HEROs

Huntington’s Enrichment Research Optimisation scheme

AN INVESTIGATION INTO

THE EFFECTS OF ENVIRONMENTAL ENRICHMENT

ON CLINICAL MEASURES OF DISEASE PROGRESSION

AND QUALITY OF LIFE IN PATIENTS WITH

HUNTINGTON’S DISEASE

FINAL Project Report

by

Dr Jennifer Thompson
Project Manager

A/Prof Mel Ziman
Chief Investigator


Collaborative Research Partners:
Huntington’s WA (Inc)
Edith Cowan University
WA Department of Health (Neurosciences Unit)
The Brightwater Group
Cambridge Centre for Brain Repair, University of Cambridge
Monash University
The Florey Neurosciences Institutes
RESEARCH TEAM

**ECU Research Team**

- A/Prof Mel Ziman
- Dr Jennifer Thompson
- Mr Travis Cruickshank
- A/Prof Rob Newton
- Ms Zhargona Khan
- Ms Anna Reid

**Exercise Physiologists**

- Mr Luis Penailillo
- Mr Alvaro Reyes
- Mr Todd Cunning
- Ms Linda Houlty

**Occupational Therapists**

- Mr Nick Kalaitzis
- Ms Alisson Lim
- Ms Alisson James
- Ms Zara Samani

**Clinicians**

- Prof Roger Barker
- Prof Joseph Lee

**Brain Imaging**

- Clinical A/Prof Mike Bynevelt
- Ms Anne Winsor
- Mr Lincoln Codd
- Mr Randall Jones
- Prof Nellie Georgiou-Karistianis

STEERING COMMITTEE

- A/Prof Mel Ziman, Edith Cowan University
- Dr Jennifer Thompson, Edith Cowan University
- A/Prof Rob Newton, Edith Cowan University
- Ms Dee Sidhu, Huntington’s WA (Inc)
- Ms Anne Jones, Huntington’s WA (Inc)
- Ms Tracey Jones, Participant Representative
- Dr Camela Connor, Neurosciences Unit
- Dr Simon Davies, Neurosciences Unit
- Mr Steve Andrew, Neurosciences Unit
- Ms Toni Liebeck, Neurosciences Unit
- Dr Penny Flett, Brightwater Group
- Ms Janet Wagland, Brightwater Group
- Mr Timothy Lo, Brightwater Group
- Prof Gill Lewin, Silver Chain
- Ms Marion Hailes-MacDonald, Disability Services Commission
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FURTHER INFORMATION
For further information regarding this project or for copies of this report contact:

A/Prof Mel Ziman,
Leader, Huntington’s Disease Research Group,
School of Medical Sciences,
Edith Cowan University,
270 Joondalup Drive,
JOONDALUP WA 6027
T: (08) 6304 3640
E: m.ziman@ecu.edu.au

Copies of raw data are available on request.
REPORT SUMMARY

Background:
Huntington’s disease is a fatal neurodegenerative disease characterised by progressive cognitive, affective and motor deterioration over a 15-25 year period, and is associated with a spectrum of physical deficits including postural instability, gait impairments, and loss of fat and lean tissue. There is no cure for Huntington’s disease and no proven therapy for altering disease progression in patients. Therefore, an urgent need exists to identify alternate treatments capable of impacting on disease progression and improving quality of life whilst the search for a cure continues. One promising strategy is the use of an enriched environment to provide enhanced neurological stimulation. Therefore, this project will assess the impact of an enriched environment, delivered via utilisation of a targeted, multidisciplinary rehabilitation program, on the signs and symptoms of Huntington’s disease.

Aims:
The HEROs Research Project sought to determine whether a prolonged program of cognitive and physical rehabilitation would positively impact on features of the disease. We performed a rigorous assessment of changes as a result of the intervention to address this goal.

Methods:
We recruited twenty patients at early-mid stages of Huntington’s disease and comprehensively assessed them using an array of outcome measures investigating cognitive and physical performance together with affective, physiological and quality of life indicators. Based on these results, patients were assigned into two equally-matched groups, with one group initially receiving the intervention and the other group serving as a control (Arm 1). At the end of this period, the program was extended and another period of intervention was then offered to all participants (Arm 2). This study design allowed for the provision of a control group, an intervention group comprised of all participants, and a longitudinal intervention group.

The intervention consisted of 9 months of clinical gym-based exercises, and six months of home-based physical exercises and occupational therapy. Patients who received the first round of intervention then continued to receive the clinical gym component for another (longitudinal) 9 month period (Group B). Control subjects from the first intervention period received the (delayed) intervention during the second period (Group A). At the end of each intervention period, all patients were assessed using the outcome measures outlined above.

Timeline:-

<table>
<thead>
<tr>
<th>Time</th>
<th>Event</th>
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<tbody>
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<td>0-3 months</td>
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<tr>
<td>3-12 months</td>
<td>Intervention provided to Group B participants</td>
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<tr>
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Results:

In this report we detail the results of the motor, cognitive, affective, physiological, physical, functional and quality of life assessments determined during the 23 month period of the study. The intervention produced significantly reduced motor deterioration in intervention recipients relative to control subjects, which was notably maintained after longitudinal intervention. Participants also demonstrated cognitive improvement after intervention, which reached statistical significance for some aspects of cognitive function, particularly after longitudinal intervention. Participants who received the intervention also showed significant improvements in body composition, muscular strength and perceptions of mental health, together with improved physiological parameters, with less participants experiencing deterioration in postural stability. Non-intervention controls showed significant deterioration in motor and cognitive function, body composition, and postural stability.

Conclusion:

Whilst the paradigm of multidisciplinary rehabilitation implemented in this study has yet to be shown to be disease modifying, we have demonstrated its ability to significantly impact on the phenotypic expression of motor, cognitive, affective, physiological and physical features of Huntington’s disease in a cohort of individuals at early-mid stages of the disease. In comparison to a previous intermittent rehabilitation protocol over a similar timeframe, it is apparent that a continuous program of rehabilitation produces superior benefits.

Whilst cautious interpretation of these results is advised due to the pilot nature of the study, it is evident that further, larger studies are warranted to more clearly elucidate the benefits of such a program in this population.

In conclusion, our results indicate that early-to-mid stage Huntington’s disease patients can and will successfully participate in an ongoing multidisciplinary rehabilitation program as an adjunct to their normal pharmaceutical regime. They do so without any apparent adverse effects, and as a consequence demonstrate positive motor, cognitive, affective, physiological, physical and functional outcomes that are maintained or improve over longitudinal timeframes.

As a result of this study, Huntingtons’ WA are to implement a program of environmental enrichment for their members.

Research Outcomes Published to Date:

PROJECT ABSTRACT

Background:

Huntington’s disease is a fatal neurodegenerative disease characterised by progressive cognitive, affective and motor deterioration over a 15-25 year period, together with a spectrum of associated physical deficits including postural instability, gait impairments and loss of skeletal muscle (lean tissue) and fat tissue.

Cognitive features of the disease include deficits in attention, memory, cognitive flexibility, visuospatial perception and other aspects of executive function, which negatively impact on the execution of everyday activities. Affective features include dysphoria, agitation, irritability, apathy, anxiety and depression. Individuals also exhibit choreiform movements - involuntary movements that are the clinical hallmark of the disease. Throughout the course of the disease motor control progressively declines, until patients are totally incapacitated, confined to a wheelchair and unable to function independently, at which time they may be placed in an end-stage care facility. Individuals ultimately become bed-bound and demented prior to death, which may be secondary to fall-related head injuries, choking or aspiration pneumonia, or as a result of suicide.

There is no cure for Huntington’s disease and no proven therapy for altering disease progression in patients. Currently, patients are managed with pharmacological interventions aimed at symptom management. Intense research is being conducted into the genetic and molecular aspects of the disease, however whilst these approaches appear promising, they currently represent strategies for the future. Thus, in recognition of the debilitating nature of the disease summarised above, there is an urgent need to identify alternate treatments capable of impacting on disease progression and/or symptomology with the ultimate goal of improving quality of life whilst the search for a cure continues.

Despite the monogenic aetiology of Huntington’s disease, it is becoming increasingly apparent that other genes and epigenetic/environmental factors significantly contribute to the disease course. Whilst this adds further complexity to understanding the disease process itself, it presents an exciting opportunity to impact on the disease course without directly modifying gene function, which has proved enigmatic to-date.

One promising area of investigation is the paradigm of environmental enrichment, which employs increased mental, physical and social stimulation in animal models to enhance neurological input with a view to modifying the clinical course of the disease. Despite promising results in animal models of Huntington’s disease, clinical research and subsequent translation of such a program has been lacking in Huntington’s disease. One study recently explored the effects of multidisciplinary care in early-mid stage Huntington’s disease patients and found that the protocol was tolerable, feasible, and appreciated by patients, families and health providers. Another study investigated the effects of an intermittent rehabilitation program in Huntington’s disease patients over a two year period. Whilst the study
demonstrated benefits for patients related to maintenance of baseline function, outcome measures were limited and patient numbers markedly declined over the two year period, requiring caution in interpretation of the data.

**Aims:**

The HEROs Research Project sought to determine whether a prolonged program of enrichment delivered through a continuous, targeted multidisciplinary rehabilitation program would more positively impact on features of the disease. We performed a rigorous assessment of changes as a result of the intervention, with a clear intent to assess the possibility of disease modification as an endpoint.

**Methods:**

We recruited twenty patients at early-mid stages of Huntington’s disease and comprehensively assessed them using an array of outcome measures investigating cognitive, motor and physical performance together with psychological, physiological and quality of life indicators.

Participants were allocated into two groups equally matched for cognitive and motor assessment scores at baseline. These groups were analysed for differences in pertinent baseline demographics, with no significant differences found. The group to receive the intervention was randomly assigned. Group A served as a control group (Arm 1) and then received a delayed intervention (Arm 2). Group B received a longitudinal intervention (Arms 1-2).

**Arm 1** - An individualised program of intervention was designed over three months (0-3 months) targeting deficits identified during baseline assessments. The control group (A) did not receive the intervention during this period and were instructed to maintain normal daily activities, although this was not monitored. The intervention group (B) received physical, cognitive and social stimulation delivered as nine months of clinical gym exercise sessions (3-12 months), together with six months of self-directed, home-based exercises and occupational therapy programs (6-12 months). Intervention participants received social stimulation via engagement with trainers, students and other participants during weekly group attendance in the gym, and during fortnightly individualised sessions with an Occupational Therapist. All participants were re-assessed at the end of the nine-month period (first post-intervention assessment at 12 month timepoint).

**Arm 2** – At the completion of the first post-intervention assessment, the control group (A) were offered the opportunity to participate in the intervention, which was accepted by all individuals. An additional three individuals joined the program at this time, and two individuals were lost from the program (for reasons not related to the program; one deceased, one hospitalized), and thus 12 individuals received and completed the program entailing 9 months of clinical gym exercises, 6 months of home-based exercises, and 6 months of fortnightly occupational therapy, delivered in the home environment (14-23 months). For
this round of intervention, the home-based exercise component, which was essentially self-directed during the first intervention round, was optimised for Group A participants by the inclusion of expert supervision delivered in the home environment once per week by an accredited exercise physiologist, supported by students.

During this period, one individual was diagnosed with another, confounding neurological disorder and, whilst allowed to continue with the program, their data was excluded from analysis, with data from the remaining 11 individuals accepted for analysis.

Participants from the original intervention group (B) had previously indicated their desire to continue with the program, and they were therefore offered the opportunity to continue with the physical exercise component of the program. All individuals with the exception of one accepted the offer and continued with the program (14-23 months), thus allowing for a longitudinal exploration of the benefits of the program.

Medication changes throughout the study were monitored and reviewed at completion of the intervention program. As there were similar medication changes in both groups, it is unlikely that the results of the intervention are due to medication effects.

**Results:**

The program was found to be feasible and well-tolerated in early-mid Huntington’s disease patients, who demonstrated a high level of adherence to the program, with no adverse effects reported.

Clinical assessment of motor function indicated significant deterioration in control subjects, which was markedly and significantly reduced after Intervention 1 (9 months) and maintained at the reduced level after Intervention 2 (18 months).

Affective assessments targeted qualitative changes to mood and behaviour. Self-reported depression scores indicated no effect of the intervention, with a differential response from each group noted. Carer-reported changes in behaviour indicated a trend towards increased behavioural symptoms after 9 months of intervention, which was reversed after 18 months of intervention.

Cognitive changes included significant improvements in learning/memory recall and retention, task accuracy and motor and cognitive processing speed during performance of simple tasks related to visual scanning and sequencing skills. Cognitive improvements were augmented after 18 months of intervention, indicating a delayed response to the intervention. Reduced deterioration in cognitive processing speed during performance of an executive function task was also noted.

Biochemical assessment of physiological markers related to neurological, cognitive and physical factors identified changes towards more normative levels after intervention, relative to the control group. Changes in cognitive function were accompanied by an overall
reduction in the decrease in BDNF concentration after 18 months of intervention, which notably increased in males. Coincident with the changes in body composition, noted below, were physiological changes to insulin and cortisol levels after intervention, relative to control subjects. Changes to levels of these biomarkers identified group differences during 9 months intervention, which may indicate a more robust response to the optimised intervention.

From a physical perspective, noteworthy changes to body composition were detected in intervention participants, in contrast to control individuals. During the normal course of the disease, control individuals lost significant fat and total body mass, with a borderline significant lean tissue loss. In contrast, this characteristic loss of body mass was rectified in intervention participants, showing a significant between-group difference. Non-significant gains in lean muscle mass occurred throughout both rounds of intervention, culminating in significantly improved lean muscle mass after 18 months of intervention. Longitudinally, total body mass gains after 18 months, represented solely by lean tissue gain, showed a trend towards significance. Importantly, these favourable changes to body composition during 9 months of intervention were maintained after 18 months of intervention.

Further in-depth examination of regional changes to body composition demonstrated that body mass changes were predominantly mediated in the trunk, and to a lesser extent in the legs. Loss of total body mass during the control period occurred at a fat/lean ratio of 2:1. Likewise, total body mass gains after Intervention 1 occurred at a fat/lean ratio of 2:1. However, the gain in total body mass after Intervention 2 was solely represented by a gain in lean mass.

Volumetric muscle strength changes in the upper and lower body showed highly significant, progressive increases in total group muscle strength, and in male and female subgroups, and may reflect the increased muscle mass identified by body composition analysis. Whilst maximal strength gains appeared after 5-6 months of intervention in Group B (Arm 1 – included non-optimised home-based intervention), this was no longer evident in the group as a whole after 9 months of intervention. This suggests a positive effect of the optimised home-based program during Arm 2, which essentially provided an additional supervised exercise event per week. Longitudinally, strength increases continued to occur, but were generally no longer statistically significant. Collectively, this data validates the physical benefits of providing a continuous, rather than intermittent, rehabilitation program to maximise strength increases for functional improvements.

