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REVIEW ARTICLE

A Comprehensive Review of Pharmacological and Non-Pharmacological Therapies for Huntington's Disease

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ABSTRACT

Over the last years, an increased interest in pharmacological and non-pharmacological therapeutic approaches have increased in HD research, at the moment HD remains a non-curable disease and current approaches involve symptomatic disease treatment. The systematic review identifies studies that have investigated pharmacological and non-pharmacological interventions, in HD individuals and mouse models to identify the clinical impact, regarding i) silencing and reduction of mHTT, ii) safety, efficiency and toxicity of antisense nucleotides, zinc finger proteins, CRISPR-Cas9 and transcription activator-like effector nuclease, iii) genetic modifiers contributing to early or delayed HD onset and, iv) importance of physical activity on motor and cognitive function. A systematic search of PubMed and Directory of Open Access Journal was performed by two independent reviewers using specific search term criteria for studies. The search period included studies from 2000 until 2024. The identified abstracts were screened and studies within the review fulfilled predetermined inclusion criteria. Furthermore, reference screening of included studies was also conducted. A total of forty-two studies were included. Most pharmacological studies identified that oligonucleotides, zinc finger proteins and transcription activator-like effector nuclease, reduced mHTT and HD mRNA, decreased toxicity and allele-specific targeting to prevent wild-type HTT targeting. In addition, studies involving medication such as Tetrabenazine showed to suppress HD-related chorea symptoms, while Atomoxetine did not suppress symptoms and Pridopidine showed an efficiency regarding motor symptoms. The safety and efficacy of these drugs are similar to that of other studies. Physical activity studies demonstrated that HD patients showed improvement in their gait and motor functions, an increase in cognitive function and quality of life, and a decrease in anxiety and depression, this indicates the beneficial health effects of physical activity in HD. However, further research is required to assess the full impact of pharmacological and non-pharmacological approaches in larger clinical trials assessing safety, feasibility and efficacy within HD mouse models and the HD population.

Keywords: Huntington's disease; Pharmacological; Non-pharmacological; Antisense Oligonucleotides; Genetic Modifiers; Biomarkers; Physical Activity.

Introduction

Huntington's Disease (HD) is a rare monogenic, autosomal dominant inherited progressive neurodegenerative disease (ND) with striatal pathology being the primary etiology[1]. HD is characterised by brain atrophy and neuronal degeneration of the caudate nucleus and putamen, which make up the striatum[2], specifically the medium spiny neuron (MSNs) of the basal ganglia being affected in HD[3]. Clinical characteristics involve movement, behavioral and cognitive impairments[3,4].

HD aetiology is a result of a CAG trinucleotide repeat expansion in the Huntingtin (HTT) gene, in exon 1 located on the short arm of Chromosome 4 (4p63)[5,6], responsible for encoding the HTT protein[5], this repeat expansion induces a polyglutamine tract (polyQ) at the N-terminus of the protein, resulting in a mutant HTT protein (mHTT)[3,5]. The number of trinucleotide repeats is the main predictor of age of onset (AOO) and disease severity[6], with the normal CAG repeat length between 10-35 times[3,6]. While individuals, between 36-39 CAG repeats, may or may not develop HD, thus indicating reduced penetrance[3,6] lastly, individuals with 40 or more CAG repeats will always develop HD[3,6]. The typical AOO is 40 years, with a life expectancy of 17-20 years[3,4].

HD prevalence in the Caucasian population is between 5-10 per 100,000, while Asian populations (Hong Kong, Japan and Taiwan) have lower prevalence of approximately 1-7 per million[4,5]. Moreover, in South Africa, a lower prevalence is seen in the African population in comparison to the Caucasian and Mixed-Race populations[5]. The difference regarding disease prevalence across various ethnic groups, related to genetic differences in the HTT gene[5], populations with a high prevalence have on average, longer CAG repeat lengths.

In spite of decades of intense research, there is still a lack of therapies for ND as in the case of HD, resulting in no existing cure, current treatments are symptomatic or delay disease progression, without truly affecting the underlying cause of disease[7,8]. Treatment options are categorized into pharmacological and non-pharmacological therapies, as the name suggests this involves disease treatment through the administration of medication via different routes (intravenous and orally)[9,10] and non-invasive intervention based on physical activity, sleep improvement and dietary habits respectively[11]. The current treatment options, offered to patients includes symptomatic treatment options such as Tetrabenazine which is to control the involuntary choreic movements of HD patients[7,8].

Current pharmacology therapies and research conducted in HD includes, i) gene therapy, defined as the treatment of a disease by the transfer of genetic material into cells[12], this includes non-viral gene delivery such as synthetic polymers, natural polymers, while viral gene delivery includes the use of viral vectors such retrovirus, adenovirus and adeno-associated virus (AAVs)[12], and ii) genetic modifiers and SNPs that can delay disease progression or can interact with small interfering RNAs (siRNAs) to inhibit mHTT expression. It is noteworthy to mention that several studies are currently investigating gene therapy as a potential cure for HD.

Non-pharmacological therapies in HD, mainly includes physical activity, where there has been an exponential increase over the past decade, indicating the efficacy of physical activity (PA) intervention in HD patients[13]. Numerous studies have verified the benefits of PA, in improving motor and gait function in HD patients[13–15]. There is growing research interest in non-pharmacological therapies as these are factors that patient and their caregivers can intervene resulting in the improvement of the quality of life (QoL) of patients.

There is mounting evidence focusing on pharmacological and non-pharmacological interventions and their potential effect on motor, cognitive and behavioral symptoms observed in HD. However, the benefits or non-beneficial effects of these two approaches remains to be extensively highlighted and a detailed investigation is warranted. The aim of this study, attempts to summarize studies,

evaluate and investigate the pharmacological and non-pharmacological therapies currently available in HD in both HD patients and HD mouse models, as the majority of studies involving viral vectors are conducted in mouse models.

Methods

2.1 SEARCH STRATEGY AND STUDY SELECTION

To appropriately investigate and examine the correction between pharmacological therapies of antisense oligonucleotides (ASOs), viral vectors, genetic modifiers and SNPs and non-pharmacological approaches such as physical activity conducted in HD patient derived cells or HD patients were included in the review. In addition, we included HD mouse models in the review as the majority of studies conducted are initially performed in animal models, we believe this was important information to include within this systematic review.

A literature review was initially conducted by utilizing the electronic databases of PubMed and Directory of Open Access Journal (DOAJ). The search traversed; studies published until 2000. The ensuing search terms applied were "Gene therapies AND Huntington's Disease", "Immunotherapy AND Huntington's Disease", "Clinical trials AND Huntington's Disease" and "Physical exercise AND Huntington's Disease".

A total of 3,921 articles was retrieved from the search conducted, 30 of which were duplicates. Abstracts were screened independently by two investigators; if the studies were thought to be relevant the full articles were then revised. The search period included studies from 2000 until 2024. Data was extracted from the identified studies. The cited references of the encompassed studies were further searched for any additional relevant publications. The article selection process is illustrated in (Figure 1). In addition, the PRISMA checklist has been included in the Supplementary Data (Table S3). Forty-two studies were included in the final review.

2.2 RISK OF BIAS ASSESSMENT OF STUDIES

Quality assessment tools offer researchers to focus on the concepts of importance in relation to the study's validity. This is accomplished, in order to avoid risk of bias and to provide a clear insight of understanding in regards to the limitations of the selected studies. The risk bias and quality of studies were critically assessed using the Cochrane risk of bias (https://www.cochrane.org/) tool, to assess the quality of randomized controlled trials (RCT), as the majority of studies included within our systematic review are RCTs, were an intervention either physical activity or medication were given to HD participants or HD mouse models. The domains covered in the Cochrane assessment of RCT and nested RCTs, are illustrated in (Figure S4 and Figure S5), while the Cochrane quality of analysis of RCT in (Table S7) and nested RCT (Table S8). The Newcastle-Ottawa risk assessment was used for cohort studies, prospective cohort studies and retrospective cohort studies (Figure S6), while the risk bias assessment is in (Table S9).

Results

3.1 SEARCH RESULTS

An overview of the studies included within this review, regarding studies that recruited HD participants are outlined in the Supplementary Data (Table S1), this includes, i) study reference, ii) study country, iii) study type, iii) range of participants, iv) age range and v). While an overview of studies conducted in HD mouse models (Table S2) includes i) study reference, ii) study country, iii) cell line or mouse model and iv) methodological approach.

The study types included in this review are i) genetic analysis studies ii) RCTs, iii) nested RCT, iv) randomized feasibility study, v) a cohort study, vi) cross-over controlled single blinded study, vii) prospective intervention study, viii) intervention study and ix) a retrospective cohort study. In total, 42 relevant studies were included in this review, these studies are illustrated in Tables 1-5 respectively.

To refine the study selection process, rigorous exclusion criteria were applied to ensure study relevance, applicability and quality. This includes studies that were i) non-English publications, ii) systematic reviews, meta-analysis, narrative reviews and conference proceedings, iii) full articles that could not be obtained and iv) studies not investigating the relationship between HD and gene therapies or physical activity or genetic modifiers in HD patients or HD mouse models. The above-mentioned exclusion criteria were intended to prevent the duplication of findings, and to warrant that the review is solely focused on original research studies.

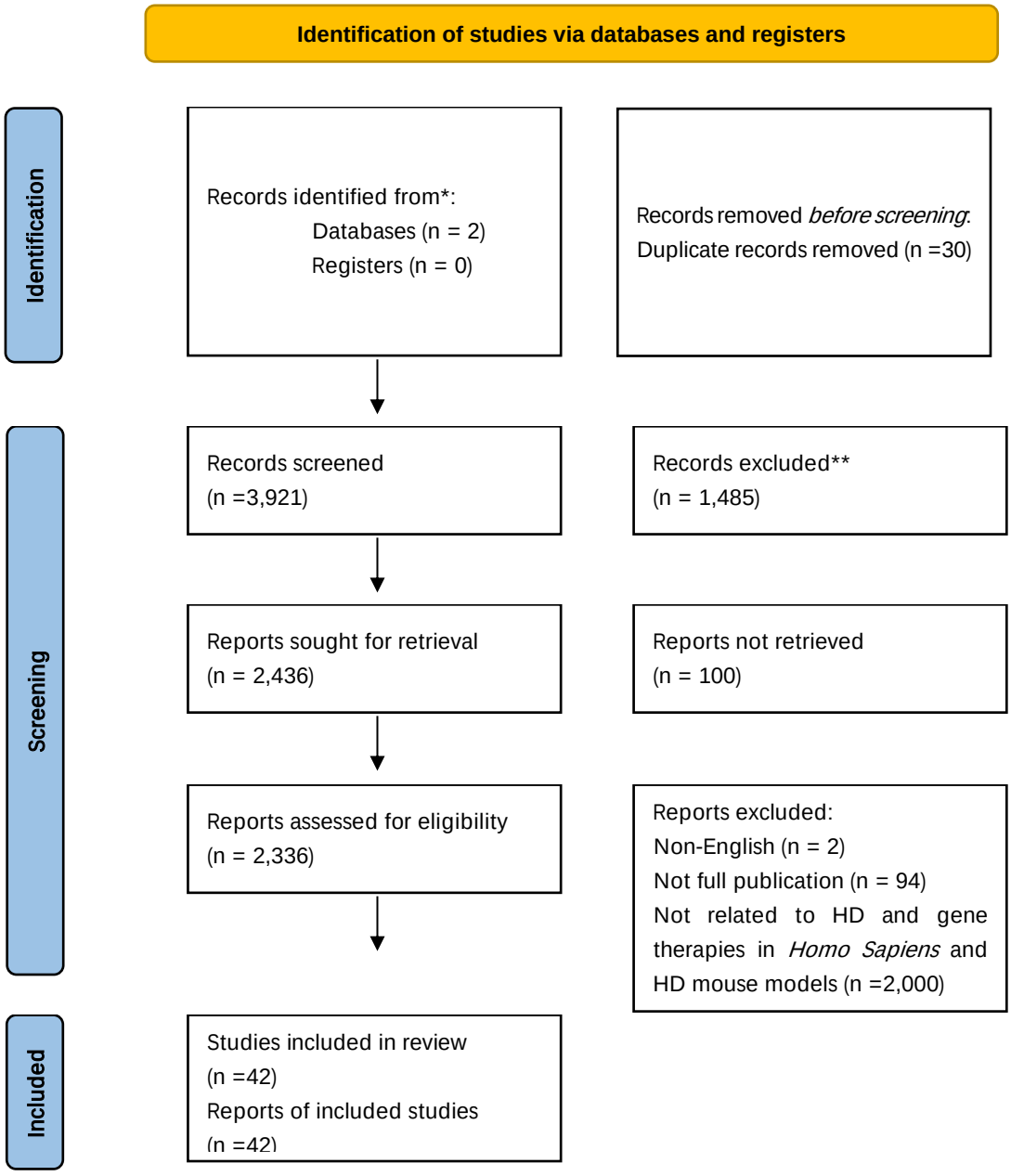

Figure 1. PRISMA article selection workflow.

Table 1. Characteristics of studies investigating the association of SNPs, Antisense oligonucleotides in Huntington's disease.

First Author, Year, and Country	Subjects and Ethnicity, N	Mean Age at Blood Collection SD Age Range (Years)	Sample Type & Methodological Approach	Clinical Outcome, Analysis & Effect Estimation	p-value	Cofounders	Clinical Conclusions
Kay et al., 2019, (Canada)[16]	Genetic Analysis Study Total HD patients (n=773) Ethnicity Swedish (n=51) French (n=53) Italian (n=88) Peruvian (n=62) Latin American (n=17) Brazilian (n=33) South Asian (n=21) East Asian (n=55) Middle Eastern (n=14) Black African (n=41) Caucasian South African (n=30) Mixed South African (n=25) Canadian (n=283)	NR	DNA	The A2 HTT haplotype is most common HD haplotype in subjects from South Asia, the Middle East, and Southern Europe. Targeting A2 HTT haplotype allows allele-specific treatment of HD-affected subjects in diverse population groups worldwide. The A2 haplotype can be selectively silenced with ASOs in patient-derived cells. ASOs targeting the A2 defining SNPs (rs362313) show significant HTT suppression *in vitro*. Novel LNA configurations of ASOs show improved HTT suppression in vitro.	0.0001* 0.01* 0.001* 0.0001* 0.0001* 0.01* 0.001*	NR	HD mutation and haplotypes A1 and A2 HTT are significantly associated, thus the possibility of targeting said haplotypes could prove to be a potential gene silencing treatment in allele-specific genes in HD populations worldwide.

First Author, Year, and Country	Subjects and Ethnicity, N	Mean Age at Blood Collection SD Age Range (Years)	Sample Type & Methodological Approach	Clinical Outcome, Analysis & Effect Estimation	p-value	Cofounders	Clinical Conclusions
Bachoud-Lévi et al., 2004 (France)[17]	Participants with either HD1 or HD2 (n=6) Female (n=3) Male (n=3)	Female 34-48 years old Male 32-40 years old	Surgery (Right Lateral ventricle), Neurophysiological tests BHK cell line as an encapsulated cell vector to release CNTF Trial period 2 years	No statistically significant benefit attributed to the effect of surgery or intracerebral release of CNTF, based on neurological, neuropsychological, and motor assessments. Mean declines in TFC and UHDRS motor score were, 0.27 _ 0.7 and 2.9 _ 7.8 at 6 months when active capsules were implanted and 0.5 _ 1.1 and 0.08 _ 5.9 with inactive capsules. Positive electrophysiological changes revealed better function of intracerebral neural circuits in three subjects.	0.1	NR	No sign of CNTF-induced toxicity was observed. Depression in three subjects after removal of the capsule, which may correlate with lack of any future therapeutic option.
Fiszer et al., 2016 (Poland)[18]	Patients with polyQ diseases (HD, SCA3, DRPLA).	NR	Fibroblasts, synthesized ONs	Huntingtin Levels A2F and WF treatments decreased HTT levels to approximately 50% of the control level. Normal Allele Expression (7 Qs) The expression of the normal allele encoding 7 Qs remained at the control level or was upregulated by A2 treatment.	< 0.05* < 0.05* < 0.05*	NR	Selected siRNAs (siRNA duplex, ss-siRNAs, sd-siRNAs) showed efficient and selective down-regulation of mHTT, ataxin-3 and atrophin.

