DOI:10.14753/SE.2024.3035

SEMMELWEIS EGYETEM

DOKTORI ISKOLA

Ph.D. értekezések

3035.

BERENTE-KERESTÉLY DALIDA BORBÁLA

Klinikai neurológiai kutatások
című program

Programvezető: Dr. Kovács Tibor, egyetemi docens

Témavezető: Dr. Horváth András Attila, egyetemi adjunktus

DOI:10.14753/SE.2024.3035

THE ROLE OF VISUOSPATIAL AND VISUOMOTOR FUNCTIONS IN THE DIAGNOSIS OF ALZHEIMER'S DISEASE

PhD thesis

Dalida Borbála Berente-Kerestély, MD

Semmelweis University Doctoral School

János Szentágothai Neurosciences Division

Supervisor: András Attila Horváth, MD, Ph.D

Official reviewers: Tibor Kovács, MD, Ph.D
Dénes Zádori MD, Ph.D

Head of the Complex Examination Committee:
János Réthelyi MD, Ph.D

Members of the Complex Examination Committee:
Zoltán Hidasi MD, Ph.D
Gábor Barcs MD, Ph.D

Budapest
2024

DOI:10.14753/SE.2024.3035

Table of Contents

DOI:10.14753/SE.2024.3035

DOI:10.14753/SE.2024.3035

DOI:10.14753/SE.2024.3035

List of Abbreviations

Aβ: amyloid-beta

AD: Alzheimer's disease

ACE: Addenbrooke's Cognitive Examination

ACE total: total score of Addenbrooke's Cognitive Examination

a-MCI: amnestic mild cognitive impairment

APP: amyloid precursor protein

BDI: Beck Depression Inventory

CDR: Clinical Dementia Rating Scale

fMRI: functional MRI

HC: healthy control

MCI: Mild Cognitive Impairment

MMSE: Mini Mental State Examination

MRI: Magnetic Resonance Imaging

pTau: phosphorylated Tau

RAVLT: Rey Auditory Verbal Learning Test

RAVLT sum-5: sum of the first five trials' scores of the Rey Auditory Verbal Learning Test

RAVLT7: delayed recall score of the Rey Auditory Verbal Learning Test

sMRI: structural MRI

STAI: Spielberger State-Trait Anxiety Inventory

STAI-S: state anxiety version of the Spielberger State-Trait Anxiety Inventory

STAI-T: trait anxiety version of the Spielberger State-Trait Anxiety Inventory

TMT: Trail-Making Test

TMT-A: part A of the Trail-Making Test

TMT-B: part B of the Trail-Making Test

VLOM: ratio of the sum of verbal fluency and language subscores and the sum of the orientation and memory subscores of the Addenbrooke's Cognitive Examination

VS: visuospatial

VM: visuomotor

DOI:10.14753/SE.2024.3035

I. Introduction

I.1. Prevalence of Alzheimer’s disease

As the world’s population is aging, the prevalence of major neurocognitive disorders (NCD) is rapidly increasing. Currently, approximately 55 million people have dementia worldwide, and the number of patients is expected to triple by 2050. Alzheimer's disease (AD) is the most common type of major neurocognitive disorders in the elderly, accounting for two-thirds of dementia cases in this age group worldwide (1). It is estimated that by 2050 there will be 106.8 million people with Alzheimer's Disease globally, and NCDs will be the leading cause of morbidity (2, 3). Together with the growing prevalence, the financial burden of the disease is also rapidly increasing. However, the available treatment options are limited and, as of now, not curative. It is apparent that immediate effort must be made to decrease the socioeconomic burden of the disease (1). A model by Brookmeyer at al. estimated, that delaying the disease onset by a year would result in a 11.8 million decrease in patients worldwide, while a 2-year delay in disease onset would lead to almost the double of that, at 22.8 million cases less (2). Furthermore, previous studies have proposed that the first apparent cognitive symptoms are preceded by the structural and functional alterations in the nervous system that had started decades earlier (4). Additionally, when the first symptoms arise, the brain has suffered permanent damage from the pathology that started several years before (5). These factors resulted in a shift in aging research to identifying cases as early as possible, preferably in prodromal phases that might represent a critical window for interventions (6).

I.2. History of Alzheimer’s disease

The first documented case of an Alzheimer’s patient goes back to 1901, when Alois Alzheimer, a German psychiatrist and neuropathologist, first met Auguste D his 51-year-old future patient at the Hospital for the Mentally Ill and Epileptics, in Frankfurt am Main. The patient’s cardinal symptoms included impaired memory and comprehension, disorientation, unstable behaviour, and impaired psychosocial functioning. Upon following the disease course of the patient, he made a precise description of the symptoms, examinations, and

interactions with the patient. Among others, his observations included disorientation, memory impairment, difficulty in speech during evening hours, difficulty in naming objects and reading. He followed Auguste D's case until the patient died on the 8th of April 1906 (7). Alzheimer performed a post-mortem study of the patient's brain and submitted his findings for the 37th Conference of South-West German Psychiatrists in Tübingen in 1906. Later, he also published his findings describing plaques and neurofibrillary tangles (8). In 1910, Emil Kraepelin was the first to use the term Alzheimer's disease in the chapter of "Senile and Presenile Dementias" of the 8th edition of Handbook of Psychiatry (9).

I.3. Pathogenesis of Alzheimer's disease

While numerous factors contribute to the pathogenesis of Alzheimer's disease, the two principal ones are amyloid-beta (Aβ) and phosphorylated Tau (pTau). Aβ has a leading role in amyloid plaque formation, while pTau constitutes neurofibrillary tangles. Aβ is produced through the cleavage of amyloid precursor protein (APP), a transmembrane protein whose definite physiological function is still uncertain. It has been proposed to have a role in cell-to-cell adhesion and neuronal signalling. APP is processed by three proteolytic enzymes: α-, β-, and γ-secretases. Regarding the cleavage products, we distinguish the amyloidogenic and non-amyloidogenic pathways of APP processing. The non-amyloidogenic pathway includes the α- and γ-secretases and results in the production of sAPPα, the soluble APPα. The sAPPα has a well-described function: it has an essential role in neuronal survival and plasticity and has been found protective against neural toxicity caused by Aβ. On the other hand, the amyloidogenic pathway is composed of the β- and γ-secretases. First, as a result of the β-secretase's action, sAPPβ, the soluble APPβ is released. Consequently, the proteolytic cleavage of the APP's carboxy-terminal fragment by the γ-secretase results in peptides with various chain lengths, including Aβ40 and Aβ42 in the brain. Of the two, Aβ42 has a greater tendency to aggregate. It has a role in synaptic dysfunction, neurotoxicity, and is the central component of amyloid plaques in AD. The process of Aβ aggregation starts with Aβ monomers accumulating and forming oligomers, which then aggregate to form protofibrils, protofibrils then form fibrils which ultimately form Aβ plaques by aggregating.

DOI:10.14753/SE.2024.3035

The other key protein in AD pathology is tau, a microtubule binding protein, which has a role in axonal transport and in maintaining the stability of microtubules. Tau function is regulated by different kinds of post-translational modifications of which phosphorylation is the most important in AD. A characteristic hallmark of AD is the presence of hyperphosphorylated tau. Hyperphosphorylation results in the protein's detachment from the microtubule and augments its aggregation and potential to form neurofibrillary tangles. Similarly to Aβ, the stages of tau aggregation include monomers, oligomers, fibrils, filaments, and finally neurofibrillary tangles (10). However, the localization and propagation of Aβ and tau aggregation is different. As described by Braak and Braak, six stages are distinguished: first, the prodromal phase is associated with the transentorhinal stages (I-II), early to moderate disease phase is linked to the limbic stages (III-IV), while moderate to late AD is associated with the isocortical stages (V-VI) (Figure 1.) (11).

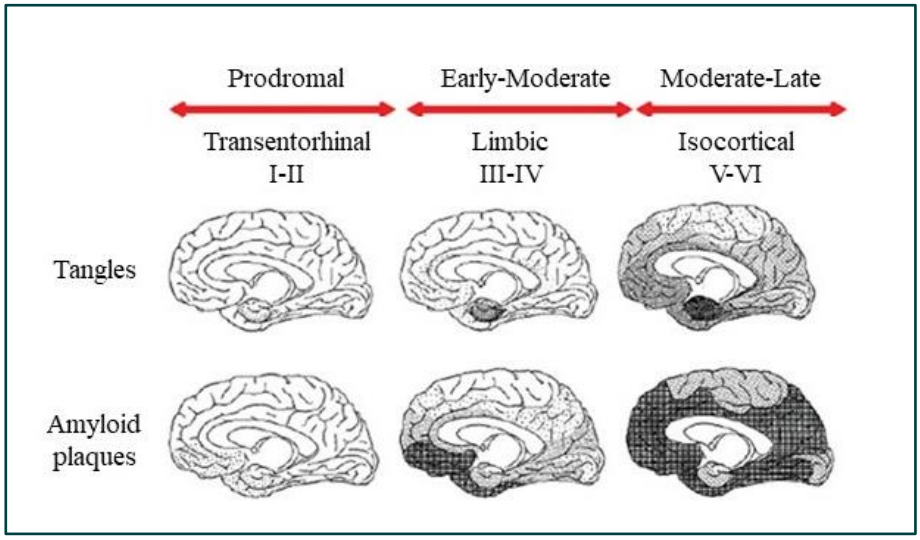

Figure 1. Progression of neurofibrillary tangles and amyloid plaques through the AD continuum. Modified figure based on Braak and Braak (11).

I.4. Causes and risk factors of Alzheimer's disease

Although a clear and exact cause of AD is still unknown, there are many factors that contribute to its pathogenesis. It is also important to distinguish between early and late onset AD. Clinically, the symptoms are the same, however, the aetiology is different. Late Onset

Alzheimer's Disease (LOAD) accounts for more than 95 percent of AD cases. Symptoms appear in people over the age of 65 and numerous factors are thought to contribute to its pathogenesis. On the other hand, Early Onset Alzheimer's Disease (EOAD) accounts for 1-5 percent of AD cases and starts earlier, usually in the fourth–fifth decades of life. The disease course for EOAD is generally shorter as this form is more severe and progresses more rapidly (12).

Regarding the genetic risk factors of the early onset type, mutations of the APP and presenilin genes PSEN1 and PSEN2 have been identified as a key factors in the pathophysiology of the disease. While APP has been described in chapter 1.3, presenilin is a component of the γ-secretase complex, responsible for APP cleavage. The inheritance of these mutations is principally autosomal dominant with high penetrance, over 85 percent. The presence of these mutations is regarded as diagnostic for EOAD (12).

As for the late onset AD, the genetic risk does not follow the Mendelian inheritance. However, individuals whose first-degree relative had LOAD are twice as likely to develop the disease themselves in their lifetime. The most well-known genetic risk factor is linked to APOE, a lipid-binding protein. Of the different isoforms, APOE ε4 poses the risk: one allele results in a 2–3-fold increase in AD risk, while homozygous APOE ε4 carriers have a 5-fold increase in the risk of developing Alzheimer's. Being an APOE ε4 carrier is also linked to memory problems, developing mild cognitive impairment (MCI), and the progression of MCI to AD. Moreover, it has been proposed that 20–30% of AD risk is attributable to APOE ε4 (12).

Considering dementia risk, 35 percent of that risk is attributable to potentially modifiable risk factors. These potentially modifiable risk factors are categorized according to life stages, such as early, mid, and late life risk factors. The early life modifiable risk factor for dementia is less education, referring to lack of secondary school education. Midlife risk factors include hearing loss, hypertension, and obesity. Finally, late life risk factors include smoking, depression, lack of physical activity, social isolation, and diabetes (13). The importance of these modifiable risk factors lies in the possibility of dementia prevention. Within the framework of the Finnish Geriatric Intervention Study to Prevent Cognitive Impairment and

Disability (FINGER), researchers examined the effect of multidomain intervention on cognitive functioning of elderly people at risk of cognitive impairment in a randomized controlled trial. The domains of the intervention included the five “fingers” of dementia prevention allocated to four components during the intervention: 1) dietary advice, 2) physical activity, 3) cognitive training and social activity, and 4) management of vascular risk factors (14). They found that the improvement of the intervention group’s overall performance on the neuropsychological test battery was 25% higher than the control group’s performance (15). In another study, Barnes et al. investigated the impact of reducing the effect of seven risk factors: midlife hypertension, diabetes, midlife obesity, depression, physical inactivity, smoking, and cognitive inactivity. They found that reducing the prevalence of these seven risk factors by 10% would result in a 1.1 million decline in the number of AD patients, while a 25% risk reduction would lead to 3.0 million fewer AD cases worldwide (16). These findings also emphasize the importance of early recognition of individuals at risk of dementia or those in the preclinical or prodromal phases.

I.5. Alzheimer’s disease continuum

Nowadays Alzheimer's disease is considered a spectrum called the Alzheimer’s disease continuum. The AD continuum is both a biological and clinical spectrum, ranging from the preclinical stage comprising individuals with confirmed AD pathology who do not have any cognitive symptoms, to patients with severe AD dementia (17).