Quantitative assessment of postural stability identified no significant changes to equilibrium scores in enriched subjects relative to control subjects, with deterioration noted in both groups. Deterioration after 9 and 18 months of intervention, however, occurred at a progressively reduced rate relative to control subjects. Over the period of the study, Group A participants demonstrated a significant deterioration after 18 months relative to baseline, whilst Group B participants did not. The lack of statistical significance in the short term is likely to be the result of high within-group variability, consistent with the variable nature of Huntington’s disease, and the small sample size of the cohort. This is further confounded by
between-group differences throughout the study period. To explore this further, additional analyses were conducted to remove the size of the effect on postural changes, and to quantitatively assess for the presence of changes to postural stability. Results indicated a stepwise reduction in the number of people experiencing deterioration in postural stability after 9 months of intervention, which culminated in a significant difference in the number of people maintaining or improving postural stability after 18 months of intervention compared to after the control period. This suggests that statistical significance for equilibrium score changes may be found in larger sample sizes.

The reduced deterioration in motor function, and improvements in cognitive and physical function, body composition and physiology detailed in this report are likely to impact on performance of everyday activities. Changes to quality of life perceptions were analysed as an end-point indicator of the effect of the program. The intervention produced perceived improvements in several areas related to QoL for participants. In particular, the intervention group perceived minor improvements in general health, energy/vitality, and social functioning which were evident after longitudinal intervention, together with a significant improvement in perceptions of mental health after longitudinal intervention.

The results presented in this report clearly demonstrate that a prolonged and continuous enrichment program is well-tolerated by early-mid Huntington’s disease patients. Participation in such a program produces adaptive physiological changes and improves motor, cognitive, affective, physiological and physical condition with maintenance of postural stability. Further research will be required to understand the neurological and physiological mechanisms underpinning these favourable responses to the intervention. Most significantly, analysis of brain imaging of participants should prove informative as to the potential neurological basis of these improvements or, at the very least, the effect of the intervention on brain volume, function and integrity.

**Conclusion:**

Our results indicate that Huntington’s disease patients at early-mid stages of the disease can successfully participate in an ongoing enrichment program as an adjunct to their normal pharmaceutical regime, and the data further suggest that engagement in such a program may delay the natural history of this condition and reduce the need for adjuvant pharmacological treatment commonly associated with negative side effects. It is apparent that cognitive improvements are delayed relative to physical improvements, and the overall trend towards greater cognitive improvement longitudinally greatly endorses the provision of a continuous program for maximal benefits.

Given the incurable and severe nature of the disease in question, together with the non-pharmacological nature of this therapeutic strategy (and thus no medication-related side effects) and the absence of any adverse events, the authors believe that such a strategy of rehabilitation is worthy of further multi-site investigation in a larger sample population. Such
large-scale investigations should be conducted with a view towards embedding such a program within the existing clinical framework for the management of Huntington’s disease. Implementation of a targeted multidisciplinary rehabilitation program into the Huntington’s disease population is anticipated to reduce dependence on the health system due to a reduction in the utilisation of health services as a result of a reduction in fall-related injuries, and may delay placement into residential and end-stage facilities by encouraging and maintaining patient independence.
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1 INTRODUCTION AND LITERATURE REVIEW

The HEROs Research Project undertook an in-depth investigation into the effects of an enriched environment on clinical measures of disease progression and quality of life in patients with early-mid stage Huntington’s disease. The study utilised a program of targeted, multidisciplinary rehabilitation as a non-pharmacological means to enhance neurological input, thus modulating brain activity and various physical parameters to ultimately impact on the signs and symptoms of Huntington’s disease.

Huntington’s Disease

Huntington’s disease is a fatal, neurodegenerative disease of genetic aetiology. It is a fully penetrant, autosomal dominant disorder caused by an expanded cytosine-adenine-guanine (CAG) trinucleotide repeat in exon 1 of the Huntingtin gene, which produces a mutant huntingtin protein\(^1\). Individuals with the expanded Huntingtin gene may be referred to as premanifest (before appearance of overt clinical symptoms) or manifest (after appearance of overt clinical symptoms), and are universally diagnosed as manifest based on the unambiguous presence of motor symptoms in accordance with the Unified Huntington’s Disease Rating Scale\(^2\). Onset typically occurs within the fourth to fifth decade of life\(^3\), and is inversely correlated with CAG repeat number\(^4,5\).

Huntington’s disease is characterised by progressive cognitive, affective and motor deterioration over a 15-25 year period, together with a spectrum of associated physical deficits including postural instability\(^6-10\), gait impairments\(^11-16\) and lean and fat tissue loss\(^17,18\). It is now well-documented that cortical degeneration preceeds clinical features, which further implicates long-term neuronal dysfunction in pathology\(^5,19-21\). Neuronal loss, particularly in the striatum and cortex, leads to additional neurological and motor deficits\(^22,23\). The death of striatal neurons subsequently alters output to other components of the basal ganglia and the thalamus, and thus to areas of the frontal cortex, compounding cortical deficits\(^24,25\). Hypothalamic degeneration, including neuronal loss in specific sub-nuclei, has been documented >15 years before predicted clinical onset\(^26-28\), and results in hypothalamic-pituitary-adrenal (HPA) axis dysregulation, producing circadian rhythm disturbances and metabolic imbalances unfavourably impacting on homeostasis and body weight\(^17,29-41\).

Cognitive features of the disease include deficits in attention, memory, cognitive flexibility, visuospatial perception and other aspects of executive function\(^42-44\). Affective features include dysphoria, agitation, irritability, apathy, anxiety and depression\(^45-48\), and symptoms may also precede clinical diagnosis\(^47\). Motor control progressively declines, affecting performance of daily activities. Individuals exhibit choreiform movements\(^49\), involuntary movements that are the clinical hallmark of the disease. Patients eventually become totally incapacitated, confined to a wheelchair and unable to function independently, at which time they may be placed in an end-stage care facility. Individuals ultimately become bed-bound.
and demented prior to death, which may be secondary to fall-related head injuries, choking or aspiration pneumonia, or as a result of suicide.

There is no cure for Huntington’s disease and no proven therapy for altering disease progression in patients. Currently, patients are managed with pharmacological interventions aimed at symptom management. Intense research is being conducted into the genetic aspects of the disease with a view to modifying or correcting the genetic mutation, either by gene silencing/interference, replacement of the expanded CAG repeat or by use of molecular chaperones to alter downstream effects. Whilst these strategies are promising, and are moving closer to clinical trials, they currently represent strategies for the future. Thus, there is an urgent need to identify alternate treatments capable of impacting on disease progression with a clear intent to improve quality of life whilst the search for a cure continues.

Epigenetic/Environmental Influences on Huntington’s Disease

In spite of the monogenic aetiology of Huntington’s disease, it is becoming increasingly apparent that other genes and epigenetic/environmental factors significantly contribute to variation in phenotype and age of onset. This has been amply illustrated by variation in disease manifestation and onset in monozygotic twins (either reared together or apart) and most notably in the Venezuelan kindred. Whilst this adds further complexity to understanding the disease process, it presents an exciting opportunity to impact on the disease course via non-pharmacological means, and without the need to directly modify gene function, which has proved enigmatic to-date.

One promising area of investigation is the use of an enriched environment to modify features of the disease and its clinical course. The paradigm of environmental enrichment employs increased mental, physical and social stimulation in animal models to enhance neurological input, thereby modulating brain activity and physical parameters to impact on disease progression. A number of studies have shown the benefits of such an approach in Huntington’s disease transgenic animal models, with evidence for it being able to delay the onset of motor abnormalities, slow disease progression, reduce impairments to weight loss, balance and physiological factors, and increase both neurogenesis and CNS levels of neurotransmitter receptors, neurotrophins and factors related to synaptic signal transduction pathways.

Despite these promising results, further clinical research and subsequent translation of such a program has been lacking, with only a few studies investigating this therapeutic approach in humans with Huntington’s disease. One study found that a program of multidisciplinary care in early-mid stage Huntington’s disease patients was tolerable, feasible and appreciated by patients, families and health care providers. Another study revealed immediate short-term
improvements in motor performance and daily activities, and highlighted that an intensive exercise rehabilitation program in this population was tolerable and beneficial. The intervention protocol utilised in the above study was of an intermittent nature, delivering six short, intense periods of enrichment (3 weeks each) over a two year period. However, as limited outcome measures were utilised, and patient compliance decreased considerably in subsequent sessions, the overall results should be interpreted with caution. Nevertheless, whilst improvements were not maintained between sessions, the patients maintained baseline levels of function over the two year period. The authors concluded that a continuous program of rehabilitation was likely to produce more significant results.

HEROs Research Project

In the present study, we sought to determine whether a prolonged and targeted program of multidisciplinary rehabilitation would more positively impact on the features of Huntington’s disease. To this end, we undertook a rigorous assessment of changes as a result of the intervention. We recruited twenty patients at early-mid stages of Huntington’s disease and comprehensively assessed them using an array of outcome measures investigating cognitive and physical performance, together with affective and quality of life indicators. We then designed and implemented a targeted and continuous 9 month program of multidisciplinary rehabilitation in half of the patients (Group B), whilst the remainder served as control subjects (Group A). The intervention provided in the HEROs Research Project was delivered during two separate intervention periods (Arms 1 and 2). High rates of compliance were noted in intervention participants.

Arm 1: The intervention consisted of 9 months of weekly clinical gym exercises together with 6 months of weekly, self-directed home-based exercises. Occupational therapy was delivered fortnightly on an individualised basis in the home environment for six months. The physical and cognitive stimulation components were designed to address key areas of deficit identified during baseline assessment in order to achieve functional and quality of life improvements. At the end of the intervention period, all patients were re-assessed to determine the effects of the intervention.

Arm 2: On completion of the first round of intervention, the intervention recipients (B) were given the option of either remaining in the program to receive a second period of physical rehabilitation, which consisted of supervised weekly clinical gym sessions and self-directed home-based exercises, or acting as a post-intervention control group. All participants with the exception of 1 agreed to receive the intervention. The control participants (A) were provided with an optimised rehabilitation program which also consisted of supervised weekly clinical gym sessions, a home-based exercise program and fortnightly occupational therapy. The physical intervention program, however, was optimised by the inclusion of weekly supervision of the home-based exercise program by an accredited exercise physiologist in the home environment.
The results presented in this Project Report clearly demonstrate that a continuous and targeted multidisciplinary rehabilitation program is well-tolerated by early-mid Huntington’s disease patients. Furthermore, participation in such a program improves motor, cognitive, affective and physical condition and produces adaptive physiological changes, including reduced deterioration in postural stability, relative to non-participation. For quality of life assessment, participants reported significant improvements in perceptions of mental health.

Additional research will be required to understand the neurological and physiological mechanisms underpinning these favourable responses to the intervention. Most significantly, analysis of brain imaging of participants should prove informative as to the potential neurological bases of these improvements or, at the very least, the effect of the multidisciplinary rehabilitation program on brain volume, function and integrity.
2 METHODOLOGY

Participant Recruitment

Patients at early-mid stages of Huntington’s disease were recruited via the Neurosciences Unit of the North Metropolitan Area Mental Health Service (Perth, Australia). Inclusion criteria included a positive test for the expanded Huntingtin gene, a clinical diagnosis of disease manifestation (Unified Huntington’s Disease Rating Scale Total Motor Score [UHDRS-TMS] ≥5), an ability to follow instructions and the physical capacity to perform the exercises. Exclusion criteria included evidence of recent substance abuse or an unstable psychiatric state, and the presence of other neurological disorders. Twenty-five participants were initially recruited, of which three withdrew for medical reasons prior to commencement (delusions/falls/increased frailty), and two withdrew consent (one participant/one control subject). Of the five that withdrew, age was the only demographic found to be different (64.9-71.5 in withdrawing participants vs mean of 54.7 ± 9.3 years). Twenty participants (9 intervention / 11 control) completed the initial intervention program, from which one was removed from analysis due to the diagnosis of an additional neurological disorder (Figure 1).

![Diagram of Participant Recruitment for Arm 1 of the Program]

*Figure 1: Participant Recruitment for Arm 1 of the Program.*
After completion of the initial 9 month period (Arms 1), all control participants were offered an optimised 9-month intervention program. The original intervention recipients were offered an additional 9 months of physical rehabilitation. The offer was accepted by all control participants and all except one initial intervention recipient. An additional three individuals joined the program at this point, and one individual from the original intervention group withdrew from the program after approximately one month to pursue other activities (Arm 2). Throughout the study, two individuals were lost to follow up from the optimised intervention group (not related to the intervention: one was deceased, one had major surgery). A cohort of age- and sex-matched, gene-negative family members were also tested to provide control data where relevant.

![Diagram](image)

**Figure 2: Participants accepted into Arm 2 of the Program.**

**Group Assignment**

Participants were initially allocated into two groups (A = Control; B = Intervention;) equally matched for cognitive and motor assessment scores at baseline. Specifically, the UHDRS-TMS was converted into a z-score for each individual. Cognitive assessment scores for each participant were converted to z-scores for each test, and then averaged to form an overall cognitive z-score per individual as an indication of cognitive function. Groups were derived by matching mean motor and cognitive z-scores to assign patients into two equally-matched groups. These groups were subsequently altered due to withdrawal of two of the patients, however the resultant mean z-scores per group remained similar (Table 1).
Table 1: Baseline cognitive and motor assessment scores per group.

<table>
<thead>
<tr>
<th>Characteristic</th>
<th>Intervention Group B (mean z-score [range])</th>
<th>Control Group A (mean z-score [range])</th>
</tr>
</thead>
<tbody>
<tr>
<td>Cognitive Assessment (overall z-score)</td>
<td>0.063 [-0.56 to 0.45]</td>
<td>-0.068 [-0.49 to 0.25]</td>
</tr>
<tr>
<td>Motor Assessment (z-score)</td>
<td>-0.125 [-1.710 to -1.398)</td>
<td>-0.157 [-1.478 to -1.312]</td>
</tr>
</tbody>
</table>

The group to receive the intervention was randomly assigned. Groups were statistically analysed for baseline differences in pertinent demographics, however no significant differences were found for any parameter (p>0.05) (Table 2).

Table 2: Baseline characteristics of the participants.

<table>
<thead>
<tr>
<th>Characteristic</th>
<th>Control Group A (mean ±SEM)</th>
<th>Intervention Group B (mean ±SEM)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Number of participants</td>
<td>10</td>
<td>9</td>
</tr>
<tr>
<td>Gender M/F</td>
<td>6/4</td>
<td>4/5</td>
</tr>
<tr>
<td>Age (years)</td>
<td>50.8 ± 2.5</td>
<td>53.7 ± 2.9</td>
</tr>
<tr>
<td>Age at Diagnosis (years)</td>
<td>48.2 ± 2.2</td>
<td>49.5 ± 2.8</td>
</tr>
<tr>
<td>Disease Duration (years)</td>
<td>2.6 ± 0.8</td>
<td>4.3 ± 1.2</td>
</tr>
<tr>
<td>CAG Number</td>
<td>44.1 ± 0.6</td>
<td>43.1 ± 1.1</td>
</tr>
<tr>
<td>CAG Index</td>
<td>427.9 ± 22.2</td>
<td>399.1 ± 53.7</td>
</tr>
<tr>
<td>Body Mass Index (kg/m^2)</td>
<td>27.0 ± 1.4</td>
<td>26.4 ± 1.4</td>
</tr>
</tbody>
</table>

Participants in the intervention group did, however, show slightly greater disease duration relative to the control group, which may serve to slightly underestimate any effect of the intervention.

