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A Review on Huntington's Disease

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Authors' contributions

This work was carried out in collaboration between both authors. Author VKG was involved in conception and design of the study. Author AK wrote the first draft of the manuscript. Both the authors participated in revising the manuscript critically. Both the authors read and approved the final manuscript.

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Review Article

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ABSTRACT

Huntington's Disease (HD) is a progressive neurodegenerative disorder that gradually declines cognitive skills, impair memory and normal movements of affected individuals. This disease affects cerebral cortex and basal ganglia of the brain and those parts of the brain which are associated with the memory retention and motor movement. HD is an autosomal dominant inherited disease caused by the elongation of CAG repeats on the short arm of chromosome 4 at 4p16.3 in huntingtin gene (Htt gene). The mechanisms by which mutant Htt (mHtt) gene causes HD have not been known yet, however mHtt gene can impair mitochondrial function by deregulation of transcriptional processes, calcium imbalance and defective mitochondrial bioenergetics. mHtt gene of neurons affected by HD induces intracellular Ca2+ which enters in the mitochondria and opens the mitochondrial permeability transition pores (mPTP), leading to decrease in mitochondrial ATP, and neuronal cell death. The mean age of onset of this disease is 40 years but the earlier onset of the disease in childhood is known as Juvenile onset HD. HD is characterized by the motor symptoms, cognitive symptoms and psychiatric symptoms. This disease is diagnosed on the basis of symptoms found in the affected individuals and confirmation is done by the genetic testing in

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which CAG repeats are counted. When the CAG repeat is 40 and above, then the individual is said to be suffering from this disease.

Keywords: Huntington's disease; 3-Nitropropanoic acid; huntingtin gene; neurodegenerative disorder; quinolinic acid; SSRIs; transglutaminase inhibitors.

1. INTRODUCTION

HD is a devastating inherited neurodegenerative disorder which is characterized by weight loss, impairment of motor function, cognitive dysfunction, and neuropsychiatric disturbances. [1]. HD predominantly affects striatum, cerebral cortex and other areas of the brain controlling motor coordination and memory storage [2]. HD affects most commonly the basal ganglia which are a group of nerve cells at the base of the brain. The basal ganglia are involved in various functions including control of voluntary motor movements. The main components of the basal ganglia are the dorsal striatum comprising of caudate nucleus and putamen, ventral striatum comprising of nucleus accumbens and olfactory tubercle, globus pallidus, ventral pallidum, substantia nigra, and subthalamic nucleus [3]. It has been demonstrated that mitochondria are the key factors in cell survival by controlling energy metabolism, apoptosis pathways and Ca^{2+} homeostasis [4-5]. The brain is acutely dependent on energy supplies for normal functioning and mitochondria are the intracellular source of brain energy supplies. Any changes in functional alterations in these essential cellular energy dynamos can lead to insidious pathological changes in neurons [6]. 3-Nitropropanoic acid (3-NPA) is an irreversible inhibitor of succinate dehydrogenase that inhibits both the Tricarboxylic acid cycle (TCA cycle) and complex II activity. Selective lesioning of the striatum occurs after administration of 3-NPA in animal models. Low doses of 3-NPA administered chronically to both rodents and non-human primates resulted in pathology and symptomatology resembling to HD [7].

Pharmacological intervention in the treatment of the movement disorder of HD is aimed at restoring the balance of neurotransmitters in the basal ganglia [8]. Cognitive deficits are core neuropsychiatric problem among HD patients. In the early stage of illness HD patients also suffer from impaired attention and visuospatial processing. The complexity and unpredictability of this disease is a great problem in management and treatment for health and social care professionals [2]. Chorea and loss of balance are the early symptoms of the disease which are first noticed by the patients, although some families also notice cognitive or personality changes before the early symptoms.

HD is an autosomal dominant inherited disease caused by the repetition of elongated CAG on the short arm of chromosome 4 at 4p16.3 in the Htt gene [9]. As long as the CAG repeat increases, the onset of the disease is earlier. HD can be defined as a movement disorder with a heterogeneous phenotype which is characterized by involuntary motor impairment with cognitive, psychiatric deficits and bio energetic inadequacy [10].

The mean age of onset of symptoms is 40 years, but the symptoms also starts in childhood which is called juvenile onset (<20 years) and later known as older onset (>70 years) forms. These forms are well recognized [11].

2. EPIDEMIOLOGY

The disease occurs among all racial groups but is most common in people of northern European origin. Its prevalence in the Western hemisphere is 7-10/100000 [12]. Genetic confirmation of the CAG repetition is the hallmark of epidemiological measure of HD. Prevalence studies incorporating both genetic and clinical diagnostic standards show that 10.6–13.7 individuals per 100,000, or 1 in 7,300, are affected in Western populations. The incidence of HD is estimated to be 4.7–6.9 new cases per million per year in Western populations. In Japan, Hong Kong and Taiwan, HD is diagnosed in only 1–7 individuals per million, approximately one-tenth as frequently as in Europe and North America [13].

Epidemiological data from other populations in Africa and Asia are limited. Germ line instability of intermediate alleles increases with CAG repeat length, indicating that longer CAG repeats in the general population results in higher CAG expansion rate and higher prevalence of HD [13].