First Author, Year, and Country	Subjects and Ethnicity, N	Mean Age at Blood Collection SD Age Range (Years)	Sample Type & Methodological Approach	Clinical Outcome, Analysis & Effect Estimation	p-value	Cofounders	Clinical Conclusions
				Mutant Allele Expression (111 Qs) In a homozygous cell line containing two mutant HTT alleles encoding 111 Qs, A2 and WF treatments efficiently downregulated huntingtin, reducing levels to 40–45% of the control level			
Pfister et al., 2009 (USA)[19]	HD participants (n=109) Non-HD controls (n=116)	NR	PCR amplicons	U isoform of rs362307 comprised 26% of HTT alleles (patients) and 6% (controls). SNP site rs363125 occurs as either A or C, a single mismatch provided high selectivity for the fully matched target A isoform >27-fold selectivity (IC50 mismatch > 20 nM; IC50 match = 0.74 ± 0.40 nM). C isoform High selectivity for SNP rs362273, occurs as either A or G, siRNA targeting A isoform demonstrated ~30 fold selectively. Fold selectivity A isoform (IC50 mismatch = 0.59 ± 0.08 nM; IC50 match = 0.02 ± 0.003 nM).	NR	NR	Targeted reduction using specific siRNAs of mHTT mRNA could prove to be an efficient method for treating HD

First Author, Year, and Country	Subjects and Ethnicity, N	Mean Age at Blood Collection SD Age Range (Years)	Sample Type & Methodological Approach	Clinical Outcome, Analysis & Effect Estimation	p-value	Cofounders	Clinical Conclusions
				G isoform ~4.9-fold selectivity (IC50 mismatch = 0.74 ± 0.11 nM; IC50 match = 0.15 ± 0.04 nM).			
Fink et al.,2016 (USA)[20]	Allele-specific reduction of mHTT	NR	Human HD Fibroblasts	No differences in toxicity between pPGK-empty vector and TALEs targeting SNP sites or CAG regions. No significant between-group differences were found in ubiquitin protein aggregation expression [F(4, 14) = 1.343]. Reduced protein aggregation observed in several TALEs. TALEs designed to target CAG collapse or gene silencing via SNP targeting effectively reduced mutant allele expression [F(4, 17) = 3.889]. TALEs were highly potent when delivered to cells, selectively reducing mutant allele expression without targeting the healthy allele.	0.320 0.383 0.027* 0.007* 0.009* 0.021* 0.010*	NR	TALEs can be used to reduce the mutant allele in the human HD fibroblasts and prove as a potential treatment method..

First Author, Year, and Country	Subjects and Ethnicity, N	Mean Age at Blood Collection SD Age Range (Years)	Sample Type & Methodological Approach	Clinical Outcome, Analysis & Effect Estimation	p-value	Cofounders	Clinical Conclusions
Yu et al., 2012 (USA)[21]	HD patient-derived cells and HdhQ150 heterozygous knock in HD-model mice (n=5)	NR	HD patient-derived fibroblasts cells, injections.	Allele-selective inhibition of HTT in Q150 HD mouse model ss-siRNA WT vs. mHTT Binding Mechanism The association of ss-siRNA with the expanded mutant repeat likely involves multiple binding events. Frontal cortex, Ipsilateral striatum, Contralateral cortex, Contralateral striatum, Thalamus, Cerebellum, Brainstem Q-PCR Findings No decrease in HTT mRNA levels in animals treated with ss-siRNA. Protein Expression ss-siRNA 537775 did not reduce expression of other proteins containing trinucleotide repeats.	< 0.01* <0.05* <0.05* <0.05* <0.001* <0.05* <0.05* < 0.01* < 0.001*	NR	Phosphonate ss-siRNA 537775 and phosphate ss-siRNA 553822, which contain mismatches at position P9, do not reduce RNA levels, potentially blocking protein translation rather than degrading mRNA.
Zhang et al., 2009 (USA)[22]	Cell lines of WT and mHTT genes	NR	HD fibroblasts (Δ2642) and neuroblastoma cells Transfections	mHTT Reduction siRNA s1 reduced mutant huntingtin by 32%. siRNA s4 reduced mutant huntingtin by 43%. siRNA 1-htt, which recognizes both alleles, reduced levels of	< 0.01* < 0.001*	NR	RNA interference can be used to specifically known down, expression of mHTT while preserving WT-HTT.

First Author, Year, and Country	Subjects and Ethnicity, N	Mean Age at Blood Collection SD Age Range (Years)	Sample Type & Methodological Approach	Clinical Outcome, Analysis & Effect Estimation	p-value	Cofounders	Clinical Conclusions
				both wild-type and mutant huntingtin. HD mRNA Reduction siRNA s4 lowered mutant HTT mRNA by 51%. siRNA s2 lowered HTT mRNA by 38%. siRNA s3 lowered HTT mRNA by 31%. Reduction of Toxicity siRNAs that specifically silence the mutant HD allele reduce H_2O_2 toxicity in host fibroblasts.			
Xu et al., 2017 (Singapore[2][3]	HD Patient-Derived hiPSCs	NR	CRISPR/Cas9 and piggyBac-based gene-editing method.	Expression of normal HTT observed in corrected hiPSC clones' post-excision. No detectable mutations were found in all ten regions examined. OT activity following CRISPR-Cas9-mediated genome editing. No mutant HTT-containing aggregates detected in neurons differentiated from HD hiPSCs. HD neurons exhibited consistent sensitivity to growth factor withdrawal.	0.038* < 0.01* < 0.05* < 0.001* < 0.01* < 0.001*	NR	Study shows the correction of HD hiPSCs and associated phenotypic abnormalities, and the importance of isogenic controls for disease modeling using hiPSCs.

First Author, Year, and Country	Subjects and Ethnicity, N	Mean Age at Blood Collection SD Age Range (Years)	Sample Type & Methodological Approach	Clinical Outcome, Analysis & Effect Estimation	p-value	Cofounders	Clinical Conclusions
				Dysregulated CHCHD2 levels were identified in HD human embryonic stem cells.			
Kordasiewicz et al., 2012 (USA)[24]	Mouse Model YAC128 and BACHD mouse models	NR	Transient infusion of HuASO (20 nucleotides in length)	Significant reduction in human HTT mRNA and protein levels, at 6 weeks. Sustained reduction in mHTT mRNA and protein, remaining suppressed to 42% and 44%, respectively. Reduction in HTT mRNA and protein was maintained in cortex and striatum, both ipsilateral and contralateral. Human HTT mRNA suppression remained at 25% in 8-month-old mice, similar to earlier reductions. ASO treatment effects lasted longer than the direct target suppression, persisting up to 15 months. HTT mRNA levels in the anterior and posterior cortex of ASO-infused animals significantly reduced to 47% and 63%, immediately after infusion.	0.005* < 0.001* 0.0012* 0.024* 0.0057* 0.0002* 0.015*	NR	Transient ASO-mediated diminition of HTT synthesis can prove to be an ideal method for maintaining HD disease reversal. ASOs suppress HTT throughout the rodent and non-human primate CNS. Suppression of mutant huntingtin reverses disease independent of wild-type levels.

First Author, Year, and Country	Subjects and Ethnicity, N	Mean Age at Blood Collection SD Age Range (Years)	Sample Type & Methodological Approach	Clinical Outcome, Analysis & Effect Estimation	p-value	Cofounders	Clinical Conclusions
Sathasivam et al., 2013 (UK)[25]	Mouse Model Knock-in HD mouse models (HdhQ20, Q50, Q80, Q100, Q150, and zQ175) (n=6) YAC128 (n=1)	NR	RT-PCR assays	Exon 1 HTT protein, detected in zQ175, HdhQ150, HdhQ100, HdhQ80, and YAC128 mouse brains, but not in WT littermates or IgG controls. A small exon 1–intron 1 polyadenylated mRNA transcript was identified in the brains of all HD mouse models expressing mHTT (mouse) or HTT (human). The pathogenic process in HdhQ150 mice could be driven by the same exon 1 HTT fragment.	0.05* 0.01* 0.001* < 0.01*	NR	RNA-targeted therapeutic approaches designed to lower the levels of HTT using ASOs, RNAi, or small hairpin RNAs could prove to be a potential therapeutic method.

ASO: Anti-sense Oligonucleotides, BDI: Beck Depression Inventory II, CNS: Central Nervous System, CNTF: Ciliary Neurotrophic Factor, DEGs: Differentially Expressed Genes, DRPLA: Dentatorubral Pallidoluysian Atrophy, GAS: Goon Attainment Scale, GFP: Green Fluorescent Protein, GO: Gene Ontology, HuASO: Human Huntingtin mRNA, HD: Huntington's Disease, hiPSCs: Human Inducted Pluripotent Stem Cells, HTT: Huntingtin, mHTT: Mutant Huntingtin, NR: Not Reported, ONs: Oligonucleotides, OT: Low Off Target, PCR: Polymerase Chain Reaction, PolyQ: Polyglutamine Diseases, Sd-siRNA: Self-duplexing siRNAs, siRNA: Short-interfering RNAs, ss-siRNA: Single Stranded Short Interfering RNA, TALE: Transcription activator-like Effector, TFC: Total Functional Capacity, UHDRS: Unified Huntington's Disease Rating Scale, WT: Wild-Type. Significant p-values: <0.05 are indicated in bold*.

Table 2. Characteristics of studies investigating the association of viral vectors in Huntington's disease mouse models.

First Author, Year, and Country	Mouse Model (N) and Vectors	Age Range (Weeks/Months)	Methodological Approach	Clinical Outcome, Analysis and Effect Estimation	p-Value	Cofounders	Clinical Conclusions
Boudreau et al., 2009, (USA)[26]	Mouse Model HD mouse model (HD-N171-82Q) and WT mice (C57BL/6) Recombinant AAV serotype 2/1 vectors (AAV1-hrGFP, AAV1-mi2.4, AAV1-sh2.4, and AAV1-sh8.2)	7 weeks old.	Transcriptional profiling Injections of AAV1 vectors on WT or HD-N171-82Q	Injection with AAV1 vectors expressing sh2.4 or mi2.4 and hrGFP reduced neurotoxicity (DARPP-32 loss and microglial activation) at 3 months. QPCR Analysis showed increased CD11b mRNA and decreased HTT proteins. Northern blot analysis showed AAV1-sh2.4 generated more HD2.4. HD-N171-82Q mice injected with AAV1-mi2.4 showed improvements in rotarod performance and reduced HTT mRNA levels at 20 weeks old.	0.001* 0.01* 0.002* 0.02* 4.678e-07* 0.0001*	Weight WT mice > HD-N171-82Q mice	HD behavioral abnormalities in the mouse model, seem to significantly improve, using Nonallele-specific silencing of mutant and wild-type HTT.
Ki Cho et al., 2019, (USA)[27]	Mouse Model HD mice (N171-82Q) and WT mice (B6C3F1/J) Total (n=56) Ten Groups WT/NT (n = 5) WT/Sham (n = 6) WT/WT (n = 6) WT/HD-NPC	NR	Injections Stem cells and gene therapy in HD mouse model N171-82Q) using Rhesus monkey neural progenitor cells (NPCs).	Rotarod performance Improved with WT-NPC injection Grip strength Improved with WT-NPC and HD-shHD-NPC injection Rotarod and grip strength progression No difference between HD/HD-NPC and HD/Sham HTT expression Reduced in HD-shHD-NPC compared to WT-NPC and HD-NPC Lifespan Increased in HD-shHD-NPC group and WT-NPC group	0.7106 0.5356 0.003* 0.0001* 0.0249* 0.0482*	NR	Combining stem cell and gene therapy could provide a potentially efficient therapeutic technique for HD patients. Combination of gene and stem cell therapy can ameliorate HD symptoms in HD mice.

First Author, Year, and Country	Mouse Model (N) and Vectors	Age Range (Weeks/Months)	Methodological Approach	Clinical Outcome, Analysis and Effect Estimation	p-Value	Cofounders	Clinical Conclusions
	(n = 6) WT/shHD-HD-NPC (n = 6) HD/NT (n = 7) HD/Sham (n=5) HD/WT-NPC (n=5) HD/HD-NPC (n=5) HD/shHD-HD-NPC (n = 5)						Combination of stem cell and gene therapy as a viable therapeutic option for HD treatment.
Harper et al., 2005 (USA)[28]	Mouse Model HD mouse model (HD-N171-82Q) and WT mice (C57BL/6) Recombinant AAV vectors AAV serotype 1 vectors	5.5 months	QPCR and DNA Analysis.	HTT-targeted shRNA, shHD2.1, reduced HD-N171-82Q mRNA levels by 85% and protein levels by 55% compared to control. shHD2.1 demonstrated directed gene silencing of endogenous HTT mRNA and protein. HD-N171-82Q mRNA was reduced by 51–55% in AAV.shHD2.1-injected HD mice, compared to AAV.shLacZ-treated or untreated HD mice. AAV.shHD2.1 reduced mHTT expression and prevented the formation of disease-associated neuronal inclusions. Significant improvements in stride length measurements (front strides	< 0.0001* < 0.0001* < 0.05* < 0.0008*	Weight	AAV.shHD2.1-treated HD mice showed dramatic behavioral improvements compared to control-treated HD mice, with only a modest, non-significant 3% drop in rotarod performance

First Author, Year, and Country	Mouse Model (N) and Vectors	Age Range (Weeks/Months)	Methodological Approach	Clinical Outcome, Analysis and Effect Estimation	p-Value	Cofounders	Clinical Conclusions
				improved by 13%, rear strides by 15%) and rotarod deficits were noted. By 10 weeks, untreated and AAV.shLacZ-injected HD mice showed impaired performance compared to all other groups, which continued to worsen. No significant decline in stride length or rotarod performance in AAV.shHD2.1-treated HD mice.			between 10 and 18 weeks.
Ekman et al., 2019, (USA)[29]	Mouse Model R6/2 HD mice B6CBAF1 mice (Female) AAV Vector AAV1-hSyn-SaCas9-HTT or AAV1-hSyn-SaCas9-mRosa26	4-week-old and 8-week-old	Plasmid-AVV Injection. Cas9 nuclease from Staphylococcus aureus, a small Cas9 ortholog that can be packaged alongside a single guide RNA into a single AAV vector	Mutant HTT Protein Inclusions R6/2 mice infused with AAV1-SaCas9-HTT had 40% fewer mutant HTT protein inclusions in dual SaCas9+ and DARPP-32+ cells compared to mice injected with AAV1-SaCas9-mRosa26. Total Mutant HTT Protein Western blot analysis revealed that mice treated with CRISPR-Cas9 had 50% less total mutant HTT protein in whole striatal lysate compared to control animals. Survival R6/2 mice injected with AAV1-SaCas9-HTT displayed a 15% increase in mean survival compared to control animals, with lifespans ranging from 80 to 106 days, compared to 66 to 98 days for control mice. Motor Function R6/2- SaCas9 treated showed improved motor function.	0.003* 0.008* 0.01* 0.01* < 0.05*	Weight	The CRISPR-Cas9 technology could prove efficient in treating HD by potentially reducing mutant HTT protein and enhance motor deficits and increase survival rates.