Medication

As the intervention being trialled in this study is an adjunct treatment to be delivered in addition to the patients’ normal pharmacological therapy, patients remained on their normal medication regime throughout the study period, which was adjusted where necessary by their physicians. Some individuals in the intervention and control groups commenced new medication, including anti-psychotics, anti-depressants, anxiolytics and anti-dyskinetics, as detailed in Table 3 (page 22).
Table 3: Changes in patient medication and supplements throughout the trial period.

<table>
<thead>
<tr>
<th>Medication and Supplements Taken</th>
<th>Adjustments Arm 1</th>
<th>Adjustments Arm 2</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Group A Ctl</td>
<td>Group B Int 1</td>
</tr>
<tr>
<td>Anti-Psychotics</td>
<td></td>
<td></td>
</tr>
<tr>
<td>olanzapine</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>aripiprazole</td>
<td>3</td>
<td>2</td>
</tr>
<tr>
<td>haloperidol</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>quetiapine</td>
<td>0</td>
<td>1</td>
</tr>
<tr>
<td>Anti-Depressants</td>
<td></td>
<td></td>
</tr>
<tr>
<td>citalopram</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>escitalopram</td>
<td>1</td>
<td>0</td>
</tr>
<tr>
<td>sertraline</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>mirtazapine</td>
<td>1</td>
<td>0</td>
</tr>
<tr>
<td>venlafaxine</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>fluoxetine</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>paroxetine</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>Anxiolytics</td>
<td></td>
<td></td>
</tr>
<tr>
<td>clonazepam</td>
<td>0</td>
<td>1</td>
</tr>
<tr>
<td>lorazepam</td>
<td>1</td>
<td>0</td>
</tr>
<tr>
<td>nitrazepam</td>
<td>1</td>
<td>0</td>
</tr>
<tr>
<td>Anti-Dyskinetics</td>
<td></td>
<td></td>
</tr>
<tr>
<td>amantadine</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>benzhexol</td>
<td>-1</td>
<td>0</td>
</tr>
<tr>
<td>tetrabenazine</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>pramipexole</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>baclofen</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>Anti-Hypertensives</td>
<td></td>
<td></td>
</tr>
<tr>
<td>telmisartan</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>candesartan</td>
<td>-1</td>
<td>0</td>
</tr>
<tr>
<td>propanolol</td>
<td>-1</td>
<td>0</td>
</tr>
<tr>
<td>prazosin</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>aspirin</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>ramipril</td>
<td>0</td>
<td>1</td>
</tr>
<tr>
<td>atenolol</td>
<td>0</td>
<td>1</td>
</tr>
<tr>
<td>clopidogrel</td>
<td>0</td>
<td>1</td>
</tr>
<tr>
<td>quinapril</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>Anti-Hypercholesterol</td>
<td></td>
<td></td>
</tr>
<tr>
<td>atorvastatin</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>simvastatin</td>
<td>1</td>
<td>-1</td>
</tr>
<tr>
<td>Anti-Hypothyroid</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Thyroxine</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>Miscellaneous</td>
<td></td>
<td></td>
</tr>
<tr>
<td>analgesics</td>
<td>1</td>
<td>-1</td>
</tr>
<tr>
<td>gabapentin</td>
<td>1</td>
<td>0</td>
</tr>
<tr>
<td>multivitamins</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>vitamin D</td>
<td>0</td>
<td>1</td>
</tr>
<tr>
<td>risedronate</td>
<td>0</td>
<td>1</td>
</tr>
<tr>
<td>CoQ10</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>folate</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>iron</td>
<td>1</td>
<td>0</td>
</tr>
<tr>
<td>fishoil</td>
<td>-1</td>
<td>0</td>
</tr>
<tr>
<td>creatine</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>solifenacin</td>
<td>1</td>
<td>0</td>
</tr>
<tr>
<td>rabeprazole</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>pantoprazole</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>macrogol</td>
<td>1</td>
<td>0</td>
</tr>
<tr>
<td>loperamide</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>salbutamol</td>
<td>1</td>
<td>0</td>
</tr>
<tr>
<td>varenidine</td>
<td>-1</td>
<td>0</td>
</tr>
<tr>
<td>dimebon</td>
<td>-1</td>
<td>0</td>
</tr>
</tbody>
</table>

Figures represent the number of participants commencing (+) or no longer taking (-) each medication, per group and per timepoint. Columns represent the Control period (Ctl)*, Intervention 1 [9 months] (Int 1)* and Intervention 2 [18 months] (Int 2)* periods.
As can be determined from Table 3, medication changes occurred in both groups throughout the study period and present as a confounding factor to this research. When assessing the overall changes in anti-psychotics, anti-depressants, anxiolytics and anti-dyskinetics collectively, however, it is apparent that net changes in medication were reduced over the period of the study (Individuals experiencing medication changes: Control period: Group A=6; Intervention period 1, Group A=4, Group B=4; Delayed Intervention period 2, Group B=0). It is therefore unlikely that the positive effects of the intervention are due to medication. Medication effects are further discussed where relevant within the results section.

Assessments

All participants were assessed at baseline (0 months) and then after Arm 1 (12 months) and after Arm 2 (23 months) using an array of tests/assessments probing motor, cognitive and affective function, and physical parameters (body composition and postural stability) and quality of life perceptions. Muscular strength was assessed regularly throughout the intervention.

Clinical Motor Assessment:

A motor assessment was performed by a clinician using the UHDRS-TMS, which contains 17 scoring domains (Table 4). Higher scores depict greater impairment. All assessments were performed by the same clinician, who declared all participants fit to undertake the program.

**Table 4: Motor examination subscale of the Unified Huntington’s Disease Rating Scale.**

<table>
<thead>
<tr>
<th>Domain</th>
<th>Range of Score</th>
</tr>
</thead>
<tbody>
<tr>
<td>1. Ocular pursuit</td>
<td>0 - 4</td>
</tr>
<tr>
<td>2. Saccade initiation</td>
<td>0 - 4</td>
</tr>
<tr>
<td>3. Saccade velocity</td>
<td>0 - 4</td>
</tr>
<tr>
<td>4. Dysarthria</td>
<td>0 - 4</td>
</tr>
<tr>
<td>5. Tongue protrusion</td>
<td>0 - 4</td>
</tr>
<tr>
<td>6. Finger taps</td>
<td>0 - 4</td>
</tr>
<tr>
<td>7. Pronate/Supinate hands</td>
<td>0 - 4</td>
</tr>
<tr>
<td>8. Luria</td>
<td>0 - 4</td>
</tr>
<tr>
<td>9. Rigidity-Arms</td>
<td>0 - 4</td>
</tr>
<tr>
<td>10. Bradykinesia-Body</td>
<td>0 - 4</td>
</tr>
<tr>
<td>11. Maximal dystonia</td>
<td>0 - 4</td>
</tr>
<tr>
<td>12. Maximal chorea</td>
<td>0 - 4</td>
</tr>
<tr>
<td>13. Gait</td>
<td>0 - 4</td>
</tr>
<tr>
<td>14. Tandem walking</td>
<td>0 - 4</td>
</tr>
<tr>
<td>15. Retropulsion pull test</td>
<td>0 - 4</td>
</tr>
<tr>
<td>16. Weight</td>
<td>actual weight</td>
</tr>
<tr>
<td>17. Diagnosis confidence level</td>
<td>0 - 4</td>
</tr>
<tr>
<td><strong>Total</strong></td>
<td><strong>Sum of scores</strong></td>
</tr>
</tbody>
</table>
Affective Assessments:

Changes to psychological and behavioural state were assessed using patient- and carer-reported instruments which assessed patients’ perceptions of their own depressive symptoms and attitudes, and carers’ perceptions of behavioural changes in the patient.

*The Beck Depression Inventory-Second Edition,* currently used for assessment in Huntington’s disease\(^8\), is a self-report instrument encompassing 21 items of depressive symptoms and attitudes, providing an estimate of overall depression severity (Table 5).

**Table 5: The Beck Depression Inventory-II assesses 21 items of depressive symptoms and attitudes.**

<table>
<thead>
<tr>
<th>Variable</th>
<th>Range of Score</th>
</tr>
</thead>
<tbody>
<tr>
<td>1  Sadness</td>
<td>0-3</td>
</tr>
<tr>
<td>2  Pessimism</td>
<td>0-3</td>
</tr>
<tr>
<td>3  Past Failure</td>
<td>0-3</td>
</tr>
<tr>
<td>4  Loss of Pleasure</td>
<td>0-3</td>
</tr>
<tr>
<td>5  Guilty Feelings</td>
<td>0-3</td>
</tr>
<tr>
<td>6  Punishment Feelings</td>
<td>0-3</td>
</tr>
<tr>
<td>7  Self-Dislike</td>
<td>0-3</td>
</tr>
<tr>
<td>8  Self-Criticalness</td>
<td>0-3</td>
</tr>
<tr>
<td>9  Suicidal Thoughts or Wishes</td>
<td>0-3</td>
</tr>
<tr>
<td>10 Crying</td>
<td>0-3</td>
</tr>
<tr>
<td>11 Agitation</td>
<td>0-3</td>
</tr>
<tr>
<td>12 Loss of Interest</td>
<td>0-3</td>
</tr>
<tr>
<td>13 Indecisiveness</td>
<td>0-3</td>
</tr>
<tr>
<td>14 Worthlessness</td>
<td>0-3</td>
</tr>
<tr>
<td>15 Loss of Energy</td>
<td>0-3</td>
</tr>
<tr>
<td>16 Changes in Sleeping Pattern</td>
<td>0-3</td>
</tr>
<tr>
<td>17 Irritability</td>
<td>0-3</td>
</tr>
<tr>
<td>18 Changes in Appetite</td>
<td>0-3</td>
</tr>
<tr>
<td>19 Concentration Difficulty</td>
<td>0-3</td>
</tr>
<tr>
<td>20 Tiredness or Fatigue</td>
<td>0-3</td>
</tr>
<tr>
<td>21 Loss of Interest in Sex</td>
<td>0-3</td>
</tr>
<tr>
<td>Total</td>
<td>Sum of Scores</td>
</tr>
</tbody>
</table>
The Cambridge Behavioural Inventory (Revised) evaluates performance under 10 functional domains associated with activities of daily living, and has been validated as an effective assessment tool within the Huntington disease population\textsuperscript{86}. This questionnaire assesses multiple aspects of behaviour within each of the following domains:

1. Memory and Orientation
2. Everyday Skills
3. Self-Care
4. Abnormal Behaviour
5. Mood
6. Beliefs
7. Eating Habits
8. Sleep
9. Stereotypic and Motor Behaviours
10. Motivation

Carers report on the frequency of occurrence of problems associated with the patients’ behavioural aspects, on a 5-point scale (0=never/1=a few times per month/2=a few times per week/3=daily/4=constantly) with higher scores thus depicting more affected behaviour.

Cognitive Assessments:

Cognitive assessments validated for use in Huntington’s disease looked for changes in cognitive processing speed, learning, memory and executive function (eg cognitive flexibility and inhibition)\textsuperscript{43, 87}.

Symbol Digit Modalities Test: assesses cognitive flexibility and processing speed within a set time period. This test comprises a series of symbols to be substituted for a corresponding number and was performed in both oral and written forms and analysed for the number of symbols substituted within the given time period, and task accuracy.

Hopkins Verbal Learning Test-Revised: assesses learning, recall, and recognition. Subjects must recall as many words as possible immediately and after a 20 minute delay, and are assessed for total and delayed recall (number of words), retention (percentage of words retained) and recognition.

D-KEFS Colour Word Interference Test: assesses colour naming, word reading and inhibition to reveal a subject’s ability at the primary inhibition task, as a measure of executive function.

D-KEFS Trail Making Trials: assesses cognitive flexibility/executive function and comprises five separate tests. The primary task assesses number-letter switching, whilst the remaining four tasks assess individual components of visual scanning, literacy, numeracy and motor skills to reduce confounding factors not related to executive function.
Physiological Assessment:

Assessment targeted specific physiological deficits reported in Huntington’s disease populations that are likely to be affected by a multidisciplinary rehabilitation program. These factors included brain-derived neurotrophic factor (BDNF) and cortisol, which are also mediators of the HPA axis, and insulin.

**BDNF** is expressed within the hypothalamus and also in the hippocampus, from where the BDNF protein is transported to the striatum and promotes survival of striatal neurons. Reduced BDNF levels are associated with Huntington’s disease and are suspected of contributing to striatal pathology. BDNF levels are believed to be activity-dependent (increased neuronal activity increases BDNF output). Exercise induces increased BDNF expression in rodents and in human populations and cortical expression is upregulated in animal models, including models of Huntington’s disease, after exercise or environmental enrichment. Upregulation or over-expression of BDNF can rescue some symptoms of Huntington’s disease in animal models. Consistent with the expression patterns noted above, BDNF is associated with memory and learning and with body weight and energy homeostasis at both central and peripheral levels.

BDNF was extracted from sonicated, non-fasting whole blood samples to minimise protein degradation, and assayed with the Enzyme-Linked Immunosorbent Assay (ELISA) methodology using a commercially available ELISA Kit (BEK-2002-2P sandwich ELISA Kit, Biosensis), as per the manufacturer’s instructions. Each sample was run on two independent ELISA assays utilising internal standards provided with the kit and one standard sample present on all plates.

**Cortisol** production is also regulated by the hypothalamus, which releases corticotrophin releasing hormone, thereby stimulating the pituitary gland to signal for increased synthesis of cortisol from the adrenal cortex. Patients with Huntington’s disease may experience elevated cortisol levels and progressive increases in cortisol have been noted in Huntington’s disease animal models. Exercise has been shown to affect cortisol levels in humans, although this response is mediated by factors including the type of exercise, intensity of exercise and diurnal rhythm.

