LIBERTY UNIVERSITY

NEXUS: The Liberty Journal of Interdisciplinary Studies

Volume 1
Issue 2 *Spring 2024*

Article 1

June 2024

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Myotonic dystrophy is an autosomal dominant disorder that can be most easily recognized by myotonia, progressive muscular weakness, cataracts, frontal balding and atrophy.[1] Myotonia itself, the condition from which Myotonic dystrophy got its name, is a neuromuscular condition where the muscles are unable to relax.[2] Individuals with this condition are often left unable to release their grip on objects, and as a result experience a lower quality of life. Myotonic dystrophy has been separated into types 1 and 2 based largely on the gene affected. The focus of this paper, myotonic dystrophy type 1 (DM1), results from a mutation in the DMPK gene which is responsible for the production of the protein dystrophia myotonica protein kinase.[3] Dystrophia myotonica protein kinase has been found to be important to the proper function of cells in the brain, heart, and muscle, and has been theorized to play a role in intracell communication.[1] The DMPK gene is found on the 19th chromosome at the q13.32 band, and is relatively small with only 12,782 nucleotides and 15 exons.[4] DMPK can be observed in detail on the annotated gene map provided in **Figure 3**. The processed mRNA from the DMPK gene spans 2784bp.[5] In order to better understand the mutations that often result in such severe symptoms, it is important to first explain the details of the DMPK gene and its resulting protein.

Normal Biology

As the field of modern genetics is still relatively new, dystrophia myotonica protein kinase, a serine/threonine kinase that if mutated can cause DM1, remains a poorly understood protein in terms of function.[6] Many forms of DMPK exist, and the longer ones are known to be associated with the mitochondria, although their role there remains unclear. The DMPK protein is known officially as DM1 protein kinase, and is made up of 629 amino acids.[7] The only isoform of DMPK to be studied in depth, isoform A, is known to be integral to the function of the outer mitochondrial membrane.[6] The protein's kinase activity helps to guard the cell from oxidative stress, and therefore prevents the opening of the mitochondria's permeability transition

pore which causes irrevocable cell damage and death.[6] Additionally, it has been found that lowering DMPK gene expression levels in vitro hinders the appropriate appearance of muscle markers in myogenesis.[6] The research on this indicates that DMPK most likely forms a multimolecular complex with hexokinase (HK II), and tyrosine kinase (SRC), that is an antioxidant responsible for functions relating to the development and differentiation of muscle fibers.[6] One important feature of the DMPK gene that should be noted is the CTG trinucleotide repeat sequence seen in the 3' untranslated region. This sequence can be seen in further detail in **Figure 3** (highlighted grey), and is 5-38 copies long in the unmutated gene.[7] Research has shown that, within the normal cell, the two DMPK genes both undergo transcription, mRNA processing in the nucleus, and are successfully transported to the cytoplasm for translation.[8] Once in the cytoplasm, they bind with a unique RNA binding protein, CUG-BP, before being translated into the DM1 protein kinase enzyme.[8] This CUG-BP binding process can be seen in contrast with the mutated DMPK protein in **Figure 1**. Some articles claim a link has been made between DMPK and the phosphorylation of the voltage gated sodium ion channels of specific muscles therefore regulating their excitability.[8,15] These sodium ion channels result in the ability of the muscles to contract and relax on demand. Furthermore the DMAHP gene, in unmutated instances, is found centromeric to the DMPK gene.[6] This is important because the DMAHP gene includes a homeobox domain relevant to brain and muscle development throughout childhood.[8] The homeobox region being found in DMAHP suggests that this gene may very well have a role in early pattern establishment.[8] Although the specific mechanics and purpose of normal DM1 protein kinase is still not well understood, the results from mutations in the 3' untranslated region of this gene have been more closely examined due to the severity of the disorder they often cause.