First Author, Year, and Country	Mouse Model (N) and Vectors	Age Range (Weeks/Months)	Methodological Approach	Clinical Outcome, Analysis and Effect Estimation	p-Value	Cofounders	Clinical Conclusions
Dey et al., 2010 (USA, France)[30]	Mouse Model YAC 128 transgenic (n=65) and WT mice (n=12) Groups YAC 128 mice that received: BDNF MSCs YAC + BDNF (n = 13) NGF MSCs YAC + NGF (n = 13) Both BDNF and NGF MSCs YAC + BDNF/NGF (n = 13) MSCs with an empty vector YAC + MSC (n = 14) DMEM vehicle solution YAC + DMEM (n = 12)	4 months old	Intrastriatal transplantation of MSCs that produce BDNF or NGF	BDNF expression Genetically engineered MSCs produced 6.8-fold more BDNF than non-transfected MSCs NGF expression Genetically engineered MSCs produced 4.6-fold more NGF than non-transfected MSCs Rotarod performance YAC + BDNF and YAC + NGF mice had longer latencies on the rotarod than YAC + DMEM mice at 15 rpm Clasping behavior YAC + DMEM mice clasped significantly more than all other groups NeuN-positive cells WT + DMEM and YAC + BDNF mice had significantly more NeuN-positive cells than YAC + NGF, YAC + BDNF/NGF, YAC + MSC, and YAC + DMEM mice GABAergic neurons WT + DMEM mice had significantly more GABAergic neurons than YAC + NGF, YAC + BDNF/NGF, YAC + MSC, and YAC + DMEM mice DARPP-32 positive cells No significant difference in the number of DARPP-32 positive cells between WT + DMEM and YAC + BDNF mice	< 0.05* < 0.05* < 0.05* < 0.05* < 0.05* < 0.05* > 0.05	NR	Intrastriatal transplantation of MSCs with over-expression of BDNF can provide results of slower neurodegenerative processes and show behavioral sparing in YAC 128 HD mouse model. Further research on the long-term safety and efficacy of this approach is needed before its potential clinical utility can be comprehensively assessed.

First Author, Year, and Country	Mouse Model (N) and Vectors	Age Range (Weeks/Months)	Methodological Approach	Clinical Outcome, Analysis and Effect Estimation	p-Value	Cofounders	Clinical Conclusions
	WT-mice injections of DMEM vehicle solution WT + DMEM (n = 12)						

First Author, Year, and Country	Mouse Model (N) and Vectors	Age Range (Weeks/Months)	Methodological Approach	Clinical Outcome, Analysis and Effect Estimation	p-Value	Cofounders	Clinical Conclusions
Pollock et al.,2016 (USA)[31]	Mouse Model YAC128 and R6/2 HD mice models Vector Lentiviral vector (pCCLc-MNDU3-BDNF-WPRE)	NR	Lentiviral transduction and transplantation	BDNF Transgene Stability The BDNF transgene was continuously present and structurally stable after integration. BDNF Production Increased BDNF production by MSC/BDNF was observed as the multiplicity of infection (MOI) increased. MSC Detection In immune-suppressed mice, MSCs could be detected for up to 28 days post-implantation in some mice. Motor Deficits and Anxiety No motor deficits were observed in YAC128 mice during open-field testing, but HD mice displayed higher levels of anxiety. Striatal Atrophy Non-modified MSCs reduced striatal atrophy, with MSC/BDNF having a more pronounced effect, indicating reduced striatal atrophy. Neuronal Marker Expression Increased expression of the immature neuronal marker doublecortin was observed in the subventricular zone of mice receiving MSC or MSC/BDNF treatment compared to vehicle-treated controls.	0.009* 0.007* 0.002* 0.038* 0.032* 0.002* 0.009*	NR	This stem cell-based delivery system for BDNF sets the foundations for cell-based therapy in the brain and prove to be an efficient modifier in HD

First Author, Year, and Country	Mouse Model (N) and Vectors	Age Range (Weeks/Months)	Methodological Approach	Clinical Outcome, Analysis and Effect Estimation	p-Value	Cofounders	Clinical Conclusions
DiFiglia et al., 2007, (USA)[32]	Mouse Model Viral transgenic mouse model of HD (AAVHtt100Q) with rapid onset.	NR	Unilateral striatal injection of AAVHtt18Q or AAVHtt100Q	AAVHtt100Q-infected mice Degenerating and shrunken HTT-labeled neurons in cortex layers 5 and 6 and the dorsal striatum. Neurons expressing AAVHtt100Q were significantly smaller in somal cross-sectional area compared to neurons expressing AAVHtt18Q. AAVHtt100Q-infected regions of the striatum had fewer neurons compared to non-injected striata. Mice co-treated with cc-siRNA-HTT had significantly more neurons in AAVHtt100Q-infected striata compared to those treated with cc-siRNA-Luc. Neurons infected with AAVHtt100Q showed strong diffuse nuclear labeling or intranuclear aggregates with anti-HTT antisera. cc-siRNA-HTT treatment significantly reduced the size of nuclear inclusions in both striatal and cortical neurons in AAVHtt100Q-infected mice. Density of anti-HTT-labeled striatal neurons with inclusions was higher in cc-siRNA-HTT-treated mice than in cc-siRNA-Luc-treated mice.	0.003* < 0.01* < 0.004* < 0.002* < 0.02* < 0.03*	NR	This method could prove an efficient therapy for HD. Improvements in neuropathology and behavior were associated with silencing of the AAVHtt100Q gene by cc-siRNA-Htt. cc-siRNA treatments did not affect animal weight or temperature.

First Author, Year, and Country	Mouse Model (N) and Vectors	Age Range (Weeks/Months)	Methodological Approach	Clinical Outcome, Analysis and Effect Estimation	p-Value	Cofounders	Clinical Conclusions
Agustín-Pavón et al., 2016 (UK)[33]	Mouse Model R6/1 HD & R6/2 HD mouse models and WT mice. Vector rAAV2/1-GFP, rAAV2/1–ZF–KOX1 and rAAV2/1–mZF–KRAB	NR	rAAV2/1-ZF-KOX1 and rAAV2/1-mZF-KRAB Steretaxic injections and pNSE	A significant negative correlation was found between RNA levels, 2 weeks after treatment, indicating that higher expression of ZF-KOX1 results in lower levels of mHTT, consistent with previous results. ZF-KOX1 resulted in a ~35% reduction in mHTT after 2 weeks, falling within the therapeutic range. Repression of mHTT was sustained at ~20% at 4- and 6-weeks post-injection. mZF-KRAB was shown to be functionally active in repressing mHTT at 6 weeks and demonstrated specificity against its target. The combination of mZF-KRAB with the pNSE promoter enabled targeted repression of mHTT in the whole brain for an extended 6-month period.	0.04* 0.05* 0.09 0.04* 0.05*	NR	Reduction of mutant HTT levels by approximately a quarter in the whole brain after a period of 6 months. Importantly, repressions of 48 % and 23 % were still detected after 12 and 24 weeks, respectively, indicating that longer term effects are possible

First Author, Year, and Country	Mouse Model (N) and Vectors	Age Range (Weeks/Months)	Methodological Approach	Clinical Outcome, Analysis and Effect Estimation	p-Value	Cofounders	Clinical Conclusions
Zeitler et al.,2019 (USA) [34]	Mouse Model HdhQ50 and R6/2 HD mouse model (n=3) Vector pression of ZFPs, plasmids were modified from the AAV vector pAAV-6P-SWB71. ZFP-B and ZFP-D	5 weeks old.	Patient-derived fibroblasts and neurons	ZFP-TFS displayed HTT repression with clear evidence of allele selectivity. ZFP-A and -B selectively downregulated mHTT mRNA and protein levels, while ZFP-C repressed both mutant and normal alleles. In NSCs, ZFP-B achieved >90% repression of the mutant allele over a 100-fold dose range, with no repression of normal HTT. ZFP treatment did not affect body weight, grip strength, or rotarod performance. Regression analysis revealed a significant negative relationship between ZFP expression and mHTT transcript levels and a positive relationship between ZFP expression and all MSN markers tested. ZFP treatment did not impact zQ175 whole-brain, striatal, and cortical volumes as assessed by MRI.	< 0.01* < 0.0001* < 0.05* < 0.01* < 0.0001*	NR	Allele-selective ZFP-TFs can be preventions and reversal towards the loss of a translational marker of HD progression. Demonstrated improvements in a range of molecular, histopathological, electrophysiological and functional endpoints. Our findings support the continued development of an allele-selective ZFP-TF for the treatment of HD

ASO: Antisense Oligonucleotides, AVV: Adenovirus Vector, BDNF: Brain Derived Growth Factor, cc-siRNAs: Cholesterol Conjugating Short Interfering RNAs, CNS: Central Nervous System HD: Huntington's Disease, HTT: Huntingtin, HuASO: Human Huntingtin mRNA, mHTT: Mutant Huntingtin, MSCs: Bone Marrow Mesenchymal Stem Cells, MRI: Magnetic Resonance Imaging, MSN: Medium Spiny Neurons, NGF: Nerve Growth Factors, NPCs: Neural Progenitor Cells, NR: Not Reported, QPCR: Quantitative Polymerase Chain Reaction, RNAi: RNA Interference, WT: Wild-Type, ZFP-TFS: Zinc Finger Protein Transcription Factors. Significant p-values: <0.05 are indicated in bold*.

Table 3. Characteristics of studies investigating the association of medication in Huntington's disease.

First Author, Year, and Country	Subjects and Ethnicity, N	Mean Age at Blood Collection SD Age Range (Years)	Medication Type	Clinical Outcome, Analysis and Effect Estimation	p-Value	Cofounders	Clinical Conclusions
Frank., 2009 (USA)[35]	RCT Participants with HD (n=75) Female (65%) Ethnicity Caucasian (95%)	50.9 ± 11.5	TBZ dosing for 13-weeks Mean dosage 63.4 (week 80)	UHDRS dysarthria score increased by 0.4 (SD 0.8) UPDRS units. No significant changes in HAM scale scores, UHDRS dysphagia score, or BAS. Mean TMC score decreased by 4.6 (SD 5.5) UHDRS units at week 80. TMC score increased by 5.3 (SD 3.2) UHDRS units at week 81. Baseline chorea score predicted a greater reduction in chorea. Significant change in CGI score by 0.3 (SD 0.7). No significant change in total motor score. No differences in TMC scores, chorea change, best dosage, or any other measure between placebo and TBZ. Reduction in total number of adverse events and somnolence during maintenance.	< 0.002* < 0.001* 0.013* 0.0054 < 0.002 < 0.01	NR	TBZ effectively suppresses HD-related chorea for up to 80 weeks.
Beglinger et al., 2009 (USA)[36]	RCT Participants with mild HD (n=20)	NR	Atomoxetine dosing for 10 weeks 80mg/d	Adverse Effects 56% of participants on Atomoxetine reported adverse effects. Commonly reported adverse effects included dry mouth (39%), loss of	0.02* 0.04* 0.05* 0.07	NR	No evident benefit of Atomoxetine in this study.

First Author, Year, and Country	Subjects and Ethnicity, N	Mean Age at Blood Collection SD Age Range (Years)	Medication Type	Clinical Outcome, Analysis and Effect Estimation	p-Value	Cofounders	Clinical Conclusions
	Female:Male Ratio 14:6			appetite (22%), insomnia (22%), and dizziness (17%). Other adverse effects reported by 11% of the sample were weight loss, headache, nausea, urinary trouble, and constipation. Cardiovascular Effects Statistically significant, mild increases in heart rate (mean increase of 9 beats/min) and diastolic blood pressure (mean increase of 5 mm Hg).			Safety and tolerability similar to other studies.
De Yebenes et al., 2011 (Germany)[37]	RCT Participants with HD (n=437) from 32 European clinics (Austria, Belgium, France, Germany, Italy, Portugal, Spain, and the UK). Female (n=222) Male (n=215) Ethnicity Caucasian Asian Latin American	50.6 (10-5)	mMS analysis and observation of 26 weeks. 45 mg/day (placebo) or 90 mg/day Pridopidine	mMS difference with 90 mg/day Pridopidine −0.99 points (97.5% CI −2.08 to 0.10). mMS difference with 45 mg/day Pridopidine −0.36 points (CI −1.44 to 0.72). Overall reduction in mMS −1.29 points (CI −2.47 to −0.12). No significant differences in non-motor endpoints.	0.042* 0.046* 0.014*	NR	Although tolerated in HD patients, Pridopidine does not have evident effect, but potential efficacy regarding the motor phenotype of HD.

First Author, Year, and Country	Subjects and Ethnicity, N	Mean Age at Blood Collection SD Age Range (Years)	Medication Type	Clinical Outcome, Analysis and Effect Estimation	p-Value	Cofounders	Clinical Conclusions
Ferrante et al., 2002 (USA)[38]	Mouse Model R6/2 and N171–82Q transgenic HD mouse models (Males)	21 days places on diet	Diets containing 0.2% coenzyme Q10 or 0.007% Remacemide, or the combination	Survival Coenzyme Q10-treated R6/2 mice showed a 14.5% increase in mean survival. Remacemide in the diet increased survival by 15.5%. Combined treatment with coenzyme Q10 and Remacemide extended survival by 31.8% in R6/2 mice and also significantly extended survival in N171–82Q mice. Motor Performance Oral administration of coenzyme Q10, Remacemide, and their combination significantly improved motor performance in R6/2 mice. Combined coenzyme Q10/Remacemide treatment showed the greatest improvement in motor behavior. Body Weight and Brain Weight Combined treatment resulted in less body weight loss compared to coenzyme Q10 or Remacemide treatment alone, with significant attenuation in N171–82Q mice. Significant decrease in brain weight was delayed until late in the disease process	< 0.001* < 0.0001* < 0.05* < 0.01* < 0.03*	Weight	Improvements of clinical and neuropathological phenotypes in the transgenic mice, from Remacemide and coenzyme Q10, indicate that multicombination therapies targeting differing pathogenic mechanisms can provide therapeutic effects.

First Author, Year, and Country	Subjects and Ethnicity, N	Mean Age at Blood Collection SD Age Range (Years)	Medication Type	Clinical Outcome, Analysis and Effect Estimation	p-Value	Cofounders	Clinical Conclusions
				with each treatment paradigm in R6/2 mice. Neuronal Atrophy and Aggregates Striatal neuron atrophy in R6/2 mice showed a 38% decrease in area measurements. Aggregate counts in the striatum at 9 and 13 weeks were significantly reduced compared to untreated R6/2 mice.			

HD: Huntington's Disease, mMS: Modified Motor Score, NR: Not Reported, UHDRS: Unified Huntington's Disease Rating Scale, TBZ: Tetrabenazine. Significant p-values: <0.05 are indicated in bold*.

Table 4. Characteristics of studies investigating the association of physical activity in Huntington's disease.

First Author, Year, and Country	Subjects and Ethnicity, N	Mean Age at Blood Collection SD Age Range (Years)	Neuropsychological and physical activity Scales or Physical activity	Clinical Outcome, Analysis and Effect Estimation	p-Value	Cofounders	Clinical Conclusions
Zinzi et al., 2007 (Italy)[39]	Intervention Study Early and Mid-stage HD disease onset (n=40) Female (n=23) Male (n=17)	Age at onset of 43.2 [10.7] years, (21 to 61 years)	Clinical Assessment Zung Depression Scale, MMSE, Barthel Index, Tinettis Scale and Physical Performance test UHDRS	Motor Performance and Daily Life Activities Each three-week treatment period led to significant improvements in motor performance and daily life activities. No carry-over effect was observed between treatment periods, but patients showed no motor decline	0.001*	NR	Intensive rehabilitation treatments may provide positive changes in maintaining

First Author, Year, and Country	Subjects and Ethnicity, N	Mean Age at Blood Collection SD Age Range (Years)	Neuropsychological and physical activity Scales or Physical activity	Clinical Outcome, Analysis and Effect Estimation	p-Value	Cofounders	Clinical Conclusions
			Treatment Programme Respiratory exercises, speech therapy, physical and occupational therapy and cognitive rehabilitation exercises.	over two years, maintaining stable functional, cognitive, and motor performance. The average score increase was 4.7 for the Tinetti test and 5.21 for the Physical Performance Test. Stability of Scores No significant changes were found in the three measures across admissions, indicating that patients maintained their initial level and did not deteriorate beyond baseline scores over time.			functional and motor performance in HD patients.
Busse et al., 2013 (UK)[40]	Randomized Feasibility Study Participants with HD (n=31) Participants who completed study (n=22) Intervention group (n=16) Control group (n=15)	50.4 [11.4] years	Clinical Assessment UHDRS-TMS and TFC Intervention group Weekly supervised gym exercise (stationary cycling and resistance exercises and twice weekly independent home-based walking programme)	Adverse Events No related adverse events were reported. Retention and Adherence Retention rate was 81%. Seven participants attended more than 75% of the gym sessions. Adherence rate among participants was 82%. Baseline Scores Baseline mean (SD) TFC scores: 8.4 (2.6) in the intervention group and 8.9 (3.1) in the control group. Baseline mean (SD) UHDRS TMS): 32.4 (15.5 in the intervention group and 35.2 (20.5) in the control group. Outcomes	0.046* 0.08 0.14 0.32 0.34 0.93	Weight	A structured exercise program can be beneficial for HD patients.