3. PATHOPHYSIOLOGY

Htt gene is located at chromosome 4 at 4p16.3 that encodes the protein huntingtin, the normal

function of which is unknown. Htt gene is abundantly expressed in the brain and testes with moderate expression observed in other organs such as liver, heart, and lungs [14]. The length of the CAG tri nucleotide repeat that encodes Poly Q segment of varying length can be determined in any individual (normal, at risk or clinically diagnosed with HD). This repeated sequence of CAG is polymorphic in the normal population in the range of 6–35 units; when expanded to ≥40 units, the mutation occurs at higher rate, which initiates the disease process that leads to the onset of motor symptoms that can be diagnosed. Repeats of 36–39 CAG units show reduced penetration rate, as some individuals with these CAG lengths have HD, whereas others live a normal lifespan without being clinically diagnosed. The CAG repeat shows instability through meiotic transmission that can be seen in the intermediate CAG repeat range 27–35 units; this instability increases with increasing in CAG length. CAG repeat typically increases or decreases in length by one or few CAGs.

The length of the CAG repeat in Htt gene determines whether an individual will develop HD or not; it is also the primary determinant of the rate of pathogenesis leading to the characteristic motor signs that underlie the clinical diagnosis [15]. Importantly, with respect to these motor signs, the timing of onset is determined by the allele with the longer CAG repeat in a completely dominant manner; the second Htt allele, regardless of its length (normal or otherwise), does not alter the rate of the process that leads to a clinical diagnosis [16]. The precise nature of the pathogenic trigger that conforms to these genetically defined criteria (CAG length dependence and allele dose in dependence) is not known, but the demonstration that the length of the CAG repeat, even in the normal range, correlates with measures in some cellular assays (for example, cellular energy charge suggests that it might involve a gain of function that acts through augmentation or deregulation of one or more normal functions of huntingtin. Although several genes — including ADORA2A, ATG7, CNR1, GRIK2, GRIN2A, GRIN2B, HAP1, PPARGC1A, MAP2K6, MAP3K5, NPY, NPY2R, OGG1, PEX7, TP53 and UCHL1 — have been proposed as genetic modifiers of HD [13].

Basal ganglia, is constitute by the major portion of striatum which integrates signals from the cerebral cortex, and is the centre for appropriate selection of behavioral action. Its dysfunction has been associated with classical motor disorder such as HD, dystonia and depression. The main barrier for such interrogation has been its apparent homogeneity, which obscures its inner compartments. The identification of two intermingled populations of medium spiny neurons (MSNs), expressing either dopamine receptor D1 or D2 is an important step in understanding of striatal subdivisions. The efferent projections of these molecularly defined populations constitute two separate pathways in the basal ganglia; a direct pathway (D1) projecting to the substantia nigra pars reticulata (SNr) and the internal globus pallidus (GPi), and an indirect pathway (D2) projecting to the external globus pallidus (GPe) [17].

The mechanism of selective degeneration of striatal neurons suggests that reduced trophic support renders striatal neurons more vulnerable to the toxic actions of mhtt gene. Numerous *in vitro* and *in vivo* studies have shown that striatal neurons require brain-derived neurotrophic factor (BDNF) for their survival and functioning. A deficiency in BDNF-mediated signaling alone is sufficient to cause dendritic abnormalities and neuronal loss in the cerebral cortex and striatum. Moreover, reduced levels of striatal BDNF were detected in both HD animal model and HD patients [18].

3.1 General Mitochondrial Defects in HD Patients

3.1.1 Mitochondrial enzymes

In biochemical studies defects in respiratory chain is found in HD individuals. The activity of complex II/III are greatly decreased in comparison to complex IV in HD patients but pre symptomatic patients showed no changes in activity of complex II, III and IV. Minor changes were observed in respiratory chain enzymes of cerebral cortex but no changes were observed in blood cells. The other enzymes of oxidative metabolism were also reported with reduced activity in the striatum. The levels of aconitase and pyruvate dehydrogenase complex were also significantly decreased in HD individuals. These decreased enzymes levels were observed in symptomatic patients having atrophy of striatum.

3.1.2 Mitochondrial membrane potential

Mitochondria which is isolated from mHtt gene expressing cells, showed decreased membrane

potential. Sawa et al. first reported that stress induced apoptotic cell death were increased in lymphoblast derived from lymphocyte of HD patients. The mitochondrial membrane depolarization was increased in lymphoblast of HD patients as compared to control lymphocyte, when subjected to apoptotic stress. This loss of potential was correlated with CAG repeat expansion. The neural cells showed high sensitivity to Ca^{2+} induced permeability transition. The loss in mitochondrial membrane potential and permeability transition was demonstrated by directly interaction of recombinant mHtt gene with the outer mitochondrial membrane.

3.1.3 Mitochondrial Ca^{2+} buffering capacity

Increased cytoplasmic Ca^{2+} levels are toxic to neurons. The Ca^{2+} buffering capacity of cells expressing mHtt gene can be reduced [19].

4. STRUCTURE AND FUNCTIONS OF HUNTINGTIN PROTEIN

Huntingtin protein has a normal polyQ repeat length of 23 glutamines (Q23) containing a total of 3,144 amino acids with a molecular weight of 348 kDa [20-21]. Huntingtin is expressed throughout the body but in varying proportion depending on cell type. Forms of this type of protein can be found in the nucleus and cytoplasm, and huntingtin can shuttle between these compartments. The normal functions of huntingtin are not known yet [22]. Some broad biological functions of the normal protein have been uncovered, including its critical role in the development of the nervous system, its ability to influence BDNF production and transport, and its role in cell adhesion [23-24]. Huntingtin is involved in transcription, regulation by interacting with an array of transcriptional factors and other proteins involved in the regulation of mRNA production [25-26]. Loss or modulation of normal huntingtin function in response to poly Q repeat expansion might also have a role in HD [27-28]. Huntingtin is linearly organized as a series of ordered domains interspersed with intrinsically disordered segments. Further folding may occur as a result of interactions between folded domains. The known ordered domains are clusters of α-helical HEAT (Huntingtin, elongation factor 3, protein phosphatase 2A and TOR1) repeats that are also found in several other proteins, in which there are binding sites for macromolecules. There is uncertainty about the exact number and location of the HEAT repeats and their roles in binding to the very large number of huntingtin interaction partners. Separating the clusters of HEAT repeat are expanses of disordered structure, the only known functions of which are as regions for post-translational modifications (PTMs) such as proteolytic cleavage, phosphorylation and glycosylation [29-30].