Mutation Description

Myotonic dystrophy is an autosomal dominant disorder which can be caused by the presence of one of two different mutations, a CTG repeat expansion in the 3'UTR region of DMPK, or a CCTG repeat expansion in intron 1 of zinc finger protein 9 (ZNF9).[9] These two mutations occur on different genes found on different chromosomes, chromosomes 19 and 3 respectively, and are therefore referred to separately as type 1 and type 2 myotonic dystrophy mutations. Types 1 and 2 myotonic dystrophy occur with approximately the same frequency, although some reports say that DM1 is slightly more common. As the focus of this paper is type 1 myotonic dystrophy (DM1), the CTG repeat expansion in the 3' UTR region of DMPK will be described in greater detail. Healthy DMPK genes contain a CTG repeat sequence with between 5-38 repetitions, however patients with DM1 have been observed to have anywhere between 50-5000 CTG trinucleotide repetitions.[1] The severity of the DM1 patients symptoms is directly correlated with the number of repeats present, and patients with over 1000 repeats are often terminal.[2] The cause of expansion of the CTG repeat sequence in humans remains largely unknown, however, recent studies in mice have shown that expansion is driven by 7,8-dihyro-8-oxoguanine DNA glycosylase (OGG1) in its function on the base excision repair pathway.[10,13] This CTG repeat extension is important because, in the cells of those suffering from DM1, it hinders the expression of the DMPK gene by binding with the mRNA transport proteins. This unwelcome binding with transport protein CUG-BP prevents the transport proteins from successfully moving the mutant alleles to the cytoplasm for translation.[8] The CUG-BP proteins in patient cells are therefore sequestered in the nucleus whereas, in healthy cells, they remain predominantly in the cytoplasm (see **Figure 1**).[8] This can interfere with other mRNA in the nucleus causing a trans-effect on RNA metabolism.[8] Additionally, the extended 3' untranslated region on DMPK inhibits the neighboring DMAHP gene, and leads to an approximately 20% deficiency in DMPK protein production. Due to several factors such as variation in length of the

CTG repeat sequences, age of onset, and even the sex of the patient, those diagnosed with DM1 exhibit a wide variety of symptoms which can vary widely in severity.[11]

Pathology

The overall toxicity, or pathology, of the extended trinucleotide repeat sequence in the DMPK gene can be simplified to three main parts; the presence of excessive CUG-BP transport protein in the nucleus, the approximate 20% deficiency in DMPK protein production, and the hinderance of the neighboring DMAHP gene expression. All three combined result in a variety of clinical symptoms including muscle weakness, cardiac and cognitive complications, dysphagia, sleep disorders, neuromuscular respiratory problems, and many more, however, a few symptoms can be directly linked to a specific part.[12] For example, the CUGBP family, along with another important splicing regulator the MBNL family, are integral to the regulation of skeletal and cardiac muscle formation.[14] However, these regulators attack a variety of pre-mRNA molecules, and therefore the retention of these regulators in the nuclear foci is bad for pre-mRNA processing.[14] The CTG repeat sequence mutation is especially bad in this instance because not only does it prevent the regulators from completing their normal functions with heart and skeletal muscle, but it also traps them in the nuclear foci where they harm pre-mRNA processing.[16] This hinderance of the MBNL and CUGBP families, alongside the drop in DMPK, has been linked to the myotonia symptoms the disease is named for, as well as several other muscular system failures.[14] On another note, the hinderance of the DMAHP genes transcription results in haploinsufficiency of the DMAHP protein.[8] The insufficiency of the DMHAP protein has been linked to mental retardation and dysmorphology.[8] However, one must keep in mind that the pathophysiological mechanism for the multisystem degeneration that characterizes advanced DM is very poorly understood.[17] DMPK knockout mice or mice that have been given an overabundance of DMPK have displayed only minor histopathological problems.[17] This fact

leads researchers to believe that DMPK activity itself doesn't provide the sole explanation for the diseases severity.[17] While some muscular symptoms can be treated, the nature of neurological symptoms, such as those caused by the hinderance of DMAHP significantly impact the difficulty of effectively treating this disorder.