First Author, Year, and Country	Subjects and Ethnicity, N	Mean Age at Blood Collection SD Age Range (Years)	Neuropsychological and physical activity Scales or Physical activity	Clinical Outcome, Analysis and Effect Estimation	p-Value	Cofounders	Clinical Conclusions
				Treatment group had a mean MCS score 7 points higher than the control group. Some evidence of a treatment benefit for UHDRS cognitive scores. Moderate effect sizes were observed for cognitive scores and the 6-minute walk test.			
Kloos et al., 2012 (USA)[41]	Cohort Study Participants with HD (n=21)	49.3 ± 11 (25–66 range)	GAITRite walkaway under seven conditions (using no AD, a cane, a weighted cane, a standard walker, and 2, 3 or 4 wheeled walker)	Compared to no AD, using ADs decreased mean velocity. The 4WW and 3WW produced statistically equivalent gait patterns with the highest velocity, longest stride length, and narrowest BOS. 4WW produced the lowest percent time in double support. StW produced the most dissimilar gait pattern, with significantly decreased velocity and stride length, and increased percent time in double support. Standard cane and 2WW significantly reduced gait speed and stride length. 3WW resulted in the highest percent time in double support. Wheeled walkers significantly narrowed the BOS.	0.05* 0.01*	NR	4WWs prescriptions instead of other ADs for gait impairments and fall prevention can be more efficient for individuals with HD.

First Author, Year, and Country	Subjects and Ethnicity, N	Mean Age at Blood Collection SD Age Range (Years)	Neuropsychological and physical activity Scales or Physical activity	Clinical Outcome, Analysis and Effect Estimation	p-Value	Cofounders	Clinical Conclusions
Kloos et al.,2013 (USA)[42]	Cross-over controlled single blinded study Ambulatory individuals with HD (n=18)	50.7 [14.7]	Dance Dance revolution video game performance (supervised) and handheld game (unsupervised) for 45 minutes, two days per week for six weeks	Improvements in Performance Participants demonstrated significant improvements in the mean proportion of correct steps. Mean maximum correct combinations from the first two to the last two sessions. Significant improvement was observed in double support percentage during forward walking and backward walking. Significant interaction was noted for forward heel-to-heel base of support. Participant Feedback The majority of participants (17 out of 18, 94%) reported that the game was fun, motivating, and more mentally than physically challenging. Safety No falls or incidents requiring medical attention were reported related to Dance Dance Revolution gameplay. No Significant Differences No significant differences were found between treatments in forward and backward walking step time, stride length, swing time, and double support time coefficients of variation.	< 0.0001* 0.0063* 0.03* 0.01* 0.05*	NR	Dance Dance Revolution seems to be a feasible, motivating, and safe exercise intervention for HD patients. Six-week intervention improved support during walking.
Piira et al., 2013, (Norway)[43]	Prospective Intervention Study Early to mid-stage HD participants (n=37)	52.4±13.1 years	Clinical Assessment MMSE UHDRS ADL	Motor Function: Gait Significant improvement in gait was observed: TUG: -1.32 seconds.	0.03* 0.05* 0.001* 0.024*	NR	A multidisciplinary intensive rehabilitation

First Author, Year, and Country	Subjects and Ethnicity, N	Mean Age at Blood Collection SD Age Range (Years)	Neuropsychological and physical activity Scales or Physical activity	Clinical Outcome, Analysis and Effect Estimation	p-Value	Cofounders	Clinical Conclusions
	Female 51.4% (n=19)		BMI 1 year rehabilitation program consisting of three admissions of three weeks each and 5-day evaluation of 3 months after last rehabilitation admission	10MWT: -0.27 m/seconds. 6MWT: +68.71 meters Overall improvement in balance was reported. ADL Function No change was observed in ADL function. Cognitive Function MMSE showed a minor improvement of +0.67 points. FAS test (cognitive regulation) showed a minor improvement in mean score (+0.89). SDMT: measures psychomotor speed and cognitive effectiveness, showed a significant decline with a mean change of -2.87 points. No significant change was observed in mean UHDRS cognitive scores. Stroop color naming (psychomotor speed) showed a minor decline of -1.35 points. Stroop word reading showed an insignificant increase of +1.50 points. Anxiety and Depression Anxiety and depression were significantly reduced by 3.54 points BMI There was a change in BMI of +0.72 units, indicating weight gain.			program in patients with early and middle stage HD is associated with improved balance, gait function, physical quality of life and reduced depressive and anxiety symptoms.

First Author, Year, and Country	Subjects and Ethnicity, N	Mean Age at Blood Collection SD Age Range (Years)	Neuropsychological and physical activity Scales or Physical activity	Clinical Outcome, Analysis and Effect Estimation	p-Value	Cofounders	Clinical Conclusions
Piira et al., 2014, (Norway)[44]	Prospective Intervention Study Participants with early to middle stage HD (n=10) Female 50% (n=5)	50.0 ±14.0	Clinical Assessment TUG 10MWT 6MWT BBS ABC ADL MMSE SF-36 2-year intervention Rehabilitation program	Gait Slight decline or stable function with TUG +2.5 seconds, 10MWT -0.17 m/s, and 6MWT +4.3 meters (NS). Balance Minor declines in balance, BBS -1.4 points, ABC scale -8.7 points (NS). ADL-function Minor decline of -0.2 points (NS). Cognitive function MMSE improved by +1.3 points, UHDRS Cognitive assessment declined (NS). Anxiety and depression Continuous reduction. Quality of life Physical component improved by 13.0 points, mental component by 10.3 points. BMI Increase by 2.4 units, indicating weight gain.	0.85 0.12 0.88 0.03* 0.58 0.97 0.03*	NR	Participating in an intensive rehabilitation program is well tolerated among motivated early to mid-stage HD patients.
Thompson et al., 2013 (Australia)[45]	RCT Participants with early to middle stage HD (n=20)	Intervention Group 53.8 (2.9) Control Group 52.2 (2.6)	Multidisciplinary rehabilitation of Clinical and home-based exercise	Adherence to Therapy High adherence to clinical exercise and occupational therapy sessions (85%). Moderate adherence to the home-based exercise program (56%). Effectiveness of Intervention Medium–large effect on UHDRS-TMS. Small-to-medium effects noted for	0.015* > 0.05 >0.05 >0.05 >0.05 0.077 0.024*	NR	Prolonged multidisciplinary rehabilitation programmes in early to middle stage HD patients is efficient, well-tolerated and

First Author, Year, and Country	Subjects and Ethnicity, N	Mean Age at Blood Collection SD Age Range (Years)	Neuropsychological and physical activity Scales or Physical activity	Clinical Outcome, Analysis and Effect Estimation	p-Value	Cofounders	Clinical Conclusions
				D-KEFS HVLT-R BDI-II QOL assessments Postural stability Walking around the house Walking up or downstairs SDMT Written Correct Incorrect Oral Correct Incorrect Goal Achievement GAS revealed partial or complete achievement of goals in 7 out of 9 participants.	>0.05 0.042* >0.05 >0.05 >0.05		therapeutically beneficial.
Quinn et al.,2014 (UK)[46]	RCT Multi-Site UK Study Participants with mid stage HD Females (n=17) Males (n=13) Total (n=30) Ethnicity NR	57.0 (10.1)	Intervention task specific training performances, focusing on walking, sit-to-stand transfers and standing twice a week for 8 weeks.	Minimal loss to follow-up Excellent adherence in the intervention group, with 96.6% adherence. 92% of goals were achieved by the end of the intervention. 46% of participants displayed better-than-expected results. Effect density was small across all aspects.	NR	NR	The excellency of this method seems to have potential but the design of this intervention is possibly unable to provide systematic conclusions.

First Author, Year, and Country	Subjects and Ethnicity, N	Mean Age at Blood Collection SD Age Range (Years)	Neuropsychological and physical activity Scales or Physical activity	Clinical Outcome, Analysis and Effect Estimation	p-Value	Cofounders	Clinical Conclusions
Quinn et al., 2016 (UK)[47]	RCT HD participants assessed (n=314) Not meet inclusion criteria (n=248) Declined (n=34) HD participants recruited (n=32) Six multi-sites	Intervention Group 53 (11) (22-69) Control Group 51 (17) (19-76)	Exercise Program-Intervention was 12 weeks, three per week (aerobic), upper and lower body strengthening exercises. Intervention Group (n=17) Control Group (n=15)	92.9% (13/14) participants completed >75% of required sessions (95% CI [64.2, 99.6]). Improvement in predicted VO2 in the intervention group. UHDRS motor score Intervention arm showed a 2.9-point lower score (95% CI [5.42, 0.32]). Weight Intervention arm was 2.25 kg lighter.	0.02* 0.03*	NR	Short-term exercise intervention is safe and beneficial, especially when the exercise intervention is structured, intense, balanced and monitored.
Quinn et al., 2022 (UK)[48]	Nested RCT ENROLL-HD participants from Germany, Spain and USA Total (n=59) RCT Control (n=26) RCT Intervention (n=26) Ethnicity Caucasian (88%)	Cohort 52.4 (11.1) RCT Control 57.1 (9.8) RCT Intervention 54.5 (10.5)	Clinical Assessment UHDRS-TMS & UHDRS-TFC FA SDMT PBA-s HADS-SIS SF-12 TUG 30sCST Intervention 12-month trial of Physical activity	Performance-based measures, including predicted VO2max, 6-minute walk test, and TUG, as well as self-reported PA, all declined over the 12-month period in the cohort group. Effect sizes for VO2max, 6-minute walk test, and IPAQ in the cohort group were −0.27, −0.40, and −0.29, indicating small-to-moderate declines in all measures. In the control group, effect sizes for these measures were −0.06, 0.07, and −0.32, indicating minimal change or a small negative effect.	NR	NR	PACE-HD displays strong foundations for future, holistic trials.

First Author, Year, and Country	Subjects and Ethnicity, N	Mean Age at Blood Collection SD Age Range (Years)	Neuropsychological and physical activity Scales or Physical activity	Clinical Outcome, Analysis and Effect Estimation	p-Value	Cofounders	Clinical Conclusions
	Latino, American Indian, African, mixed or other (12%)			In the intervention group, effect sizes were 0.08, 0.25, and 0.07, indicating no change or a small positive effect, suggestive of improvement. Decline rates for UHDRS-TMS, TFC, and SDMT in the cohort group were consistent with previous studies.			
Dawes et al., 2014 (UK)[49]	RCT Participants with HD (n=30) Healthy control group (n=20) Total (n=50)	HD group 51± 11 (29-71) Healthy group 51± 11 (30-71)	12-week gym and walking Exercise training program	Gradual increase in HR throughout the exercise. No linear relationship DBS and HR changes between minutes two-three or eight-nine. HD vs Control Group Work Rate phase a min 3 RPE three (3±2(0-7) vs 1±1(0-4)) Work Rate phase b min 9 (7±3(1-10) vs 5±2(2-9)) Heart rate phase b min 9 RPE phase b min Weak relationship between UHDRS motor score and HR change between minutes two and three, no relationship between minutes eight and nine.	0.10 0.003* 0.00 0.04* 0.65 0.03*	NR	Individuals with HD failed to achieve a steady-state HR during the first phase of exercise. Reduced aerobic metabolism capacity in people with HD.

First Author, Year, and Country	Subjects and Ethnicity, N	Mean Age at Blood Collection SD Age Range (Years)	Neuropsychological and physical activity Scales or Physical activity	Clinical Outcome, Analysis and Effect Estimation	p-Value	Cofounders	Clinical Conclusions
Khalil et al., 2012 (UK)[50]	Exploratory mixed-method design Participants with HD (n=15)	53.6 (25-69)	Home-Based Exercise DVD	The UHDRS cognitive subscale score and IMI pressure/tension subscale score were significantly different between participants who adhered to the program and those who did not. No significant differences were observed in the TFC score or other IMI subscales. Adherence rate was significantly correlated with the TFC score, UHDRS cognitive subscale score, and IMI pressure/tension subscale score. Correlations of adherence rate with IMI subscales such as interest/enjoyment, perceived competence, effort/importance, and value/usefulness were not significant.	0.003* 0.04* 0.34 0.85 0.14 0.23 0.41	NR	The DVD shows to be a supportive and suitable method to engage HD patients in exercise at different types of environments (home/therapies/outside).
Cruickshank et al., 2015 (Australia)[51]	Intervention study Participants with HD Total (n=15)	43.6± 2.2	Clinical Assessment HVL Test UHDRS-TMS CWIT D-KEFS 9-month Multidisciplinary exercise	Increased gray matter volume in right caudate and bilaterally in dorsolateral prefrontal cortex after 9 months of multidisciplinary rehabilitation. Improvements in verbal learning and memory. A significant association was found between gray matter volume increases in the dorsolateral prefrontal cortex and performance on verbal learning and memory	<0.05* 0.0130*	NR	Multidisciplinary rehabilitation positively impacts gray matter changes and cognition which relates to verbal learning and memory.

First Author, Year, and Country	Subjects and Ethnicity, N	Mean Age at Blood Collection SD Age Range (Years)	Neuropsychological and physical activity Scales or Physical activity	Clinical Outcome, Analysis and Effect Estimation	p-Value	Cofounders	Clinical Conclusions
Cruickshank et al., 2018 (Australia)[52]	RCT Assessment 1 (n=25) Randomisation (n=22) Participants with HD Total (n=18) Females (n=9) Males (n=9)	Training group 53.8 ± 2.9 Control group 51.2 ± 2.7	Clinical Assessment BBS 10MWT 10MWT-SP 6MWT Multidisciplinary therapy performances	No adverse events occurred throughout the intervention. High adherence to the supervised exercise program (80.5%) and cognitive therapy sessions (84.2%). Moderate adherence to the home exercise program (56.0%). Significant difference in NBT outcomes between groups, with deterioration in the usual care group and no changes in the training group. Significant improvements in knee extension and flexion strength in the training group.	0.042* 0.008* 0.001* 0.002* 0.019* 0.616 0.844 0.250 0.123 0.332 0.174	NR	Outpatient multidisciplinary therapy has potential due to the positive effects displayed on muscle strength and manual dexterity. But no effects on mobility, cardiorespiratory endurance and balance in HD patients.

10MWT: 10 Meter Walk Test, 6MWT: 6 Meter Walk Test, 30sCST: 30 Second Sit to Stand Test, 2-WW: 2-Wheel-walker, 3-WW: 3-Wheel-Walker, AD: Assistive Device, ADL: Activities of Daily Living, BBS: Berg Balance Scale, BDI-II: Becks Depression Inventory-II, BMI: Body-Mass Index, BOS: Base of Support, CWIT: Colour Word Interference Test, DBS: Deep Brain Stimulation (DBS), D-KEFS: Delis-Kaplan Executive Function System, FA: Functional Assessment, GAS: Goal Attainment Scaling, HADS-SIS: Hospital Anxiety and Depression Scale-Snaith's Irritability Scale, HD: Huntington's Disease, HR: Heart Rate, HVLT-R: Hopkins Verbal Learning Test Revised, IMI: Intrinsic Motivation Inventory, MMSE: Mini-Mental State Examination, NR: Not Reported, PBA-s: Problem Behaviours Assessment, QoL: Quality of Life, RCT: Randomized Controlled Trial, RPE: Rated Perceived Exertion, SDMT: Symbol Digit Modalities Test, SF-12: Short Form Survey 12, SF-36: Short Form Health Survey 36, TUG: Timed up and Go Test, UHDRS: Unified Huntington's Disease Rating Scale, UHDRS-TMS: Unified Huntington's Disease Rating Scale-Total Motor Score, UHDRS-TFC: Unified Huntington's Disease Rating Scale-Total Functional Capacity. Significant p-values: <0.05 are indicated in bold*.