5. INHERITANCE OF HD

Everybody has two copies of the Htt gene, but only one changed copy of the gene can develop the disorder. The normal copy cannot compensate for the effects of the copy that is mutated. When people who have Htt gene with mutation have offspring, they can pass on either their normal copy of the gene or the copy with the mutation to their offspring. This means there is a 1 in 2, or 50% chance of their child inheriting the gene with mutation. There is also a 1 in 2 or 50% chance of their child inheriting the normal copy of the gene [31]. A person who inherits the HD gene, and survives long enough, will sooner or later develop the disease. In some families, all the children may inherit the Htt gene; in others, none do. If one child inherits the gene has no bearing on whether others will or will not share the same fate. A small number of cases of HD are sporadic, that is, they occur even though there is no family history of the disorder. These cases are thought to be caused by a new genetic mutation or alteration in the gene that occurs during sperm development and that brings the number of CAG repeats into the range that causes disease [12].

6. MECHANISM OF CORTICAL AND STRIATAL NERVE CELLS DEATH IN HD PATIENTS

The exact function of normal huntingtin protein is not yet known by the scientists but it is very important for development and remains active in the whole body. The symptoms of HD individuals might be due to regular interaction of huntingtin protein with protein found only in the brain and this changed form of huntingtin protein disrupts this interaction leading to nerve cell death. Various studies showed that huntingtin protein interacts with two proteins; huntingtin's interactor protein (HIP-1) and huntingtin's associated protein (HAP-1) present only in the brain and this is the reason that HD affects only the brain. The interaction of HIP & HAP with huntingtin protein depends on the number of CAG repeats in Htt gene. The increase number of CAG repeat leads

to binding of huntingtin protein more to HAP-1 than HIP-1 [3].

7. CAUSES OF HD

HD results from genetically programmed degeneration of nerve cells in Striatum, Cerebral cortex and other areas of the brain responsible for memory storage. Degeneration of neurons causes uncontrolled movements, loss of intelligence, and emotional disturbance. Specifically affected neurons are the cells of basal ganglia, which are deep within the brain that have many important functions, including coordination movement. Within the basal ganglia, HD especially targets neurons of the striatum, particularly those in the caudate nuclei and the pallidum cortex; also gets affected which controls thought, perception, and memory [32].

It has been postulated that an excitotoxin is involved in the pathogenesis of HD. Quinolinic acid (QA) is an excitotoxin which can produce axon sparing lesions in a dose of 60nmol similar to those found in HD. The lesions results in depletion of neurotransmitters present within striatal spiny neurons. Scientists observed that lesions of HD are similar to the lesions caused by quinolic acid and results in marked depletion of both Gamma Amino Butyric Acid (GABA) and Substance P and selectively spare Somatostatin/ neuropeptide Y neurons. It was concluded that QA or a similar compound could be responsible for degeneration of neurons in HD [33].

8. CLINICAL FEATURES

The disease was named as Huntington's chorea after George Huntington, who gave first detailed description in 1872. The name has changed to HD because chorea is not the only predictive symptom of the disease, there may be many non-motor symptoms which may be more disabling and distressing than the motor symptoms. Imaging and postmortem studies have shown that the disease is characterized by cerebral atrophy. Atrophic changes are initially seen most prominently in the striatum (part of the basal ganglia) and later become widespread to all muscles.

8.1 Psychiatric Symptoms

Depression is one of the most common psychiatric symptoms of the disease and it occurs as part of the disease. Other psychiatric symptoms include obsessive-compulsive symptoms and psychosis. It is important to recognize psychiatric symptoms in HD so that symptomatic treatment can be given to the patient. This may be difficult to recognize in the disease later because diagnoses may be unidentified by other features of the disease. For example, depression may be difficult to detect in patients who has altered facial expressions and tone of voice [34].

8.2 Motor Symptoms

The motor symptoms of HD can be divided into two categories: 1. Added involuntary movements. Example - chorea and 2. Impaired voluntary movements, which cause limb in coordination and impaired hand function. Motor symptoms get worsened when postural reflexes are lost. The motor symptoms changes over time, chorea decline as the disease progresses while dystonia, rigidity, and bradykinesia becomes more marked in the later stage.

8.3 Cognitive Symptoms

Cognitive impairment includes slowing of thought processes like problem solving and deterioration of executive functions. Typically, patients suffer from difficulty with multitasking, concentration, and short term memory [35]. People with HD are often impulsive and develop psychomotor perseveration. Visuospatial perceptions are also impaired [36]. Cognitive dysfunction in HD, not only impairs long-term memory but also impairs executive functions such as organizing, planning, checking, or adapting alternatives, and delays the acquisition of new motor skills. These features are worsened over time; speech deteriorates faster than comprehension [37].

8.4 Metabolic Symptoms

Both animal and human studies indicate that some of the peripheral symptoms of HD, including weight loss and alterations in appetite, could be linked to endocrine and metabolic alterations. These alterations may be reflected in plasma levels of carbohydrate, lipid or protein metabolites and in hormones related to energy metabolism.