The importance of the preclinical stages lies within the phenomenon that pathological processes responsible for AD start several years prior to the appearance of the first symptoms, providing a critical window for intervention. Two variants of the preclinical states have been defined by the international working group. One is the presymptomatic phase referring to cognitively intact individuals carrying monogenic mutations with autosomal dominant inheritance. For them, it is essentially unavoidable to develop AD in their lifetime. The other variant is the asymptomatic at-risk state. These individuals have at least one AD biomarker positivity, however, they lack clinical AD symptoms (18). The next phase of the continuum is the prodromal phase, called mild cognitive impairment. Mild cognitive impairment has

several subtypes according to the affected cognitive domains. The two major subtypes are amnestic MCI (a-MCI) and non-amnestic MCI (na-MCI). Both a-MCI and na-MCI can further be specified as single or multi domain subtypes based on the number of cognitive domains affected (19). Of special importance is amnestic MCI, as this subtype has the highest conversion rate to AD (18). The prodromal phase is also accompanied by diverse non-cognitive symptoms, such as changes in personality and behaviour, depression, impaired motor function, and sleep disorders. Recognizing patients in this phase of the continuum is also highly important for early intervention (20). In a study, Wilson et al. found that cognitive decline accelerated moderately around 4–6 years prior to MCI diagnosis and showed a more marked increase 5–6 years before dementia diagnosis (5). Alzheimer's disease has three different clinical stages, namely mild, moderate, and severe AD. In mild AD, patients are disoriented in place and time, have memory impairment, and have difficulties with the duties of everyday life. In moderate AD, patients have difficulty in identifying friends and family members, and have altered speaking, reading, and writing abilities. In severe AD, cognitive and functional impairment is so extensive that patients are confined to a bed, unable to recognize family and may also lose their ability to eat and swallow (21). The rate of progression varies between individuals, however, for most patients, life expectancy after AD diagnosis is 4-8 years (1).

I.6. Diagnosis of Alzheimer's disease

Despite the expanding availability of diagnostic methods, post-mortem histopathological examination remains the sole method for a definite diagnosis of AD. With the available clinical and biological methods, the most precise diagnosis one might reach is probable AD. Regarding the diagnostic protocol for AD, multiple changes have been implemented during the past decades. The first diagnostic criteria were established by McKhann et al. in 1984 and focused on clinical findings: progressive impairment of cognition, including memory and other cognitive domains. Laboratory tests were advised as supplementary methods to exclude other conditions that might cause progressive cognitive impairment (22). These criteria were revised in 2011 by The National Institute on Aging and the Alzheimer's Association (NIA-AA). Core criteria include impaired functioning of at least two of the following areas:

memory, language, visuospatial abilities, poor judgement, and changes in behaviour and personality, accompanied by disruption of the usual level of functioning in tasks of everyday life. However, a new paradigm emerged suggesting the use of magnetic resonance imaging and proving the presence of AD pathophysiological processes via positron emission tomography (PET) or cerebrospinal fluid (CSF) analysis (23). Both amyloid and tau tracers are available for PET, with around a 96% sensitivity (24). CSF analysis includes measuring the quantity of Aβ42, total tau and phosphorylated tau, and the ratio of Aβ42/ Aβ40 (25).

According to current recommendations, the diagnosis of Alzheimer's disease should be based on clinical and biological proof simultaneously. This includes the presence of specific clinical AD phenotype and biomarker positivity. Biomarker positivity can either be established via cerebrospinal fluid analysis showing concurrent elevated pTau levels with decreased Aβ42 levels, and Aβ42/Aβ40 ratio or via increased amyloid and tau tracer retention in PET (26). Clinical examination includes structural magnetic resonance imaging (MRI) and neuropsychological evaluation. As for MRI, bilateral entorhinal and hippocampal atrophy are principal MRI findings in AD (27). Regarding neuropsychological evaluation, there are several tests in use, including Mini Mental State Examination (MMSE), Addenbrooke's Cognitive Examination (ACE), Montreal Cognitive Assessment (MoCA) and Alzheimer's Disease Assessment Scale-Cog (ADAS-Cog) (28). Despite the serious efforts aimed at timely diagnosis, 50% of dementia patients remain undiagnosed (29). One of the possible causes of difficulty with diagnosis is the limited availability of screening: only 16 percent of adults aged 65 years or older receive routine cognitive evaluation (30). Furthermore, neuropsychological tests fail to identify early-stage cognitive impairment due to lack of sensitivity (31). Additional hindering factors of early diagnosis include the limited availability of trained medical personnel and the geographical constraint of these examinations since the patient must be physically present at the examination. A possible solution to tackle these problems could be telemedicine, making dementia screening more readily available to a greater number of patients, including those living at more remote locations (32). There is an increasing number of electronic cognitive appliances available, such as CogState and E-MoCA, for both domestic and clinical use (33, 34). However, these appliances carry the risk of nonadherence due to unfamiliarity with digital tools, vision

problems and cognitive tests being too difficult and tiring, among others (35, 36). These issues point to the possible benefit of an automated, electronic, self-administered screening tool that uses plain tasks aimed at evaluating few cognitive domains.

I.7. Treatment of Alzheimer's disease

Unfortunately, curative treatment is not available for AD. Thus, therapeutic interventions aim at slowing the progression of the disease to decelerate the cognitive decline. The two groups of drugs approved for AD treatment are acetylcholinesterase inhibitors (AchEIs)—donepezil, galantamine and rivastigmine—and memantine, a noncompetitive N-methyl-d-aspartate (NMDA) receptor antagonist. The shared aim of the two groups is to counteract the disturbed equipoise of neurotransmitters. AchEIs do so by raising the level of acetylcholine in synapses. Their use comes from the cholinergic hypothesis of AD. According to this hypothesis, limbic and neocortical structures, crucial for numerous higher brain functions, such as learning and memory, lose their cholinergic innervation. In addition, neuronal loss in the basal forebrain also contributes to the disruption of cholinergic innervation. As for memantine, as an antagonist of NMDA receptors, it acts against glutamate toxicity caused by abnormally high glutamate levels. Both groups are approved for both monotherapy and combination therapy. It is important to not only treat cognitive symptoms but also address concomitant factors such as behavioural and psychological symptoms of dementia (BPSD). For the treatment of anxiety and depression, selective serotonin reuptake inhibitors (SSRIs) are the favoured. Antipsychotics might also be used in the treatment of BPSD, for instance, to address aggressive behaviour (37).

Disease modifying treatments targeting Aβ are emerging: aducanumab and lecanemab have been approved by the Food and Drug Administration (FDA) in the United States, while the approval process is currently ongoing for donanemab. These drugs are aimed at slowing the cognitive decline via the reduction of Aβ in the brain. It is important to note that the two FDA-approved drugs are authorized not only in mild AD but also in MCI due to AD. Although reduction in Aβ levels has been confirmed, further studies are needed to investigate their effect on cognitive decline (38, 39). Their approval for MCI also points to the

importance of accurate early diagnosis. Furthermore, previous studies have suggested that complementing drug therapies with lifestyle interventions in MCI could lessen the risk of advancement of cognitive decline (14).

I.8. Visuospatial abilities and Alzheimer's disease

Visuospatial abilities include visual perception, construction, and visual memory (40). These are important skills in everyday life, for instance, for route learning (41). Recognition of a stimulus along with its location are key components of visuospatial function. Several cortical areas are activated while recognizing and localizing an object: premotor areas, superior parietal cortex, posterior parietal cortex and parieto-occipital junction (42). Two substantial pathways are distinguished in visuospatial processing, the ventral "What" pathway and the dorsal "How" pathway. Primate and human studies show that the dorsal pathway includes three different pathways, namely a parieto-prefrontal, a parieto-premotor and a parieto-medial temporal pathway. The parieto-prefrontal pathway connects the ventral intraparietal, the lateral intraparietal, the medial superior temporal, and the middle temporal areas to the caudal part of the banks of the principal sulcus and the prearcuate region. In humans, this pathway is associated with spatial working memory. Two parallel projections constitute the parieto-premotor pathway: one connects a part of the parieto-occipital area and the medial intraparietal area to the dorsal premotor cortex, while the other connects the ventral intraparietal area and the ventral premotor cortex. In humans, the parieto-premotor pathway is associated with reaching for and grabbing objects. The parieto-medial temporal pathway connects the rostral and caudal parts of the inferior parietal lobule to the medial temporal lobe. Its function is associated with navigation (43). Literature also suggests that the ventral and dorsal pathways are interconnected via inferior temporal and lateral intraparietal areas (44).

Studying the alteration of visuospatial abilities in MCI and AD is an area of increasing interest in AD research. Quental et al. found that visuospatial impairment is present already in the early phase of AD (42). Furthermore, Beretta et al. reported temporoparietal hypometabolism on F-18 fluorodeoxyglucose (FDG) PET recordings in AD patients (45).

Similarly, compared to healthy controls, AD patients showed reduced activation in premotor areas, parieto-occipital junction, and superior parietal lobule on visuospatial task-based functional MRI (fMRI) recordings. On the contrary, AD patients had increased activity in the inferior parietal lobule (46). Even though visuospatial impairment is less frequently noted among the symptoms in AD, Salimi et al. suggest that the assessment of visuospatial abilities could have diagnostic potential in the diagnosis of AD (40).

I.9. Visuomotor abilities and Alzheimer's disease

Visuomotor abilities include visual and motor processes as well as the conversion between the two (47). Prefrontal, premotor, and parietal cortical areas and cerebellar and striatal networks have been found to participate in visuomotor transformation (48). Reciprocal interactions are required between brain areas associated with motor and visual processing to expedite the coordination between movement and vision. The impact of visual information on movement processing and the impact of movement information on visual processing is called visuomotor integration (47).

There are numerous studies investigating the impairment of visuomotor function along the AD continuum. Previous research suggests that visuomotor impairment is advancing simultaneously with the cognitive decline in early AD (49). Furthermore, expanding literature points to the alteration of visuomotor functions already in MCI (50). Moreover, severely impaired visuomotor function was observed in older adults at high risk of AD, compared to younger adults and older adults with low AD risk. This study points to the possible benefit of using visuomotor ability-based tasks for identifying individuals at risk of AD prior to the onset of apparent cognitive decline, as well as for monitoring the conversion of preclinical AD to MCI (51). A previous study of Mollica et al. proposed that a computerized task based on evaluating visuomotor functions could find a faint alteration in visuomotor coordination that otherwise would remain unnoticed (52). These findings point to the possible benefits of a visuomotor ability-based screening tool for the AD continuum.

DOI:10.14753/SE.2024.3035

II. Objectives

With a series of studies, we aimed to explore the alteration of the visuospatial and visuomotor functions in Alzheimer's disease patients and in mild cognitive impairment compared to age-matched, cognitively healthy control elderly participants, to better understand their association with disease stages and progression. These objectives served as the basis of our work towards establishing visuomotor ability-based screening methods for MCI and Alzheimer's disease.

Specific objectives:

- To assess the involvement of different cognitive domains—orientation, attention, memory, verbal fluency, language, and visuospatial skills—in the cognitive impairment of AD patients with different disease duration.
- To evaluate the possible role of the above cognitive domains in the early recognition of AD.
- To evaluate the possible benefit of assessing these cognitive domains in monitoring the progression of cognitive impairment.
- To analyse the structural integrity of the visuospatial network in amnestic MCI patients on structural MRI recordings.
- To characterize the functional connectivity of the visuospatial network in amnestic MCI via functional MRI recordings.
- To ascertain the discriminatory potential of small amplitude hand movements, recorded via a visuomotor ability-based paradigm, in distinguishing between cognitively healthy individuals and MCI patients in a clinical environment.

DOI:10.14753/SE.2024.3035

III. Methods

III.1. Participants

For the three studies that serve as the base of this thesis, we recruited participants from the AlzEpi Cohort Observational Library (ACOL database) of the National Institute of Mental Health, Neurology and Neurosurgery, Budapest, Hungary. This database is incorporated in the Euro-Fingers international database (http://www.eufingers.com). The cohort is composed of cognitively healthy elderly individuals, patients with MCI or dementia. The diagnosis of MCI and AD patients was determined by a multidisciplinary team based on a comprehensive dementia protocol described in detail below. For the neuroimaging study, participants from another database of a collaborating research laboratory were also included. This database is introduced in Chapter III.4. Every participant from both databases gave their informed written consent for the examinations and the use of data for research purposes, among others. Every study event was conducted consonantly with the relevant regulations and guidelines. Our experimental protocols were authorized by The Hungarian Medical Research Council (reference number of ethical approval: 024505/2015).

Every study participant underwent the very same comprehensive dementia protocol. The protocol included the collection of medical history from the participants themselves and from the relatives and caregivers of the patients affected by MCI or AD. Moreover, the protocol included detailed neuropsychological and neurological evaluation conducted by neurologists, neuropsychologists, and trained neuroscientists. Structural and resting-state functional MRI acquisition and blood tests were also included. Additionally, some but not all participants completed the visuomotor ability-based Precognize paradigm, that will be described in detail in Chapter III.5.1. Every study participant was native Hungarian.