Table 5. Characteristics of studies investigating the association of genetic modifiers and potential biomarkers in Huntington's disease.

First Author, Year, and Country	Subjects and Ethnicity, N	Mean Age at Blood Collection SD Age Range (Years)	Methodological Approach	Clinical Outcome, Analysis and Effect Estimation	p-Value	Cofounders	Clinical Conclusions
Becánović et al., 2015 (USA)[53]	Genetic Analysis Study Participants with HD Danish Cohort (n=36) B-haplotype (n=8) Ethnicity Danish UBC Cohort (n=NR) Ethnicity Canadian and European EHDN Registry (n=NR)	NR	Reporter Assays	The construct 5 variant of the NF-κB binding site significantly reduced transcriptional activity. Sequence changes at both the first and last positions of the NF-κB binding site also led to a significant reduction in transcriptional activity of the HTT promoter. TNFα and CAPE stimulations, as well as siRNA knockdown of NF-κB, modulated the HTT promoter's transcriptional activity. HTT protein levels were reduced by approximately 50%. The rs13102260 regulatory variant altered NF-κB binding, reduced HTT protein levels, and was associated with modulation of AOO in patients.	< 0.0001* < 0.0001* 0.0183* 0.0495* 0.0169*	NR	rs13102260:G > A, impaired NF-kB binding and reduced HTT transcriptional activity and HTT protein expression. Presence of the rs13102260 minor (A) variant associated with delayed age of onset in familial cases HTT gene silencing displays promising results for HD treatment.

Goold et al., 2019 (UK)[54]	Genetic Analysis Study GeM Cohort (n=4,082) HD progression Track+Registry Cohort (n=2,078) Control Dorsolateral prefrontal Cortex (Common Mind Consortium) (n=452)	NA	TWAS & PCR	Endogenous FAN1 expression significantly slowed CAG repeat expansion in cells (FAN1+/+: 1 CAG unit increase per 17.7 ± 1.4 days). Cells expressing GFP–FAN1 at near endogenous levels showed similar stabilization (1 CAG unit increase per 14.8 ± 1.1 days). Tet-induced overexpression of GFP–FAN1 further decreased the CAG repeat expansion rate (1 CAG unit increase per 21.3 ± 1.8 days). Knockdown (KD) of FAN1 significantly increased the CAG expansion rate to 1 repeat every 9.1 ± 0.5 days. FAN1 expression was reduced at both mRNA and protein levels in knockdown experiments.	0.000269* 0.000257* 0.00167* 0.0806	NR	TWAS data strongly suggest that FAN1 expression plays a role in modifying HD onset and progression. FAN1 affects somatic expansion of CAG repeat through a nuclease-independent mechanism. Increased FAN1 expression is significantly associated with delayed AAO of HD.
Kay et al., 2018 (Canada)[55]	Genetic Analysis Study HD participants (n=3,907) Ethnicity North Americans Hispanic American African-American or East Asian North of Scotland, Swedish, Danish, Finnish, Italian, African	NR	Saliva-extracted DNA samples	Mean CAG Repeat Length Highest in self-identified Hispanic individuals from the U.S., lowest in African South Africans. Higher in Scottish Europeans, Causcian South Africans, and North Americans than in all African ancestry groups. Consistently ≥18.0 in all European ancestry populations. Consistently <17.5 in African, Amerindian, and East Asian ancestry groups. Intermediate Alleles	0.0013* 0.0202* 0.0002* 0.0053* 0.0091* 0.0029* 0.0244* 0.0072*	NR	Differences of HD prevalence worldwide can be explained from new frequent HD mutations in populations as a function of IA frequency and haplotype composition. Overall HD new mutation rate in the Western population was 0.01745%, or 1

				Frequent in Northern Scottish and North American cohorts compared to mixed ancestry South Africans, Black South Africans, and East Asians. Higher IA frequency in Northern Scottish population than in North Americans. IAs occurred more frequently on A1 haplotype in Northern Scottish cohort compared to North Americans. Haplotype and Mutation Rate The HD-associated A3a haplotype found in European ancestry populations was absent in mixed ancestry and Black South Africans. Scottish Europeans had a high IA frequency (3.71%), leading to a high inferred new mutation rate of 0.0307% HD expansion events, or 1 in every 3,257 births. African South Africans and East Asians had the lowest IA frequency and inferred new mutation rates (<1 new HD expansion per 25,000 births). Populations with a higher A1 haplotype frequency may have a higher new mutation rate for HD.		in 5,372 births expected to have a paternally transmitted new mutation.

Byrne et al., 2017 (UK)[56]	Retrospective Cohort Study 3-year TRACK-HD study Mutation carriers (n=201) Healthy Controls (n=97) Total (n=298) CSF Cohort Mutation carriers (n=23) Healthy Controls (n=14) Total (n=37)	NR	NR	NfL levels in plasma at baseline were significantly higher in HTT mutation carriers compared to controls. Baseline NfL concentration in plasma correlated significantly with subsequent cognitive decline, TFC, and brain atrophy. NfL concentration in plasma at baseline was associated with subsequent clinical onset during the 3-year follow-up period. Concentrations of NfL in CSF and plasma were correlated in HTT mutation carriers.	< 0.0001* < 0.0001* 0.0033* 0.0264* 0.0036* 0.602	NR	NfL in plasma can be a potential prognostic blood biomarker of disease onset and progression in HD. NfL quantification of concentrations in plasma to be included in future observational and therapeutic trials for HD.
Moss et al., 2017 (UK)[57]	GWAS Study TRACK-HD (n=218) EHDN REGISTRY (n=1,773)	NR	TRACK-HD and EHDN REGISTRY Phenotypic data. GWAS TRACK-HD (n=216) EHDN REGISTRY (n=1,773)	TRACK-HD Longitudinal motor, cognitive, and imaging scores were correlated with each other. Locus on Chr 5 spanning DHFR, MSH3, and MTRNR2L2. MAGMA Analysis showed significance in MTRNR2L2, MSH3, DHFR Progression measures in TRACK-HD and REGISTRY were correlated with each other and with age at onset. A meta-analysis of progression in TRACK-HD and REGISTRY revealed a genome-wide significant signal on chromosome 5, spanning three genes: MSH3, DHFR, and MTRNR2L2.	1.12×10^{-10}* 5.8×10^{-8}* 2.15×10^{-9}* 2.94×10^{-8}* 8.37×10^{-7}* $2{\cdot}94 \times 10^{-8}$* $8{\cdot}37 \times 10^{-7}$* 2.15×10^{-9}*	NR	The MSH3 gene could probably be a modifier of disease progression in HD.

				Genes in this locus were associated with progression in TRACK-HD. In TRACK-HD, each copy of the minor allele at the identified SNP was associated with a 0.4 units/year reduction in the rate of change of the UHDRS Total Motor Score (95% CI 0.16–0.66) and a 0.12 units/year reduction in the rate of change of UHDRS TFC score (95% CI 0.06–0.18).		

AO: Antisense Oligonucleotides, CSF: Cerebrospinal Fluid, EHDN: European Huntington's Disease Network, FAN1: FANCI Associated Nuclease 1, GFP: Glial Fibrillary Acidic Protein, GWAS: Genome Wide Association Studies, HD: Huntington's Disease, HTT: Huntingtin, KD: Knockdown, NF-κB: Nuclear Factor Kappa B, NfL: Neurofilament Light, TFC: Total Functional Capacity, TNFα: Tumor Necrosis Factor Alpha, TWAS: Transcriptional Wide Association Study, UBC: University of British Columbia, UHDRS: Unified Huntington's Disease Rating Scale. Significant p-values: <0.05 are indicated in bold*.

3.3 ANTISENSE OLIGONUCLEOTIDES

The momentum in ASOs advancement, have allowed their use, in a clinic setting[58]. Several studies[16,17,59], have been conducted regarding ASOs in HD patients or HD cell lines, these studies are described below.

A genetic analysis study in several ethnic populations affected by HD was conducted by Kay et al., 2019[16], to investigate how ASOs can affect HD-related haplotypes within specific populations. The study, that targeting the A2 HTT haplotype, allowed for allele specific treatment of most HD affected participants in a wider diverse population[16], this highlights A2 haplotype may be selectively silenced. ASOs targeting A2, proved significant HTT suppression *in vitro*. This indicates the potential of ASO targeting of specific HD-related haplotypes to suppress mHTT in affected populations[16].

Ciliary neurotrophic factor (CNTF) is a hypothalamic neuropeptide, promoting survival of neurons and oligodendrocytes *in vitro* and *in vivo*. Experimental studies have shown the capacity of CNTF and its role in protecting MSN[17,59]. The study by Bachoud-Levi et al., 2004 [17], investigated CNTF as a neuroprotective gene therapy approach in HD participants via encapsulated cell release of CNTF. However, no statistically significant results were detected in HD individuals either through surgery or intracerebral release of CNTF, based on neurological, neuropsychological and motor assessments performed. Although no noteworthy results were found this gene therapy approach exhibited safety, tolerability and feasibility in study participants [17].

Fiszer et al.,[18], investigated gene silencing on fibroblasts using synthesized oligonucleotides (ONs) in HD, Spinocerebellar Ataxia 3 (SCA3) and Dentatorubtal Pallidoluysian Atropy (DRPLA) which are all polyglutamine disease. In this review, we mainly focused on the HD aspect of this research work. Fiszer et al.,[18], observed that in homozygous cell lines with two mHTT alleles encoding for 11Qs, that treatments with A2 and WF ONs treatments, efficiently down-regulated HTT. This concentration was approximately 40-45% of that of control levels. Regarding normal allele expression (7Qs), this remained either at the control concentration or was up-regulated by A2 ON treatment. The study illustrates that ONs prove to be efficient as a potential therapeutic treatment in HD[18].

The study by Pfister et al.,[19], investigated siRNAs that may well target SNPs in the HTT gene in HD participants and non-HD controls [19]. The rs362307 SNP comprised 26% of HTT alleles among patients but only 6% among controls. The SNP rs363125 is high selectivity for the fully matched target of the HTT allele. The study demonstrates high selectivity for SNP rs362273, at either A or G. siRNA targeting the A isoform, was shown to have ~30 fold selectively. Targeted reduction using specific siRNAs of mHTT mRNA could verify to be an effective method for treating HD[19].

Allele-specific decrease of mHTT using transcription activator like effectors (TALEs) are designed to target SNPs in mutant alleles and packaged into a vector backbone containing a KRAB to promote transcriptional repression of the disease associated allele as investigated by Fink et al.,2016[20]. TALEs were designed to silence and target CAGs via SNP binding, thus effectively decreasing mutant allele expression in human HD fibroblasts[20]. It is noteworthy to mention that TALEs are highly potent and displayed selectivity in reducing mutant allele expression, without targeting the healthy allele. This indicates that TALEs provide a stepping stone for targeted treatment in HD[20].

The silencing of mHTT using techniques such as ASOs or ONs would ultimately eliminate the root cause of HD. Yu et al., 2012[21] developed single-stranded antisense oligonucleotides (ss-siRNAs) to target HD patient and HD mouse model, fibroblast cells [21]. ss-siRNAs are potent and allele selective inhibitors of mHTT expression in HD patient derived cells. Moreover, they are optimised to discriminate between mutant and WT alleles. Study findings[21], showed no decrease in HTT mRNA levels in HD mouse models treated with ss-siRNAs. Furthermore, the ss-siRNAs of phosphonate ss-siRNA 537775 and phosphate ss-siRNA 553822 do not decrease

RNA levels, resulting in potentially blocking protein translation rather than degrading mRNA.

An additional study, involving allele specific silencing using siRNA of mHTT in HD fibroblasts and neuroblastoma cells was conducted by Zhang et al., 2009[22]. The study reported that siRNA s1 reduced mHTT levels by 32% and siRNA s4 by 43%. However, the siRNA s1 recognizes both HTT alleles, therefore, reducing the mutant and WT HTT alleles[22]. siRNA s4 lowered mHTT mRNA levels by 51%, siRNA s2 by 38% and siRNA s3 by 31%[22]. The following gene silencing approach, proves to be effective across a variety of cells, including neuronal cells. This approach shows great potential as it may lead to personalized HD gene therapy approaches.

CRISPR/Cas9 is a gene editing technology involving two vital components, the guid RNA to match the desired target gene and the Cas9, endonuclease enzyme that causes a ds-DNA break, permitting modifications to the genome[60]. To this end, Xu et al., 2017[23], aimed to reverse phenotypic abnormalities using CRISPR/Cas9 in HD patient derived induced pluripotent stem cells (hiPSCs). Expression of WT-HTT was detected in corrected hiPSCs. Furthermore, no mHTT aggregates were detected in neurons, differentiated from HD hiPSCs[23]. The study by Xu et al., 2017[23], demonstrated the potential of CRISPR/Cas9 to be used in rare neurological diseases such as in HD, to correct HD hiPSCs and phenotypic abnormalities associated with disease[23].

Kordasiewicz et al., 2012[24], were interested to observe the effect of ASO, by infusion into the CSF of YAC128 and BACHD mouse models[24]. mHTT mRNA and protein, remained suppressed at 42% and 44%, respectively. A decrease in HTT mRNA and protein was maintained in cortex, striatum, ipsilateral and contralateral brain regions. HTT mRNA levels in the anterior and posterior cortex of ASO-infused animals were significantly reduced to 47% and 63%, respectively, directly after infusion[24]. Human HTT mRNA suppression remained at 25% in 8-month-old mice, similar to that seen in earlier reductions [24]. ASO treatment lasted up to 15 months, therefore, rather than requiring continuous treatment, these findings can launch a therapeutic strategy for continuous HD disease reversal[24].

HTT fragments have been implicated in HD pathogenesis, they have been observed in HD post-mortem brain tissue, these nuclear inclusion fragments can only be detected by antibodies. Therefore, there is an unmet need to determine the precise length and mechanisms by which these fragments are generated, which still remains unknown. A study by Sathasivam et al., 2013[25], were interested to investigate how splicing of exon 1 may results to HD pathogenesis in knock-in HD mouse models (HdhQ20, Q50, Q80, Q100, Q150, zQ175 and YAC128)[25]. The study, exemplified that aberrant splicing of exon 1, leads to a short polyadenylated mRNA that is encoded into the HTT protein[25]. The polyadenylated mRNA transcripts were observed in all HD mouse models, furthermore, the exon 1 HTT protein was detected in all HD mouse models apart from the WT and IgG control mice[25]. RNA-targeted therapeutic approaches are designed to lower HTT levels using ASOs, RNAi, or small hairpin RNAs that could attest to be a potential therapeutic technique for HD and other NDs[25].

3.4 VIRAL VECTORS IN HD MOUSE MODELS

The studies[26–33], identified that investigated viral vectors in HD mouse models are described below. Boudreau et al., 2009[26], conducted a study in HD-N171-82Q and WT mice (C57BL/6) HD mouse models, where a recombinant AAV1 vector was injected in WT and HD mouse models. Researchers[26], observed that HD-N171-82Q mice injected with AAV1-mi2.4 showed improvements in rotarod performance and reduced HTT mRNA levels at 20 weeks old. Injection with AAV1 vectors expressing sh2.4 or mi2.4 and hrGFP reduced neurotoxicity at 3 months. The study by Boudreau et al., 2009[26], demonstrated that non-allele silencing of mHTT and WT-HTT exhibits therapeutic efficacy in HD mice.