8.4.1 Carbohydrate metabolism

Carbohydrate metabolism in patients with HD has shown significant changes in various monosaccharide levels, particularly glucose, between HD gene carriers and studies have

shown impaired glucose tolerance and increased prevalence of diabetes in HD patients**.** Insulin sensitivity studies in HD patients have shown both a decrease in insulin sensitivity and impaired insulin secretion capacity in normoglycemic subjects.

8.4.2 Lipid metabolism

HD patients display changes in body fat stores, as indicated by decreased visceral and peripheral adiposity**.** In addition, altered fatty acid metabolism and changes in various markers of fatty acid breakdown have been reported in HD**.** Dysfunction of the cholesterol biosynthetic pathway and reduction in cholesterol precursors and their metabolites have also been shown in HD patients.

8.4.3 Protein metabolism

Muscle wasting is a common feature of HD and abnormal skeletal muscle energy generation has been shown in both symptomatic patients with HD and presymptomatic mutation carriers**.** Several studies of amino acid metabolism in patients with HD have found a decrease in the concentrations of neutral amino acids (especially alanine, valine, leucine and isoleucine) in HD plasma.

8.4.4 Gastric and adipose hormones

Ghrelin, an orexigenic peptide of gastric origin, and leptin, a peptide hormone secreted by adipose tissue, are two peripherally produced hormones that exert effects on the hypothalamus in the regulation of body energy homeostasis. Leptin induces weight loss by suppressing food intake and stimulating metabolic rate, whereas Ghrelin stimulates appetite and increases adiposity [38].

9. DIAGNOSTIC TEST FOR HD

The onset of HD symptoms starts with motor symptoms like clumsiness, tremor, balance trouble and jerkiness. The earliest symptom of HD is chorea or choreoathetosis i.e. continuous and irregular writhing and jerking movements. The most prominently affected parts are limbs and trunk but respiratory, laryngeal, pharyngeal, oral and nasal musculature are also affected. Impaired visual acuity, slow, poorly coordinated motor movement, dysarthria & dysphagia, rigidity and ataxia are frequently observed in HD individuals.

Memory impairment begins with movement abnormalities and causes an increase in the loss of voluntary movement capacity. Like the other sub cortical dementias, aphasia and agnosia are less evident than in Alzheimer's disease where as cognitive speed and efficiency are more impaired but dementia becomes more global as the disease progress. Within 10-15 years of onset of disease, about 80% of HD patients develop non cognitive psychiatric disorder. Mood swing and personality changes like irritability or apathy increases the suicide rate in such individuals. Psychiatric symptoms of HD patients may be treated more effectively.

9.1 Differential Diagnosis

The differential diagnosis plays an important role in evaluation of clinically affected individuals who do not have HD mutation. So for diagnosis of these conditions some specific treatments are available.

9.1.1 Basic laboratory evaluation

The presence of mutant allele may be determined by the development of PCR based assay. The length of CAG repeat may be calculated by the length of PCR product which controls both the CAG repeat and CCG and CCT repeat containing seven and two triplet respectively. It may be shown that CCG repeat ranges between 7-12 triplets in length and CCT repeat in between 2-3 triplets in length in which 2 triplet lengths is common and 3 triplet lengths is rare. Therefore it is recommended for laboratories to use only a PCR assay containing only the CAG length, to prevent the overestimation of length of CAG repeat.

Polyacrylamide gel electrophoresis is used to evaluate the size of PCR product. The length of PCR product is detected by use of a radiolabelled product like (CAG)n or (CTG)n oligonucleotide which is generated by radiolabelled primer to probe a southern blot of the gel or fluorescent product is detected by an automated genotyping system by using fluorescent tagged primer.

9.1.2 Laboratory evaluation of apparent homozygosity

The potential source of diagnostic confusion is the detection of single repeat. The result of PCR reflects repeat length homozygosity. The CAG repeat length homozygosity is confirmed by comparing the original PCR assay for repeat

length which includes both CAG and CCG/CCT repeat. Two different PCR products will be distinguishable when the length of CCG repeat is heterozygous. The length of CAG repeat may be confirmed by second PCR which amplifies only CCG (and CCT) repeat.

9.1.3 "Sporadic" HD

Recently it has been shown that the rate of genetically confirmed symptomatic individuals of HD is 8% more than all HD individual. This may reflect denovo expansion with unstable repeat in the range of 27-35 triplets. It also includes some other factor like anticipation (age of onset of symptoms in child before parent), early death or misdiagnosis of affected parents, adoption and false paternity. Therefore the family history cannot be excluded for the diagnosis of HD.

9.1.4 Presymptomatic testing: Special issues

As the presymptomatic testing gains an important consideration, the experience and research protocols for HD served as a basis for presymptomatic testing of other disorders.

Pretesting genetic counseling is so informative that an individual can decide the risk and benefit of testing protocols. The main result of these testing is that:

a) Only a few individuals desire testing.
b) Some individuals get traumatized by getting the result.

The other problem is a negative result in the absence of genetically confirmed family history of HD. In such type of cases it may be possible that disease in the family of at risk individual is not HD. The recent findings demonstrate that HD like 2 caused by different mutation is clinically and pathologically indistinguishable from HD and it also demonstrates that even pathologically diagnosed HD does not prove that the familial disease is caused by HD mutation. Researchers need to recognize the limitations of a negative HD presymptomatic test in family members in whom HD mutation has not been demonstrated.