Every healthy control participant involved in our studies had no cognitive complaints and upon the completion of the dementia protocol, no alterations were found in their neurological status, neuropsychological test results, brain MRI scans, and blood tests.

III.1.1. Exclusion criteria

None of the studies included participants with known risk factors of cognitive decline. These risk factors are as follows: vitamin B12 deficiency without treatment, hypothyroidism, liver disorder, renal insufficiency, significant systemic medical conditions, considerable brain injury such as white matter disease, significant white matter damage affecting the periventricular areas, and cortical stroke. Patients with a history of hydrocephalus, demyelinating conditions, and previous head injury accompanied by loss of consciousness were also excluded. Further exclusion criteria included previous central nervous system infection, HIV and syphilis infection, major depression, electroconvulsive therapy, schizophrenia, psychoactive drugs that alter cognitive functioning, and substance or alcohol abuse. Specifically, for the Precognize study, we established further exclusion criteria for conditions that could disturb the motor control of the upper extremity. Such conditions include Parkinson's disease and parkinsonism, tremor disorders, cortical lesions in motor areas, motoneuron and neuromuscular disorders, conditions affecting the peripheral nerves of the upper limb and the cervical region of the spinal cord.

III.2. Neuropsychological evaluation

Every participant underwent comprehensive neuropsychological evaluation administered by neurologists, neuropsychologists, or trained neuroscientists. Our test battery consisted of diverse tests assessing cognition, mood, and anxiety as well. We chose the validated Hungarian version of the Addenbrooke's Cognitive Examination (53) as a tool to assess global cognitive functioning. It is considered a highly specific and sensitive test in neurocognitive disorders (54). The evaluation of the following six cognitive domains is possible with this test: orientation (10 points), attention (8 points), memory (35 points), verbal fluency (14 points), language (28 points), and visuospatial abilities (5 points) resulting in a total score of 100. The orientation domain contains questions assessing both the spatial and temporal orientation of the participant. The attention part contains tasks for listening to and remembering three words as well as counting backwards by 7, starting at 100. The memory tasks assess both anterograde and retrograde memory as well as delayed recall. The

verbal fluency domain assesses phonemic (words with the letter M) and categoric (animals) fluency. The language part contains tasks for evaluating reading, writing, naming object, and following commands, among others. And finally, the visuospatial domain consists of three tasks: copying overlapping pentagons, copying a cube, and drawing a clock face with numbers, followed by setting the clock hands at a specific time. It also includes the Mini Mental State Examination with a possible maximum score of 30, for which we chose the age and educational background adjusted cut-off scores represented in Table 1. (55). However, contrary to MMSE, ACE is thought to indicate cognitive impairment at the prodromal, MCI, stage (56). For the ACE, we set 83 as our cut-off score, which was shown to have a sensitivity of 82% at the age of 65 years and over (57). Furthermore, the VLOM ratio—calculated by dividing the sum of the verbal fluency and language subscores by the sum of the orientation and delayed recall tasks ((V+L)/(O+M))—signals frontotemporal (VLOM<2.2) or AD type (VLOM>3.2) dementia. The normal value of the VLOM ratio is, therefore, between 2.2 and 3.2.

For the precise differentiation of MCI patients from cognitively healthy control individuals, we selected the Hungarian version of the Ray Auditory Verbal Learning Test (RAVLT) (58). Its sensitivity in detecting a-MCI has been formerly established with readily available cut-off scores (Table 1.) (59-62). The RAVLT test has two parts: the immediate recall and the delayed recall tasks. In the first part, participants are asked to listen to 15 words (list A) read out loud by the examiner and memorize as many as possible. When the examiner finishes reading, the participant is asked to list every word they remember. The examiner marks the correctly repeated words on the test sheet, with each word worth one point in the test result. This process is repeated four times. The scores of these five trials are then summarized, resulting in a maximum of 75 points called the RAVLT sum-5 score. Afterwards, the examiner reads list B, a list of 15 different words. The participant's task is the same as before: memorize and recall as many words as possible. Instantly after that, the participant is asked to call to mind the words from list A. The second part of the RAVLT, the delayed recall, is recorded after a 30-minute waiting period. In this part of the test, the participant is aksed to enumerate every word they remember from list A without the examiner reading them out

loud. The correctly recalled words are then counted, resulting in a maximum of 15 points called the RAVLT7 score.

We selected the Trail-making test (TMT) to assess attention (part A) and cognitive flexibility (part B) (63). In part A (TMT-A) participants are asked to connect numbers in circles from 1 to 25 in an ascending order. In part B (TMT-B) letters in circles are also present. For this task, the participants are asked to connect the numbers in ascending order, the letters in alphabetic order while alternating the two: 1-A-2-B-3-C, etc. The time required to complete the task is registered in seconds. The less time needed to complete the task, the better.

In order to control for mental states known to affect cognitive function such as depression or anxiety (64, 65), our neuropsychological test battery included the Hungarian version of the Beck Depression Inventory II (BDI) and the Hungarian version of the Spielberger State-Trait Anxiety Inventory (STAI) (66, 67). For the BDI test, a cut-off score of 13 was used for minimal depression. Mild depression was marked by scores of 14–19, scores of 20–28 indicate moderate depression, while a score of 29 or above indicates severe depression. We excluded participants whose BDI scores were 13 or higher. We applied the Trait version of the STAI (STAI-T) to estimate the participants everyday level of anxiety, while the State version of the Inventory (STAI-S) served to measure the participants' anxiety level right before the start of the visuomotor experimental protocol. A cut-off score of 45 points was used to mark low-level anxiety, therefore, we did not include participants with a STAI-T or STAI-S score of 45 or higher.

We selected the Clinical Dementia Rating Scale (CDR) Sum of Boxes to assess the independence and everyday functioning of the participants (68). For the CDR Sum of Boxes, the examiner evaluates six domains: personal care, home and hobbies, community affairs, judgement and problem solving, orientation and memory. These areas are rated on a scale of five: 0 signals no impairment, 0.5 means questionable impairment, 1 equals mild impairment, moderate impairment is marked as 2, while 3 indicates severe impairment.

III.3. Evaluating the potential role of visuospatial abilities in diagnosing Alzheimer's disease

110 individuals with clinically defined AD (69 male and 49 female, with a mean age of 73.1±6.6 years) were recruited for this study together with 45 cognitively healthy control participants (16 male and 29 female, with a mean age of 68.6 ±7.40 years) from the Department of Neurology at the National Institute of Mental Health, Neurology and Neurosurgery in Budapest, Hungary. Patients with Alzheimer's Disease were diagnosed based on the guidelines of the National Institute on Aging and the Alzheimer's Association (23). Every participant underwent detailed neurological and neuropsychological evaluation, including the MMSE and ACE tests. We created three groups based on disease duration and assigned the participants accordingly. We calculated the disease duration from the day the clinical diagnosis of AD was established. However, based on heteroanamnestic data provided by caregivers, we excluded participants who had prolonged cognitive complaints for at least two years preceding the diagnosis. Group 1 included 36 individuals with a disease duration of up to two years, Group 2 was composed of 44 participants with a disease duration of 2 to 4 years, while patients with a disease duration of at least four years (n=30) belonged to Group 3. Furthermore, we created a fourth group, Group 0 containing 45 cognitively healthy control individuals. Altogether, we included 155 subjects in this study. Previously, de Boer et al. found significant differences in total MMSE scores as well as MMSE subscores, assessing various cognitive domains, between three groups of AD patients (n=125) with different disease duration (69). We conducted power calculations to determine the appropriate sample size for achieving at least 80% probability of finding between-group differences (α=0.05) in ACE total score and subscores. On the basis of our calculations and the results of de Boer et al., a sample size of 150 was determined sufficient.

Regarding statistical analysis, we applied the Shapiro-Wilk test to assess the distribution of data. Based on the distribution of the data, we used one-way ANOVA for parametric and Kruskal-Wallis test for non-parametric data to perform intergroup comparisons in demographic variables such as school years and age. We applied Bonferroni correction with the statistical significance of $p<0.01$ to counteract multiple comparisons. We tested the association between global cognition represented by ACE total score and disease duration

(in years) with Spearman's rho, as the data distribution was non-parametric. We assessed the intergroup differences in ACE subscores with ANCOVA and Kruskal-Wallis test with age, sex, and disease onset as covariates, followed by Tukey's post-hoc test. We applied Spearman's rho to test the association between the subscores of the ACE test and disease duration. We normalized the ACE subscores after which within-group analysis was completed with Wilcoxon signed-rank test. The normalization was done by dividing the achieved scores for each subscore by the total score of the given subscore (e.g., 5/10 in orientation subscore gives a normalized score of 0.5). For the statistical analysis, we used the IBM SPSS 20 software (https://www.ibm.com/support/pages/ibm-spss-statistics-20-documentation).

III.4. Neuroimaging study of the visuospatial system in mild cognitive impairment

In total, 78 individuals were involved in our study: 32 patients with multidomain a-MCI and 46 cognitively healthy control (HC) participants. We obtained the data by the collaboration of two independent research centres through the Euro-Fingers Consortium (www.eufingers.com). The two centres used an established, identical protocol for medical imaging examination for clinical and research purposes. We included data of 19 MCI patients and 26 healthy control individuals from the aforementioned AlzEpi Cohort Observational Library (ACOL), the database of the National Institute of Mental Health, Neurology and Neurosurgery. In addition, data of 33 individuals (13 MCI patients and 20 healthy controls) was selected from the Semmelweis MCI Neuroimaging Cohort (SMNC) database of the Department of Psychiatry and Psychotherapy, Semmelweis University. The diagnosis of MCI patients was determined by a multidisciplinary team based on the Petersen criteria: presence of subjective cognitive complaints—fortified by objective neuropsychological evaluation—that do not interfere with activities of independent daily living (19). Patients at the dementia stage of the disease continuum were excluded from this study.

Table 1. Thresholds for determining amnestic MCI patients and patients already at dementia stage for exclusion. Scores are adjusted for school years and age.

MMSE: Mini Mental State Examination, a-MCI: amnestic mild cognitive impairment, RAVLT: Rey Auditory Verbal Learning Test, a: the total number of words learnt in the first 5 tries (out of the maximum 75), b: the number of words the participant remembered after 30 minutes of delay (out of a maximum 15) (70).

MMSE cutoff scores for the exclusion of dementia								
Education\Age	50-54	55-59	60-64	65-69	70-74	75-79	80-84	85+
5-8 years	23	23	23	23	23	21	21	17
9-12 years	25	25	25	25	24	24	21	21
>12 years	27	27	27	27	25	25	25	24
RAVLT sum-5 cutoff scores for a-MCI[a]								
Age					Score			
50-59 60-69 70+					39 35 29			
RAVLT7 cutoff scores for a-MCI[b]								
Age					Score			
50-59 60-69 70+					6 5 4			

III.4.1. Structural and functional magnetic resonance imaging

Every participant in this study underwent high resolution structural and functional MRI acquisition. Two different MRI scanners were used at the two participating research centres. A 3T Siemens Magnetom Verio scanner (Siemens Healthcare, Erlangen, Germany) was used at the National Institute of Mental Health, Neurology and Neurosurgery with the standard 12 channels head coil. The imaging protocol included a T1-weighted 3D magnetization prepared rapid gradient echo (MPRAGE) anatomical imaging (TR (time resolution)=2.300ms; TE (time echo)=3.4ms; TI (inversion time)=100ms; flip angle: 12°; voxel size: 1.0 × 1.0 × 1.0mm) and a 10-minute eyes-closed resting-state functional MRI recording with echo planar imaging (EPI) (TR=2.000ms; TE=30ms; flip angle=79°; voxel size = 3 × 3 × 3mm). The other research centre, the MR Research Center at Semmelweis University, used a Philips Achieva 3.0 T MRI scanner (Philips Medical Systems, Best, The Netherlands). The head coil

used was a SENSE coil with 8 channels. The imaging protocol included a 3-dimensional T1 weighted Turbo Field Echo sequence for anatomical imaging (TR=9.7ms; TE=4.6ms; flip angle=8°; FOV (field-of-view) of 240 × 240mm; voxel size of 1.0 × 1.0 × 1.0mm). The T2*-weighted EPI-based resting-state MRI recordings (TR=2.0 s; TE=30ms; flip angle=70°, FOV of 240 ×240mm; voxel size of 3.0 × 3.0 × 4.0mm; number of slices = 36) took approximately 8.5 minutes, with the subjects fixating on a cross in the middle of the screen. Foam padding was applied to minimize head motion during the recording. After the recordings, the participants were asked whether they had fallen asleep during the recording, all of them negated doing so. Furthermore, in order to recognize any potential pathological alterations, both research centres applied FLAIR, T2-weighted and DTI sequences as well.

III.4.2. Neuroimaging analysis

The open source Freesurfer 6.0 software package (http://surfer.nmr.mgh.harvard.edu/) was used for volumetric segmentation and cortical reconstruction of T1-weighted anatomical images. Cortical surface models were created with the default settings of the processing stream called “recon-all”. We applied the CONN MATLAB toolbox for the analysis of resting-state functional MRI data (71). Detailed description of our complete imaging analysis protocol has been published previously (62).