The study by Ki Cho et al., 2019[27], aimed to demonstrate the efficacy of a stem cell and gene therapy combination in HD mice (N171-82Q) using Rhesus monkey neural progenitor cells (NPCs),

whereas, the WT-NPCs, transgenic HD monkey (HD-NPCs) and genetically modified HD-NPCs have decreased mHTT levels, these were grafted into the striatum of WT and HD mice[27]. Motor and grip strength improved in WT-NPC and HD-shHD-NPD grafts in HD mouse models. Mice that received HD-shHD-NPD grafts, presented a significant increase in their lifespan in comparison to sham injection and HD mice groups[27]. Study results, suggest a combination of stem cell and gene therapy, ameliorates HD symptoms in HD mice, allowing the following a viable therapeutic option for HD treatment.

Harper et al., 2005[28], were interested in the role of RNAi using a AAV serotype vector in an HD-N171-82Q HD mouse model[28]. The study showed that HTT-targeted shRNA, shHD2.1, reduced HD-N171-82Q mRNA levels by 85% and protein levels by 55% compared to control (C57BL/6). Direct gene silencing of endogenous HTT mRNA and protein was seen for shHD2.1[28]. It is noteworthy to mention, that sustained expression and proper processing of shRNAs were observed in the striatum 3 weeks following transduction. AAV.shHD2.1-treated HD mice showed behavioral improvements compared to control-treated HD mice[28]. This indicates the potential for further development of a RNAi direct therapeutic approach.

Ekman et al., 2019[29] used CRISPR-Cas9 within a plasmid-AAV vector to effectively deliver it in the striatum of the R6/2 HD mice. The R6/2 mice infused with AAV1-SaCas9-HTT had 40% fewer mutant HTT protein inclusions compared to mice injected with AAV1-SaCas9-mRosa2. HD mice, treated with CRISPR-Cas9. Had 50% fewer total mHTT protein in comparison to control HD mice. Furthermore, the lifespan and motor function improved in R6/2 mice treated with SaCas9[29]. This illustrates the use of CRISPR-Cas9 technology to potentially treat HD and other trinucleotide repeat expansion diseases[29].

The aim of the study by Dey et al., 2010[30], was to evaluate the therapeutic properties of transplanted mesenchymal stem cells (MSCs), which are genetically engineered to over-express brain derived neurotrophic factor (BDNF) or nerve growth factor (NGF) to observe their effect on motor impairment and neuronal degeneration in YAC128 HD vs WT mice[30]. The study identified that BDNF expression increased by approximately 7-fold and NGF by 5-fold in genetically engineered MSCs cells in comparison to non-transfected MSCs. During rotarod performance, the YAC 128+BDNF, have longer latencies and minimum neuronal loss within the striatum[30]. An additional study by Pollock et al., 2016[31], intended to investigate genetically engineered MSCs to over-express BDNF using lentiviral vectors for transduction and transplantation in YAC128 and R6/2 HD mice models[31]. An increase in BNDF concentration was observed as the multiplicity of infection increased. In regards to motor and behavioural impairment, no motor deficits were observed in YAC128 mice, while the R6/2 HD mice displayed increased levels of anxiety. Moreover, an increase in the lifespan of R6/2, MSC/BDNF treated mice was observed. The study by Dey et al., 2010[30] and Pollock et al., 2016[31], illustrates that intrastriatal MSCs transplantation and over-expression of BDNF generates an environment in the striatum, that slows down neuronal loss and neurodegenerative process in HD mouse models.

Cholesterol-conjugated small interfering RNA duplexes (cc-siRNA) which target the human HTT mRNA (siRNA-HTT) were injected into a transgenic mouse model of HD AAVHtt100Q or AAVHtt18Q. The aim of the study by DiFiglia et al., 2007[32], observed whether siRNA attenuates striatal and cortical pathology and behavioral impairment in HD. Initially, cc-siRNA-HTT treatment significantly decreased the size of nuclear inclusions in striatal and cortical neurons in AAVHtt100Q-infected mice. Mice treated with cc-siRNA-HTT had significantly more neurons in AAVHtt100Q-infected striata compared to those treated with cc-siRNA-Luc. Additionally, treatment of cc-siRNA-HTT in mice with mHTT, prolonged striatal neuron survival, reduced aggregates, diminished inclusion size, and lowered occurrence of clasping and foot-slips on the balance beam[32]. The findings indicate that siRNA targeting HTT, results in mHTT silencing which

leads to improvement of behavioural impairment and decreases neuropathology associated with HD[32].

A host-matched analogue of ZF–KOX1(MZF-KRAB) in combination with an rAAV vector was developed by Agustin-Pavón et al., 2016[33] where it was used to safely deliver the ZF to R6/1 HD and R6/2 HD mouse models[33]. A negative correlation was seen between RNA levels. 2-weeks of ZF-KOX1 treatment, indicated increased ZF-KOX1 expression and lower mHTT levels[33]. A 35% decrease of mHTT was observed following treatment. A combination of mZF-KRAB and pNSE promoter, permitted repression of mHTT in the whole brain for a 6-month period, therefore, signifying the long-term effects are imaginable[33]. Another study by Zeitler et al.[34], engineered zinc finger protein transcription factors (ZFP-TFs), to directly target CAG trinucleotide repeats, while selectively lowering mHTT[33]. ZFP-TFs were transported using an AAV vector, and injected into HdhQ50 and R6/2 HD mouse models[33]. The study demonstrated, ZFP-TFs and more specifically, ZFP-B selectively repress >99% of HD-causing alleles, while maintaining at least >86% of the WT-alleles. In addition, ZFP-A and -B selectively downregulated mHTT mRNA and protein levels, while ZFP-C repressed both mutant and normal alleles. Upon examination, the HD mouse models demonstrated improvements at both the molecular, histopathological, motor and electrophysiological variables[33]. These findings support the notation that allele-selective ZFP-TFs may be used for HD treatment.

3.5 MEDICATION DOSING

Drug development in HD has increased over the past years with focuses ranging from symptomatic treatment to therapies targeting the gene to delay disease progression. Studies[35–38] that involved intervention using medication using Tetrabenazine, Pridopidine and Remacemide were investigated in either HD participants or HD mouse models. These studies are further discussed below.

Tetrabenazine (TBZ) is a vesicular monoamine transporter 2 (VMAT) inhibitor used for symptomatic management of involuntary movements seen in HD and it is currently the only FDA approved drug for HD[61]. Frank[35], conducted a 13-week RCT in HD participants. The mean TMC score decreased by 4.6 units from baseline to week-80. In addition, a reduction in the total number of adverse events and somnolence during maintenance was observed[35]. The study illustrates that TBZ suppresses HD movement impairments for a period of 80 weeks. However, patients should be monitored as an increase in parkinsonism, dysarthria, dysphagia, sleep disturbance, depression and anxiety were among the common adverse events experienced in patients[35].

Atomoxetine is a selective norepinephrine reuptake inhibitor, it is used to increase attention span, while decreasing impulsiveness[61]. HD patients experience cognitive and behavioural impairments which progressively become worse as the disease progresses. Therefore, due to its mode of action, Atomoxetine was investigated as a potential drug for cognitive impairment for a week-10 RCT study in HD participants by Beglinger et al.[36]. However, no significant cognitive improvement was observed while on Atomoxetine compared to placebo. Moreover, 56% of participants reported an adverse effect such as, dry month, dizziness, increased heart rate[36]. No beneficial effect of Atomoxetine was observed in HD patients.

Pridopidine is a small molecule that is a Sigma-1 receptor (S1R) agonist, a chaperone protein at the endoplasmic reticulum, responsible for modulating calcium signalling via the IP3 receptor[61,62]. Pridopidine is an investigational drug, currently under development for HD treatment[61]. The study by De Yebenes et al., 2011[37] investigated the effect of Pridopidine on the modified motor score (mMS) in HD patients across multiple clinical sites[37]. A -0.99 mMS difference from baseline was seen in HD patients who were administered 90mg/day in comparison to -0.36 mMS difference in patients administered the placebo. Furthermore, no significant changes in cognition and behavioural endpoints in either dose was observed[37]. Although well tolerated in HD patients, the potential effect of Pridopidine on motor symptoms, requires further investigation.

Lastly, coenzyme Q10 and Remacemide rich diets were investigated in the R6/2 and N171– 82Q transgenic HD mouse models by Ferrante et al., 2002[38]. Q10 is a naturally occurring coenzyme and antioxidant produced by the human body[63], while Remacemide acts as a low-affinity NMDA antagonist, which can block sodium channels[61]. The study identified an improvement in motor performance, following administration of Q10 and Remacemide in R6/2 mice. In addition, a 14.5% and 15.5% increase in mean survival was observed in R6/2 for Q10 and Remacemide diets respectively. While a combination of Q10 and Remacemide prolonged survival by 31.8% in R6/2 mice while also significantly extending survival in N171–82Q mice[38]. The improvements of clinical and neuropathological phenotypes in transgenic mice with the use of Q10 and Remacemide, indicate the combination therapies, prove to be beneficial and should be further investigated[38].

3.6 PHYSICAL ACTIVITY

Over the past decade, there has been a steady increase of studies demonstrating the advantage of physical activity in neurodegenerative diseases including HD. Numerous studies[14,39–47,49–52], have investigated the beneficial effects of physical activity in HD patients.

The study by Zinzi et al., 2007[39], investigated a three-week treatment programme consisting of respiratory exercise, speech therapy, physical and occupational therapy along with cognitive rehabilitation exercises in patients who are in the early and mid-stage HD disease onset [39]. A significant association was observed in the motor performance and daily activities, no motor decline was seen for a period of 2 years in HD patients, thus indicating that continuous level of functional, cognitive and motor performance was maintained[39].

Busse et al., 2013[40], conducted a 12-week randomized feasibility study, investigating the benefit of a community-based exercise program in HD participants vs controls[40]. The intervention consisted of a weekly supervised stationary cycling and resistance exercise, along with an independent home-based walking program, twice weekly[40]. No adverse event was reported and the intervention was well-tolerated by most study participants[40]. The study demonstrated that a structured exercise programme has a potential benefit on the total functional capacity (TFC) and total motor score (TMS) of HD patients.

A cohort study by Kloos et al., 2012[41] investigated the impact of various assistive devices on gait measures and safety in HD patients. Gait and balance impairment are often seen in HD, and result in frequent falls and injuries in HD patients[41]. Spatial and temporal gait parameters were examined under several conditions (no assistive device, use of assistive devices (AD) such as canes, weight cane, a walker or a 2,3 or 4 wheeled walker) (2WW) (3WW) (4WW)[41]. It was observed that the indicator of a fall decreased when using a 4WW in comparison to other ADs forms. Moreover, HD patients who used a 4WW, showed to have fewer stumbles and falls. It is noteworthy, to mention that HD patients, walked significantly slower when using an assistive device in comparison to patients who had no AD across the GAITRite[41]. The following study, showed that gait stability and safety improved with the use of 4WW, while on the other hand these were made worse with the use of other ADs such as 3WW and standard walker[41].

Another study by Kloos et al., 2013[42], conducted a cross-over controlled single blinded study, where the video game, Dance Dance Revoultion was used as a potential exercise therapy for 45 minutes, two days per week for six weeks in HD patients[41]. Researchers observed improvements in their performance regarding the number of correct steps obtained during game play[41]. However, no significant improvement was found in forward and backward walking step time, stride length and swing time[41].

A prospective intervention study by Piira et al., 2013[43], investigated the effects of a one-year intensive multidisciplinary rehabilitation program in early to mid-stage HD stage patients[41]. A significant improvement was observed in balance, gait function, QoL, anxiety, depression and BMI of

HD patients[41]. A minor improvement was seen in the mini mental state examination (MMSE) of participants, while the symbol digit modalities (SDMT) cognitive test showed a significant decline. However, the Unified Huntington's Disease Rating Scale (UHDRS) cognitive scores were no significant[41]. The following study, indicated that a multidisciplinary intensive rehabilitation program demonstrates an improvement. An additional study by Piira et al., 2014 [44] investigated the effects of a two-year intensive multidisciplinary rehabilitation program involving six admissions of three weeks in early to mid-stage HD patients motor and cognitive impairment of study participants was examined. No significant improvements were seen in the QoL, anxiety, depression and body mass index (BMI). Furthermore, activities of daily living (ADL) function persisted to be stable but with no significant decline[44]. The MMSE did not show any significant decline in HD participants. Four participants demonstrated motor function improvement, whereas a decline in motor function was seen in two participants[44]. The study shows that an intensive rehabilitation programme seems to be well-tolerated among early to mid-stage HD patients and an improvement regarding gait function, physical quality of life, anxiety and depression[44].

Thompson et al., 2013[45], were interested in investigating the effects of a multidisciplinary rehabilitation in early to mid-stage HD patients which included clinical and home-based exercises[45]. The intervention group participants had a significant decrease in their motor and postural stability deterioration, whereas, minor improvement in their depression, cognitive function and QoL were seen[45]. The results from this pilot study, suggest that a prolonged multidisciplinary rehabilitation programme in early to mid-stage seems to not only be feasible but also well-tolerated among study participants.

A RCT study by Quinn et al., 2014[46], aimed to assess the feasibility and safety addressing mobility limitations for individuals with mid-stage HD. Intervention involved, task specific training, focusing on walking, sit-to-stand transfers and standing, twice a week for 8 weeks. The study, showed that 46% of study participants, displayed better than expected results in regards to their mobility[45]. However, there was a lack of measurements regarding walking endurance in this group of patients. The initial results of the study illustrated the safety and adherence to home-based and task specific training. However, the frequency, intensity and specificity in terms of the intervention may not have been sufficient enough to illustrate a response[45]. Therefore, a larger-scale and longer study duration is needed to assess these intervention requirements. An additional RCT study by Quinn et al., 2016[47]consisted of exercise intervention and control groups that were assessed at three-time points (baseline, 13 and 26 weeks)[49]. Intervention was initiated at week 12, three times per week consisting of a progressive exercise programme such as stationary cycling and upper and lower body strengthening[49]. The intervention group demonstrated a decreased score and overall improvement in the UHDRS motor score[49]. The above-mentioned study demonstrated that even short-term exercise is both safe and feasible, while also demonstrating the potential benefit of a structured exercise and intensity programme in HD patients. PAGE-HD is a nested RCT study by Quinn et al., 2022[14], were interested in determining both physical activity and exercise outcomes in HD patients over a 12-month duration period. Performance-based measures such as VO2max, 6-minute walk test, timed up and go (TUG) test, and self-reported PA, all declined over the 12-month period in the cohort group[14]. Investigators observed a small positive effect, in terms of effect size in the intervention group, suggestive of improvement. Rates of the UHDRS-TMS, TFC and SDMT in the cohort group were reliable with previous studies identified by the same group[14]. PAGE-HD, provides the footing for a physical activity and exercise cohort trial with an extended duration in HD participants.

The aim of Dawes et al., 2014[49], was to investigate, exercise response in participants with HD in

comparison to a healthy control group. Intervention consisted of a 12-week gym and home walking exercise programme. HD participants vs controls, achieved a lower work rate at nine minutes[49]. A higher rate of perceived exertion (RPE) at three and nine minutes which are both statistically significant. Moreover, HD participants displayed an increased lactate concentration at three 2.5±2.5(1.1-8)mmo.L-1 and nine 3.8±1.9(1.2-6.6)mmo.L-1 minutes and respiratory exchange ratio at three and nine minutes[49]. While after exercise training, no change was seen in the HR or RPE response during the exercise test. A large variability was observed by researchers regarding the metabolic and physiological response to exercise in HD participants[49]. Therefore, suggesting HD participants may require alternative exercise programmes.