Another complication of presymptomatic testing arises when the adult grandchild of HD affected individual's show his interest in presymptomatic testing where as the other middle aged child of that individual is not interested in presymptomatic testing. In such cases exclusion testing is carried out by genotyping markers which determine that grandchild received an allele either from affected grandparents or from the unaffected spouse of those grandparents. If an allele is inherited from unaffected grandparents the risk of HD is low but if it is inherited from affected grandparent the risk of HD is 50% because it is not known that the allele which has inherited is with or without mutation. No information is obtained from this testing which can change the risk of the child of affected individual. Exclusion testing and direct test of the length of HD mutation have been successfully applied in *in vitro* and in *in utero* fertilization. Interpretation of test result is same like other forms of presymptomatic testing with only the exception that pre implantation genetic diagnosis (PGD) requires DNA preparation from a single cell. Without disclosing the result of HD test, PGD can be performed in at risk individual who do not want to know his/her gene status. Parents should be advised that they should not consider inutero exclusion testing until they are not prepared to terminate the pregnancy for a positive test. Recently an alternative exclusion testing has been developed for PGD which involves the use of two markers in a duplex PCR protocol [39].

9.1.5 Genetic testing

It can be diagnostic or predictive depending on the circumstances. Most people have about 15-25 CAG repeats in each copy of the gene. However, if a person has a Htt gene with more than 39 repeats, they will develop HD at some point in their life, because large CAG repeats send message to our cells to make the huntingtin protein that is harmful. Now-a-days it is possible to find out exactly how many repeats an individual has in each of their Htt genes - and this is the basis for genetic testing. If a patient has features of HD, the most useful confirmatory diagnostic test is CAG repeat testing [40]. The repeating CAG fragment is longer on the HD chromosome than on the normal chromosome and is unstable. Lengths of CAG repeat changes when passed to offspring. Worldwide experience suggests the following interpretations for the results of HD genetic testing [41].

Table 1. Interpretation of genetic testing in HD patients

CAG repeat size	Interpretation
26 & below	Normal
27-35	Normal but partially unstable
36-39	Abnormal with variable penetration
40 & above	Huntington's disease

9.2 Other Diagnostic Tests for HD

9.2.1 Predictive testing

In the United States, predictive testing is requested by a small proportion of people at-risk for HD. The reasons commonly given by those who undergo predictive testing include future planning regarding marriage, reproduction, career, finances, or simply a need to relieve uncertainty. The decision to take a predictive test for HD should always be an informed, carefully considered, and freely chosen personal decision. Individuals should not be forced into testing by the spouse, another family member, a physician, an insurance company, or an employer.

9.2.2 Prenatal testing

Individuals or couples considering prenatal testing are advised to seek Genetic counseling prior to becoming pregnant. Many reproductive options are available to individuals affected with or at risk for HD, of which prenatal testing is one. Samples for prenatal analysis of the HD gene may be obtained in two ways, by chorionic villus biopsy at 10-11 weeks of pregnancy, or by amniocentesis at 14-18 weeks. Some couples may also desire pre-implantation testing of a fertilized embryo. This requires the use of fertility drugs and other procedures available only at specialized *in vitro* fertilization centers [41].

10. MANAGEMENT OF HD

10.1 Pharmacological Treatment

There are no disease-modifying treatments available for HD in routine clinical use, and current treatment is therefore given on the basis of symptoms. Many trials were conducted mainly by concentrating on the mechanisms and outcomes associated with movement disorder. Patients reported that their quality of life is decreased by psychiatric manifestations of their condition, including depression, irritability and apathy. Rates of depression may be as high as 40%, and suicide may occur in as many as 10%. Obsessive compulsive symptoms are also common in HD patients [42]. Drugs of different categories like anti-psychotics, sedatives and anti- depressants etc. are given after monitoring the symptoms.

10.1.1 DA depleting agents

A major neurological symptom associated with HD is chorea. The use of tetrabenazine (TBZ), a specific inhibitor of vesicular monoamine transporter, is approved for chorea in HD patients [43]. TBZ exerts its antichoreic effect by reducing the level of dopamine in brain by preventing its release from vesicle and inhibiting the uptake of monoamines.

Uptake of monoamines in vesicle is regulated by vesicular monoamine transporter (VMATs) proteins present on vesicular membrane. TBZ binds to these VMATs and inhibits the storage of dopamine in vesicles and thus prevents the further release of dopamine in to synapse. The highest binding density for TBZ is in the caudate nucleus, putamen, and nucleus accumbens, areas known to bear the brunt of pathology in HD. In addition, Secondly, TBZ reduces dopamine by blocking dopamine receptors, as it has been shown in *in vitro* studies [44]. TBZ may also act by binding to the receptor of receiving nerve cells and thus blocks the binding of dopamine to these receptors and thus blocks the signaling of neurons [45]. Long-term feeding with TBZ (combined with levodopa) alleviated the motor deficits and reduced the striatal neuronal loss in the mouse model of HD, thus suggesting a neuroprotective effect [46].

10.1.2 Neuroleptics

Both typical and atypical neuroleptics can be used for the treatment of chorea and psychosis in HD. Both are DA D_2 receptor antagonist; however the newer atypical antipsychotics are preferred because they cause less extra pyramidal side-effect. Atypical antipsychotics are often used in the therapy, but evidences showed that clozapine have fewer efficacies with significantly more side effects .While olanzapine gives better results from small open-label studies.

10.1.3 Anti-glutaminergic agents

Remacemide, a non-competitive NMDA receptor antagonist showed an overall improvement in chorea level.

10.1.4 Acetylcholinesterase inhibitors

Acetyl cholinesterase inhibitors are used to treat cognitive decline, a common symptoms of HD.