Cortisol was extracted from saliva collected using Salivette tubes. This methodology accurately reflects the free fraction of plasma cortisol and provides for easy collection of saliva, allowing for repeated sample collection throughout the day to generate a day curve. Other benefits include the stability of saliva at room temperature, and the absence of stress due to venipuncture. After oral and written instruction, participants were asked to chew on a cotton swab until saturated. The swab was then placed inside the Salivette tube labelled with identification codes and immediately placed in a freezer at -20°C. Participants were asked to refrain from eating or drinking for 1 hour or smoking for 30 minutes prior to each sample, and to refrain from alcohol consumption 12 hours prior to the first sample until
completion of the sampling period. Samples were taken at six timepoints over a 36 hour period (Day 1, 8.00pm; Day 2, approximately 8.00am [at awakening], 12.00 noon, 4.00pm, 8.00pm; Day 3, approximately 8.00am [at awakening]) to account for the diurnal rhythm, and samples were assayed using a commercially available ELISA kit (High Sensitivity Salivary Cortisol ELISA Kit 1-3002, Salimetrics). The average morning and evening samples were calculated from the two samples taken, providing four timepoint values per day. Daily cortisol values were calculated by summing the average value for each of the four timepoints. All samples were run in duplicate within the same assay.

**Insulin** levels have been reported to be altered in Huntington’s disease, and may occur subsequent to insulin resistance, reduced insulin sensitivity, transcriptional dysregulation of insulin or reduced insulin secretion. Exercise has been shown to increase insulin sensitivity and attenuate insulin resistance in adult and ageing rats after exercise. Likewise, insulin levels, insulin resistance and glucose tolerance may be favourably impacted by exercise in ageing and diabetic populations.

Insulin was indirectly quantified by assaying insulin-C peptide from plasma extracted from fasting blood samples utilising a commercially available ELISA kit (DE1293 ELISA Kit, Demeditech). All samples were run in duplicate within the same assay.

**Physical Assessments:**

Assessment of physical changes targeted known deficits in Huntington’s disease using predominantly quantitative outcome measures.

**Body Composition:** Whole body composition was assessed on a Hologic Dual Energy X-ray Absorptiometry (DEXA) Scanner (Hologic Discovery W, Waltham MA), with the participants lying relaxed in a supine position. Low-intensity x-rays filtered into two energy spectra, which are differentially absorbed by bone, lean and fat tissue, were used to quantify lean, bone and fat mass (measured in kilograms), and body mass index and bone mineral density were calculated. All DEXA scans were performed by the same operator throughout the study.

**Muscular Strength:** Increases in upper and lower body strength were determined by calculating session training volumes at regular intervals throughout the intervention period. Results are reported as volume of load (weight lifted (kg) × number of repetitions × number of sets) per training session for seated row, lat pull down, chest press, leg press, leg flexion and leg extension exercises, together with percentage change values. These exercises were chosen as they represent a broad assessment of upper and lower body muscle strength.

**Postural Stability:** Postural stability was assessed during performance of the Sensory Organisation Test® (SOT) utilising the NeuroCom Smart Balance Master computerised dynamic posturography apparatus. The SOT consists of six measures, using sway-referencing and eyes open/closed conditions, which systematically degrade or eliminate
somatosensory and visual information. Results are given as a percentage equilibrium score and an overall composite score (generated by NeuroCom Balance Master Version 8.3 software) (refer Appendix 1 for detailed information). SOT has previously been used to assess postural stability in various neurological populations\textsuperscript{134-136} and has demonstrated postural stability deficits in manifest and pre-manifest Huntington’s disease individuals\textsuperscript{7, 9, 10}. Differences in overall performance and for each test condition were assessed between groups, with higher scores depicting better balance. All Neurocom assessments were performed by the same assessor throughout the study.

**Quality of Life (QoL) Assessment:**

Changes to quality of life perceptions were assessed using both patient- and carer-reported instruments.

The SF-36 Health Questionnaire has been validated for use in Huntington’s disease populations\textsuperscript{137}. The revised SF-36v2 assesses QoL under 8 health dimensions (physical functioning; physical role limitations; body pain; general health; energy/vitality; social functioning; emotional role limitations; mental health) and two summary groups (physical health summary; mental health summary) rated on a 0 to 100 scale (0=severe impairment; 100=no impairment) and provides a summary assessment of physical and mental health-related QoL. SF-36v2 assessments were analysed using QualityMetric Health Outcomes\textsuperscript{TM} Scoring Software 3.0 (Quality Metric Inc., Lincoln, USA).

The Huntington’s disease Quality of Life Battery for Carers is a psychometrically-sound, disease-specific tool for assessing QoL in spousal carers of Huntington’s disease individuals\textsuperscript{138}. This instrument assesses QoL under four sections including 1) demographic and objective information; 2) aspects of caring; 3) satisfaction with life, and 4) feelings about life. We used sections 2-4 to assess the effect of the intervention on carers’ QoL. Analysis was performed for each question within each section, for section totals and for overall total scores.

**The Intervention Protocol**

**Arm 1** - An individualised program of intervention was designed over three months (0-3 months) targeting deficits previously reported and subsequently identified during baseline assessments. The control group (A) did not receive the intervention and were instructed to maintain normal daily activities, although this was not monitored. The intervention group (B) received physical, cognitive and social stimulation delivered as nine months of clinical gym exercise sessions (3-12 months), together with six months of self-directed, home-based exercises and occupational therapy programs (6-12 months). All participants were re-assessed at the end of the nine-month period (12 months).

**Arm 2** - After the first intervention period, all participants were offered the opportunity to participate in an additional round of intervention. All control participants (A) accepted the
offer to receive the intervention. Whilst the effects of a wash-out period (in which intervention recipients (B) then no longer receive the intervention and act as post-intervention controls for 9 months) may have provided valuable information as to the long-term effects of the intervention, it was evident that these participants did not wish to discontinue the intervention, and it was deemed unlikely that participants would abstain from activity during this period. Group B participants were therefore offered an additional round of the supervised clinical gym exercise program to allow a longitudinal investigation of the effect of the intervention program.

**Physical Stimulation:**

Based on physical performance at baseline, an exercise rehabilitation program was formulated which included a clinical gym component (Table 6) and a home-based component (Table 7). The program was supervised by trained exercise physiologists directly in the clinical gym environment and indirectly by reviewing the home-based program.

**Table 6: Clinical Gym-based Exercise Rehabilitation Program**

<table>
<thead>
<tr>
<th>Clinical Gym Exercise Rehabilitation Program</th>
</tr>
</thead>
<tbody>
<tr>
<td>Resistance Exercises:</td>
</tr>
<tr>
<td>Program:</td>
</tr>
<tr>
<td>---</td>
</tr>
<tr>
<td>3</td>
</tr>
</tbody>
</table>

Reps = repetitions; 1RM = 1 Repetition Maximum

The gym program consisted of weekly, sub-maximal resistance training that could be safely performed on resistance equipment. Training included exercises targeting major muscle groups responsible for the maintenance of gait, balance, postural stability and performance of everyday activities. Specifically, patients performed 3 sets of 8-12 repetitions with an inter-set rest of 1 minute. Training loads (weight) were set for each repetition at 65-80% of the individual’s maximum capability, and were increased when tolerated, with one 6RM (repetition maximum) testing session every six weeks to gauge strength improvements. High rates of compliance to the program were noted, with compliance at 77% for the first 9 months and 87% for the second 9 months.

In addition to the gym program, a home-based exercise program was performed for 1 hour, 3-4 days a week. Gross and fine motor exercises were chosen to broadly improve motor functioning, muscular strength and fine motor skills (Table 7). Multiple progressions and training loads were individualised for participants to cater for motor performance variability.
between subjects, and were increased when tolerated at the discretion of the exercise physiologist. Participants completed a diary of their weekly home-based program to monitor compliance, as the program was performed in a self-directed manner during Arm 1, with a compliance rate of 56% noted.

Throughout Arm 2, the home-based exercise program was optimised for the second round of intervention for Group A participants by the provision of an accredited exercise physiologist in the home environment once per week to provide supervision and to maximise efficacy and compliance, with a compliance rate of 73% noted. Group B participants continued to perform the home-based exercise program in a self-directed manner, although compliance was not monitored for this period.

Table 7: Home-based Exercise Rehabilitation Program.

<table>
<thead>
<tr>
<th>Home-based Exercise Rehabilitation Program</th>
</tr>
</thead>
<tbody>
<tr>
<td>Resistance Exercises:</td>
</tr>
<tr>
<td>Program:</td>
</tr>
<tr>
<td>-------------------------------------------</td>
</tr>
<tr>
<td>3</td>
</tr>
<tr>
<td>Additional Exercises:</td>
</tr>
<tr>
<td>Fine Motor Exercises:</td>
</tr>
</tbody>
</table>

Reps = repetitions; 1RM = 1 Repetition Maximum

Cognitive Stimulation:

A program of personalised cognitive stimulation was designed for each intervention recipient, targeting areas of deficit based on neurocognitive assessment of executive function, learning, memory and cognitive processing speed prior to intervention.

Based on results of the neurocognitive assessments at baseline, Neuropsychologists (blinded to the patient’s identification) made recommendations on an individualised patient basis to guide the Occupational Therapists in the key areas of deficit per patient, with recommendations for therapy.
Occupational therapists initially completed an Interest Checklist with patients, to determine key areas of interest that would be compatible with a cognitive stimulation program and would maximise engagement and compliance. Occupational therapists then visited patients within their home environment for one hour on a fortnightly basis to devise and deliver the program. Targeted goals were conceived within the area/s of interest and included planning and/or execution of tasks involving cooking, holidays, social activities and personal management together with stimulating tasks such as Sudoku and electronic games.

**Social Stimulation:**

Intervention participants received social stimulation via engagement with trainers, students and other participants during weekly group attendance in the gym, and during fortnightly individualised sessions with an Occupational Therapist. Control subjects did not receive these components of the intervention.

All study participants received social stimulation via interaction with researchers, students and health professionals during performance of testing procedures, assessments, tissue sampling and brain imaging, however the confounding effects of these components are controlled for within this study as stimulation was equally provided in both groups.
**Statistical Analysis:**

**Reporting:**

Comparisons were conducted between groups for baseline assessments, and for the mean change in values or scores over the assessment periods of the study (Figure 3). Mean change in values or scores refers to the change in values or scores during Assessment Period 1 (Analysis Point (AP) 1 – AP0; indicates the natural progression of the disease during the control period), Assessment Period 2 (AP2 – AP1; indicates the effect of the [single] Intervention 1) or Assessment Period 3 (AP3 – AP2; indicates the effect of the [longitudinal] Intervention 2). Within-group comparisons were conducted to directly assess changes within the same individuals in each cohort due to the effect of the intervention relative to no intervention (Group A; change during Assessment Period 2 versus change during Assessment Period 1) and the effect of a longitudinal intervention relative to a single intervention period (Group B; change during Assessment Period 3 versus change during Assessment Period 2). Results are reported as mean ± standard error of the mean (SEM) unless otherwise stipulated.

![Diagram](image)

**Figure 3:** Diagrammatic representation of Assessment Periods throughout the study. Diagram indicates Analysis Points and calculation of change in scores or values after Assessment Periods to determine changes after the Control, Intervention 1 and Intervention 2 periods.
Continuous variables:

**Between-Groups Comparisons:** Baseline differences and changes in values for continuous variables were assessed using a Student’s independent *t*-test (two-tailed). Differences in the ratio of participants per group experiencing a decline in postural stability versus no decline in postural stability after each timepoint were examined using the Two Sample Test of Proportion.

**Within-Group Comparisons:** Increases in training volume were assessed using a Student’s paired *t*-test (two-tailed). Changes within each group for other continuous variables were assessed using either a Two Way Analysis of Variance (ANOVA) or Repeated Measures ANOVA, where appropriate.

Ordinal Variables:

Mann Whitney U tests were used to assess ordinal variables, or where parametric assumptions were violated.

**Statistical Significance Values:**

Values of *p*<0.001 were considered statistically highly significant.

Values of *p*<0.05 were considered statistically significant.

Values of *p* >0.05 and ≤0.06 were considered borderline significant.

Values of *p* >0.06 and ≤0.100 were reported as a trend towards significance.

**Statistical Software:**

Statistical analyses were conducted using STATA version 9.1. (Stata Corp, 4905 Lakeway Dr, Texas 77845 USA). Effect size calculations (Cohen’s *d*) were performed in G*Power Software Version 3.0.10\(^{139}\).
3 RESULTS

The program was found to be feasible and well-tolerated in early-mid Huntington’s disease patients, with a high level of adherence to the program demonstrated. Assessments were performed at baseline and participants were further examined after control and intervention periods (Int 1 = overall group [A +B] after one intervention, Int 2 = Group B after second intervention). Results for these assessment periods are reported accordingly.

Clinical Motor Assessment:

To assess the effect of the intervention at a clinical/functional level, using the disease-specific UHDRS-TMS, which assesses a broad range of motor functions.

During the normal course of the disease process (control period), control individuals demonstrated a significant deterioration in motor scores ($p=0.003$; Table 8). After 9 months of intervention, clinical assessments showed significantly reduced deterioration (~65%) in motor scores for the entire group after Intervention 1; this deterioration was no longer significant ($p>0.05$; Table 8; Figure 4). Collectively, this represents a significant difference between groups due to the 9 month intervention ($p=0.020$). Of considerable importance is the further demonstration that this significantly reduced rate of motor deterioration is maintained after Intervention 2, producing a significant reduction in deterioration over an 18 month period ($p=0.028$).

Table 8: UHDRS Total Motor Score assessment indicating reduced motor deterioration after intervention, relative to the control group.

<table>
<thead>
<tr>
<th>Timepoint (n)</th>
<th>Difference in values</th>
<th>Within Groups ($p$)</th>
<th>Between Groups ($p$)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Ctrl (10)</td>
<td>Int 1 (13)</td>
<td>Int 2 (7)</td>
</tr>
<tr>
<td>UHDRS (TMS)</td>
<td>15.4 ±2.9</td>
<td>5.6 ±1.6</td>
<td>4.2 ±4.1</td>
</tr>
</tbody>
</table>

Values are reported as mean difference in score ± Standard Error of the Mean (SEM). Higher scores denote increasing impairment. Ctl = Control; Int 1 = Intervention 1; Int 2 = Intervention 2. Highlighting denotes statistical significance or a trend towards significance.
Figure 4: Change in Mean UHDRS Total Motor Score for participants after control and intervention periods (n; Control =11; Int 1=23; Int 2=7). Higher scores denote increasing impairment.

Although these results are confounded by alterations in medication in some patients, this is unlikely to account for the changes as both groups of patients had similar changes in medication over the period of the study, with no changes in anti-dyskinetic drugs occurring during Intervention 2 (Table 3).

Affective Changes:

Changes in measures of reported depressive symptoms and behaviour were assessed using the Becks Depression Inventory-II (self-reported) and the Cambridge Behavioural Inventory (Revised) (carer-reported).

There were no statistically significant differences between groups for depression scores at baseline (Group A 12.91 ±2.63; Group B 10.78 ±3.17; p>0.05). After the control period, control subjects demonstrated an uncharacteristic decrease in depression scores with a trend towards significance relative to 9 months intervention (p=0.089; Table 9). After 9 and 18 months of intervention, depression scores increased marginally (Table 9; Figure 5).

Table 9: Beck Depression Inventory-II Score indicating the change in score after intervention, relative to the control group.

