds A-G-2; c.678A>G	Spastic paraplegia	Lossos et al. [41]
<i>IBA57</i>	Small insertion	c.87_88ins11	Leukodystrophy with acute psychomotor regression	Torraco et al. [39]
<i>NFU1</i>	Missense	c.62G>C; p.Arg21Pro	NFU1 deficiency	Ahting et al. [42]
<i>NFU1</i>	Missense	c.544C>T; p.Arg182Trp	NFU1 deficiency	Ahting et al. [42]
<i>NFU1</i>	Missense	c.565G>A; p.Gly189Arg	Leukoencephalopathy with cysts and hyperglycinaemia	Nizon et al. [43–45]

Gene	Mutation type	Mutation	Disease and phenotype	References
<i>NFU1</i>	Missense	c.568G>A; p.Gly190Arg	NFU1 deficiency	Ahting et al. [42]
<i>NFU1</i>	Missense	c.572A>T; p.Asp191Val	Multiple mitochondrial dysfunctions syndrome	Bai et al. [46]
<i>NFU1</i>	Missense	c.622G>T; p.Gly208Cys	Fatal infantile encephalopathy and/or pulmonary hypertension	Navarro-Sastre et al. [47]
<i>NFU1</i>	Missense	c.629G>T; p.Cys210Phe	Leukoencephalopathy	Invernizzi et al. [44]
<i>NFU1</i>	Splice	c.302+3A>G; p.Val56Glyfs*	NFU1 deficiency	Ahting et al. [42]
<i>NFU1</i>	Splice	c.545G>A; Skipping exon 6	Deficiency of the 2-oxoacid dehydrogenases accompanied by respiratory chain defects	Cameron et al. [18]
<i>NFU1</i>	Splice	c.545+5G>A; Skipping exon 6	Fatal infantile encephalopathy, pulmonary hypertension	Navarro-Sastre et al. [47]
<i>NFU1</i>	Microdeletion	c.90delC; p. Tyr30*	Multiple mitochondrial dysfunctions syndrome	Bai et al. [46]
<i>NFU1</i>	Microdeletion	c.146delC; p.Pro49LeufsX8	Spastic paraplegia	Tonduti et al. [45]
<i>NFU1</i>	Large deletion	55.6 kb region covering exons: 4–8	NFU1 deficiency	Ahting et al. [42]
<i>ISCA1</i>	Missense	c.259G>A; p. Glu87Lys	Multiple mitochondrial dysfunctions syndrome	Shukla et al. [19]
<i>ISCA2</i>	Missense	c.229G>A; p.Glu77Ser	Multiple mitochondrial dysfunctions syndrome	Al-Hassnan et al. [26] and others [27, 28]
<i>ISCA2</i>	Deletion and Missense	c.295delT and c.334A>G; p.Phe99Leufs*18 and p.Ser112Gly	Multiple mitochondrial dysfunctions syndrome	Toldo et al. [29]

Table 2. Published mutations in some Fe-S cluster genes.

6. Conclusion

Iron-sulfur clusters are indispensable inorganic cofactors for biological function and involve in numerous cellular processes such as respiration and DNA repair. The cluster's assembly is complex and requires sophisticated protein machinery for its maturation and insertion into apoproteins. Since mitochondria is the main site for ISC biogenesis in human, any defect disturbing the biogenesis leads to a pathological outcome mostly appears as an mitochondrial entity in human. Currently, genetic alterations in several genes involving in ISC assembly and maturation have been linked to autosomal recessive mitochondrial human diseases known as multiple mitochondrial dysfunction syndromes. It is expected that more genes and alterations

will appear in the literature related to ISC pathways. Moreover, there is still need to fully elucidate the phenotypic consequences of these genetic alterations and alteration of ISC pathways during the ISC related pathogenesis in human.

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