Prevention and Therapies

Sadly, those affected with DM1 are not always treatable with the medications and techniques of today, however, there are some things that have been tried or theorized by doctors to help ease the symptoms of DM1. One such treatment that is often tried is the antidiabetic drug Metformin, which is pharmaceutical in nature, and is believed to help with the cellular aging aspect associated with DM1.[18] Additionally, for patients suffering from milder cases of DM1, treatment for the namesake myotonia symptom is available through the antimyotonia drug Mexiletine.[19] On the other hand, several therapeutic treatments for the disorder through methods of gene therapy have been theorized, and may soon be available to the public.[14] One such therapeutic treatment would be the use of CRISPR/Cas 9 nucleases to induce deletions, mutations, or contractions in the CTG repeat region.[14] Even additional mutations that break the CTG pattern would be beneficial as studies have shown that patients with variant repeats - GGC, CCG, or CTC interruptions in the CTG repeat chain – stabilize the segment and exhibit much milder phenotypes.[14] However, as many of these treatments are still theoretical, or are not applicable to all patients affected, the safest route remains to place focus on prevention. The most effective form of prevention is to consult with a genetic counselor before having children, especially if DM1 runs in the family, as DM1 is autosomal dominant and worsens from parent to child in 95% of cases.[20]

Ethical Implications

The ethical considerations surrounding the treatment and prevention of DM1 are complex. Given that DM1 is an autosomal dominant disorder, where even a single mutated gene can lead to the disease, the ethical discussions often revolve around genetic counseling, prenatal testing, and potential gene therapies, including the use of CRISPR/Cas9 for gene editing.

Genetic Counseling and Testing

Genetic counseling plays a crucial role in helping at-risk individuals or families understand the nature of DM1. Patients often struggle to understand aspects of DM1 such as its mode of inheritance and implications for current or future offspring. Ethical considerations arise regarding the right to know or not know about one's genetic predisposition to DM1, confidentiality, and the potential for genetic discrimination by employers or insurance companies. There's also the challenge of conveying complex genetic information in an understandable manner, ensuring informed consent is truly informed.

Prenatal Testing

Prenatal testing for DM1 presents ethical dilemmas around the decision-making process for prospective parents. If a fetus is found to carry the DM1 mutation, parents are faced with the difficult decision of whether to continue the pregnancy. This raises ethical questions about the value placed on lives affected by disabilities and the societal pressures that might influence personal choices. Additionally, there's the concern about the "slippery slope" of selecting which genetic conditions warrant prenatal testing and the implications for society's view on diversity and inclusion.

Gene Editing

The potential for gene editing, particularly through technologies like CRISPR/Cas9, to correct the mutation responsible for DM1 before birth or in affected individuals is an area of significant ethical debate. While the promise of curing genetic diseases is highly appealing,

ethical concerns include the long-term safety and unforeseen consequences of editing the human genome, the potential for creating "designer babies," and the accessibility and equity of such treatments. There is also the broader philosophical question of what it means to be human and the role of genetics in shaping our identity.

Conclusions

In all these areas, the ethical considerations demand a balance between the benefits of advanced genetic technologies, treatments, and the protection of individual rights and societal values. Navigating these ethical challenges it is crucial to engage a wide range of perspectives, including affected individuals, families, healthcare providers, ethicists, and policymakers, in open dialogue to ensure that any advances in treating and preventing DM1 are pursued responsibly and equitably.

Figures

Figure 1: The processing of DMPK and DMAHP genes in normal cells (left) and DM1 patient cells (right). In the unmutated cells both DMPK alleles are transcribed, bind with CUG-BP, and are transported to the cytoplasm for translation.[8] In the mutated cells, from MD1 patients, the DMPK gene is transcribed by remains for the most part within the nucleus. This is due to the binding of CUG-BP to the higher number of CUG repeats.[8] Furthermore, the length of the additional repeats inhibits the proper expression of the neighboring DMAHP gene. [8] Figure modified from 8.