<table>
<thead>
<tr>
<th>Timepoint (n)</th>
<th>Difference in values</th>
<th>Within Groups (p)</th>
<th>Between Groups (p)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Ctl</td>
<td>Int 1</td>
<td>Int 2</td>
</tr>
<tr>
<td></td>
<td>Ctrl (10)</td>
<td>Int 1 (19)</td>
<td>Int 2 (7)</td>
</tr>
<tr>
<td>BDI-II Score</td>
<td>-4.3 ±1.8</td>
<td>1.4 ±2.6</td>
<td>1.3 ±0.9</td>
</tr>
</tbody>
</table>

Values are reported as mean difference in score ± Standard Error of the Mean (SEM). Higher scores denote increasing symptoms. Ctl = Control; Int 1 = Intervention 1; Int 2 = Intervention 2. Highlighting denotes statistical significance or a trend towards significance.
Groups were further explored to assess differential effects of the intervention on depression scores. Whilst it was initially apparent that intervention recipients had a greater improvement in depression scores after one round of intervention relative to control subjects (refer Report 1), this was not the case after the delayed intervention in Group A participants. As indicated below (Figure 6), depression scores increased after intervention in Group A participants in contrast to the decrease in Group B participants. Thus, a differential response to depression scores after intervention was noted between groups, with Group B participants also showing a slightly decreased score after 18 months relative to baseline.

When depression scores were examined relative to assessment periods (Figure 7) it was apparent that depression scores decreased at midpoint relative to baseline, with an increase at final assessment relative to the midpoint, irrespective of the nature of the control and intervention assessment periods.
This pattern of change in scores indicates the possibility that other factors may have influenced the self-reported scores at these assessment points. Specifically, participation or impending participation in the intervention program may have influenced self-reported scores at midpoint prior to the delayed/longitudinal intervention. In contrast, the completion of the intervention program may have influenced self-reported scores at final assessment. Future studies would benefit from altering depression assessment timepoints to cater for this, with additional assessment tools and greater frequency of assessment to more effectively elucidate any effects of such an intervention on depression scores.

For the Cambridge Behavioural Inventory (Revised), there were no statistically significant differences between groups for overall scores at baseline (Group A 44.86 ±18.34; Group B 43.86 ±6.90; $p>0.05$). After one round of intervention there was a trend towards a significant deterioration in overall scores ($p=0.099$; Table 10; Figure 8). After 18 months of intervention, however, this trend was reversed, with carers reporting maintained scores for participants relative to scores after 9 months intervention, suggesting a delayed response to the intervention (Table 10; Figures 8-9).
Table 10: Cambridge Behavioural Inventory (Revised) Score indicating the change in score after intervention, relative to the control group.

<table>
<thead>
<tr>
<th>Timepoint (n)</th>
<th>Difference in values</th>
<th>Within Groups (p)</th>
<th>Between Groups (p)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Ctl (5) Int 1 (14) Int 2 (6)</td>
<td>Group A</td>
<td>Group B</td>
</tr>
<tr>
<td>CBI Score</td>
<td>-10.2 ±4.3 10.4 ±3.7 -1.1 8.8±</td>
<td>0.808 0.565 0.798 0.886 0.568 0.099 0.173</td>
<td></td>
</tr>
</tbody>
</table>

Values are reported as mean difference in score ± Standard Error of the Mean (SEM). Higher scores denote increasing symptoms. Ctl = Control; Int 1 = Intervention 1; Int 2 = Intervention 2. Highlighting denotes statistical significance or a trend towards significance.

Figure 8: Total score for the Cambridge Behavioural Inventory (Revised) after control and intervention periods. (n; Control=5; Int 1=14; Int 2=6). Higher scores denote increasing behavioural symptoms.

Figure 9: Total score for the Cambridge Behavioural Inventory (Revised) per group after control and intervention periods. (n; Control=5; Int 1=14; Int 2=6). Dashed line indicates the control period. Higher scores denote increasing behavioural symptoms.
Cognitive Changes:

Cognitive changes included significantly improved memory recall and retention and task accuracy, and reduced deterioration in cognitive processing speed, as detailed below.

The Symbol Digit Modalities Test was used as a measure of cognitive flexibility and processing speed, and task accuracy (Table 11).

Table 11. Results of the Symbol Digit Modalities Test assessing changes to cognitive flexibility, processing speed and task accuracy after intervention, relative to controls.

<table>
<thead>
<tr>
<th>Timepoint (n)</th>
<th>Difference in values</th>
<th>Within Group (p)</th>
<th>Between Groups (p)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Ctrl (10)</td>
<td>Int 1 (18)</td>
<td>Int 2 (7)</td>
</tr>
<tr>
<td>Correct Oral</td>
<td>2.4 ±1.59</td>
<td>-3.78 ±1.86</td>
<td>0.14 ±2.58</td>
</tr>
<tr>
<td>Correct Written</td>
<td>-2.3 ±1.27</td>
<td>-1.0 ±1.59</td>
<td>-0.85 ±1.49</td>
</tr>
<tr>
<td>Incorrect Oral</td>
<td>-0.2 ±0.49</td>
<td>-1.0 ±0.52</td>
<td>0.43 ±0.72</td>
</tr>
<tr>
<td>Incorrect Written</td>
<td>1.0 ±0.77</td>
<td>-1.18 ±0.58</td>
<td>0.57 ±0.92</td>
</tr>
</tbody>
</table>

Values are reported as mean difference in the number of responses decoded ± Standard Error of the Mean (SEM). Lower values for correct responses and higher values for incorrect responses denote greater impairment. Ctrl = Control; Int 1 = Intervention 1; Int 2 = Intervention 2. Highlighting denotes statistical significance or a trend towards significance.

Assessment of the correct number of oral responses (Figure 10) indicated a minor improvement after the control period followed by a significant deterioration after the delayed intervention for Group A participants (p=0.006) and for the overall group (p=0.035), relative to controls. Group B participants showed minor deterioration after one round of intervention and maintained levels after a second round of intervention. This may represent a delayed response to intervention as deterioration occurring after provision of one round of intervention is arrested after longitudinal intervention, however large within-group variability precludes a statistically significant result. Neither group showed significant changes at final assessment relative to baseline.
Figure 10: The difference in the number of correctly decoded symbols after oral performance of the Symbol Digit Modalities Test (SDMT) after intervention, relative to the control period. (n; Control =10; Int 1=18; Int 2=7). Lower scores denote greater impairment.

Assessment of the correct number of written responses (Figure 11) indicated that Group A participants had non-significant deterioration after the control period which was reduced after one round of intervention, with a trend towards significant deterioration over the two year period (p=0.083). Group B participants maintained levels throughout both intervention periods, and thus maintained their baseline score.

Figure 11: The difference in the number of correctly decoded symbols after written performance of the Symbol Digit Modalities Test (SDMT) after intervention, relative to the control period. (n; Control =10; Int 1=17; Int 2=7). Lower scores denote greater impairment.

Analysis of the incorrect number of oral responses (Figure 12) revealed that task accuracy was maintained after the control period but improved non-significantly in both groups after one round of intervention, which showed a trend towards significance in Group B (p=0.087). This was maintained after longitudinal intervention. Both groups had less oral errors overall relative to baseline, however this was not significant.
Figure 12: The difference in the number of errors during oral performance of the Symbol Digit Modalities Test (SDMT) after intervention, relative to the control period. (n; Control =10; Int 1=18; Int 2=7). Higher scores denote greater impairment.

Analysis of the incorrect number of written responses (Figure 13) identified more errors after the control period in contrast to improved accuracy after one round of intervention, which approached significance in Group B (p=0.063) and was significant for the overall group, relative to controls (p=0.033). Despite a minor increase in errors after a second round of intervention, Group B participants maintained baseline scores (Table 11).

Figure 13: The difference in the number of errors during written performance of the Symbol Digit Modalities Test (SDMT) after intervention, relative to the control period. (n; Control =10; Int 1=17; Int 2=7). Higher scores denote greater impairment.

Collectively, the data indicates that cognitive processing speed during performance of an executive function task was not significantly impacted by the intervention. There was an overall trend, however, towards either reduced deterioration or improvement in cognitive processing speed over time, despite differences between groups. There was a significant improvement in written task accuracy and a trend towards improved oral task accuracy after intervention, both of which were maintained at baseline levels after longitudinal intervention.
Changes to memory and learning were assessed using the Hopkins Verbal Learning Test-Revised. Despite a borderline significant deterioration after 9 months intervention in Group B participants ($p=0.056$), there was a trend towards a significant improvement in total recall after 18 months intervention relative to 9 months intervention ($p=0.072$; Table 12; Figure 14).

Table 12: Changes in memory and learning as indicated by changes in the Hopkins Verbal Learning Test-Revised.

<table>
<thead>
<tr>
<th>Timepoint (n)</th>
<th>Difference in values</th>
<th>Within Group ($p$)</th>
<th>Between Groups ($p$)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>Group A</td>
<td>Group B</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Ctl Int 1 Int 2</td>
<td>18 mths Ctl v Int 1 v Int 1</td>
</tr>
<tr>
<td>Ctl (10)</td>
<td>Int 1 (20)</td>
<td>Int 2 (7)</td>
<td></td>
</tr>
<tr>
<td>Total Recall (n)</td>
<td>0.60 ±1.15</td>
<td>-1.25 ±0.83</td>
<td>1.86 ±1.47</td>
</tr>
<tr>
<td>Delayed Recall (n)</td>
<td>-1.0 ±0.52</td>
<td>0.3 ±0.42</td>
<td>1.43 ±1.15</td>
</tr>
<tr>
<td>Retention (%)</td>
<td>-19.75 ±10.0</td>
<td>1.48 ±5.2</td>
<td>22.93 ±10.2</td>
</tr>
<tr>
<td>Recognition Discrimination Index (score)</td>
<td>1.0 ±0.68</td>
<td>-0.95 ±0.53</td>
<td>0.71 ±0.52</td>
</tr>
</tbody>
</table>

Values are reported as mean difference in score ± Standard Error of the Mean (SEM). Lower scores denote greater impairment. Ctl = Control; Int 1 = Intervention 1; Int 2 = Intervention 2. Highlighting denotes statistical significance or a trend towards significance.

![Figure 14: The difference in mean value for total recall, as a subcomponent of the Hopkins Verbal Learning Test-Revised (HVLT-R), after control and intervention periods. (n; Control =10; Int 1=20; Int 2=7). Lower scores denote greater impairment.](image-url)
There was a significant decline in delayed recall ($p=0.032$) and retention ($p=0.017$) after the control period, which contrasts with maintenance of ability after one round of intervention ($p>0.05$; Table 13). This produced a trend towards a significant difference in delayed recall ($p=0.075$; Figure 15) and a significant difference in retention ($p=0.046$; Figure 16) relative to the control period. After a second round of intervention, Group B participants demonstrated a trend towards a significant improvement in delayed recall ($p=0.095$) with a significant improvement in retention ($p=0.035$) compared to results after one round of intervention. Overall, 18 months of intervention in Group B participants rendered a borderline significant improvement in retention relative to all intervention recipients after 9 months of intervention ($p=0.055$).

**Figure 15:** The difference in mean value for delayed recall, as a subcomponent of the Hopkins Verbal Learning Test-Revised (HVLT-R), after control and intervention periods. ($n; \text{Control }=10; \text{Int } 1=20; \text{Int } 2=7$). (Int 1 = overall intervention group; Int 2 = longitudinal intervention group). Lower scores denote greater impairment.

**Figure 16:** The difference in mean value for retention, as a subcomponent of the Hopkins Verbal Learning Test-Revised (HVLT-R), after control and intervention periods. ($n; \text{Control }=10; \text{Int } 1=20; \text{Int } 2=7$). Lower scores denote greater impairment.
For the Recognition Discrimination Index, which provides a further assessment of memory retention, there were no significant within-group differences for either group, however a significant decline was noted after 9 months of intervention relative to the control period ($p=0.038$), which was partially reversed after a second round of intervention ($p=0.094$), inferring a delayed response to the intervention (Figure 17).

![Recognition Discrimination Index (HVLT-R) Difference in Mean Value](image)

**Figure 17**: The difference in mean value for the Recognition Discrimination Index, as a subcomponent of the Hopkins Verbal Learning Test-Revised (HVLT-R), after control and intervention periods. ($n; \text{Control}=10; \text{Int 1}=20; \text{Int 2}=7$). Lower scores denote greater impairment.

Taken together, these results indicate significant improvements in memory and learning ability, which is further augmented over longitudinal timeframes.
Executive function was assessed using the D-KEFS Colour Word Interference Test and the D-KEFS Trail Making Trials. The D-KEFS Colour Word Interference Test identified a number of significant improvements in cognitive processing speed (Table 13).

**Table 13:** Mean changes in cognitive processing speed and executive function detected after control and intervention periods using the D-KEFS Colour Word Interference Test.

<table>
<thead>
<tr>
<th>Timepoint (n)</th>
<th>Difference in values</th>
<th>Within Group (p)</th>
<th>Between Groups (p)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Ctl (10)</td>
<td>Int 1 (20)</td>
<td>Int 2 (7)</td>
</tr>
<tr>
<td>Colour naming</td>
<td>5.8 ±2.9</td>
<td>4.4 ±2.5</td>
<td>4.4 ±3.7</td>
</tr>
<tr>
<td>Word Reading</td>
<td>0.7 ±1.7</td>
<td>1.8 ±1.6</td>
<td>5.14 ±1.87</td>
</tr>
<tr>
<td>Inhibition</td>
<td>-1.8 ±4.1</td>
<td>3.1 ±4.8</td>
<td>12.83 ±6.8</td>
</tr>
</tbody>
</table>

Values are reported as mean difference in score ± Standard Error of the Mean (SEM). Lower scores denote greater impairment. Ctl = Control; Int 1 = Intervention 1; Int 2 = Intervention 2. Highlighting denotes statistical significance or a trend towards significance.

Assessment of colour naming ability (Figure 18) demonstrated non-significant improvements overall after 9 months of intervention and after an additional round of intervention in Group B participants, which culminated in a significant increase after 18 months of intervention, relative to baseline (p=0.039). No significant differences were seen for Group A.

![Colour Naming Ability](image)

**Figure 18:** The difference in mean value for colour naming ability, as a subcomponent of the D-KEFS Colour Word Interference Test, after control and intervention periods. (n; Control =8; Int 1=17; Int 2=7). Lower scores denote greater impairment.
Assessment of word reading ability (Figure 19) revealed a trend towards a significant improvement in Group A after nine months of intervention ($p=0.080$). Group B participants demonstrated a significant improvement after 18 months of intervention relative to 9 months ($p=0.011$), and a significant improvement after 18 months of intervention relative to baseline ($p=0.019$).

**Figure 19:** The difference in mean value for word reading ability, as a subcomponent of the D-KEFS Colour Word Interference Test, after control and intervention periods. ($n$; Control =9; Int 1=18; Int 2=7). (Int 1 = overall intervention group; Int 2 = longitudinal intervention group). Lower scores denote greater impairment.