We determined the regions-of-interest (ROI) of the visuospatial network using previous literature. Visuospatial processing consists of spatial perception, followed by recognition and visual input analysis (72). Frontal, temporal, and parietal brain regions have been identified as parts of the visuospatial network based on activation during visuospatial task-based fMRI acquisition (73-78). More precisely, the involvement of the caudal part of the superior frontal and middle frontal gyri, insula, precentral gyrus, inferior temporal gyrus, temporal pole, occipitotemporal gyri, posterior part of the superior parietal lobule, angular gyrus, supramarginal gyrus, and calcarine cortex was described (72-83) (Figure 2.). Furthermore, literature points to the predominance of the subdominant hemisphere in the coordination of visuospatial functions (84, 85). We applied seed-based connectivity (SBC) (86) to investigate functional connectivity of the aforementioned regions of interest at the group level and at the

subject level as well. Fischer-transformed bivariate correlation coefficients between an ROI BOLD time series and each voxel BOLD time series gives the SBC maps. We included age and sex as covariates in the analysis. We used a paired t-test for intergroup comparison of functional connectivity. For parametric statistics, we applied the following parameters: cluster threshold: $p < 0.05$, cluster-size: p-FDR corrected, voxel threshold: $p < 0.001$ uncorrected (87).

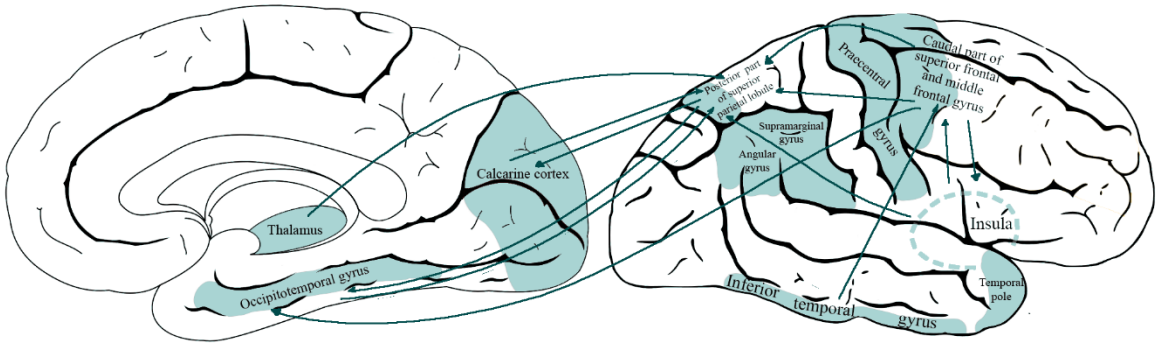

Figure 2. Region-of-interest (ROI) areas of the visuospatial network. Regions included: caudal part of the superior frontal and middle frontal gyri, insula, precentral gyrus, inferior temporal gyrus, temporal pole, occipitotemporal gyri, posterior part of the superior parietal lobule, angular gyrus, supramarginal gyrus, and calcarine cortex.

We applied intergroup comparisons for demographic data, such as sex, age, and years of education in the HC and MCI group. We used independent samples t-test for continuous variables with parametric distribution, whereas we used Mann-Whitney U test for non-parametrically distributed data. We applied the Chi-square test for categorical variables. We analysed the intergroup differences in structural MRI recordings and neuropsychological test results with ANCOVA test (covariates: age, sex). Subsequently, Benjamini-Hochberg correction was applied to counteract multiple comparisons. We reported the effect sizes with Cohen's d with values 0.2–0.5 indicating small effect, values of 0.5–0.8 indication medium effect while values over 0.8 signal large effect. We used the IBM SPSS 20 software (https://www.ibm.com/support/pages/ibm-spss-statistics-20-documentation) for statistical analysis excluding the analysis of functional MRI results.

DOI:10.14753/SE.2024.3035

III.5. Investigating the differentiating potential of a visuomotor paradigm in distinguishing MCI patients from healthy controls

In this study, we solely recruited right-handed participants who reported themselves as regular computer and internet users with active email addresses and use. Handedness in this study was established based on the participants' own admission. Altogether, 68 individuals participated in this study: 29 male and 39 female. We assigned the participants to two groups: the first group included 46 healthy controls without cognitive complaints, while the second group was composed of 22 patients with MCI. The diagnosis of MCI was given based on the revised Petersen criteria: subjective cognitive complaints should be confirmed by objective neuropsychological tests, however, the objectively confirmed impairments should not interfere with the patient's ability to live an independent everyday life (19). MRI acquisition of patients also showed reduced cortical thickness in hallmark regions, such as the entorhinal cortex, together with a reduced volume of total grey matter. Patients already at the dementia stage were not included in this study.

We selected the Kolmogorov-Smirnov test to analyse the distribution of the data. We applied the Mann-Whitney U test or independent samples t-test to assess continuous variables of demographic and cognitive data. We used the Chi-square test for categoric variables. ANCOVA test was applied to assess group effect on motor data with gender, age, and anxiety level as covariates. We used the Benjamini-Hochberg correction to counteract the effect of multiple comparisons on the results. We determined the effect sizes in Cohen's d with values 0.2–0.5 indicating small effect, values of 0.5–0.8 indication medium effect while values over 0.8 signal large effect. We used Pearson correlation to analyse the relation between neuropsychological test results and visuomotor characteristics that were significantly different between the two groups based on p-values, as these characteristics might have discriminative potential.

DOI:10.14753/SE.2024.3035

III.5.1. Precognize paradigm

In close collaboration with researchers and medical professionals from the National Institute of Mental Health, Neurology and Neurosurgery, Precognize Ltd. developed a visuomotor paradigm called Precognize in the framework of the National Brain Research Program II (2017-2021). The paradigm was developed based on part A of the Trail-Making Test. In this case, instead of connecting the numbers in ascending order, the participants are asked to click on each number on the screen with a computer mouse in ascending order as speedily as they can. The numbers (1–9) are presented in circles in fixed positions on the screen (Figure 3.). Solely correct clicks are accepted, that are the next in order and are on the margin of or within the circle surrounding the number. The program will not let the participant advance until any of the above two are violated.

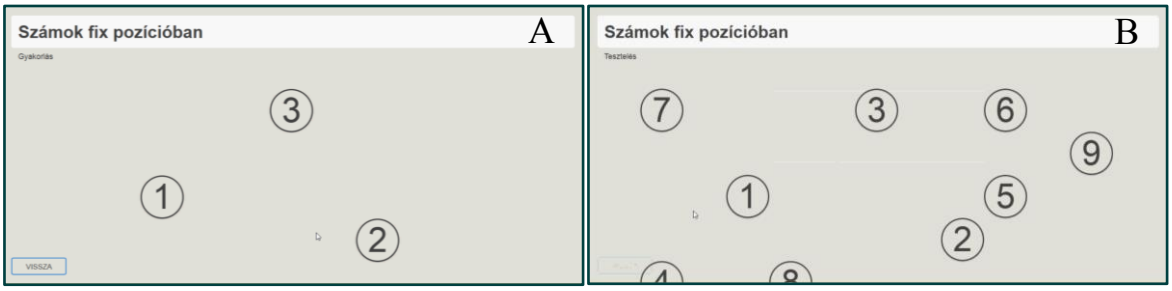

Figure 3. Screenshot images of the Precognize paradigm. The paradigm starts with a mock part (A) to confirm that participants understand the task, followed by the live test (B).

In order to limit the influence of factors other than the underlying disease, we used the same laptop, computer mouse, chair, table and time of examination (4–6 PM) during the completion of the Precognize paradigm. Once the participant took their seat comfortably and confirmed that they were ready for the paradigm to begin, we entered research codes into the program for both the participant and the examiner. This ensured that the data was anonymized, so no personal information could be associated with the data. After the examiner had saved the research codes, the recording of the mouse actions started, thus, from that moment, only the participant was allowed to touch the computer mouse. The tasks were preceded by a thorough description followed by a simplified trial version of the task to confirm that the participant knew what to do. The participants were asked to complete the

DOI:10.14753/SE.2024.3035

task with both dominant and subdominant hands, one after the other. The primary mouse button was adjusted so that the participants could control it with the index finger of each hand.

During the task solving, the program recorded every mouse action made by the participants. Since the task description instructed the participants to complete the paradigm as quickly as they can, their focus was shifted from operating the computer mouse. The recorded computer mouse movement parameters were stored in log files named after the above-mentioned research codes. For data analysis, we extracted the computer mouse movement parameters for the two hands separately. We divided the task completion into 9 sections: each section represented the interval between two numbers, e.g. section 0 contained all the mouse movement parameters from the beginning of the task until the participants clicked on the first number. Section 1 represented the information between the numbers 1 and 2, etc. This adds up to 18 sections altogether, considering both the dominant and subdominant hands. We extracted the following parameters: entropy, distance, time, number of tries, velocity and speed described in detail in Table 2. We then averaged the values of these parameters from the 9 sections for both hands separately. Overall, we had one average value for each of the parameters for each hand separately.

Table 2. Description of mouse movement parameters. While *y* and *x* indicate the *y* and *x* coordinates of the screen, *t* signals time, and *n* refers to the given section between two numbers (88).

Motor parameter	Description	Formula
Distance	The overall distance from the mouse movements	$\sum_{i=1}^{n} \sqrt{(x_n - x_{n-1})^2 + (y_n - y_{n-1})^2}$
Entropy	The Shannon entropy of the mouse movements, where the underlying distribution (random variable) is the two-dimensional coordinate of the location of the mouse on the screen	$-\sum_{i=1}^{n} f(x;y)_i \times lg\, f(x;y)_i$
Number of tries	The sum of all mouse clicks	$\sum all\ clicks$
Time	The time required to complete the task	$t_n - t_0$
Speed	The speed of the mouse movements	$\frac{distance}{time}$
Velocity	The velocity of the mouse movements	$\sum_{i=0}^{n} \frac{distance_n - distance_{n-1}}{time_n - time_{n-1}}$

III.6. Summary of methods

Table 3. Summary of methods.

HC: healthy control, AD: Alzheimer's Disease, ACE: Addenbrooke's Cognitive Examination, RAVLT: Rey Auditory Verbal Learning Test, TMT: Trail-making Test, STAI: Spielberger's State-Trait Anxiety Inventory, BDI: Beck Depression Inventory, CDR: Clinical Dementia Rating scale, MCI: Mild Cognitive Impairment, sMRI: structural Magnetic Resonance Imaging, fMRI: functional Magnetic Resonance Imaging

Number of study	Number of participants	Applied methods
I	45 HC, 110 AD	ACE, RAVLT, TMT, STAI, BDI, CDR
II	46 HC, 32 MCI	ACE, RAVLT, TMT, STAI, BDI, CDR, sMRI, fMRI
III	46 HC, 22 MCI	ACE, RAVLT, TMT, STAI, BDI, CDR, Precognize

DOI:10.14753/SE.2024.3035

IV. Results

IV.1. Evaluating the potential role of visuospatial abilities in diagnosing Alzheimer's disease

IV.1.1. Demographics and neuropsychological test results

Overall, 155 participants were involved in this study, 77 males (49.7%) and 78 females (50.3%). The participants' mean age was 71.8±7.1 years. The median time frame of their education was 12 (12.0–17.0) years. Out of the 155 individuals, 45 were cognitively intact control participants while 110 were diagnosed with clinically defined AD. Cortical atrophy affecting the bilateral frontal and temporal cortices and hippocampi was a distinctive feature observed in AD patients on MRI scans of the brain. All patients had an MTA score of at least 3. Group 1 was composed of 36 individuals whose disease duration was no longer than two years. The mean age of these participants was 70.7±7.4 years, 23 of whom were male (63.89%) and 13 were female (36.11%). Group 2 included 44 individuals with a disease duration of 2–4 years. 25 (56.8%) of these participants were male and 19 female (43.2%) with a mean age of 74.1±6.2 years. In group 3, the disease duration of the participants was longer than 4 years. 30 individuals belonged to this group, 13 (43.3%) of whom were male and 17 (56.7%) were female. Their mean age was 74.6±5.4 years. Group 0 was composed of 45 control individuals with no cognitive impairment. The mean age of participants in Group 0 was 68.6±7.4 years, 16 of whom were male (35.6%) and 29 were female (64.4%). Between-group differences were tested for age, sex, disease duration, age at the beginning of their AD, school years, VLOM ratio, ACE subscores, and ACE total score (Table 4). We found significant differences ($p<0.001$) in all parameters, excluding sex and age at disease onset.

DOI:10.14753/SE.2024.3035

Table 4. Demographics and neuropsychological test results.

a: mean ± standard deviation; b: median followed by interquartile range (IQ1–IQ3); c: Chi-square test, d: ANCOVA, e: Kruskal-Wallis test, ACE: Addenbrooke's Cognitive Examination, VLOM: (verbal fluency + language subscores of ACE) / (orientation + memory subscores of ACE), MMSE: Mini Mental State Examination (89).