An individualized exercise programme on a regular basis may be ideal for HD patients, however, it is vital to facilitate engagement in independent exercise programmes such as home-based exercises. One study by Khalil et al., 2012[50], aimed to explore the use and perception of home-based DVD exercises by HD participants and their caregivers. A significant difference was observed between participants who adhere to the exercise programme in comparison to those who did not, in terms of their UHDRS cognitive subscale score[50]. The adherence rate was also significantly correlated with TFC score, UHDRS cognitive and IMI pressure/tension subscale scores[50]. No significant correlation between adherence rate with IMI subscales (enjoyment, perceived competence, effort, importance, value and usefulness) were identified. It is noteworthy, to mention that, HD participants and their caregivers, reported barriers in terms of using the exercise DVD this include i) physical factors (a caregiver was needed at all times, as patients struggled due their movement and gait impairment), ii) cognitive factors (difficulty to understand what they doing and if they were doing it correctly) and iii) lack of motivation[50]. However, the majority of participants and their caregivers perceived the DVD as suitable and supportive to engage HD individuals in home-based exercise[50].

An intervention study by Cruickshank et al., 2015[51], aimed to investigate the effect of multidisciplinary rehabilitation on brain structure and cognition in HD individuals[51]. Previous studies have shown a positive impact on the functional capacity, QoL and depression with multidisciplinary rehabilitation, which encompasses both motor and cognitive intervention. A significant increase in gray matter volume was observed in the right caudate and bilaterally in the dorsolateral prefrontal cortex following 9 months of multidisciplinary rehabilitation. Moreover, study participants displayed an improvement in verbal learning and memory, which is due to the volumetric gray matter increase[51]. The study indicates the positive impact of multidisciplinary rehabilitation on gray matter and cognitive function[51]. Another study by Cruickshank et al., 2018[52], set out to investigate the effects of a multidisciplinary therapy on physical function in HD for a duration of 9 months. Participants demonstrated a high adherence to both a supervised exercise program and cognitive therapy sessions. In addition, the intervention group showed significantly enhanced dexterity and lower extremity muscle strength in comparison to those in the control group[52]. No significant difference in balance, mobility, upper extremity strength and cardiorespiratory endurance outcomes, between groups following intervention[52]. A multidisciplinary rehabilitation therapy has a positive effect on dexterity and muscle strength. However, a larger randomized controlled trial is required to confirm these findings.

3.7 GENETIC MODIFIERS & POTENTIAL BIOMARKERS

Over the past decade, there has been an exponential increase in high-throughput sequencing, which have transformed the discovery of genes responsible for being the causative effect of rare Mendelian disease, including HD[64]. The use and need for biomarkers have increased over the past

years, with their use in basic and clinical research being of great importance[65]. Some studies have investigated the role of genetic modifiers[53–55,57] and potential HD-related biomarkers[56].

A genetic analysis study by Becánović et al., 2015[53] investigating the SNP rs13102260:G > A, in the HTT promoter region that alters nuclear factor Kappa B subunit 1 (NF-κB) binding[53]. In addition, the SNP was further investigated to determine its disease-modifying effects. NF-κB is a transcription factor (TF), and is well-thought-out to be a regulator of the innate immune system (Albensi., 2019)., it plays a role in numerous biological processes such as inflammation, immunity, differentiation, cell growth, apoptosis and tumorigenesis (Albensi.,2019). The study identified rs13102260, a non-coding SNP to impair NF-κB and reduced HTT transcriptional activity and protein expression[53]. Additionally, SNP rs13102260 minor (A) variant was associated with a delay AOO in familial HD cases, whereas on the other hand, the presence of the rs13102260 (A) variant on the WT-allele was associated with an earlier AOO in HD patients. The correlation between allele-specific effects of rs13102260 on HTT expression and AOO, may pave the way for HTT silencing treatments.

One of the most-well studied, HD genetic modifiers is FANCI associated Nuclease 1 (FAN1), which plays a role in DNA intrastand cross-link repair[66]. A genetic analysis study by Goold et al., 2019[54], was conducted to investigate how FAN1 modifies HD progression by stabilizing the CAG repeat expansion. The study, demonstrated that FAN1 expression, significantly reduced CAG repeat expansion in cells. Knockdown experiments of FAN1, illustrated an increase of CAG repeat expansion rate[54]. Moreover, FAN1 expression was decreased in both the mRNA and protein levels[54]. The study, demonstrated how genetic modifiers may alter disease progression, providing new insights to therapeutic interventions in trinucleotide repeat diseases.

Another genetic analysis study, was conducted by Kay et al., 2018[55], the study aimed to perform a comprehensive haplotype targeting strategy for allele-specific HTT suppression[55]. Haplotypes (A1m A2 and A3a) are associated with HD, these represent allele specific gene silencing targets, for effective treatment of HD of Northern European and Indigenous South American ancestry[55]. The A3a haplotype was identified in populations of European ancestry. Therefore, future approaches may target SNPs on A1 and A2 haplotypes in populations expressing those haplotypes. Moreover, the study illustrated that in a comprehensive haplotype analysis, HD affected individuals may be treated with three to four alleles targeting different populations worldwide.

Neurofilament light chain (NfL), is a neuronal cytoplasmic protein, expressed by large myelinated axons[67]. NfL concentration, increases in the CSF and blood, correspondingly to the degree of axonal damage in cerebrovascular, traumatic, inflammatory and neurodegenerative diseases[67] A 3-year retrospective cohort study was conducted by Byrne et al.,[56] to investigate the NfL protein as a potential HD biomarker [56]. The researchers, observed that NfL plasma concentration was significantly higher in HTT carriers in comparison to controls. In addition, baseline NfL concentration, was associated with a decline in cognition, TFC and brain atrophy. Moreover, NfL concentration in the CSF and plasma were correlated in HTT carriers[56]. This study, indicates the potential of NfL as a prognostic blood biomarker of HD disease progression and onset.

Lastly, a genome-wide association study (GWAS) study that includes data from the TRACK-HD and EHDN registry was conducted by Moss et al., 2017[57], for identification of genetic variants that may be associated with HD progression[57]. MTRNR2L2, MSH3 and DHFR were identified to be correlated with AOO in both datasets. In the TRACK-HD data, the minor allele of SNP (rs557874766) was correlated with a decrease rate of change in the UHDRS motor score and TFC score; these associations continued to be significant following adjustment for AOO[57]. Previous studies,

showed that a knockout of MSH3 in an HD mouse model, reduced somatic expansion, therefore, suggesting this mechanism as an area of interest for future therapeutic investigation.

Discussion

Over the past decade the field of potential HD therapeutics, genetic modifiers and biomarker discovery is rapidly advancing and embraces exciting future results. Presently, particular focus is given on lowering mHTT via DNA and RNA targeted therapies, this includes genome editing approaches such as CRISPR/Cas9, which are based at the pre-clinical stages due to ethical considerations in terms for allowing the editing of germline DNA[68]. While, on the other hand, ASOs, currently stand as the most rapidly advancing potential therapy in HD research, both of these approaches are invasively delivered via intrathecal or intracranial injections[68]. Therefore, less invasive therapies such as stem cell therapy, or biomarkers that may delay disease onset and progression are currently under investigation[68]. All these approaches hold significant promise, however, they are not without their strengths and limitations, which need to be further researched and understood, to have the optimal safety and efficacy in HD mouse models and study participants.

During the last couple of years, there has been an exponential increase in research regarding non-pharmacological interventions such as lifestyle changes that include dietary habits and physical activity. It is well-known that physical activity improves overall health, well-being, mental status and physical movement[69]. In individuals suffering from NDs, physical activity has proven to be beneficial by improving neurotransmitter, neurotrophic factors and hormone production [69]. Furthermore, physical activity promotes neuronal survival, neuroplasticity by enhancing neuroendocrine and physiological response to stress[69].

The purpose of this review was to recognize the pharmacological and non-pharmacological interventions, genetic modifiers and potential biomarkers in HD participants and mouse models, to identify if these factors have a positive association on motor, cognitive or behavioral impairments, delay disease onset and progression.

ASO research and development is rapidly growing, with a special interest in neurological diseases such as HD. ASOs are versatile molecules of a single-stranded deoxyribonucleotide that is complementary to the mRNA target[70]. ASOs are designed to precisely bind, target and modify RNA transcripts[71] and modulate protein expression[70]. The goal of ASOs is the down-regulation of the molecular target, typically achieved by RNase H endonuclease activity, which cleaves the RNA-DNA heteroduplex resulting in a significant decrease of the target gene translation [70], this may halt or slow down progression of rare genetic disease[72].

Over the past decade, attention has moved from targeting downstream processes to targeting the HTT gene itself via potential HD therapies, this includes lowering mHTT, via allele-specific ASOs or allele non-specific ASOs that target both mHTT and wild-type HTT (wtHTT)[68]. ASO therapies hold momentous attention as a potential HD therapy, with multiple ASOs at the moment in the clinical trial stage, some past ASOs studies include Tominersen (non-allele specific), WVE-120101 and WVE-120102 (allele-specific ASOs). However, not all of these lead to successful results due to issues regarding either safety, efficacy or no effect was observed between the intervention and placebo groups[68].

Other therapies, currently under investigation as potential HD treatments include ZFP, TALEN and CRISPR/Cas9 therapies. As mentioned above, there are DNA targeting therapies which affect the mHTT gene by presenting protein coding sequences that bind to specific DNA regions, these however, require a viral vector for delivery and are administered intracranially [68]. ZFPs refers to a group of proteins, where the structural features are wrapped around a zinc io, which then binds to the Cysteine and Histidine amino acids, therefore, resulting in a finger-like domain with the ability to

integrate in the groove of the unfolded DNA helix[73]. Current ZFPs bind to the CAG trinucleotide expansion, averting the mHTT gene's transcription, thus lowering mHTT protein concentration without changing the gene itself[74]. However, this poses an issue as ZFPs can alter the genetic material allowing the mHTT gene to either be corrected or disrupted[73]. The ZFP-TAK-686, targets expanded CAG repeats which successfully reduced mHTT in knock-in HdhQ50 mice. Moreover, it reduced mHTT by 62% resulting in behaviour improvement[73]

TALENS contain DNA binding domains which have repeated peptides that bind to DNA nucleotides[73], they cause double stranded break using artificial nucleases that allow for segment correction or deletion of the mutated gene[73]. TALENS research and development for HD, remains in the pre-clinical or drug discovery stages, as further research is required to understand their impact on the phenotype, safety, toxicity and an effective delivery system to ensure decrease of gene expression[73]. Studies have demonstrated that a TALE-SNP complex reduced mHTT expression without altering normal HTT expression, indicating their potential use in HD[73].

CRISPR/Cas9 has been used as a genome editing tool over the past decade, it functions through single-guide RNA binding to the target DNA, the double-stranded breaks are induced by Cas9 nucleases[73]. The double stranded breaks are repaired by non-homologous end joining, instigating frameshifts which can impair gene expression[73]. Viral vectors are used for the delivery of the CRISPR/Cas9 system, while they are administered via an intracranial injection [73]. In HD, CRISPR/Cas9 is used to remove the CAG repeat expansion, correcting for HD alleles, HD associated alleles or targeting the HTT gene; there are various research groups investigating the potential of CRISPR/Cas pre-clinically in HD[73].

Viral vectors have proven to be the utmost effective means of gene transfer to modify certain cell types or tissues, they can be manipulated to express the therapeutic genes of interest[74]. Numerous viruses such as adenoviruses (Ads), lentiviruses and adeno-associated viruses, are currently under investigation for their safety and successful gene delivery to cells[74]. The achievement of gene therapy depends on the safety and effective delivery of the genetic information to target cells, which depends on various factors such as i) choice of virus, ii) ease of production, iii) safety, toxicity and stability and iv) efficiency of transgene expression. Numerous studies are investigating viral vectors as a potential HD therapy.

There are constant improvements in genomic-based approaches, with the aim to identify even more disease-causing variants. There are genetic variants known as genetic modifiers, which are defined as genes that may alleviate or exacerbate the phenotypic and molecular expression of other genes[64]. Gene modifiers identified for HD include MRMR10, FAN1, RRM2B, UBR5, MLH1 and PMS2[75], with some of these influencing AOO and clinical characteristics of the disease. For instance, FAN1 and PMS2, are involved in DNA repair, in HD they can delay disease onset by 1.4 years, whereas RRM2B can result in early disease onset of 6.1 years[75]. Therefore, to truly understand the disease, an investigation is needed to recognize how disease-causing variants interact with other genes.

Modifiable factors such as physical activity have gained increased momentum over the past years due to mounting evidence supporting its role in maintaining brain health by slowing disease progression and improving disease prognosis [76]. Moderate physical activity promotes regulation of neuroplasticity and cell death and neurotransmitter release. Moreover, physical activity such as aerobic exercise, moderate neurotransmitter release and the neural circuitry, this demonstrates the positive effects of motor, behavioural and cognitive function improving the QoL of patients as a non-pharmacological therapeutic based approach[76]. As technology advances, so does its role in medicine and research, this is evident with the use of virtual reality (VR), which aims to apply traditional exercise within virtual environments, this makes it possible

to monitor motor and cognition in an easier and precise way[77]. Studies, using VR, where balance and gait function were monitored in Parkinson's Disease (PD) patients, showed improvement in these motor functions, therefore, indicating the potential use of VR in the setting of physical activity and physiotherapy[77]. Physical activity, along with dietary intake[78,79] show potential as modifiable non-pharmacological approaches in NDs including HD.

The studies encompassed within this review are not without their limitations this includes a small sample size in both HD patients and mouse models studies, lack of follow-up studies particularly regarding physical activity related studies, which results in a lack of inferring confidence as to whether physical activity affects the HD clinical phenotype. An additional limitation that was shared among many studies was that the study duration period was too short although significant results were obtained in regard to motor symptoms as significant changes in disease progression could not be easily detected. However, the benefits of this study include, correction of missing data performed in all studies, the inclusion of various studies involving pharmacological therapeutic approaches, ranging from genetic modifiers and potential biomarkers to ASOs and viral vectors and non-pharmacological approaches such as physical activity.

Conclusion

The outcomes of this review suggest an increase in research involving pharmacological and non-pharmacological therapeutic approaches such as ASOs, viral vectors, genetic modifiers and physical activity respectively. The majority of studies identified a decrease in mHTT protein expression with the use of ASOs and viral vectors in cell lines, fibroblasts and HD mouse models, indicating their potential as future therapeutic treatment for HD. Studies, involving HD participants, particularly focused on physical activity, an improvement in the motor score of HD individuals was observed, moreover, participants and their caregivers reported their enjoyment during these activities. Lastly, an understanding of genetic analysis in various HD populations, can provide insight into potential genetic modifiers that can delay disease onset or those that contribute to early disease onset, and pharmacological therapies can be designed to target these genes involved in early onset to delay disease progression. Although there are a variety of studies investigating potential therapies in HD, further research is required to fully understand the safety, efficacy of ASOs, viral vectors and CRISPR/Cas9 in cell lines to HD mouse models. Furthermore, although the benefits of physical activity have been established further research is needed to fully understand how motor, cognitive and behavioural improvements affect neutrophrins, neurotransmitter and hormone release and how this in turn may delay disease onset and progression.

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References:

1. Nopoulos., C. P. Huntington Disease: a single-gene degenerative disorder of the striatum. *Dialogues Clin Neurosci* 18, 91–98 (2016).

2. Jacob, E. L., Gatto, N. M., Thompson, A., Bordelon, Y. & Ritz, B. Occurrence of depression and anxiety prior to Parkinson's disease. *Parkinsonism Relat Disord* 16, 576–581 (2010).

3. Walker, F. O. Huntington's disease. *Lancet* 27, 143–150 (2007).

4. Roos, R. A. Huntington's disease: a clinical review. *Orphanet J Rare Dis* 5, 40 (2010).

5. McColgan, P. & Tabrizi, S. J. Huntington's disease: a clinical review. *European Journal of Neurology* vol. 25 24–34 Preprint at https://doi.org/10.1111/ene.13413 (2018).

6. Manoharan, S. *et al.* The Role of Reactive Oxygen Species in the Pathogenesis of Alzheimer's Disease, Parkinson's Disease, and Huntington's Disease: A Mini Review. *Oxidative Medicine and Cellular Longevity* Preprint at https://doi.org/10.1155/2016/8590578 (2016).

7. Mortada, I. *et al.* Immunotherapies for Neurodegenerative Diseases. *Frontiers in Neurology* vol. 12 Preprint at https://doi.org/10.3389/fneur.2021.654739 (2021).