The primary test of inhibition indicated that executive function (Figure 20) was maintained after the control and 9 month intervention periods. Whilst function improved after an additional round of intervention it failed to reach statistical significance, most likely due to the small sample size (Table 13). Nevertheless, these results indicate improvement in the performance of these tasks, which further improves over time.

**Figure 20:** The difference in mean value for inhibition, as a subcomponent of the D-KEFS Colour Word Interference Test, after control and intervention periods. ($n$; Control =8; Int 1=16; Int 2=6). Lower scores denote greater impairment.
Changes in motor and cognitive processing speed, and executive function were further explored using the D-KEFS Trail Making Trials assessment (Table 14).

**Table 14**: Mean changes in cognitive processing speed and executive function detected after control and intervention periods using the D-KEFS Trail Making Trials assessment.

<table>
<thead>
<tr>
<th>Timepoint (n)</th>
<th>Difference in values</th>
<th>Within Group (p)</th>
<th>Between Groups (p)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Ctl</td>
<td>Int 1</td>
<td>Int 2</td>
</tr>
<tr>
<td>Visual Scanning</td>
<td>6.2</td>
<td>4.1</td>
<td>-5.6</td>
</tr>
<tr>
<td></td>
<td>±3.9</td>
<td>±3.3</td>
<td>±7.6</td>
</tr>
<tr>
<td>Number Sequencing</td>
<td>-0.7</td>
<td>11.7</td>
<td>-10.7</td>
</tr>
<tr>
<td></td>
<td>±4.2</td>
<td>±4.3</td>
<td>±4.6</td>
</tr>
<tr>
<td>Letter Sequencing</td>
<td>-14.1</td>
<td>9.9</td>
<td>-4.9</td>
</tr>
<tr>
<td></td>
<td>±9.9</td>
<td>±4.3</td>
<td>±5.3</td>
</tr>
<tr>
<td>Motor Speed</td>
<td>-8.5</td>
<td>2.0</td>
<td>-8.0</td>
</tr>
<tr>
<td></td>
<td>±6.5</td>
<td>±6.0</td>
<td>±7.2</td>
</tr>
<tr>
<td>Number-Letter</td>
<td>-17.8</td>
<td>0.3</td>
<td>18.8</td>
</tr>
<tr>
<td>Switching</td>
<td>±17.0</td>
<td>±8.1</td>
<td>±7.3</td>
</tr>
</tbody>
</table>

Values are reported as mean difference in time taken to perform the task ± Standard Error of the Mean (SEM). Higher scores denote greater impairment. Ctl = Control; Int 1 = Intervention 1; Int 2 = Intervention 2. Highlighting denotes statistical significance or a trend towards significance.

Assessment of visual scanning ability (Figure 21) indicated that participants experienced a trend towards significant deterioration after the control period. After one intervention period, participants’ ability continued to deteriorate but at a lesser level relative to the control period, before improving after a second round of intervention. These changes, however, failed to reach statistical significance.

**Figure 21**: The difference in time taken to perform the Visual Scanning Task, as a subcomponent of the D-KEFS Trail Making Trials, after control and intervention periods. (n; Control =10; Int 1=20; Int 2=7). Higher scores denote greater impairment.
For number sequencing, whilst ability was maintained after the control period, there was a trend toward significant deterioration after one round of intervention ($p=0.076$). A significant improvement was detected after a second round of intervention ($p=0.008$; Figure 22).

**Figure 22:** The difference in time taken to perform the Number Sequencing Task, as a subcomponent of the D-KEFS Trail Making Trials, after control and intervention periods. ($n; \text{Control }=10; \text{Int 1}=20; \text{Int 2}=7$). Higher scores denote greater impairment.

Assessment of letter sequencing ability indicated an unexpected improvement that was borderline significant after the control period ($p=0.054$), followed by significant deterioration after one round of intervention ($p=0.015$). In contrast, there was a trend towards a significant improvement after a second round of intervention ($p=0.066$; Figure 23).

**Figure 23:** The difference in time taken to perform the Letter Sequencing Task, as a subcomponent of the D-KEFS Trail Making Trials, after control and intervention periods. ($n; \text{Control }=10; \text{Int 1}=20; \text{Int 2}=7$). Higher scores denote greater impairment.
For assessment of motor speed changes, participants performed faster after the control period, then slowed after one round of intervention, before improving after two rounds of intervention, however these changes were not significant (Figure 24).

![Motor Speed D-KEFS Trail Making Trials Difference in Mean Value](image)

**Figure 24:** The difference in time taken to perform the Motor Task, as a subcomponent of the D-KEFS Trail Making Trials, after control and intervention periods. (n; Control =10; Int 1=20; Int 2=7). Higher scores denote greater impairment.

The primary test of number-letter switching indicated that despite improved time to perform the cognitive flexibility (executive function) task after the control period and maintenance of ability after one round of intervention, participants took significantly longer to perform the task after the second round of intervention ($p=0.036$; Figure 25). This is confounded by approximately half the participants being unable to complete this task at each assessment point, including the baseline assessment.

![Number-Letter Switching D-KEFS Trail Making Trials Difference in Mean Value](image)

**Figure 25:** The difference in time taken to perform the Number-Letter Switching Task, as a subcomponent of the D-KEFS Trail Making Trials, after control and intervention periods. (n; Control =4; Int 1=10; Int 2=4). Higher scores denote greater impairment.
In summation, the data from the clinical neurocognitive assessments indicates a trend towards improved motor and cognitive function (but not cognitive flexibility/executive function), which appears to be maximised after longitudinal intervention, suggesting a delayed response to improvement after intervention for some aspects of cognitive function.

**Physiological Changes:**

The biomarkers assessed in this study represent factors that are known to be perturbed in Huntington’s disease, and may be expected to change due to the provision of cognitive and physical stimulation.

**BDNF:**

Quantification of BDNF levels from whole blood was performed using the BDNF BEK-2002-2P sandwich ELISA Kit, with an intra-assay coefficient of variation of 1.8%, and an inter-assay coefficient of variation of 5.6%.

Consistent with previous reports\(^9\)\(^-\)\(^10\)\(^1\), the ELISA results indicated lower levels of BDNF in the whole blood of Huntington’s disease individuals, relative to normative data (Figure 26).

![Mean BDNF Concentration at Assessment Points](image)

**Figure 26:** The mean whole blood BDNF concentration, after control and intervention periods. (n: Control =10; Int 1=17; Int 2=7). Dashed line indicates the control period. The highlighted area denotes normative values of 13,470-19,470 pg/mL\(^11\).

The assay to detect changes in mean BDNF concentration over the time period of the study revealed a non-significant reduction in mean BDNF levels after the control period \((p=0.838)\) which worsened after one round of intervention \((p=0.393)\). Although reduced levels still occurred after longitudinal intervention, this was ameliorated compared to 9 months intervention \((p=0.469; Table 15; Figure 27)\).
Table 15: Mean changes in whole blood BDNF concentration after control and intervention periods, as determined by ELISA.

<table>
<thead>
<tr>
<th>Timepoint (n)</th>
<th>Difference in values</th>
<th>Within Group (p)</th>
<th>Between Groups (p)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Ctl (10)</td>
<td>Int 1 (17)</td>
<td>Int 2 (7)</td>
</tr>
<tr>
<td>BDNF (pg/mL)</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>-144 ±652</td>
<td>-794 ±428</td>
<td>-166 ±940</td>
</tr>
</tbody>
</table>

Values are reported as mean difference in concentration ± Standard Error of the Mean (SEM).
Ctl = Control; Int 1 = Intervention 1; Int 2 = Intervention 2. Highlighting denotes statistical significance or a trend towards significance.

Figure 27: The difference in mean whole blood BDNF concentration, after control and intervention periods. (n; Control =10; Int 1=17; Int 2=7).

When assessing group differences over time, the profile of changes to BDNF levels was very similar between groups, irrespective of control or intervention periods (Figure 26). It is therefore unlikely that the ameliorated deficit after 18 months of intervention is due to existing group differences.

As differential gender responses have been previously reported, we further assessed changes to mean BDNF concentration for males and females, separately (Figure 28). A differential gender response after longitudinal intervention is noted. Whilst females demonstrate a consistent reduction in BDNF concentration throughout the study, males experienced an increase in BDNF concentration after longitudinal intervention, however large variation and a small sample size within the group precluded a statistically significant result (p=0.380). This suggests a delayed response for changes to BDNF levels in males after intervention.
Figure 28: The difference in mean whole blood BDNF concentration per gender, after control and intervention periods. (n; Control = 10 [Female 4/Male 6]; Int 1 = 17 [Female 10/Male 7]; Int 2 = 7 [Female 3/Male 4]). Dotted plots represent females, solid colour plots represent males.

Finally, to further assess factors affecting the reduced deficit in BDNF levels after longitudinal intervention, we analysed medication changes in participants. Of the three females present in the longitudinal cohort, one experienced increased levels of BDNF, without changes in medication. Of the four males present in the longitudinal cohort, three had increased levels of BDNF without any changes in medication.

The reduced BDNF change after 18 months of intervention coincides with the significant increase in memory and learning also witnessed after longitudinal intervention.

Cortisol:

Quantification of salivary cortisol levels was performed using a High Sensitivity Salivary Cortisol ELISA Kit, with an Intra-assay coefficient of variation of 1.36%, and an inter-assay coefficient of variation of 4.60%.

Analysis of mean salivary cortisol levels identified a diurnal secretion pattern, with no significant baseline differences between groups ($p>0.05$). The diurnal pattern was similar to that seen in the general population, albeit at lower levels in the first half of the day, most noticeable in the morning, with a minor increase outside the normal range in the evening (Figure 29).
Figure 29: Graph depicting the diurnal pattern of salivary cortisol concentration at baseline. \((n; \text{Group A}=11, \text{Group B}=8)\). Boxes denote normative range for each timepoint. Green series depicts midpoint of normative data from the general population\(^{140}\).

Over the period of the study the diurnal pattern of salivary cortisol concentration demonstrated similar, minor changes in the morning and evening timepoints during both control and intervention periods, but generally remained within the normative range except for the evening data after longitudinal intervention (Figures 30-31; Table 16).

Figure 30: Graph depicting the diurnal pattern of salivary cortisol concentration at midpoint. \((n; \text{Group A}=11, \text{Group B}=8)\). Boxes denote normative range for each timepoint. Green series depicts midpoint of normative data from the general population\(^{140}\).
Figure 31: Graph depicting the diurnal pattern of salivary cortisol concentration at final assessment. (n; Group A=7, Group B=7). Boxes denote normative range for each timepoint. Green series depicts midpoint of normative data from the general population. Further analysis of the changes to the diurnal cortisol pattern during the control and intervention periods is represented in Figure 32.

Quantification of overall mean daily salivary cortisol levels over the time period of the study revealed an overall increase in daily cortisol levels after the control period and after each intervention period (Figure 33). Despite some significant within-group fluctuations, increased levels were only statistically significant after longitudinal intervention, and predominantly reflect the increased levels identified in this cohort in the evening (p=0.002; Table 16; Figures 31-32).
Table 16: The difference in mean salivary cortisol concentration after control and intervention periods, as determined by ELISA.

<table>
<thead>
<tr>
<th>Timepoint (n)</th>
<th>Ctl (12)</th>
<th>Int 1 (17)</th>
<th>Int 2 (7)</th>
<th>Within Group (p)</th>
<th>Between Groups (p)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Morning</td>
<td>0.202 ±0.034</td>
<td>0.058 ±0.070</td>
<td>0.046 ±0.090</td>
<td>&lt;0.001</td>
<td>0.630</td>
</tr>
<tr>
<td>Midday</td>
<td>0.078 ±0.033</td>
<td>0.046 ±0.027</td>
<td>0.021 ±0.059</td>
<td>0.041</td>
<td>0.702</td>
</tr>
<tr>
<td>Afternoon</td>
<td>0.079 ±0.044</td>
<td>0.024 ±0.026</td>
<td>-0.036 ±0.065</td>
<td>0.103</td>
<td>0.259</td>
</tr>
<tr>
<td>Evening</td>
<td>-0.008 ±0.046</td>
<td>-0.005 ±0.024</td>
<td>0.168 ±0.068</td>
<td>0.864</td>
<td>0.357</td>
</tr>
<tr>
<td>Daily</td>
<td>0.351 ±0.115</td>
<td>0.122 ±0.107</td>
<td>0.199 ±0.174</td>
<td>0.012</td>
<td>0.394</td>
</tr>
</tbody>
</table>

Values are reported as mean difference in concentration ± Standard Error of the Mean (SEM). Ctl = Control; Int 1 = Intervention 1; Int 2 = Intervention 2. Highlighting denotes statistical significance or a trend towards significance.

**Figure 33:** The difference in mean daily salivary cortisol concentration after control and intervention periods. (n; Control=11; Int 1=15; Int 2=7).

Due to the significant within-group differences, the difference in values were further analysed and expressed as a percentage change after the [control or intervention] period relative to values at commencement of the period, detailing the different response of each group. As can be determined from Table 17, an increase in daily salivary cortisol values were experienced after the control period (193%), which continued albeit it at a reduced rate after 9 months of intervention (139%) and 18 months of intervention (128%). This is consistent with reports of progressive increases in urinary cortisol in Huntington’s disease patients which increase in parallel with disease progression.30
Table 17: Relative difference in mean salivary cortisol concentration after control and intervention periods, as determined by ELISA.

<table>
<thead>
<tr>
<th>Timepoint (n)</th>
<th>Relative difference in values expressed as a percentage*</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Morning</td>
</tr>
<tr>
<td>Post Control (Group A only)</td>
<td>279</td>
</tr>
<tr>
<td>Post Intervention 1 Overall (Group A) (Group B)</td>
<td>156</td>
</tr>
<tr>
<td></td>
<td>110</td>
</tr>
<tr>
<td></td>
<td>207</td>
</tr>
<tr>
<td>Post Intervention 2 (Group B only)</td>
<td>114</td>
</tr>
</tbody>
</table>

*Values are reported as a percentage change in mean values after control or intervention periods, relative to before control and intervention periods. Groups are also shown separately for Intervention 1 to denote group differences.

When analysed to assess changes attributable to the different groups, a differential group response to the intervention can be seen (Table 17; Figure 34). Despite increasing cortisol levels after the control period, levels are maintained at normal levels after 9 months of intervention in Group A participants. This contrasts with increased cortisol levels after 9 months intervention in Group B participants, which then decreases to near normal levels after subsequent intervention. This suggests that the cortisol response to exercise may occur in a dosage-dependent manner, and contrasts the effects of an optimised intervention (Group A) to a longitudinal intervention (Group B).