Parameter	Total	Group 0	Group 1	Group 2	Group 3	p-value
Participants (n)	155	45	36	44	30	-
Female, n (%) [c]	78 (50.3%)	29 (64.4%)	13 (36.11%)	19 (43.2%)	17 (56.7%)	0.936
Age (years) [a]	71.8±7.1	68.6±7.4	70.7±7.4	74.1±6.2	74.6±5.4	<0.001
Age at disease onset (years) [a]	70.2±6.4	–	69.2±7.3	71.1±6.2	70.0±5.6	0.43
Education (years) [b]	12.0 (12.0–17.0)	17.0 (12.0–17.0)	12.0 (12.0–16.5)	12.0 (12.0–17.0)	12.0 (10.0–15.0)	<0.001
Disease duration (years) [b]	3.0 (2.0–4.0)	–	1.0 (1.0–2.0)	3.0 (3.0–3.0)	5.0 (4.0–5.0)	<0.001
ACE total score [b]	72.0 (59.0–88.0)	94.0 (91.0–96.0)	72.0 (67.3–78.0)	66.5 (55.0–74.3)	50.0 (45.8–57.3)	<0.001
VLOM [b]	3.3 (2.9–4.0)	2.6 (2.4–2.9)	3.5 (3.3–4.1)	3.5 (3.2–4.6)	3.6 (3.3–4.7)	<0.001
MMSE [b]	22.0 (17.0–28.0)	29.0 (28.0–29.0)	24.0 (21.3–25.0)	19.0 (16.0–21.0)	15.5 (12.8–18.0)	<0.001
Orientation [b, e]	8.0 (7.0–10.0)	10.0 (10.0–10.0)	8.5 (8.0–10.0)	7.0 (6.0–8.0)	7.0 (5.0–8.0)	<0.001
Attention [b, e]	7.0 (5.0–8.0)	8.0 (8.0–8.0)	6.0 (5.0–7.0)	6.0 (5.0–7.0)	5.0 (4.0–6.0)	<0.001
Memory [a, d]	21.0±4.9	25.1±1.8	21.9±3.1	20.5±4.4	14.2±3.0	<0.001
Verbal fluency [b, e]	9.0 (7.0–12.0)	13.0 (11.0–14.0)	9.0 (8.0–10.8)	8.5 (6.3–10.0)	7.0 (6.0–8.0)	<0.001
Language [b, e]	23.0 (19.0–28.0)	28.0 (28.0–28.0)	24.0 (22.0–25.0)	20.0 (17.0–22.8)	17.5 (15.0–20.3)	<0.001
Visuospatial abilities [b, e]	4.0 (4.0–5.0)	5.0 (5.0–5.0)	4.0 (3.3–5.0)	3.0 (2.0–3.0)	1.0 (0.75–2.0)	<0.001

IV.1.2. Interrelation between disease duration and the total score of the Addenbrooke's Cognitive Examination

We used Spearman's rho to explore the relationship between the total score of the ACE test and disease duration. Significant negative correlation presented between ACE total score and disease duration ($p<0.001$; $r= -0.643$). To reinforce our finding, we applied a one-way Kruskal–Wallis test, which supported that groups significantly influenced the total score of the ACE test. ($\chi2=115.81$; $p<0.001$).

IV.1.3. Intergroup differences in the subscores of the Addenbrooke's Cognitive Examination

We applied a one-way ANCOVA test to study between-group differences in the memory subscores (Table 4.). This analysis resulted in significant differences ($F=69.11$; $p<0.001$).
We then applied the Kruskal–Wallis test to investigate the between-group differences in the subscores of attention, orientation, language, verbal fluency, and visuospatial abilities (Table 4.). Between-group differences proved to be significant for visuospatial abilities ($\chi2=113.96$; $p<0.001$), language ($\chi2=100.38$; $p<0.001$), orientation ($\chi2=96.27$; $p<0.001$), attention ($\chi2=87.11$; $p<0.001$), and verbal fluency ($\chi2=61.12$; $p<0.001$). No significant modifying effect of sex, age, and disease duration was found on between-group differences (all p-values >0.01). With Tukey's post-hoc test, we found the following group differences: regarding orientation skills, Group 1 is significantly different from Group 0, Group 2, and Group 3 (all p-values <0.001). There is a significant difference between Group 0 and Group 2 and Group 3 (all p-values <0.001). On the contrary, no significant difference was found between Group 2 and Group 3 ($p=0.779$). As for the attention subscore, Group 0, Group 1, and Group 2 are all significantly different from one another (all p-values <0.001). There were further significant differences between Group 2, Group 0, and Group 3 ($p<0.001$). On the contrary, no significant difference was found between Group 2 and Group 1 in the attention subscore ($p=0.984$). Regarding the memory subscore, we found that Group 0, Group 2, and Group 3 all differed significantly. Furthermore, Group 1 is significantly different from Group 0 and Group 3 (all p-values <0.001). However, we did not find a significant difference between Group 1 and Group 2 ($p=0.254$). As for the verbal fluency, we found that Group 0 differs

significantly from all three groups (p-values <0.001). On the contrary, we found no significant difference between Group 1 and Group 2 and Group 3 (p=0.629 and p=0.017, respectively), nor between Group 2 and Group 3 (p=0.198). Regarding the subscore of language, we found that Group 0, Group 1, and Group 3 all differed significantly from one and other (all p-values <0.001). Additionally, Group 2 significantly differs from Group 0 and Group 1 (all p-values <0.001). However, we found no significant difference between Group 2 and Group 3 (p=0.142). Concerning the visuospatial subscore, we found that all four groups differed significantly (all p-values <0.001). Compared to healthy controls (Group 0) the difference was highest in Group 1, the earliest disease stage. (Figure 4.)

DOI:10.14753/SE.2024.3035

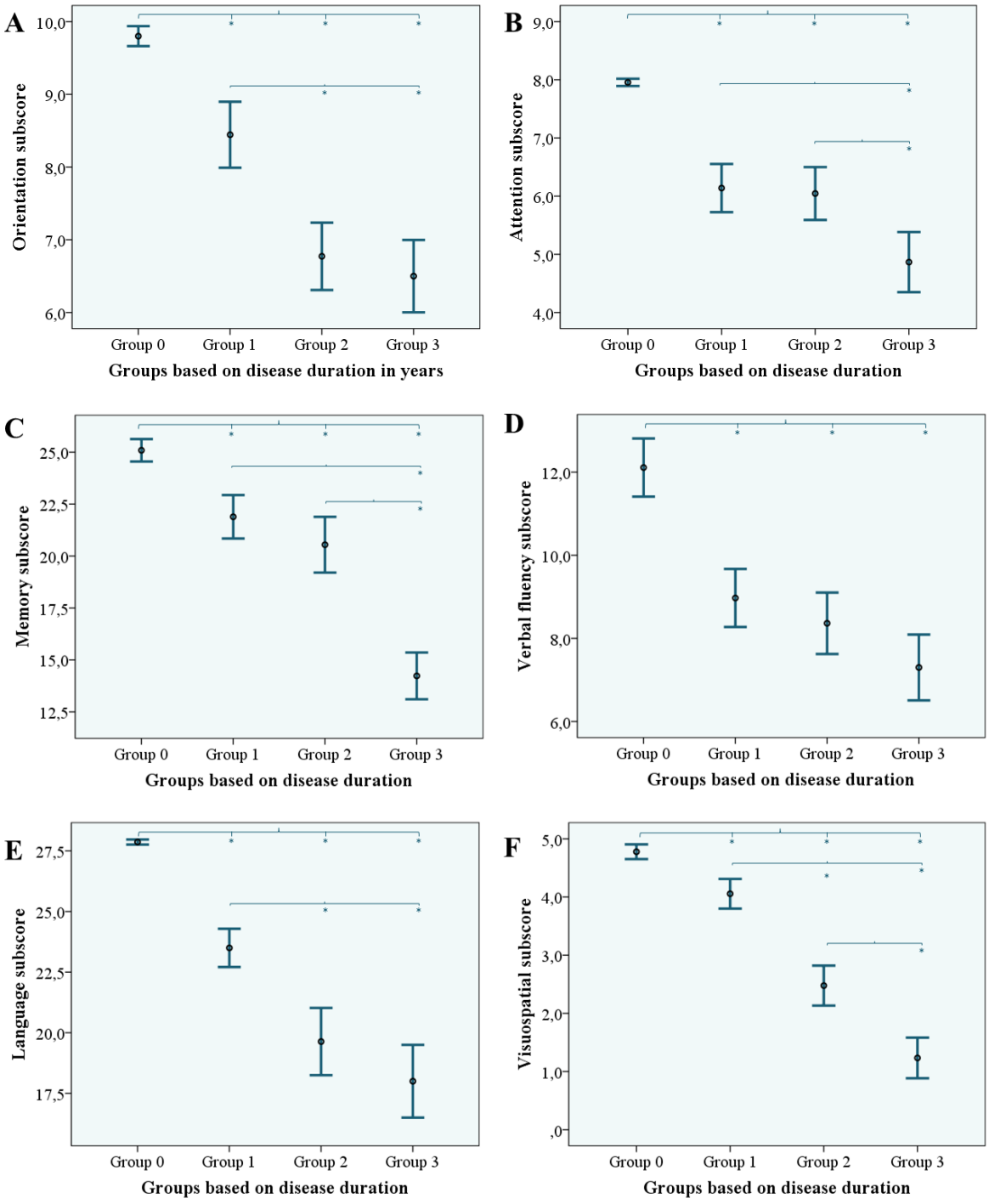

Figure 4. Intergroup differences in orientation, attention, memory, verbal fluency, language, and visuospatial abilities cognitive domains based on the subscores of the Addenbrooke's test. (A) Orientation was already impaired after less than two years of disease duration (DD)

(Group 0 vs Group 1) and demonstrated a steep decline after 4 years of DD. (B) Attention showed a biphasic decline: first, early on (Group 0 vs Group 1) and the second after 4 years of DD (Group 3 vs Group 2). (C) Similarly to attention, memory also showed a two-step decline: first early on (Group 0 vs Group 1) and then after 4 years of DD (Group 3 vs Group 2). (D) Verbal fluency was the most affected domain in the early phase; however, its subsequent decline was not significant. (E) Language subscore was affected in the early phase and declined gradually, nonetheless, the decline was not significant after 4 years of DD. (F) Visuospatial abilities were already impaired early on and followed a linear pattern of decline (every group was significantly different). Note: the points represent means while the vertical lines represent standard errors. *Signals significant differences ($p<0.01$).

IV.1.4. Interrelation between disease duration and the subscores of the Addenbrooke's Cognitive Examination

We used Spearman's rho to assess the interrelation between the six subscores of the ACE and disease duration. Figure 5. shows scatter plots illustrating the interrelation between ACE subscores and disease duration (Figure 5.).

DOI:10.14753/SE.2024.3035

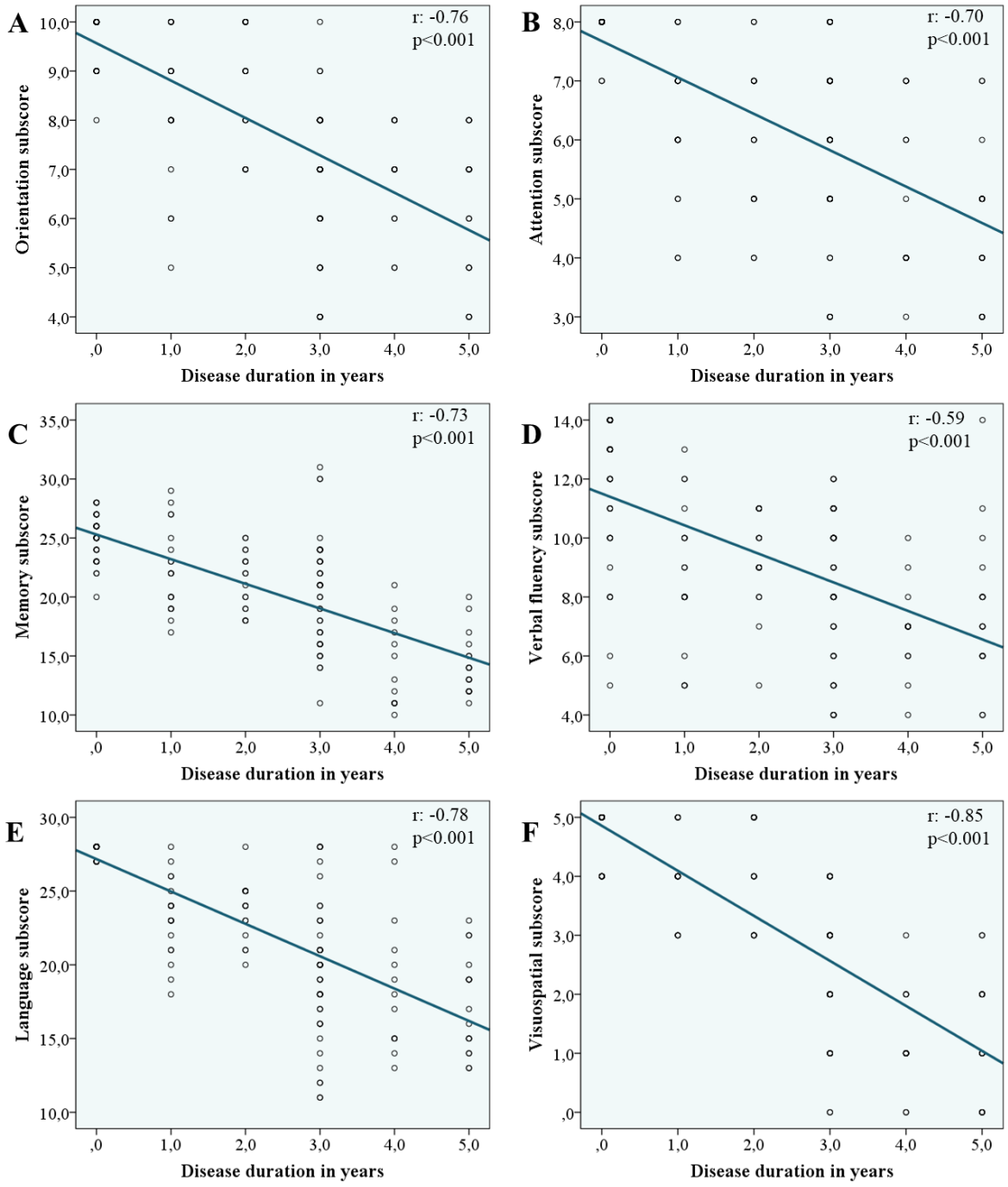

Figure 5. Results of Spearman's correlation analysis investigating the association between disease duration and ACE subscores. Every subscore of the ACE ((A) orientation, (B) attention, (C) memory, (D) verbal fluency, (E) language, and (F) visuospatial subscore) correlated significantly negatively with disease duration (p-values <0.05). The most

prominent association presented between disease duration and visuospatial abilities (r= -0.85).

