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**Utilizing CRISPR/Cas 9 for Gene Editing to Address Hereditary Movement Disorder**

**Introduction**

The Clustered Regularly Interspaced Short Palindromic Repeats (CRISPR)-Cas (CRISPR-associated proteins) technology has emerged as a ground breaking gene-editing tool over the past decade, enabling scientists to precisely and efficiently alter DNA sequences. This article delves into the multifaceted aspects of CRISPR-Cas9 technique that have revolutionized the fields of genetics, biotechnology, and medicine.

CRISPR/Cas9 was initially identified as a peculiar DNA sequence in bacteria. Paired with CRISPR-associated (Cas) proteins, this system forms a natural defence mechanism used by bacteria to protect themselves against viral infections. The Cas9 protein, in particular, acts as a molecular "scissors," which is an RNA-guided DNA endonuclease enzyme that was originally associated with the adaptive immune system of *Streptococcus pyogenes* and is now being utilized as a genome editing tool to induce double strand breaks (DSB) in DNA. This system is crucial for acquired immunity necessary to defend off invading viruses and plasmids (P Horvath 2010). In 1987, researchers discovered CRISPR in the genome of *Escherichia coli*. They identified it as a sequence of 29 nucleotides that repeated several times, interspaced with variable sequence fragments of 32 (Y Ishino 1987).

Genome editing is a form of genetic engineering that involves the precise modification of DNA through the insertion, deletion or replacement in the genome using nucleases which enable precise modification of genes by introducing DSBs (double strand break) at target location in the genome. Nucleases that can be used for in genomic editing include zinc-finger nuclease (ZFNs) and transcription activator-like effector nuclease (TALENs), that create site specific DSBs at target location. One particularly significant tool in genome editing for this purpose is the CRISPR/Cas9, which is an RNA-guided engineered nuclease (RGEN) system, which has synthetic guide RNA (gRNA) introduces a DSB at specific location in target genome (R.Barrangou et al. 2007; JE Garneau 2011). CRISPR Cas is associated with the adaptive immune system of Streptococcus pyogenes. Notably, it offers distinct advantages in clinical applicability compared to other editing technologies.

In medicine, CRISPR-Cas9 holds immense promise for treating genetic disorders. By editing disease-causing mutations, researchers aim to correct genetic defects at the root level. This technology has the potential to pave the way for groundbreaking therapies, offering hope to patients suffering from previously incurable genetic conditions.

**Mechanism of action CRISPR Cas9 system in gene editing.**

CRISPR/Cas9 system is categorized as RGEN, it recognizes specific target sequence of 23-bp length and the mechanism of action of CRISPR/cas9 is different form ZFNs and TALENs (T.gaj et al. 2013; E Deltcheva et al. 2011). CRISPR/Cas9 uses gRNA to recognize genomic DNA and utilizes Cas9 as a nuclease. (MJ Moscou et al. 2009; P Mali et al. 2013). The gRNA recognizes approximately 20-bp nt and requires protospacer adjacent motif (PAM), which recruit Cas9 (P Mali et al. 2013), where Cas9 is guided by a specific sequence of gRNA that is related to trans activating crRNA (tracrRNA) and forms the complementary DNA target sequence, resulting in site specific DSB (E Deltcheva et al 2011; JE Garneau 2010; M Jinek 2012; G Gasiunas 2012). CRISPR/Cas9 has the ability to disrupt multiple genes simultaneously, so it can be used for studying genetic interaction and making models for multigenic disorder.  Cas protein is able to target specific DNA sequences by controlling the short specific part of gRNA. Cas protein can be dual-guide RNAs or single-guide RNAs, but one major problem with Cas is presence of off-target effects, which involve the nonspecific recognition and digestion of non-targeted DNA regions. The method for avoiding off-target effects need further investigation for effective application of CRISPR/Cas9 to human disease.

**CRISPR/Cas9 as therapeutic intervention for hereditary movement disorder**

Many genes are identified to be critically involved in pathogenesis of hereditary movement disorder; hence these are potential targets for CRISPR/Cas9 system to develop disease modifying treatment strategies (Im Wooseok 2016). An increase in genetic mutations cause hereditary movement disorders such as ataxia, dystonia, chorea or spastic paraparesis, Huntington disease and Parkinson disease have been identified.

Parkinson's disease is the most common illness of the nervous system characterized by non-motor and motor indications. Since the existence of misfolded proteins which are called Lewy bodies and their chief component, α-synuclein is the usual pathology of PD, and it can be postulated that one of the main genes related to PD is the SCNA gene which encodes α-synuclein. The most common genetic cause of sporadic and familial PD is the existence of mutation in the leucine-rich repeat kinase 2 (LRRK2) that induces toxicity in the dopaminergic neuron. The amount of neurite complexity in dopaminergic neurons reduces as a result of using the CRISPR/Cas9 tools and editing of mutated LRRK2 by CRISPR/ Cas9 that reduced the incidence of the familial and sporadic form of PD.

In autosomal dominant spinocerebellar ataxia type 1, 2, 3, 6, 7 and 17, cerebellar ataxia are trinucleotide repeat disorders which have accumulation of abnormal protein with expanded polyglutamine tract. This is a common pathogenic mechanism in neurodegeneration. Although in Parkinson’s disease, amyotrophic lateral sclerosis and Alzheimer’s disease are associated with multifactorial etiological factors, and have accumulation of abnormal misfolded protein. CRISPR/Cas9 can be used to modify abnormal protein production and accumulation appears to be effective in their diseases. Some hereditary movement disorder occurs in autosomal recessive patterns, caused by the loss-of-function mutation of a particular gene. genes responsible for hereditary movement disorder can be knock in a specific transgene by CRISPR/Cas9. Some hereditary movement disorder occurs in autosomal recessive patterns, caused by the loss-of-function mutation of a particular gene. Genes responsible for hereditary movement disorder can be knock in a specific transgene by CRISPR/Cas9.

**Futuristic trends of CRISPR/Cas9**

The evolution of the CRISPR system into a gene editing tool has sparked a revolutionary shift in the life sciences. The advent of next-generation gene editing technologies has further enhanced the versatility of the CRISPR system, offering researchers potent and innovative tools to explore biological systems and study human diseases. CRISPR technologies hold significant promise as potential treatments for genetic diseases in humans. Base editing screening has emerged as a valuable method to investigate the connections between gene mutations and their effects. Its remarkable ability to knockout specific genes without causing extensive chromosomal rearrangements by introducing a premature stop codon or disrupting the splice site.

Gene editing technology has changed the meaning of gene therapy but there is still lots of fundamental and translational work to be done to understand the full potential of CRISPR/Cas9, particularly in the area of genetic diseases. In recent years, it has been seen that an increasing number of CRISPR related clinical trials and promises are being proposed worldwide. CRISPR/Cas9 has been widely used for creating related cellular or animal models of human diseases, developing the CRISPR/ Cas9 technique as a therapeutic tool for gene editing can pave the way for the treatment of human disorders. It can be postulated that the CRISPR/Cas9 system could correct or repair mutations caused by both monogenic recessive and dominant-negative disorders to reach therapeutic advantages.

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