**Figure 34:** Mean daily salivary cortisol concentration after control and intervention periods. (n; Control=11; Int 1=15; Int 2=7). Note the increase after the control period, the differential response between groups during Intervention 1, and reduced levels after longitudinal intervention. The highlighted area denotes normative values of 0.65-0.90µg/dL.
**Insulin C-peptide:**

Levels of insulin were indirectly quantified by assaying levels of plasma insulin C-peptide extracted from fasting blood samples using the Demeditec DEC1293 ELISA Kit, with an intra-assay coefficient of variation of 2.50%, and an inter-assay coefficient of variation of 4.20%.

Baseline values were at the lower end of the normative range, consistent with previous reports detailing reduced insulin levels in Huntington’s disease\(^\text{33, 40, 127-129}\), with no significant differences existing between groups (p>0.05). Analysis of the changes in mean insulin C-peptide levels revealed a progressive increase in concentration throughout the study, which was reduced after intervention (Table 18; Figure 35). Overall, the intervention significantly reduced the increase in insulin C-peptide levels relative to the control period (p=0.048), and the increase in concentration after intervention 2 was not significantly different relative to levels after intervention 1 (p=0.738).

**Table 18:** The difference in mean fasting insulin C-peptide concentration after control and intervention periods, as determined by ELISA.

<table>
<thead>
<tr>
<th>Timepoint (n)</th>
<th>Difference in values</th>
<th>Within Group (p)</th>
<th>Between Groups (p)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Ctl (7) Int 1 (16)</td>
<td>Int 2 (6)</td>
<td>Group A</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>Ctl Int 1</td>
</tr>
<tr>
<td>Insulin C-peptide (ng/mL)</td>
<td>4.06 ±0.55</td>
<td>2.41 ±0.46</td>
<td>2.66 ±0.89</td>
</tr>
</tbody>
</table>

Values are reported as mean difference in concentration ± Standard Error of the Mean (SEM). Ctl = Control; Int 1 = Intervention 1; Int 2 = Intervention 2. Highlighting denotes statistical significance or a trend towards significance.

**Figure 35:** The difference in mean insulin C-peptide concentration after control and intervention periods. (n; Control =7; Int 1=16; Int 2=6). (Int 1 = overall intervention group; Int 2 = longitudinal intervention group).
Group A participants experienced a highly significant increase in insulin C-peptide levels after the control period \((p=0.0003)\) and a significant increase after 9 months of intervention \((p=0.038)\). Group B participants experienced a highly significant increase in insulin C-peptide levels after 9 months of intervention \((p=0.0001)\) and a significant increase after a further 9 months of intervention \((p=0.030)\), which culminated in significant increases over the 18 month period relative to baseline for both groups \((A, p=0.009; B, p=0.007; \text{Table 18}; \text{Figure 36})\).

**Figure 36: Mean insulin C-peptide concentration after control and intervention periods.** \((n; \text{Control }=7; \text{Int 1}=16; \text{Int 2}=6)\). Dashed line indicates the control period. The highlighted area denotes normative values of 0.4-2.1ng/mL\(^{141}\).

Therefore, lower levels of insulin reported at baseline significantly increased over the period of the study. This increase was significantly reduced after 9 months of intervention.

**Physical Changes:**

The intervention group showed significant improvements in body composition and muscular strength, and a reduction in the number of people demonstrating deteriorating postural stability relative to the control group.

**Body Composition:**

Whilst there were no significant differences in body composition between the groups at baseline for any factor \((p>0.05)\), significant differences between groups were apparent after the initial control versus intervention period. Control participants exhibited a significant reduction in body weight relative to baseline \((p=0.013)\), as is typically seen in advancing Huntington’s disease \(^{17, 18}\), which was predominantly represented by a significant loss of fat mass \((p=0.013)\), and a loss of fat-free mass (lean + bone mineral content) which was borderline significant \((p=0.058)\). In contrast, participants receiving one round of intervention demonstrated a significant increase in total body mass \((p=0.006)\) and fat mass \((p=0.002)\), with a trend towards increased fat-free mass \((p=0.097)\) relative to control subjects (Table 19;
Figures 37-39). After a second round of intervention, participants gained additional total body mass which was essentially a gain in fat-free mass. Overall, this group maintained their body weight relative to baseline with a trend towards a gain in total body mass ($p=0.081$) represented by a significant increase in fat-free mass ($p=0.012$). Bone mineral density was unchanged in both groups (Table 19).

**Table 19:** Changes in body composition as determined by DEXA scanning.

<table>
<thead>
<tr>
<th>Timepoint (n)</th>
<th>Difference in values</th>
<th>Within Group ($p$)</th>
<th>Between Groups ($p$)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Ctl (10)</td>
<td>Int 1 (20)</td>
<td>Int 2 (7)</td>
</tr>
<tr>
<td>Fat Mass (kg)</td>
<td>-1.9 ±0.7</td>
<td>0.6 ±0.4</td>
<td>-0.1 ±1.1</td>
</tr>
<tr>
<td>Lean Mass (kg)</td>
<td>-1.0 ±0.5</td>
<td>0.3 ±0.4</td>
<td>0.8 ±0.8</td>
</tr>
<tr>
<td>BMD (g/cm³)</td>
<td>0.0 ±0.0</td>
<td>0.0 ±0.0</td>
<td>0.0 ±0.0</td>
</tr>
<tr>
<td>Lean+BMC (kg)</td>
<td>-1.0 ±0.6</td>
<td>0.3 ±0.4</td>
<td>0.8 ±0.8</td>
</tr>
<tr>
<td>Total Mass (kg)</td>
<td>-2.9 ±1.1</td>
<td>1.0 ±0.7</td>
<td>0.7 ±1.8</td>
</tr>
<tr>
<td>BMI (kg/m²)</td>
<td>-0.60 ±0.3</td>
<td>0.03 ±0.3</td>
<td>0.09 ±0.6</td>
</tr>
</tbody>
</table>

Values are reported as mean difference ± Standard Error of the Mean (SEM). Higher scores denote greater impairment. Ctl = Control; Int 1 = Intervention 1; Int 2 = Intervention 2. Highlighting denotes statistical significance or a trend towards significance.

A more in-depth analysis of body composition indicated that total body mass, fat mass and fat-free mass were predominantly lost from the trunk and to a lesser extent from the legs in control subjects, and these regions were represented by gains in these factors after intervention. Interestingly, loss of total body mass in control subjects occurred in a fat/fat-free mass ratio of 2:1. Similarly, gains in total body mass after one round of intervention identified a fat/fat-free mass ratio of 2:1. This was rectified in participants receiving a second round of intervention, who gained fat-free mass with minor loss of fat mass (Figures 37-39).
Figure 37: Total and regional changes in total body mass after intervention, relative to control subjects; a) total body mass changes; b) regional body mass changes; (n; Control =10; Int 1=20; Int 2=7). Error bars denote SEM.

Figure 38: Total and regional changes in fat mass after intervention, relative to control subjects; a) total fat mass changes; b) regional fat mass changes; (n; Control =10; Int 1=20; Int 2=7). Error bars denote SEM.

Figure 39: Total and regional changes in lean mass after intervention, relative to control subjects; a) total lean mass changes; b) regional lean mass changes; (n; Control =10; Int 1=20; Int 2=7). Error bars denote SEM.
Muscular Strength:

Volumetric muscle strength changes in the upper and lower body showed highly significant, progressive increases in total group muscle strength, and significant increases for female (p<0.03) and male (p<0.02) subgroups, in all exercises after 9 months intervention (p<0.0005; Figure 40).

Figure 40: Increases in total training volume per session for upper and lower limbs during the 9 month intervention period (n=21; values are shown for the whole group [21], and separately for female [11] and male [10] sub-groups to indicate gender response). Highly significant increases in muscle strength were achieved for each of the above exercises after 9 months intervention (p<0.0005). Percentage change values are shown for the whole group and separately for female and male subgroups.

It is particularly noteworthy that maximal strength gains after the first round of intervention in Group B participants appeared between time points 7-9, which coincided with 5-6 months of intervention (refer Report 1). This highlighted a delay in strength gains after resistance training; in normal healthy individuals strength gains occur within a few weeks of
commencement of training\(^{142}\). It is of interest to note that this phenomenon is no longer evident when assessing the total group after intervention (Groups A + B). It is plausible, therefore, that the optimised home-based intervention bolstered the training levels of individuals, and intervention recipients thus demonstrate a better, more normalised response to resistance training subsequent to greater frequency of training.

During a further 9 months of training, individuals in the longitudinal group continued to achieve strength increases, although these were generally not statistically significant, except for leg flexion ($p=0.032$).

**Postural Stability:**

Our results indicate that the multidisciplinary rehabilitation program employed in this study did not appear to significantly impact on the quantitative level of postural stability over the time period of the study ($p>0.05$; Figure 4). Both groups showed deterioration in equilibrium scores throughout the conditions tested and also for the composite balance score, which is a weighted average of all test conditions, albeit at a reduced level after intervention.

![Figure 41: Change in postural stability for the sensory organisation test (n; Control=10; Int 1=20; Int 2=7). Differences in mean equilibrium score are expressed as a percentage for each condition. Comp = mean composite equilibrium score (refer Appendix A for further details of each SOT condition).](image)

Further subgroup analysis of the difference in the mean composite score indicates a differential response to 9 months of intervention in the two groups (Figure 42).
Figure 42: Change in the mean composite score per group for the sensory organisation test \((n; C=10; I^1=20; I^2=7)\). Differences in the mean composite score are expressed as a percentage for each condition.

The results, however, are confounded by a difference between the groups for mean equilibrium score at each timepoint throughout the period of the study indicated. Nevertheless, the delayed intervention group demonstrated significant deterioration over the 18 month period (which included no intervention for 9 months; \(p=0.041\)) compared to a non-significant deterioration after 18 months of intervention in the longitudinal intervention group \((p=0.109; \text{Figure 43})\).

![Neurocom Balance Test](image)

Figure 43: Mean Composite scores indicating postural stability per group for the sensory organisation test \((n; \text{Control}=10; I^1=20; I^2=7)\). Dashed line indicates the control period.

Results were further assessed to investigate the number of participants experiencing deterioration in postural stability compared with those who maintained or improved postural stability. Maintenance was arbitrarily designated as the baseline equilibrium score \(\pm 2\) points (ie \(\pm 2\%\)). When assessing the relative number of participants in each group that demonstrated maintained or improved postural stability in contrast to deterioration after each timepoint, there was a significant reduction in the number of participants demonstrating deterioration \((p=0.046; \text{Table 20})\).
Table 20: Relative proportion of individuals demonstrating maintained or improved postural stability after control and intervention periods.

<table>
<thead>
<tr>
<th>Timepoint</th>
<th>Maintained/Improved</th>
<th>Deterioration</th>
<th>Intervention</th>
<th>(p^*)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Control Period</td>
<td>30.0%</td>
<td>70.0%</td>
<td>0 months</td>
<td>0.128</td>
</tr>
<tr>
<td>Intervention 1</td>
<td>50.0%</td>
<td>50.0%</td>
<td>9 months</td>
<td>0.163</td>
</tr>
<tr>
<td>Intervention 2</td>
<td>71.4%</td>
<td>28.6%</td>
<td>18 months</td>
<td>0.046</td>
</tr>
</tbody>
</table>

*Two sample test of proportion

These results indicate a stepwise reduction in the number of individuals experiencing deteriorating postural stability after intervention, which reached statistical significance after 18 months.

Quality of Life Assessment:

The intervention produced perceived improvements in several areas related to QoL for participants. In particular, when the QoL was assessed using the Short Form 36v2, the intervention group demonstrated minor improvements in general health, energy/vitality, and social functioning which was evident after longitudinal intervention but failed to reach statistical significance. Overall, impressions of mental health, which unexpectedly improved in the control group, experienced a minor decline after 9 months intervention before significantly improving after longitudinal intervention (\(p=0.043\)) (Figure 44).

![SF-36v2 Health Scores](image)

**Figure 44:** The difference in mean SF-36v2 Health Scores for intervention and control groups (n; I=9; C=10). Reported as 8 functional domains (PF physical functioning; PRL physical role limitations; BP body pain; GH general health; E/V energy/vitality; SF social functioning; ERL emotional role limitations; MH mental health) and two summary groups (PHS physical health summary; MHS mental health summary). Higher scores depict more favourable impressions of their health.
The data collected from the HD Quality of Life Battery for Carers was reviewed, however due to the smaller number of carers participating in the study, it was deemed that there was insufficient data for meaningful statistical analysis, and the information was therefore excluded from this report.
4 DISCUSSION

Despite numerous studies documenting the therapeutic benefit of environmental enrichment in Huntington’s disease transgenic mouse models, further research into a human Huntington’s disease population has been relatively unexplored. Here we have successfully undertaken a prolonged and continuous program of multidisciplinary rehabilitation in a cohort of early-mid Huntington’s disease individuals and have demonstrated that such a program is feasible and tolerable, with clear therapeutic benefit in motor and cognitive function, muscular strength, body composition, balance and some aspects of affective functioning. Notably, physical improvements detected as a result of the intervention typically precede changes in cognitive performance. Changes to physiological factors coincide with the abovementioned improvements, providing further evidence of the benefits of the multidisciplinary rehabilitation protocol investigated in this research project.

The clinical motor assessment conducted throughout this research project clearly identifies a statistically significant reduction in motor deterioration after 9 months of intervention, which is maintained at the significantly reduced level after 18 months of intervention. Similarly, muscular strength assessments conducted in the gym throughout the intervention identified highly significant improvements after 9 months of intervention, with continued increases occurring during longitudinal intervention. Whilst the commencement of improvement in muscle strength appears to be delayed for a few months in the non-optimised intervention (1 supervised session [gym] per week) relative to the optimised intervention (2 supervised sessions [gym/home] per week), highly significant improvements can still be detected after 9 months. These results are very encouraging given the high level of statistical significance obtained in such a small sample size, and this is highly likely to impact on the performance of everyday activities.

The improvements in gross motor function observed in this study most likely occurred subsequent to increased muscle mass and strength. Training in patients with Multiple Sclerosis supports this theory, where enhanced function occurs concomitant with muscle hypertrophy\textsuperscript{143}, indicating that training in neurodegenerative disease populations can alter the disease and improve function by increasing muscle mass. Physical therapy has also been shown to increase muscle mass in the elderly, which has been attributed to changes in muscle fibre type, dimensions and architecture\textsuperscript{144-147}, which are generally mediated by increases in mitochondrial biogenesis and function\textsuperscript{148-150}.