IV.1.5. Intragroup Differences in the subscores of the Addenbrooke's Cognitive Examination

We used the Wilcoxon signed rank test to analyse within-group differences in ACE subscores. Within-group differences in normalized subscores are detailed in Table 5 and illustrated in Figure 6.

In group 0, the normalized subscore of verbal fluency (0.87) was the lowest from the six subscores of the ACE, followed by memory (0.9) but the two did not differ significantly. However, the normalized subscore of verbal fluency was significantly lower than that of orientation ($Z= -3.95$; $p<0.001$), attention ($Z= -4.60$; $p<0.001$), language ($Z= -4.68$; $p<0.001$), and visuospatial abilities ($Z= -3.75$; $p<0.001$). Regarding memory, its normalized subscore was significantly lower than that of orientation ($Z= -4.083$; $p<0.001$), attention ($Z= -5.40$; $p<0.001$), language ($Z= -5.52$; $p<0.001$), and visuospatial abilities ($Z= -3.61$; $p<0.001$). The normalized subscore of visuospatial abilities was the third lowest (0.96), it was significantly lower than the normalized subscore of orientation ($Z= -2.10$; $p=0.036$), attention ($Z= -2.94$; $p=0.003$), and language ($Z= -2.82$; $p=0.005$).

Similarly to group 0, the lowest normalized subscore of the ACE in Group 1 was verbal fluency (0.64), followed by attention (0.77) and memory (0.78). The normalized subscore of verbal fluency was significantly lower than that of orientation ($Z= -4.79$; $p<0.001$), attention ($Z= -4.14$; $p<0.001$), memory ($Z= -4.41$; $p<0.001$), language ($Z= -5.23$; $p<0.001$), and visuospatial abilities ($Z= -4.69$; $p<0.001$). Regarding attention, its normalized subscore was significantly lower than orientation ($Z= -2.34$; $p=0.019$) and language ($Z=-5.23$; $p<0.001$), while it was not significantly different from memory and visuospatial abilities. As for memory, its normalized subscore was significantly lower than that of orientation ($Z= -2.27$; $p=0.023$) and language ($Z= -5.23$; $p<0.001$) but it did not differ significantly from visuospatial abilities.

Contrary to Group 0 and Group 1, the lowest normalized subscore of the ACE in Group 2 was visuospatial abilities (0.5). Verbal fluency (0.6) was the second, while orientation (0.68)

was the third lowest subscore. The normalized subscore of visuospatial abilities was significantly lower than orientation (Z= -4.38; p<0.001), attention (Z= -5.24; p<0.001), memory (Z= -5.25; p<0.001), verbal fluency (Z= -3.31; p=0.001), and language (Z= -4.07; p<0.001). As for verbal fluency, its normalized subscore was significantly lower than that of orientation (Z= -3.62; p<0.001), attention (Z= -5.47; p<0.001), memory (Z= -4.87; p<0.001), and language (Z= -3.55; p<0.001). Regarding orientation, it had a significantly lower normalized score than attention (Z= -3.23; p=0.001) and memory (Z= -2.19; p=0.029).

Similarly to Group 2, the lowest normalized subscore of the ACE in Group 3 was visuospatial abilities (0.25). The second lowest was memory (0.51) followed by verbal fluency (0.52). The normalized subscore of visuospatial abilities was significantly lower than that of any other cognitive domain: orientation (Z= -4.73; p<0.001), attention (Z= -4.74; p<0.001), memory (Z= -4.46; p<0.001), verbal fluency (Z= -4.47; p<0.001), and language (Z= -4.64; p<0.001). Memory had a significantly lower normalized score than orientation (Z= -3.86; p<0.001), attention (Z= -3.10; p=0.002), and language (Z= -4.32; p<0.001). The normalized subscore of verbal fluency was significantly lower than that of orientation (Z= -3.75; p<0.001), attention (Z= -2.42; p=0.016), and language (Z= -2.76; p=0.006).

DOI:10.14753/SE.2024.3035

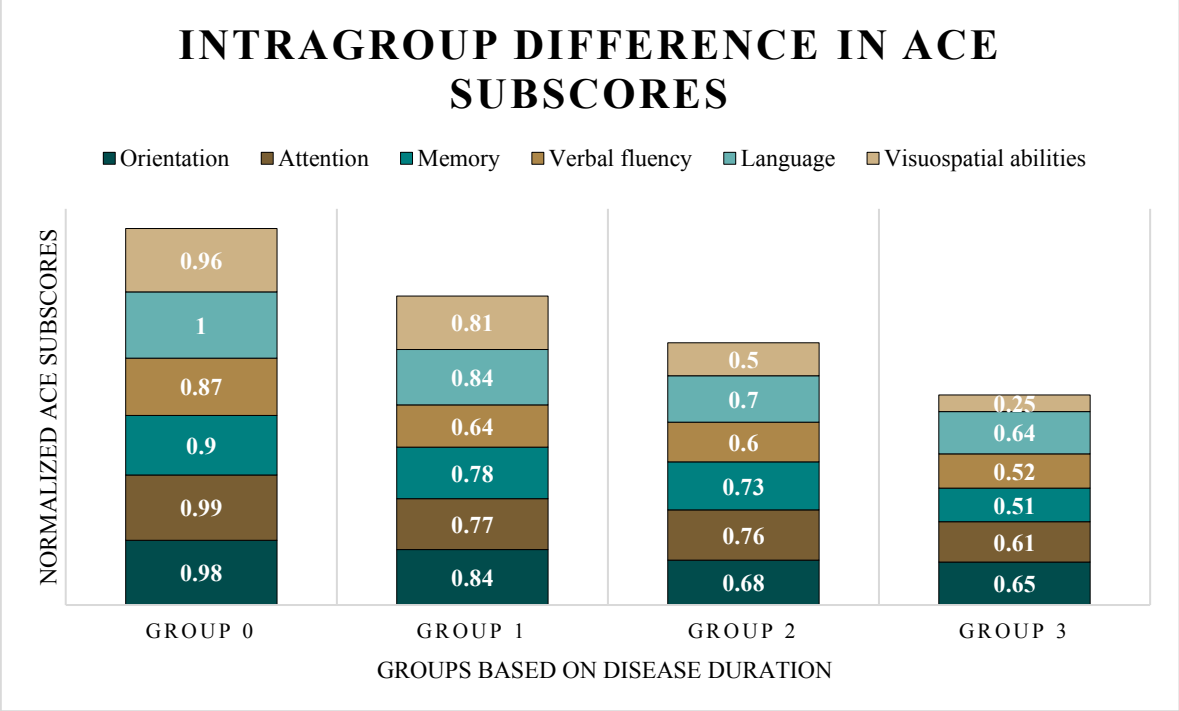

Figure 6. Intragroup differences in normalized subscores of Addenbrooke's Cognitive Examination (ACE). Verbal fluency is the most impaired subscore in Group 1 pointing to marked involvement of this cognitive domain the early phase of AD. In addition, visuospatial abilities presented a steep linear trajectory of decline with longer disease duration.

DOI:10.14753/SE.2024.3035

Table 5. Normalized attention, orientation, verbal fluency, memory, visuospatial abilities, and language subscores of the ACE per group.

Note: Normalization was carried out by dividing each subscore by the maximum score of the given subscore (e.g., 6/8 in attention subscore gives a normalized score of 0.75). Wilcoxon-signed ranked test was applied for the analysis of normalized scores. Nonsignificant differences are marked by =, whereas >, < signals significance ($p<0.05$) and the relation of the given subscores. A: attention, O: orientation, VF: verbal fluency, M: memory, VS: visuospatial abilities, L: language, SD: standard deviation (89).

Cognitive domains	Descriptive statistics	Group 0	Group 1	Group 2	Group 3
Orientation	Mean	0.98	0.84	0.68	0.65
	SD	0.05	0.13	0.15	0.13
	Differences	O=A, O>M, O>VF, O<L, O>VS	O>A, O>M, O>VF, O=L, O=VS	O<A, O<M, O>VF, O=L, O>VS	O=A, O>M, O>VF, O=L, O>VS
Attention	Mean	0.99	0.77	0.76	0.61
	SD	0.03	0.15	0.19	0.17
	Differences	A>M, A>VF, A=L, A>VS	A=M, A>VF, A<L, A=VS	A=M A>VF, A>L, A>VS	A>M, A>VF, A=L, A>VS
Memory	Mean	0.90	0.78	0.73	0.51
	SD	0.06	0.11	0.16	0.11
	Differences	M=VF, M<L, M<VS	M>VF, M<L, M=VS	M>VF, M=L, M>VS	M=VF, M<L, M>VS
Verbal fluency	Mean	0.87	0.64	0.60	0.52
	SD	0.17	0.15	0.17	0.15
	Differences	VF<L, VF<VS	VF<L, VF<VS	VF<L, VF>VS	VF<L, VF>VS
Language	Mean	1.00	0.84	0.70	0.64
	SD	0.995	0.08	0.16	0.14
	Differences	L>VS	L=VS	L>VS	L>VS
Visuospatial abilities	Mean	0.96	0.81	0.50	0.25
	SD	0.08	0.15	0.23	0.19

DOI:10.14753/SE.2024.3035

IV.2. Neuroimaging study of the visuospatial system in mild cognitive impairment

IV.2.1. Demographics and neuropsychological test results

The two study groups differed significantly in age and sex: the HC group had a higher ratio of female participants detected by Chi-square test ($\chi 2=5.128$, $p=0.024$); while the a-MCI group was significantly older ($F=6.18$, $p=0.015$). However, age and sex had no significant modifying effect ($p>0.05$). No significant difference was found between the two groups in years of education ($p=0.142$). The two groups also significantly differed in neuropsychological test results, out of which many remained significant after Benjamini-Hochberg correction for multiple comparisons (Table 6.). Compared to the HC group, the a-MCI group had lower score on the MMSE test ($F=9.098$, $p<0.001$) decreased total ACE score ($F=11.065$, $p<0.001$), reduced RAVLT sum-5 and RAVLT7 scores ($F=13.53$, $p<0.001$ and $F=11.9$, $p<0.001$, respectively). The a-MCI group needed significantly longer time to complete the TMT-A and B tests ($F=4.69$, $p=0.048$ and $F=5.51$, $p=0.021$, respectively) signalling impaired cognitive function. Regarding the cognitive domains, the visuospatial subscore of the a-MCI group was significantly lower ($F=8.32$, $p<0.001$), while the other domains were not significantly different between the two groups after correction for multiple comparisons (corrected $p>0.05$).

Table 6. Demographics and neuropsychological test results.

Note: HC: healthy control, a-MCI: amnestic mild cognitive impairment; ACE: Addenbrooke's Cognitive Examination, RAVLT: Rey Auditory Verbal Learning Test, MMSE: Mini Mental State Examination, TMT: Trail-Making Test, ACOL: AlzEpi Cohort Observational Library, SMNC: Semmelweis MCI Neuroimaging Cohort, a: independent sample t-test, b: chi-square test, c: Mann–Whitney U-test, d: ANCOVA analysis with age and sex as covariates, e: given in the form mean ± standard deviation, f: reported as percentage of female subjects in each group, g: presented in Cohen's d, * signals significant intergroup differences after Benjamini-Hochberg correction ($p<0.05$) (70).