10.1.5 Antidepressant

Mirtazapine may be preferred for depression as they have anticholinergic activity compared with some other anti-depressants.

10.1.6 Olanzapine and sertraline

These are reported to cause Improvement in depression and obsessional thinking. Risperidone and Amisulpiride may be used in the treatment of psychosis in HD, while Quetiapine is reported to help in behavioral disturbance [43].

10.1.7 Transglutaminase inhibitors

Transglutaminases belongs to a family of closely related proteins that catalyze the cross-linking of a glutamine residue of a protein/peptide substrate to a lysine residue of a protein/peptide co-substrate with the formation of a GGEL [N^{ϵ}-(γ-L-glutamyl)-L-lysine] cross-link. These bonds may be important in the formation of aggregates and the toxicity of mutant huntingtin. Example-Cystamine.

Cystamine is a transglutaminase inhibitor which improves survival, motor phenotype and neuropathology in mouse models, and preliminary dose finding and tolerability trials of Cystamine in humans have been completed.

10.1.8 Chaperones

Within cells protein are continually degraded in to amino acids and replaced by newly synthesized proteins. These newly synthesized proteins may undergo aberrant folding and aggregation. Protein misfolding can lead to the formation of toxic substances. Cells employ different processes and machinery to prevent the buildup of abnormal proteins. Chaperones and their regulators (co-chaperones) are a group of molecule that contributes to the prevention of aggregation [46]. Chaperons help proteins to attain more stable conformations and prevent their aggregation, and their production is increased when subjected to heat-shocks [47].

Chaperones assist other proteins to achieve a functionally active 3D structure and thus prevent the formation of a misfolded protein may be due to loss of function and deleterious gain of function as seen in HD. In which protein misfolding results in the formation of harmful amyloid. Natural chemical and pharmacological chaperones have been shown to be promising agents for the control of many protein conformational disorders like HD [48].

Several drugs have been identified that induce the expression of HSP-70 and other chaperones. These include the hydroxylamine derivative Biclomol and its analogue Arimoclomol and the benzoquinone ansamycin antibiotic, Geldanymycin and its derivative Alvespimycin. These compounds potentiate chaperones expression by activating heat shock transcription factor (Hsf-1) [49].

Chaperon's production can be induced by chemical initiators such as Geldanamycin which act as a protective in cell models of HD. Small-molecule of chemical chaperones such as Trehalose have a similar effect and have shown beneficial effects in mouse models of HD [47].

10.1.9 Genetic modification

Currently, a leading strategy among HD researchers are trying to suppress the expression of the mutant gene by introducing fragments of DNA meant to bind with and eliminating the ability of the gene to make the damaging protein. The goal of this approach is to prevent the mHtt from being expressed in the brain and potentially slow the progression of the disease, if not stop [50].

10.2 Non- pharmacological Treatment of HD

The aim of treatment is to manage symptoms and improve quality of life. No current treatments can slow disease progression, although promising disease modifying treatments are being tested in animal models. There are many effective options for symptomatic management, however, both drug based and non-drug based treatments are available [51]. Many non-drug based measures are effective in the management of Huntington's disease, and these are often more helpful than drugs [52].

10.2.1 Physical therapy

In the early stage of HD, people have difficulty in solving problem and making decision but they are able to care for themselves. In the middle stage peoples have difficulty in walking and may fall down. Due to difficulty in thinking and memory, they easily get frustrated and angry. But in the late stage they need help for all of their daily routine activities and requires some special arrangements for their eating, sleeping, sitting due to severe chorea and loss of all controlled movement [53].

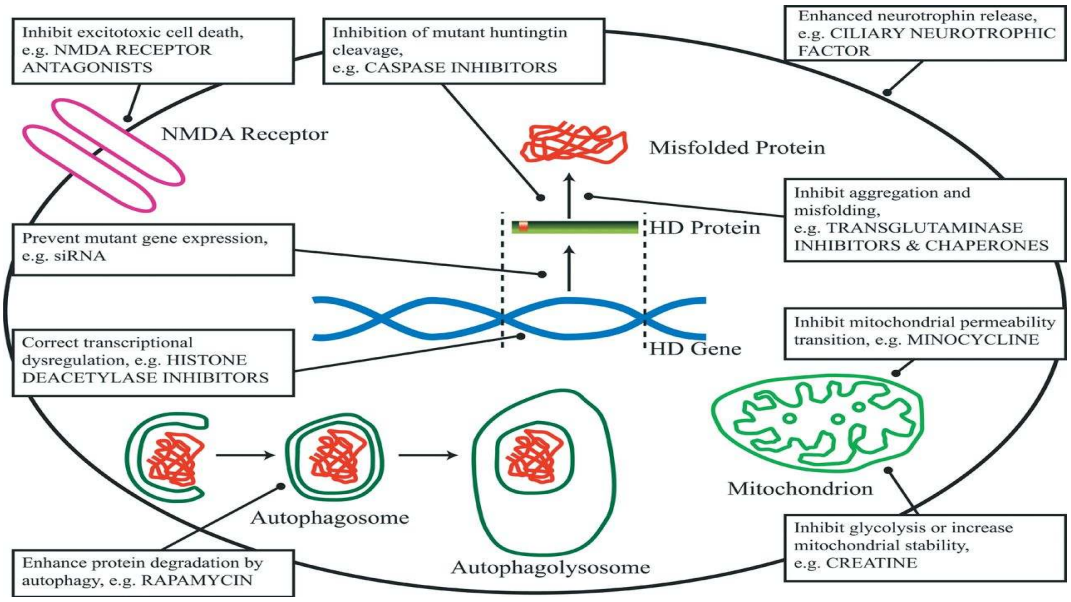

Fig. 1. Potential disease modifying strategies for HD [47]

In the early stage of disease, physical therapist (PT) can improve the health and wellness of HD people by:

a) Telling them how to increase their energy level
b) The use of exercise like aerobic exercise and stretching exercise for keeping the muscle from getting tight and strengthening exercise are used for good posture and balance exercise for improving balance.