Figure 2: The pathogenic mechanisms of DM1 and their theorized gene therapy strategies. The molecular therapies depicted for DM1 patients at different pathogenic levels are as follows.[14] (1) At the level of the DMPK gene, drugs can inhibit trinucleotide repeat transcription, and induce contraction. CRISPER/Cas9 nucleases, and others, can modify gene sequences by inducing deletions or contractions of the extended repeat.[14] (2) DMPK mRNA can be inactivated using

drugs that bind to the CUG repeats or degrade the DMPK mRNA.[14] (3) MBNL and CUG repeats can be separated through competitive binding.[14] (4) Altered pathways neighboring the DMPK transcript can be saved through modulation of splicing and miRNAs.[14] Figure modified from 14.

Figure 3: The annotated gene map for the DM1 protein kinase gene (DMPK). The official gene symbol is DMPK, official name dystrophia myotonica 1 protein kinase.[1] The DMPK gene is located on chromosome 19q13.32, and has 15 exons.[1] The cDNA is annotated as follows: TSS - aqua, 5' UTR - yellow, start codon - green, CDS - blue, exon 1 - beige, exon 2 - light pink, stop codon - red, 3' UTR - magenta, unmutated CTG repeat sequence - grey. Figure constructed and annotated by Caleb Smith.

Figure 1:

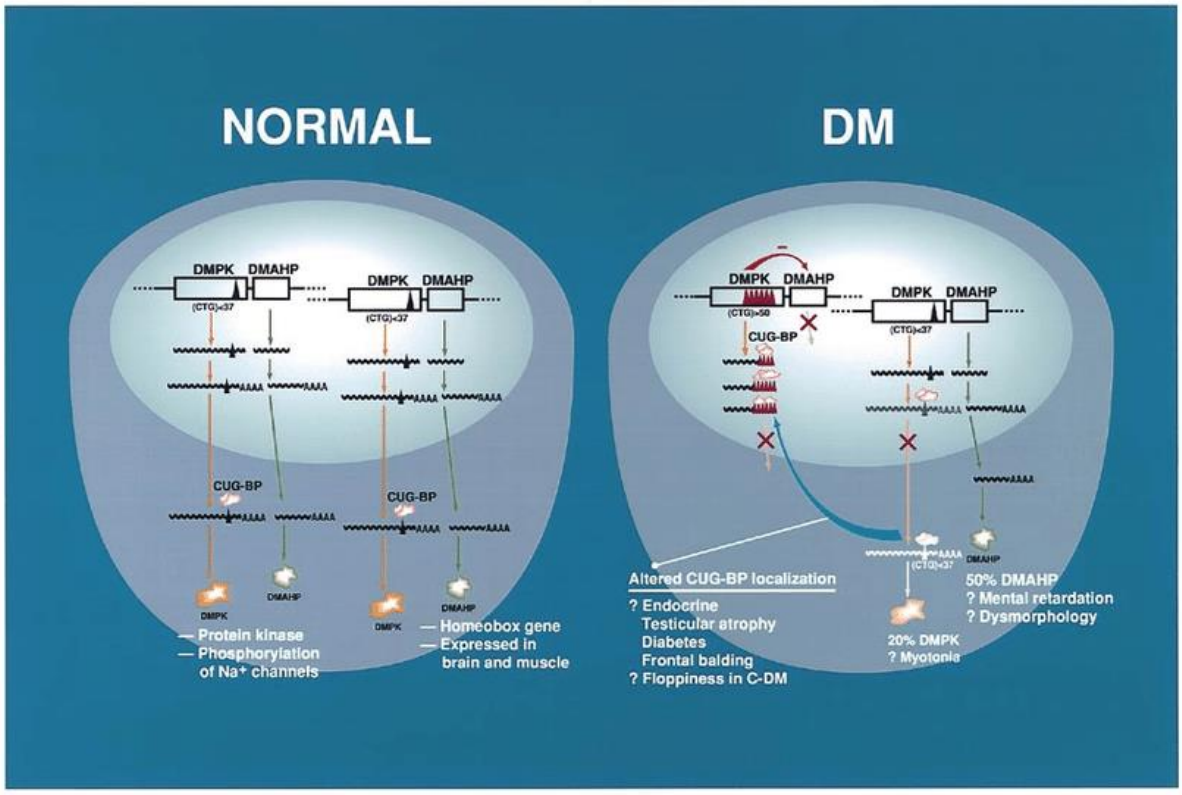

Figure 2:

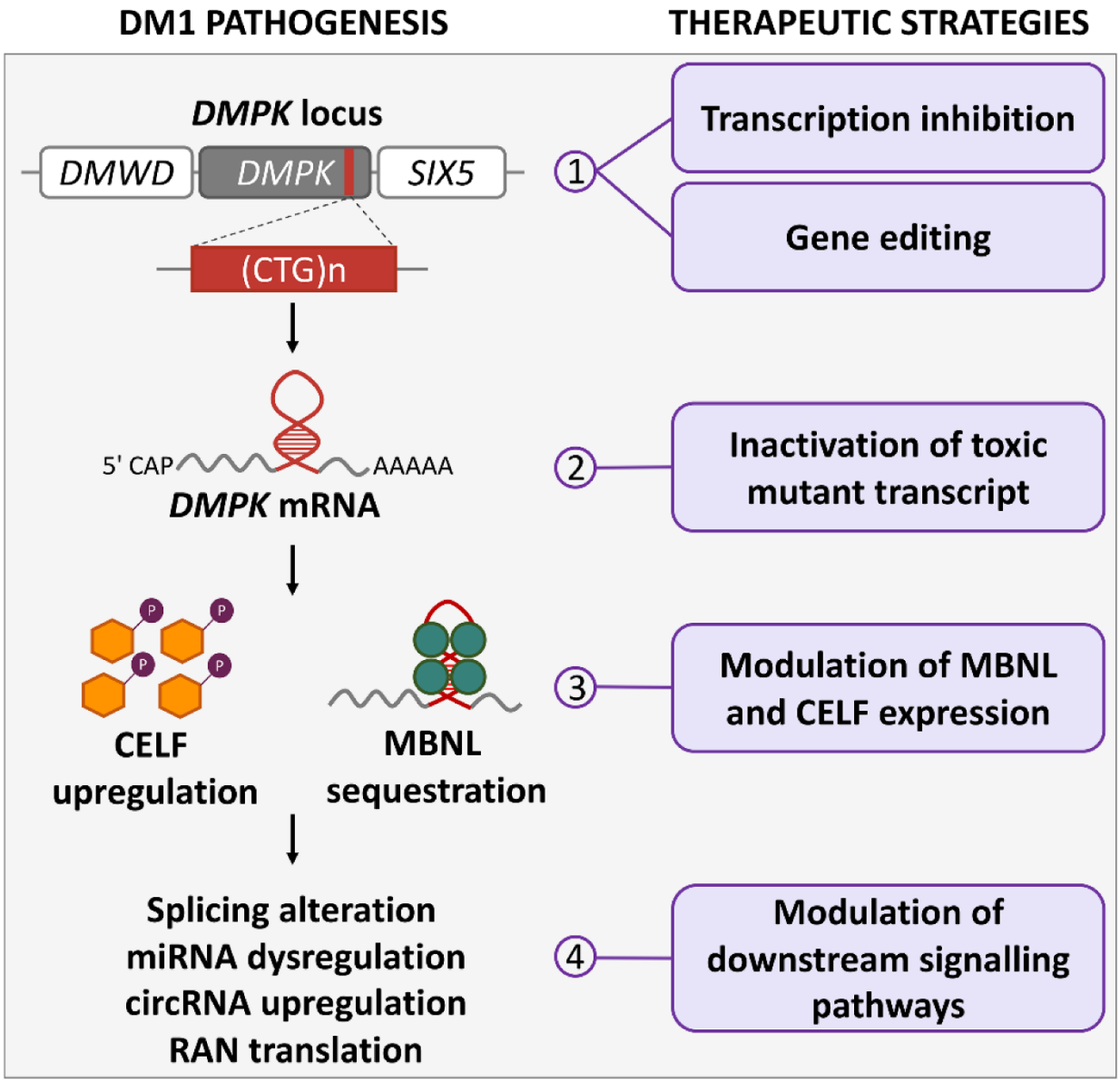

Figure 3: (Gene Map can be found in the attached PDF)

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