	HC (n:46)		a-MCI (n:32)		p-value	Effect size[g]
Demographics						
Age (years)[a,e]	67.63±7.15		70.68±9.94		0.015*	0.352
Sex (% of females)[b,f]	69.6		53.1		0.024*	–
Education (years) [c,e]	15±2.53		14.43±3.13		0.142	0.200
Neuropsychology						
MMSE [d,e]	28.52±1.13		26.87±1.62		p<0.001*	1.181
ACE Total [d.e]	93.24±3.29		82.31±7.26		p<0.001*	1.939
ACE Orientation [d,e]	9.88±0.31		9.43±0.8		0.029	0.742
ACE Attention [d,e]	7.91±0.28		7.65±0.86		0.134	0.407
ACE Memory [d,e]	30.97±2.11		24.68±5.47		0.03	1.517
ACE Verbal fluency [d,e]	11.95±2.24		9.62±2.87		0.021	0.905
ACE Language [d,e]	27.71±0.54		27.125±1.58		0.378	0.495
ACE Visuo-spatial [d,e]	4.71±0.5		3.78±1.12		<0.001*	1.072
RAVLT sum-5 [d,e]	48.43±8.69		31.15±9.4		<0.001*	1.909
RAVLT7 [d,e]	9.89±2.75		4.03±2.83		<0.001*	2.100
TMT-A [d,e]	39.62±10.58		90.41±66.98		0.008*	1.059
TMT-B [d,e]	83.13±32.67		209.33±147.31		0.003*	1.183
Demographics separately for the two centres						
	ACOL (n:29)	SMNC (n:17)	ACOL (n:18)	SMNC (n:14)		
Age (years)[e]	67.83±7.55	67.29±6.64	71.00±7.19	70.43±13.12	–	–
Sex (% of females)[f]	68.97	70.59	44.44	64.29	–	–

DOI:10.14753/SE.2024.3035

IV.2.2. Results of structural MRI acquisition

Compared to the HC groups, we found reduced cortical thickness in the a-MCI group in several cortical areas (Appendix). The cortical thickness of the right superior temporal gyrus and the left temporal pole showed the greatest F values (F=8.04 and F=5.26, respectively). These are the only areas where intergroup differences of cortical thickness remained significant after Benjamini-Hochberg correction ($p<0.001$ and $p=0.034$, respectively) (Figure 7.). We found that age had a significant modifying effect on cortical thickness of several regions: right frontal region (orbital and opercular parts of the inferior frontal gyrus $p=0.005$ and $p=0.033$, respectively, and precentral gyrus $p=0.04$), left frontal region (orbital, opercular, and triangular part of the inferior frontal gyrus $p=0.024$, $p=0.027$, and $p=0.006$, respectively, medial and lateral orbitofrontal gyrus $p=0.004$ and $p=0.008$, respectively, caudal middle frontal gyrus $p=0.049$ and precentral gyrus $p=0.04$), right temporal region (superior temporal gyrus $p=0.0015$ and temporal pole $p=0.002$), and left temporal regions (entorhinal cortex $p=0.009$, superior temporal gyrus $p=0.003$, temporal pole $p=0.022$, and parahippocampal gyrus $p=0.006$). We found that age had the largest effect on the right superior temporal gyrus (F=21.81) and the opercular part of the left inferior frontal gyrus (F=9.39). However, this effect could not be seen once Benjamini-Hochberg correction was applied ($p>0.05$). Sex did not modify the results significantly ($p>0.05$).

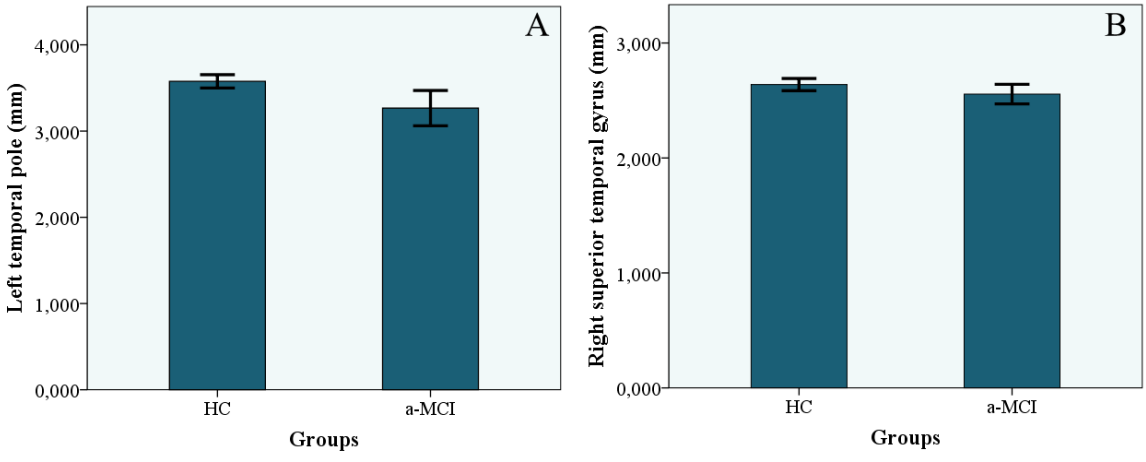

Figure 7. Intergroup comparison of cortical thickness between healthy controls and amnestic MCI patients. Cortical thickness of the left temporal pole and right superior temporal gyrus

was significantly decreased in a-MCI patients. HC: healthy control, a-MCI: amnestic mild cognitive impairment. Note: the intersection of vertical and horizontal lines represent means while the vertical lines represent standard errors.

IV.2.3. Intergroup differences in functional connectivity

Compared to the HC group, two cortical regions of the VS network showed reduced functional connectivity with seed-to-ROI analysis. We found that the right middle frontal gyrus had decreased functional connectivity to the left superior frontal gyrus, the left middle frontal gyrus, and left precentral gyrus (Figure 8A). Moreover, the right superior frontal gyrus had impaired functional connectivity to the left temporal pole, the left inferior temporal gyrus, and the left precentral gyrus (Figure 8B). However, the a-MCI group had stronger functional connectivity between the left inferior temporal gyrus and the triangular part of the left inferior frontal gyrus and the left middle frontal gyrus compared with the HC group (Figure 9.).

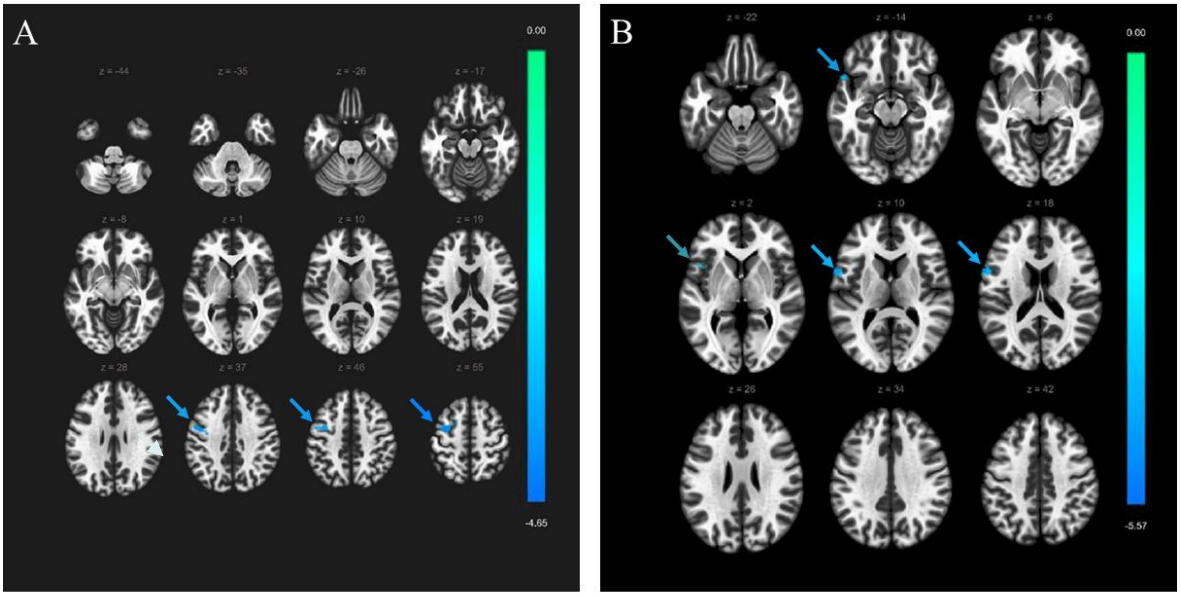

Figure 8. Weaker functional connectivity in amnestic MCI patients compared to healthy controls. Significantly reduced functional connectivity presented between the seed region called the right middle frontal gyrus and the left middle frontal gyrus, the left superior frontal gyrus, and the left precentral gyrus (part A). It also presented between the seed region called

DOI:10.14753/SE.2024.3035

the right superior frontal gyrus and the left frontal operculum, the left inferior temporal gyrus, the left temporal pole, and the left precentral gyrus (part B). The uncorrected threshold at the voxel-level was $p<0.001$, while the FDR corrected threshold at the cluster-level was $p<0.05$. Contrast: amnestic MCI > healthy controls. Colour bar depicts the Fischer-transformed correlation value.

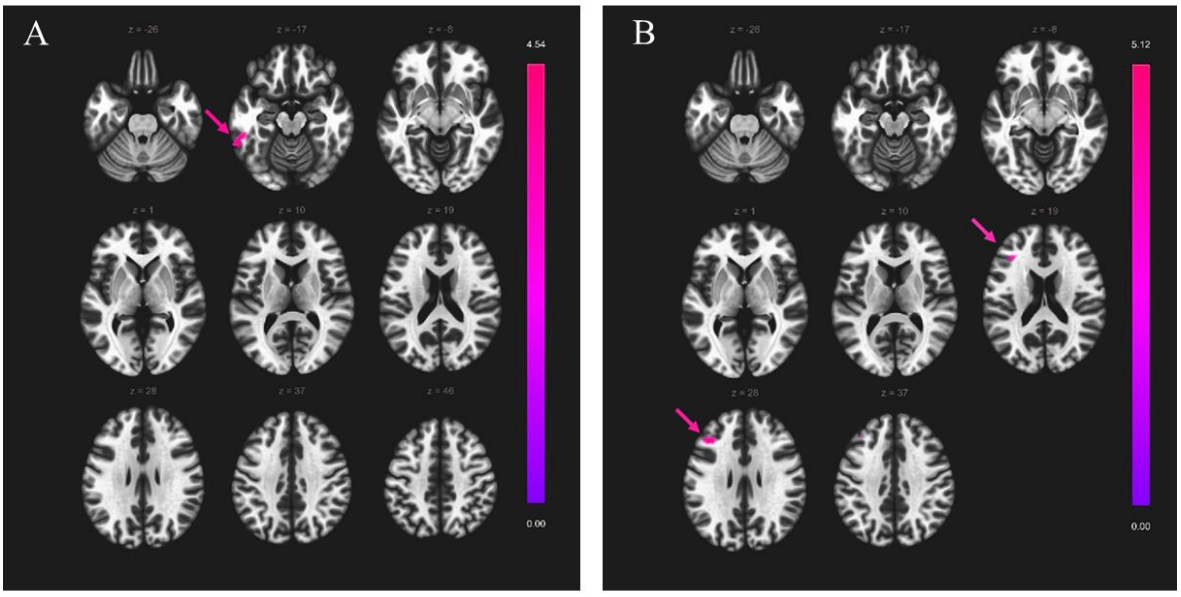

Figure 9. Stronger functional connectivity in amnestic MCI patients compared to healthy controls. Significantly increased functional connectivity presented between the seed region called the left inferior temporal gyrus (part A) and the left inferior frontal gyrus and the left middle frontal gyrus (part B). The uncorrected threshold at the voxel-level was $p<0.001$, while the FDR corrected threshold at the cluster-level was $p<0.05$. Contrast: amnestic MCI > healthy controls. Colour bar depicts the Fischer-transformed correlation values.

IV.3. Investigating the differentiating potential of a visuomotor paradigm in distinguishing MCI patients from healthy controls

IV.3.1. Demographics, neuroimaging, and neuropsychological test results.

We selected 80 individuals (49 healthy controls and 31 MCI patients) from the ACOL database who were eligible for the study based on the selection criteria. Three healthy

controls and 9 MCI patients, altogether 12 individuals denied participation. 68 individuals agreed to participate in the study: 46 healthy controls and 22 MCI patients. The MCI group was composed of 12 females (54.55%) while the HC group included 27 female participants (58.70%). We did not find significant differences in years of education (p=0.128) or ratio of sex (p=0.225). However, we found significant differences between the two groups in age (F=0.638; p=0.019) and numerous neuropsychological test results, such as the RAVLT sum-5 (F=110; p=0.015) and RAVLT7 (F=69.84; p=0.008) scores, the ACE total score (F=60.086; p<0.001) as well as its subscores: category and letter fluency (F=31.48; p<0.001 and F=5.07; p=0.028, respectively). The two groups also differed in the CDR score (Z= -4.284; p<0.001). We did not find significant intergroup differences in the results of other neuropsychological tests (p>0.05). However, we detected significant differences between the two groups in cortical thickness: compared to the HC group, MCI patients had decreased grey matter volume (F=4.21, p=0.012) and reduced thickness of the entorhinal cortex (F=3.86, p=0.017) (Table 7.).