In the middle stage, PT gives important advice about:

a) Balance improving exercise to prevent falls
b) Wheel chair or walker to move the person safely
c) Home safety to prevent falls from slippery surface [54].

In the later stage of HD to become active, PT can suggest about: some special chairs with the ability of eating, PT can suggest the care taker, how to help the individual in getting in and out of a bed or car and advice exercise to keep the muscle loose [55].

11. RECENT ADVANCES IN HD

Addition of new gene to human cells is defined as a gene therapy. Recently new technologies of genome coding have been developed which enables the scientist to manipulate the sequence of human genome to achieve a better therapeutic effect. It includes the correction of mutation which causes disease, a therapeutic gene may be added to the specific site in the genome and deleterious gene or genome sequence may be removed.

In Gene therapy the exogenous good DNA is used to replace the defective DNA in those who suffer from genetic defects. Unwanted genetic material like viral genomes or receptor cannot be completely removed by addition of exogenous genetic material. The limitations of gene edition technology can be overcome by new gene editing technology which makes the precise, targeted modification to genome sequence.

11.1 Mechanism of Gene Editing

The gene editing technology is based on the targeted DNA double strand breaks (DSBs) which stimulate the endogenous cellular repair mechanism. DNA breaks are repaired by anyone of these pathways: homology directed repair (HDR) or non-homologous end-joining (NHEJ). HDR is based on the invasion of broken end in to homologous sequence and subsequently DNA break is repaired in a template dependent manner.

NHEJ is stimulated by site- specific DSBs which disrupt the target gene in various cell types. Now

it is possible to perform a wide variety of genomic alteration in a site specific manner.

11.2 Targeted Nucleases

Gene editing by DSB is based on the cells endogenous repair mechanism and it is applicable to any cell type or organism. Precise introduction of a targeted DSB is the main critical part for implementing these genes editing method. Four major platforms are available for inducing site specific DSBs: zinc finger nucleases (ZFNs), transcription activator-like effector (TALE) - nucleases (TALENs), meganucleases and very recently CRISPR/cas system.

11.2.1 Zinc finger nucleases

Zinc finger (ZF) is the protein of most copious class of transcription factor and Cys2-His2zinc finger domain is main DNA-binding domain present in human genome. The basic structure of Zif268 is necessary for DNA recognition by zinc fingers. Zinc finger domain in the presence of Zinc atom forms a compact ββα structure with the α-helical portion of every finger making contact with 3 or 4 bp in the major groove of DNA. In the range of zinc finger, Tandem fingers wrap around the DNA to bind large target sequences such that 3f protein binds to a 9bp target site.

11.2.2 Talens

The discovery of DNA binding specificity of TALE proteins increases the possibility of modular design of novel DNA binding proteins. Each of 33-35 amino acid TALE repeat binds a single base pair of DNA with specificity dictated by two hyper-variable residues. DNA bounded by crystal structure of TALEs explains that each TALE repeat forms a double helix structure which is connected by a loop presenting the hyper variable residue into the major groove of DNA. These modular TALE repeats can be linked together to build a long range with DNA binding specificity.

11.2.3 Meganucleases

It involves the reengineering of DNA binding specificity of naturally occurring homing endonucleases. LAGLIDADG family is the largest class of homing endonucleases which includes the commonly used I-CreI and I-SceI enzymes. By the use of rational design and selection, the novel sequences can be targeted by re-engineering the homing endonucleases. Various studies indicate the use of meganucleases in genome editing but it is very difficult to separate cleavage domain of homing endonucleases and DNA binding. Therefore this difficulty of engineered protein with novel specificities limited the use of meganucleases. To overcome this limitation, chimeric proteins consisting of meganucleases, ZFs and TALEs have been engineered to produce novel monomeric enzymes which take advantages of ZFs and TALEs and the cleavage specificity of meganucleases.

The advantage of meganucleases technology is the DSB formation by these enzymes results in a 3' overhang which may be more recombinogenic for HDR than the 5' overhang generated by FoK 1 cleavage.

11.2.4 Crispr/cas nucleases

CRISPR-Cas RNA guided nucleases are derived from an adaptive immune system to defend against invading plasmids and viruses. Lots of work on investigation of CRISPR system in various microbial species proposed a mechanism of incorporation of short sequence of nucleic acid in to CRISPR loci. Then they are transcribed and processed in to CRISPR RNA which together with trans-activating CrRNAs (tracrRNAs), complex with CRISPR associated proteins to dictate specificity of DNA cleavage by Cas nucleases through Watson-crick base pairing between nucleic acids. All these studies propelled the CRISPR/ Cas9 technology in to the spotlight of genome editing field.