8. Durães, F., Pinto, M. & Sousa, E. Old drugs as new treatments for neurodegenerative diseases. *Pharmaceuticals* vol. 11 Preprint at https://doi.org/10.3390/ph11020044 (2018).

9. Li, X., Frye, M. A. & Shelton, R. C. Review of pharmacological treatment in mood disorders and future directions for drug development. *Neuropsychopharmacology* vol. 37 77–101 Preprint at https://doi.org/10.1038/npp.2011.198 (2012).

10. Scatena, R. *et al.* An update on pharmacological approaches to neurodegenerative disease. *Expert Opinion on Investigational Drugs* vol. 16 59–72 Preprint at https://doi.org/10.1517/13543784.16.1.59 (2007).

11. Castellano-Tejedor, C. Non-Pharmacological Interventions for the Management of Chronic Health Conditions and Non-Communicable Diseases. *International Journal of Environmental Research and Public Health* vol. 19 Preprint at https://doi.org/10.3390/ijerph19148536 (2022).

12. Scheller, E. L. & Krebsbach, P. H. Gene therapy: Design and prospects for craniofacial regeneration. *Journal of Dental Research* vol. 88 585–596 Preprint at https://doi.org/10.1177/0022034509337480 (2009).

13. Quinn, L. *et al.* Clinical recommendations to guide physical therapy practice for Huntington disease. *Neurology* vol. 94 217–228 Preprint at https://doi.org/10.1212/WNL.0000000000008887 (2020).

14. Quinn, L. *et al.* Physical activity and exercise outcomes in Huntington's disease (PACE-HD): results of a 12-month trial-within-cohort feasibility study of a physical activity intervention in people with Huntington's disease. *Parkinsonism Relat Disord* 101, 75–89 (2022).

15. Mueller, S. M., Petersen, J. A. & Jung, H. H. Exercise in huntington's disease: Current state and clinical significance. *Tremor and Other Hyperkinetic Movements* vol. 9 1–10 Preprint at https://doi.org/10.7916/tm9j-f874 (2019).

16. Kay, C. *et al.* A Comprehensive Haplotype-Targeting Strategy for Allele-Specific HTT Suppression in Huntington Disease. *Am J Hum Genet* 105, 1112–1125 (2019).

17. Bloch, J. *et al.* Neuroprotective gene therapy for Huntington's disease, using polymer-encapsulated cells engineered to secrete human ciliary neurotrophic factor: Results of a phase I study. *Hum Gene Ther* 15, 968–975 (2004).

18. Fiszer, A., Ellison-Klimontowicz, M. E. & Krzyzosiak, W. J. Silencing of genes responsible for polyQ diseases using chemically modified single-stranded siRNAs. *Acta Biochim Pol* 63, 759–764 (2016).

19. Pfister, E. L. *et al.* Five siRNAs Targeting Three SNPs May Provide Therapy for Three-Quarters of Huntington's Disease Patients. *Current Biology* 19, 774–778 (2009).

20. Fink, K. D. *et al.* Allele-specific reduction of the mutant huntingtin allele using transcription

activator-like effectors in human huntington's disease fibroblasts. *Cell Transplant* 25, 677–686 (2016).

21. Yu, D. *et al.* Single-stranded RNAs use RNAi to potently and allele-selectively inhibit mutant huntingtin expression. *Cell* 150, 895–908 (2012).

22. Zhang, Y., Engelman, J. & Friedlander, R. M. Allele-specific silencing of mutant Huntington's disease gene. *J Neurochem* 108, 82–90 (2009).

23. Xu, X. *et al.* Reversal of Phenotypic Abnormalities by CRISPR/Cas9-Mediated Gene Correction in Huntington Disease Patient-Derived Induced Pluripotent Stem Cells. *Stem Cell Reports* 8, 619–633 (2017).

24. Kordasiewicz, H. B. *et al.* Sustained Therapeutic Reversal of Huntington's Disease by Transient Repression of Huntingtin Synthesis. *Neuron* 74, 1031–1044 (2012).

25. Sathasivam, K. *et al.* Aberrant splicing of HTT generates the pathogenic exon 1 protein in Huntington disease. *Proc Natl Acad Sci U S A* 110, 2366–2370 (2013).

26. Boudreau, R. L. *et al.* Nonallele-specific silencing of mutant and wild-type huntingtin demonstrates therapeutic efficacy in Huntington's disease mice. *Molecular Therapy* 17, 1053–1063 (2009).

27. Cho, I. K., Hunter, C. E., Ye, S., Pongos, A. L. & Chan, A. W. S. Combination of stem cell and gene therapy ameliorates symptoms in Huntington's disease mice. *NPJ Regen Med* 4, (2019).

28. Harper, S. Q. *et al.* RNA interference improves motor and neuropathological abnormalities in a Huntington's disease mouse model. *Proc Natl Acad Sci U S A* 102, 5820–5825 (2005).

29. Ekman, F. K. *et al.* CRISPR-Cas9-Mediated Genome Editing Increases Lifespan and Improves Motor Deficits in a Huntington's Disease Mouse Model. *Mol Ther Nucleic Acids* 17, 829–839 (2019).

30. Dey, N. D. *et al.* Genetically engineered mesenchymal stem cells reduce behavioral deficits in the YAC 128 mouse model of Huntington's disease. *Behavioural Brain Research* 214, 193–200 (2010).

31. Pollock, K. *et al.* Human mesenchymal stem cells genetically engineered to overexpress brain-derived neurotrophic factor improve outcomes in huntington's disease mouse models. *Molecular Therapy* 24, 965–977 (2016).

32. Difiglia, M. *et al.* Therapeutic silencing of mutant huntingtin with siRNA attenuates striatal and cortical neuropathology and behavioral deficits. *PNAS* 104, 17204–17209 (2007).

33. Agustín-Pavón, C., Mielcarek, M., Garriga-Canut, M. & Isalan, M. Deimmunization for gene therapy: Host matching of synthetic zinc finger constructs enables long-term mutant Huntingtin repression in mice. *Mol Neurodegener* 11, (2016).

34. Zeitler, B. *et al.* Allele-selective transcriptional repression of mutant HTT for the treatment of Huntington's disease. *Nat Med* 25, 1131–1142 (2019).

35. Frank, S. Tetrabenazine as anti-chorea therapy in Huntington Disease: An open-label continuation study. Huntington Study Group/TETRA-HD Investigators. *BMC Neurol* 9, (2009).

36. Beglinger, L. J. *et al.* Randomized controlled trial of atomoxetine for cognitive dysfunction in early huntington disease. *J Clin Psychopharmacol* 29, 484–487 (2009).

37. Garcia de Yebenes, J. *et al.* Pridopidine for the treatment of motor function in patients with Huntington's disease (MermaiHD): a phase 3, randomised, double-blind, placebo-controlled trial. *Lancet Neurol* 10, 1049–1057 (2011).

38. Ferrante, R. J. *et al.* Therapeutic Effects of Coenzyme Q 10 and Remacemide in Transgenic Mouse Models of Huntington's Disease. *Journal of Neuroscience* 22, 15922–15999 (2002).

39. Zinzi, P. *et al.* Effects of an intensive rehabilitation programme on patients with Huntington's disease: A pilot study. *Clin Rehabil* 21, 603–613 (2007).

40. Busse, M. *et al.* Q12 A randomised feasibility study of a 12-week exercise programme in Huntington's disease (HD). *J Neurol Neurosurg Psychiatry* 83, A58.2-A59 (2012).

41. Kloos, A. D., Kegelmeyer, D. A., White, S. E. & Kostyk, S. K. The impact of different types of

assistive devices on gait measures and safety in Huntington's disease. *PLoS One* 7, (2012).

42. Kloos, A. D., Fritz, N. E., Kostyk, S. K., Young, G. S. & Kegelmeyer, D. A. Video game play (Dance Dance Revolution) as a potential exercise therapy in Huntington's disease: A controlled clinical trial. *Clin Rehabil* 27, 972–982 (2013).

43. Piira, A. *et al.* Effects of a One Year Intensive Multidisciplinary Rehabilitation Program for Patients with Huntington's Disease: A Prospective Intervention Study. *PLoS Curr* (2013) doi:10.1371/currents.hd.9504af71e0d1f87830c25c394be47027

44. Piira, A. *et al.* Effects of a Two-Year Intensive Multidisciplinary Rehabilitation Program for Patients with Huntington's Disease: a Prospective Intervention Study. *PLoS Curr* (2014) doi:10.1371/currents.hd.2c56ceef7f9f8e239a59ecf2d94cddac.

45. Thompson, J. A. *et al.* The effects of multidisciplinary rehabilitation in patients with early-to-middle-stage Huntington's disease: A pilot study. *Eur J Neurol* 20, 1325–1329 (2013).

46. Quinn, L. *et al.* Task-Specific Training in Huntington Disease: A Randomized Controlled Feasibility Trial. *Physical Therapy Journal* 94, 1555–1568 (2014).

47. Quinn, L. *et al.* A randomized, controlled trial of a multi-modal exercise intervention in Huntington's disease. *Parkinsonism Relat Disord* 31, 46–52 (2016).

48. Quinn, L. *et al.* Physical activity and exercise outcomes in Huntington's disease (PACE-HD): results of a 12-month trial-within-cohort feasibility study of a physical activity intervention in people with Huntington's disease. *Parkinsonism Relat Disord* 101, 75–89 (2022).

49. Dawes, H. *et al.* Exercise testing and training in people with Huntington's disease. *Clin Rehabil* 29, 196–206 (2015).

50. Khalil, H. *et al.* Adherence to use of a home-based exercise DVD in people with Huntington disease: Participants' perspectives. *Phys Ther* 92, 69–82 (2012).

51. Cruickshank, T. M. *et al.* The effect of multidisciplinary rehabilitation on brain structure and cognition in Huntington's disease: An exploratory study. *Brain Behav* 5, 1–10 (2015).

52. Cruickshank, T. M. *et al.* Effects of multidisciplinary therapy on physical function in Huntington's disease. *Acta Neurol Scand* 138, 500–507 (2018).

53. Becanovic, K. *et al.* A SNP in the HTT promoter alters NF-κB binding and is a bidirectional genetic modifier of Huntington disease. *Nat Neurosci* 18, 807–816 (2015).

54. Goold, R. *et al.* FAN1 modifies Huntington's disease progression by stabilizing the expanded HTT CAG repeat. *Hum Mol Genet* 28, 650–661 (2019).

55. Kay, C. *et al.* The molecular epidemiology of Huntington disease is related to intermediate allele frequency and haplotype in the general population. *American Journal of Medical Genetics Part B: Neuropsychiatric Genetics* 177, 346–357 (2018).

56. Byrne, L. M. *et al.* Neurofilament light protein in blood as a potential biomarker of neurodegeneration in Huntington's disease: a retrospective cohort analysis. *Lancet Neurol* 16, 601–609 (2017).

57. Moss, D. J. H. *et al.* Identification of genetic variants associated with Huntington's disease progression: a genome-wide association study. *Lancet Neurol* 16, 701–711 (2017).

58. Rinaldi, C. & Wood, M. J. A. Antisense oligonucleotides: The next frontier for treatment of neurological disorders. *Nature Reviews Neurology* vol. 14 9–22 Preprint at https://doi.org/10.1038/nrneurol.2017.148 (2018).

59. Kruttgen, A. *et al.* Human ciliary neurotrophic factor: a structure-function analysis. *Biochem Journal* 30, 215–220 (1955).

60. Redman, M., King, A., Watson, C. & King, D. What is CRISPR/Cas9? *Arch Dis Child Educ Pract Ed* 101, 213–215 (2016).

61. Wishart, D. S. *et al.* DrugBank: A knowledgebase for drugs, drug actions and drug targets. *Nucleic Acids Res* 36, 901–906 (2008).

62. Meng, F., Xiao, Y., Ji, Y., Sun, Z. & Zhou, X. An open-like conformation of the sigma-1 receptor reveals its ligand entry pathway. *Nat Commun* 13, (2022).

63. Tsai, I. C., Hsu, C. W., Chang, C. H., Tseng, P. T. & Chang, K. V. Effectiveness of Coenzyme Q10 Supplementation for Reducing Fatigue: A Systematic Review and Meta-Analysis of Randomized Controlled Trials. *Frontiers in Pharmacology* vol. 13 Preprint at https://doi.org/10.3389/fphar.2022.883251 (2022).

64. Rahit, K. M. T. H. & Tarailo-Graovac, M. Genetic Modifiers and Rare Mendelian Disease. *Genes* vol. 11 Preprint at https://doi.org/10.3390/genes11030239 (2020).

65. Strimbu, K. & Tavel, J. A. What are biomarkers? *Current Opinion in HIV and AIDS* vol. 5 463–466 Preprint at https://doi.org/10.1097/COH.0b013e32833ed177 (2010).

66. Safran, M. *et al.* GeneCards Version 3: the human gene integrator. *Database (Oxford)* 2010, 1–16 (2010).

67. Gaetani, L. *et al.* Neurofilament light chain as a biomarker in neurological disorders. *Journal of Neurology, Neurosurgery and Psychiatry* vol. 90 870–881 Preprint at https://doi.org/10.1136/jnnp-2018-320106 (2019).

68. Ferguson, M. W. *et al.* Current and Possible Future Therapeutic Options for Huntington's Disease. *Journal of Central Nervous System Disease* vol. 14 Preprint at https://doi.org/10.1177/11795735221092517 (2022).

69. Mahalakshmi, B., Maurya, N., Lee, S. Da & Kumar, V. B. Possible neuroprotective mechanisms of physical exercise in neurodegeneration. *Int J Mol Sci* 21, 1–17 (2020).

70. Fusco, D. Di *et al.* Antisense oligonucleotide: Basic concepts and therapeutic application in inflammatory bowel disease. *Frontiers in Pharmacology* vol. 10 Preprint at https://doi.org/10.3389/fphar.2019.00305 (2019).

71. Dhuri, K. *et al.* Antisense oligonucleotides: An emerging area in drug discovery and development. *Journal of Clinical Medicine* vol. 9 1–24 Preprint at https://doi.org/10.3390/JCM9062004 (2020).

72. Lauffer, M. C., van Roon-Mom, W. & Aartsma-Rus, A. Possibilities and limitations of antisense oligonucleotide therapies for the treatment of monogenic disorders. *Communications Medicine* vol. 4 Preprint at https://doi.org/10.1038/s43856-023-00419-1 (2024).

73. Cassandri, M. *et al.* Zinc-finger proteins in health and disease. *Cell Death Discovery* vol. 3 Preprint at https://doi.org/10.1038/cddiscovery.2017.71 (2017).

74. Warnock, J. N., Daigre, C. & Al-Rubeai, M. Introduction to viral vectors. in *Methods in molecular biology (Clifton, N.J.)* vol. 737 1–25 (2011).

75. Bettencourt, C. *et al.* DNA repair pathways underlie a common genetic mechanism modulating onset in polyglutamine diseases. *Ann Neurol* 79, 983–990 (2016).

76. Liao, Q., He, J. & Huang, K. Physical activities and risk of neurodegenerative diseases: A two-sample Mendelian randomization study. *Front Aging Neurosci* 14, (2022).

77. Farì, G. *et al.* The effect of physical exercise on cognitive impairment in neurodegenerative disease: From pathophysiology to clinical and rehabilitative aspects. *International Journal of Molecular Sciences* vol. 22 Preprint at https://doi.org/10.3390/ijms222111632 (2021).

78. Christodoulou, C. C., Demetriou, C. A. & Zamba-papanicolaou, E. Dietary Intake , Mediterranean Diet Adherence and Caloric Intake in Huntington ' s Disease : A Review. *Nutrients* 12, 1–25 (2020).

79. Christodoulou, C. C., Demetriou, C. A., Philippou, E. & Papanicolaou, E. Z. Investigating the Dietary Intake Using the CyFFQ Semi-Quantitative Food Frequency Questionnaire in Cypriot Huntington's Disease Patients. *Nutrients* 15, (2023).