The demonstrated changes to body composition similarly reflect changes detectable within 9 months, with significant improvements in total body mass and fat mass and a trend towards significance for a gain in fat-free (lean) mass identified. This is complimented by further significant gains in lean mass and stabilisation of fat mass after an additional round of intervention. These changes in body composition represent a striking benefit given that there
is typically progressive weight loss and musculoskeletal atrophy in individuals with Huntington’s disease. Likewise, reduced decline in postural stability is likely to afford considerable benefit to individuals with Huntington’s disease, as fall-related injuries are a significant problem for this population. Our finding that the decline in postural stability in intervention recipients is significantly reduced by the intervention, with a stepwise reduction in the number of individuals experiencing deterioration occurring after 9 months, and reaching statistical significance after 18 months, is a welcome and much anticipated outcome of this research. Similarly, another study in Huntington’s disease has identified improved balance after a home-based exercise program, as detected by significant improvement in the Berg Balance Score relative to control individuals. The reduced decline in postural stability demonstrated in this study may reflect changes in the rate of force development and the increased strength of postural musculature, as these are primary mediators of static and dynamic balance. Therapeutic changes may also have arisen due to modulation of proprioceptive feedback mechanisms, particularly at the level of muscle spindles and golgi tendon organs. Such changes have previously been reported in Parkinson’s disease and aged populations following physical therapy targeted at improving balance, although this has not been looked at previously in Huntington’s disease. Significantly, the results presented here, identifying amelioration of both weight loss and postural stability decline, reflect previous results in various animal models of Huntington’s disease after environmental enrichment.

In contrast to the physical improvements noted above, which occurred within 9 months of intervention, changes to cognitive functioning showed an overall trend towards improvement after 18 months, generally indicating a delayed response to the intervention. It is interesting to note that these cognitive improvements after longitudinal intervention occurred in the absence of the cognitive stimulation component over the final 9 months of intervention. Whilst the intervention reported here did not appear to significantly impact on some aspects of executive function, significant improvements in memory and learning, task accuracy and cognitive processing speed of visual scanning and sequencing tasks were achieved over longitudinal timeframes. This contrasts with the Zinzi et al study, which demonstrated maintenance of baseline cognitive capacity, determined using the Mini Mental State Examination, after two years of intermittent rehabilitation in individuals with early-to-mid stage Huntington’s disease, and validates the use of a continuous program of intervention for provision of maximal benefits. Furthermore, Baker et al demonstrated improved executive function performance in women with amnestic mild cognitive impairment after six months of high-intensity aerobic exercise, suggesting that optimising the program with provision of a greater aerobic component may provoke executive function improvement.

Affective changes after intervention appear to be limited in this study. Our inability to significantly impact on depression mimics that of the other multidisciplinary rehabilitation program provided to early-to-mid-stage patients with Huntington’s disease. Changes to behavioural aspects during performance of everyday activities, as reported by carers/support people, indicates a trend towards increased behavioural problems after 9 months intervention,
in contrast to maintenance of these levels after 18 months, suggesting a delayed response to the intervention consistent with that of the cognitive data.

The physiological factors examined in this study also showed a favourable response to the intervention, which appeared to be sensitive to optimisation of the home-based program and to the timeframe of the intervention. BDNF has been extensively studied as a hallmark pathological biomarker of Huntington’s disease. Whilst BDNF levels decreased throughout the control period with a further, greater reduction after the 9 month intervention period, this reduction was ameliorated to a lower level after longitudinal intervention. The longitudinal data showed a differential gender response, with an increase in 3 of the 4 males noted. This delayed increase coincides with significant improvements in memory and learning achieved after longitudinal intervention. Our results are in accord with previous evidence demonstrating reduced BDNF/BDNF levels in Huntington’s disease, in both humans and animal models\textsuperscript{95-100}, and activity-dependent increases in BDNF expression in animals\textsuperscript{77, 82, 104, 105, 109, 110} and in human populations\textsuperscript{106-108}. During the course of this investigation, however, a recent publication has questioned the utility of BDNF measurement as a biomarker for the monitoring of Huntington’s disease\textsuperscript{162}. The authors assessed BDNF/BDNF levels in plasma and serum of control, pre-symptomatic and symptomatic Huntington’s disease individuals and found that BDNF levels were not reliably different between groups, citing that intra-group variability and methodological aspects affect the measurement, and concluded that BDNF levels from blood were neither informative nor reliable, especially when associated with quantification from serum. We believe this is mitigated to a certain extent in this project by our use of repeated measurements from the same population of individuals (to assess changes within the same individuals after control/intervention periods) and from the use of whole blood to minimise degradation associated with stored serum and plasma, as detailed previously\textsuperscript{118}. Nevertheless, due to our small sample size, particularly for the longitudinal cohort, and the presence of medication changes, we encourage cautious interpretation of this data.

In contrast to previous literature, which has identified increased cortisol levels in Huntington’s disease, cortisol levels of participants were within the expected normative range throughout the study, with the exception of an emerging increase in evening values after longitudinal intervention. This may reflect within-group variation, which included individuals at early- and mid-stages of the disease, as a previous report has indicated increased cortisol levels in moderate and moderate-late stages of the disease, but not at early stages of the disease\textsuperscript{30}. The increase in evening values after longitudinal intervention contrasts with evening values after 9 months of intervention, and group differences indicate the possibility of a differential response between the optimised and non-optimised cohorts. It is also interesting to note the concomitant elevation in BDNF and cortisol levels (predominantly evening values) after longitudinal intervention. A parallel association between BDNF and cortisol levels has been observed previously after aerobic exercise in patients with mild cognitive impairment, although in this study the levels decreased after intense exercise\textsuperscript{161}. Although cortisol levels were significantly increased after the control
period, which is consistent with previous reports of increasing levels of cortisol which parallel stages of the disease, this increase occurred at a reduced rate after intervention, suggesting a favourable effect of the intervention.

Collectively, the observed changes to BDNF and cortisol levels after intervention, together with significant improvements in body composition, identify a positive influence of the intervention on the HPA axis. Other studies have demonstrated the capacity for exercise to modify the activity of the HPA axis in patients with mild cognitive impairment and in healthy aged populations.

Similar to the results for BDNF, the baseline results for insulin C-peptide reflect the current literature on Huntington’s disease, indicating lower levels of insulin relative to the general population. The increased level of insulin C-peptide over the time period of the study in this cohort may reflect disease-related changes to insulin sensitivity and insulin resistance, known to occur in Huntington’s disease. Interestingly, hyperinsulinaemia and insulin resistance have also been reported in patients with other polyglutamine repeat disorders. In this study, the intervention produced a significant reduction in the increased level of insulin C-peptide after 9 months. Furthermore, consistent with the results obtained for cortisol, it appears that the optimised intervention had a more ascertainment effect on the progressive increase in insulin C-peptide when compared to the non-optimised intervention, and may indicate beneficial changes to insulin sensitivity/resistance as a result of greater frequency of exercise. Further work will be required to investigate this in detail.

Previously, environmental enrichment has been shown to modulate CNS pathology in Huntington’s disease, demonstrated by changes in peristriatal volume and in the motor and behavioural phenotype in transgenic Huntington’s disease mouse models. Environmental enrichment influences Huntington’s disease at a cellular level by increasing the birth and maturation of adult hippocampal neurons, ameliorating a deficit known to occur in Huntington’s disease. At a molecular level, it is proposed that environmental enrichment modulates neuronal function by rectifying mutant huntingtin-mediated deficiencies in gene expression, affecting key neurotransmitter receptors and synaptic signal transduction pathways (e.g., cannabinoid CB1/glutamate/dopamine receptors, DARPP-32, PSD-95) and thus effects changes to synaptic function and experience-dependent plasticity. Such changes to signalling and synaptic molecules elicit cognitive and behavioural changes in transgenic Huntington’s disease mouse models, including enhanced spatial learning and memory, reduced task time and accuracy, and changes to exploratory behaviour. Another significant benefit of this type of intervention is its ability to endogenously provide multiple therapeutic factors at physiological dosages. Whilst symptoms of the disease may be influenced by the exogenous provision of these therapeutic factors, the mode of delivery, particularly within the CNS, remains problematic.
Our results indicate multiple, widespread benefits subsequent to provision of the HEROs multidisciplinary rehabilitation program, and provide a solid platform for further, large-scale multi-site investigations. In keeping with the above changes noted in animal models of Huntington’s disease after environmental enrichment, the neurocognitive and psychological benefits of the program implemented here similarly included significant improvements in memory and learning, task accuracy, and reduced behavioural problems. Furthermore, the results for cognitive function, body composition and postural stability identify a response to the intervention which continues to improve over time, validating the use of a continuous program for optimal benefits. Further investigations will be required to understand the neurological aspects that underpin the changes observed after intervention, and especially to decipher whether the improvements elicited by such interventions are acting at the level of the signs and symptoms of the disease, or whether they are impacting on the progression of the disease itself.

The plethora of studies performed to-date support the theory that environmental enrichment counteracts the effects of reduced cortical activity noted in Huntington’s disease\textsuperscript{78}. In humans, one of the few studies in this area has shown a direct relationship between lifestyle and cortical activity which identified an association between increased age of onset, reduced phenotypic expression and an active lifestyle in Huntington’s disease patients\textsuperscript{170}. Similarly, high levels of complex mental activity across the lifespan correlates with reduced hippocampal atrophy in healthy older individuals\textsuperscript{171}. The findings from this study, which reveal the benefits of multidisciplinary rehabilitation in individuals at early-to-mid stages of Huntington’s disease without any apparent adverse effects, together with evidence of delayed onset in animal models of Huntington’s disease after environmental enrichment, provides support for the notion that such intervention strategies would have optimum impact when provided to gene-positive individuals who are yet to undergo manifestation of the disease. Preservation of the pre-manifest state by delaying the onset of the disease for as long as possible and maintaining individuals at the best functioning level possible is likely to represent significant improvements in quality of life for individuals at-risk of Huntington’s disease and their families, together with a substantially reduced burden on the health sector.
Limitations:

Although positive, the present study has some limitations, including the confounding effects of changes to medication, although they appear to have minimal impact on this study. Moreover, it is difficult to accurately monitor patient compliance in home-based programs, although this is a common methodological problem. This was reduced as a confounding factor in the second cohort as patients received weekly home visits by exercise physiologists and completed a diary to ensure compliance.

The small cohort of Huntington’s disease subjects greatly limited the statistical power of the current study and increased the potential for type 2a errors (false-negative findings). Longitudinal studies with greater sample sizes are warranted to replicate these findings and to fully investigate the effects of multidisciplinary rehabilitation on the disease course. Furthermore, detailed investigations which measure the effects of such a program on CNS pathology will help to determine possible mechanisms underlying the observed changes in the signs and symptoms of the disease.

Future intervention studies would benefit from more frequent functional, affective and physiological assessments throughout the study period so as to more closely define the timing of these changes, which in turn may provide insight into the adaptive mechanisms. Investigation of participants after a washout period, where the intervention is withheld for a time-period, may also provide valuable information as to the ability of the intervention to impact on disease progression, rather than at the level of signs and symptoms of the disease.
5 CONCLUSION:

In conclusion, our results indicate that individuals with Huntington’s disease can be enriched by successfully participating in an ongoing multidisciplinary rehabilitation program as an adjunct to their normal pharmaceutical regime. Multidisciplinary rehabilitation programs cannot replace the need for medication in Huntington’s disease, however our data suggest that engagement in such a program may delay the natural history of this condition and reduce the need for adjuvant treatment commonly associated with negative side effects.

Whilst the paradigm of multidisciplinary rehabilitation implemented in this study has yet to be shown to be disease modifying, we have demonstrated its ability to significantly impact on motor and cognitive function, and quality of life, together with additional improvements in physical, physiological and affective factors, in a cohort of individuals at early-mid stages of the disease. Moreover, it is apparent that the continuous and longitudinal nature of the rehabilitation program provided here produced superior benefits which appear to accumulate over time for some physical and functional aspects.

Another significant forte of this study is the applicability of the intervention to other neurodegenerative or neurotrauma populations. As the intervention trialled in this research project did not directly target the genetic aetiology of the disease, but rather the signs and symptoms of the disease, it is anticipated that this intervention will be relevant for other neurological populations with only minor adaptations envisaged. Indeed, other programs delivering cognitive or physical rehabilitation programs have shown some benefit for other neurodegenerative diseases such as Parkinson’s and Alzheimer’s diseases, and taken together, these research studies are revealing an emerging trend for such programs to favourably impact on disease symptoms and quality of life for patients.

Given the incurable and severe nature of the disease in question, together with the non-pharmacological nature of this therapeutic strategy (and thus no medication-related side effects) and the absence of any adverse events, the authors believe that such a strategy of rehabilitation is worthy of further multi-site investigation in a larger sample population. These large-scale investigations should be conducted with a view towards embedding such a program within the existing clinical framework for the management of Huntington’s disease.

Implementation of such a program of targeted multidisciplinary rehabilitation into the Huntington’s disease population is anticipated to reduce dependence on the health system due to a decline in the utilisation of health services due to a diminution in fall-related injuries, and may delay placement into residential and end-stage facilities by encouraging and maintaining patient independence.
REFERENCES


155. Khalil, H., An Exploratory Study of Mobility-related Outcome Measures and an Exercise Intervention in People with Huntington’s Disease (HD), 2012, Cardiff University.


Appendix 1 – Sensory Organisation Test:

The apparatus consists of a support surface and a three-sided visual surround, each of which can be rotated individually or simultaneously in reference to the subject’s anterior-posterior (AP) postural sway. Each SOT condition comprised three trials.

Table A1: NeuroCom Smart Balance Master Sensory Organisation Test Conditions

<table>
<thead>
<tr>
<th>Condition</th>
<th>Forceplates</th>
<th>Eyes</th>
<th>Visual Surround</th>
<th>Sensory feedback</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>stationary</td>
<td>open</td>
<td>stationary</td>
<td>All feedback accessible</td>
</tr>
<tr>
<td>2</td>
<td>stationary</td>
<td>closed</td>
<td>stationary</td>
<td>Visual feedback eliminated</td>
</tr>
<tr>
<td>3</td>
<td>stationary</td>
<td>open</td>
<td>sway-referenced</td>
<td>Visual feedback degraded</td>
</tr>
<tr>
<td>4</td>
<td>sway-referenced</td>
<td>open</td>
<td>stationary</td>
<td>Somatosensory feedback degraded</td>
</tr>
<tr>
<td>5</td>
<td>sway-referenced</td>
<td>closed</td>
<td>stationary</td>
<td>Visual feedback eliminated; somatosensory feedback degraded; reliance on vestibular feedback</td>
</tr>
<tr>
<td>6</td>
<td>sway-referenced</td>
<td>open</td>
<td>sway-referenced</td>
<td>Visual and somatosensory feedback degraded; reliance on vestibular feedback</td>
</tr>
</tbody>
</table>

A trial was considered unsuccessful (ie a fall) when the participant either changed their base of support (eg stepping off plate or losing balance) or could not complete the trial duration. Performance on each trial was assessed using a percentage equilibrium score generated by comparison of the difference between the participant’s actual AP sway and the theoretical maximum AP sway of 12.5° (where 0 = fall; 100 = no sway), with higher scores depicting better balance. Overall performance is assessed by comparing an overall composite score calculated using the weighted average of all six conditions. Equilibrium score calculations were generated using NeuroCom Balance Master Version 8.3 software Neurocom International 172.