The limitation of CRISPR/Cas system derives from the necessity of a proto spacer adjacent motif (PAM) located immediately 3' to the target site. Cas9 species has specific PAM sequence. Exp. For binding and cleavage of DNA by the commonly used Cas9 from *Streptococcus pyogens*, the PAM sequence 5-NGG-3' is necessary. However, direct evolution may engineer the Cas9 variant with novel PAMs. Thus number of potential target sequence may be expanded. CrRNA and tracer RNA gets complexed with the Cas9 for a conformational change and remain associated with PAM throughout the genome and interrogating the genome sequence directly upstream to find out the sequence complementarily with gRNA. Cas9 RNA complex cleaves the target DNA by the formation of DNA RNA heteroduplex at a

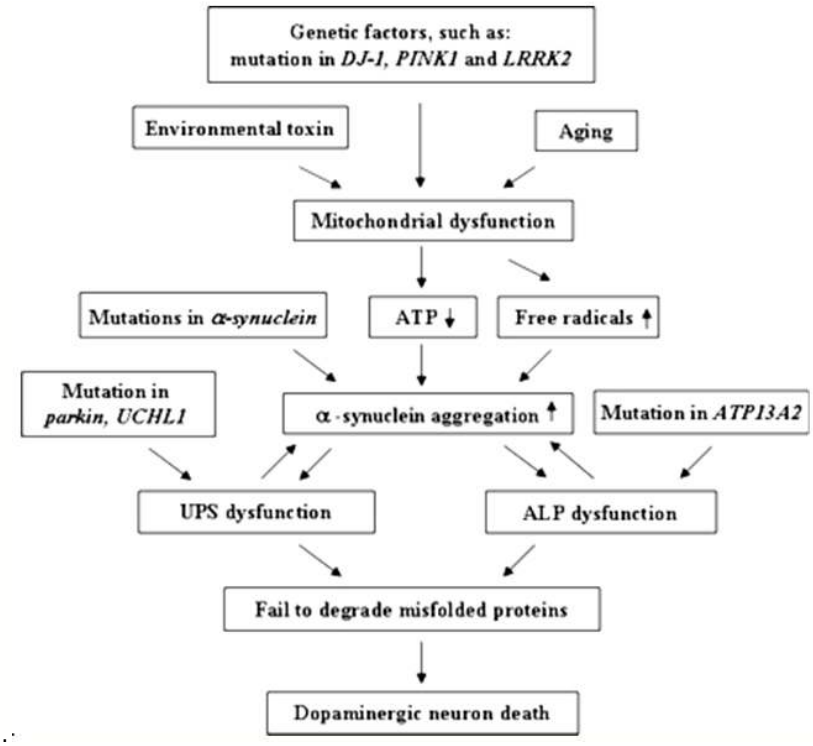

Fig. 2. Causes of protein aggregation and dopaminergic neuron death [57]

matched target site. New site can be targeted simply by altering the short region of gRNA which dictates specificity, is a highly attractive method for introducing site specific DSBs. As Cas9 is not coupled to gRNA, the DSBs can be induced at several loci by the use of multiple gRNA which makes this system highly amenable to multiplexing [56].

11.3 Autophagy

Recently, scientists have shown their interest to find out the role of ALP in neurodegeneration. Disturbances in this pathway cause neurodegenerative disease like cancer and cardiomyopathy etc.

A balance between synthesis and degradation of normal cellular protein is necessary for cell survival. Ubiquitin-proteasome system (UPS) and autophagy-lysosome pathway (ALP) degrades the most of the cytosolic and misfolded protein. Impairment of any of these pathways causes aggregation and accumulation of proteins which results in cellular toxicity and neurodegeneration like in HD.

Besides accumulation of misfolded protein, some environmental neurotoxin like Paraquat, mitochondrial proteins like Rotenone, mutant Proteins like PINK1, DJ-1 and LRRK2 increase cellular oxidative stress by mitochondrial dysfunction. Increased oxidative stress causes depletion of ATP which reduces the proteosome activity and accumulation of abnormal proteins.

Mutation of ATP13A2 which codes lysosomal ATPase and mutation of Parkin and UCH-L1, a component of UPS leads to impaired protein degradation. This impaired protein degradation leads to neuronal cell death and various other neurodegenerative disorders [57].

12. FUTURE CHALLENGES

HD is a physically, socially and psychologically devastating disorder. Most of the review studies are on motor functions in HD but there is a great impact of non-motor symptoms also. They have a major impact on patient's quality of life. Suicide is common in HD. Hence it is crucial that any behavioral symptoms that might make suicide more likely are treated as effectively and efficiently as possible. The main challenge for HD is treatment of presymptomatic individuals. Recently researchers have targeted HD and knowledge about the disease and care for the patients has increased enormously. Various

treatment options are under research and the developments are promising for the patients of HD. Still further studies and research are required for better understanding and treatment of the disease.

13. CONCLUSION

Based on the above review, it can be concluded that HD is a neurodegenerative disease, which results in the loss of motor functions, cognitive dysfunction and psychiatric disturbances. It occurs due to the mutations in the Htt gene which is located at chromosome 4 at 4p16.3 that encodes the protein huntingtin. This disease can be recognized by the psychiatric symptoms, motor symptoms and cognitive symptoms. HD can be inherited to the off springs but the chances are only 50%. But in many people this disease occurs even when there is no family history of this disease. There are diagnostic and predictive testing are available for HD. In diagnostic testing we prefer genetic testing, which is done by counting of the CAG repetition on the Htt gene. If the CAG count is more than 40 or above, then the person is said to be suffered from HD. For the HD patients no modifying treatments are available till now. Anti-depressants, transglutaminase inhibitors and chaperons are given after observing the symptoms of the disease. Chaperons are given to help the proteins to attain more stable confirmation. Currently, scientists are trying to suppress the mutant gene by the introduction of the fragments of DNA to bind with mutant Htt gene and make over the damaged protein. Symptoms of HD can also be managed by the physical therapy at all stages of the disease.

CONSENT

It is not applicable.

ETHICAL APPROVAL

It is not applicable.

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COMPETING INTERESTS

Authors have declared that no competing interests exist.

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