Table 7. Demographics, neuroimaging, and neuropsychological test results

Note: ACE: Addenbrooke's Cognitive Examination, MMSE: Mini-Mental State Examination, RAVLT: Rey Auditory Verbal Learning Test, CDR: Clinical Dementia Rating scale, TMT: Trail-Making Test, STAI-T: Spielberger State-Trait Anxiety Inventory Trait Score, STAI-S: Spielberger State-Trait Anxiety Inventory State Score, BDI: Beck Depression Inventory, MCI: mild cognitive impairment, HC: healthy control, * signals significant differences (p<0.05), a: independent samples t-test was applied, b: Chi-square test was applied, c: Mann-Whitney U-test was applied, d: mean ± standard deviation, e: median followed by interquartile range (IQ_1–IQ_3), f: higher score indicates worse cognitive functioning (88).

	HC (n=46)	MCI (n=22)	p-value	Effect size (Cohen's d)
Demographics				
Age (years) [a]	66.76±7.63	71.18±5.82	0.019*	0.65
Sex (% of females) [b]	58.70	54.55	0.225	–
Education (years) [a, d]	15.54±1.93	14.63±2.38	0.128	0.42
Neuroimaging				

Average entorhinal thickness (mm^2) [a, d]	3.43±0.25	3.11±0.23	0.017*	1.31
Total grey matter volume (mm^3) [a, d]	576828±57471	573914±57471	0.012*	0.05
Neuropsychology				
MMSE [a, d]	28.76±1.08	26.5±1.79	0.088	1.55
ACE Total [a, d]	93.93±3.85	80.22±9.7	<0.001*	1.88
ACE Orientation [a, d]	9.77±0.53	9.19±0.16	0.434	0.59
ACE Attention [c, e]	8.00 (8.00–8.00)	8.00 (8.00–8.00)	0.307	0.45
ACE Memory [a, d]	29.93±0.25	23.7±6.07	0.089	1.17
ACE Letter fluency [a, d]	5.98±1.39	4.29±2.07	0.028*	0.97
ACE Category fluency [a, d]	6.67±0.04	4.91±1.76	<0.001*	1.36
ACE Language [a, d]	27.93±0.26	26.7±1.88	0.388	0.96
ACE Visuospatial [a, d]	4.8±0.1	4.1±0.73	0.135	1.2
RAVLT sum-5 [a, d]	49.24±6.36	29.2±7.51	0.015*	2.93
RAVLT7 [a, d]	10.17±2.73	3.82±2.21	0.008*	2.59
TMT-A (in sec) [a, d, f]	39.98±11.76	85.6±55.1	0.288	1.19
TMT-B (in sec) [a, d, f]	87.83±46.17	186±117.7	0.078	1.12
CDR [c, e, f]	0 (0–0.2)	0.88 (0.4–1)	<0.001*	1.19
BDI [a, d, f]	4.3±4.02	5.61±3.15	0.32	0.28
STAI-S [a, d, f]	36.5±9.23	43.25±8.38	0.32	0.77
STAI-T [a, d, f]	41.95±9.51	42.38±7.85	0.118	0.04

IV.3.2 Between-group differences in computer mouse-movement characteristics

We found significant intergroup differences in computer mouse movement characteristics for both the right and left hand. Regarding the left hand, we detected the most significant difference in movement entropy ($F=5.24$; $p=0.001$, Cohen's $d=0.94$) while the time required for task completion and the distance of the hand movements were also significantly different ($F=4.32$; $p=0.005$ and $F=1.16$, $p=0.0134$, respectively) (Figure 10.). In regard to the right hand, we found that the entropy of the computer mouse movements proved to be the most significantly different characteristic between the groups ($F=8.46$; $p<0.001$, Cohen's $d=0.9$), while the time of task completion and the distance of the movements also proved to be significantly different ($F=4.626$; $p=0.003$ and $F=1.03$, $p=0.019$, respectively) (Figure 10.). Out of the six measures only four remained significant after the Benjamini Hochberg correction ($p<0.05$), the significance was lost for distance of computer mouse movements for both the left and right hand ($p>0.05$). Although not significant, we identified a shift in computer mouse movement characteristics of the MCI group: their speed of movement was

DOI:10.14753/SE.2024.3035

slower, and they needed more tries to successfully hit the targets; however, the velocity of their movements was also higher (Table 8.). No significant modifying effect of age, sex, and state anxiety was detected ($p>0.05$).

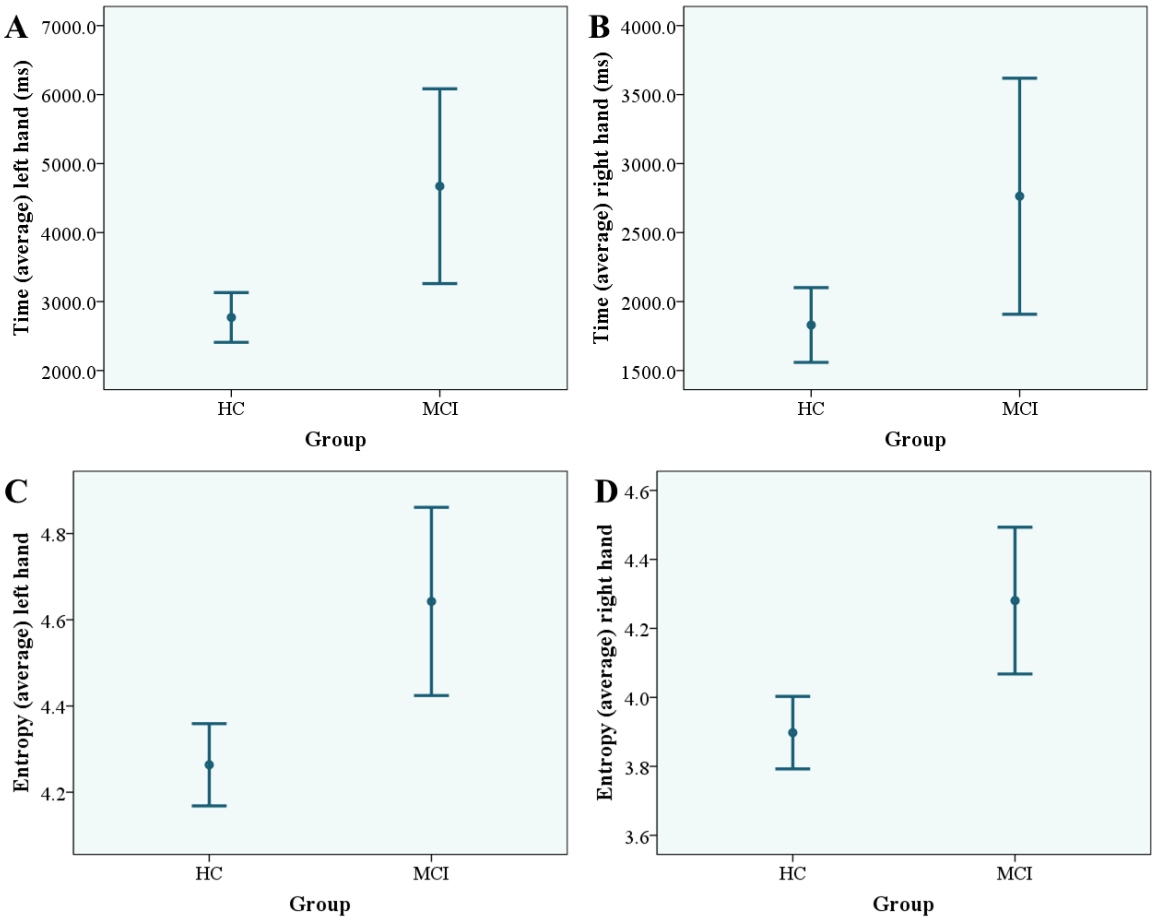

Figure 10. Intergroup comparisons of MCI patients' and healthy controls' computer mouse movement characteristics. MCI patients required longer time for task completion (part A and B) and had significantly higher levels of movement entropy (part C and D). Intergroup differences are more prominent in case of the left hand (part A and C). Every plot shows significant difference ($p<0.05$).

Note: the points represent means while the vertical lines represent standard errors. MCI: mild cognitive impairment, HC: healthy control, ms: millisecond

DOI:10.14753/SE.2024.3035

Table 8. Differences in computer mouse movement parameters between MCI and HC participants. The MCI group had significantly increased movement entropy and required time for task completion.

Note: MCI: mild cognitive impairment, HC: healthy control, *signals significant differences after Benjamini-Hochberg correction was applied to counteract multiple comparisons, ms: millisecond (88).

	HC (n=46)	MCI (n=22)	Nominal p-value	Effect size (Cohen's d)
Left hand				
Average distance of 9 routes (pixel)	1611.94±1044	2319.31±1368.18	0.0134	0.58
Average entropy of 9 routes	4.26±0.32	4.64±0.47	0.001*	0.94
Average number of tries of 9 routes	2.88±0.31	3.17±0.69	0.329	0.54
Average speed of 9 routes (pixel/ms)	0.53±0.14	0.45±0.2	0.124	0.46
Average time of 9 routes (ms)	2932.41± 1601.92	4672.21±2648.63	<0.005*	0.79
Average velocity of 9 routes (pixel/ms)	4911765± 9621912	7765693±7971712	0.389	0.32
Right hand				
Average distance of 9 routes (pixel)	1181.41±275.2	1714.32±591.35	0.0119	0.22
Average entropy of 9 routes	3.9±0.35	4.28±0.48	<0.001*	0.9
Average number of tries of 9 routes	3.2±0.62	3.66±1.76	0.97	0.34
Average speed of 9 routes (pixel/ms)	0.57±0.17	0.47±0.19	0.069	0.55
Average time of 9 routes (ms)	1830.25±901.93	3545.95±3064	0.003*	0.75
Average velocity of 9 routes (pixel/ms)	2462435± 8318595	2923976±5945440	0.5	0.06

DOI:10.14753/SE.2024.3035

IV.3.3 Interrelation between neuropsychological test results and computer mouse movement characteristics

Based on the results of the intergroup comparisons, entropy, time, and speed of computer mouse movements appeared to be the most promising measures for further analysis. We applied Pearson correlation between these measures and the neuropsychological test results (Table 9. for p-values and Figure 11. for r values). We found significant correlations between movement parameters and the scores of RAVLT, ACE, MMSE, CDR and TMT tests, while results for tests such as BDI and STAI—measuring mood and anxiety—were not significant. The correlation was positive for the tests where higher scores indicate worse performance, e.g., TMT-A and B and CDR scale with r values of 0.1 to 0.65. We detected the most significant correlation between the CDR scale and movement parameters (average $r=0.36$, all p's<0.001). With respect to the measure with the leading predictive value, we found left hand movement features to be superior: time of left-hand movements proved to be the first with an average r value of +0.62 ($p<0.001$), followed by entropy of the left-hand movement with an average r value of +0.49 ($p<0.001$). Apart from the leading measures, we found higher average r values for the left hand (0.46) in every measure compared to the right hand (0.21) (Figure 11.). Regarding the tests where higher scores indicate better cognitive performance, such as the ACE, MMSE, and RAVLT, we found a negative correlation with the computer mouse movement features, with r values of -0.14 to -0.5. We detected the most significant correlation between the ACE total score and the movement parameters (average $r= -0.37$, all p's <0.05). In regard to the predictive value of the movement features, time of the left-hand movements proved to be the leading measure with an average r of -0.38 (p-values <0.05) (Figure 12.) closely followed by entropy of left hand with and average r of -0.34 and the right hand where average r was -0.31 (all p-values <0.05). Considering the average r values of all movement parameters per hand, we did not find a significant difference between the two hands with the average r values of -0.29 for both hands. Results of the Pearson correlation analysis were log-transformed before averaging, followed by back-transformation after averaging was conducted.

DOI:10.14753/SE.2024.3035

Table 9. Pearson correlation analysis between computer mouse movement parameters and neuropsychological test results (p-values). The analysis resulted in several significant correlations between mouse movement characteristics and neuropsychological test results assessing cognitive functioning. However, no significant association was found between movement parameters and mood and anxiety measurements.

Note: ACE: Addenbrooke's Cognitive Examination, MMSE: Mini-Mental State Examination, TMT: Trail-Making Test, RAVLT: Rey Auditory Verbal Learning Test; CDR: Clinical Dementia Rating scale; STAI-T: Spielberger State-Trait Anxiety Inventory Trait Score, STAI-S: Spielberger State-Trait Anxiety Inventory State Score, BDI: Beck Depression Inventory, a: Lower score signals better cognitive performance, *Signals significant correlation ($p<0.05$) (88).

	Left hand: Distance (pixel)	Left hand: Entropy	Left hand: Time (in ms)	Right hand: Distance (pixel)	Right hand: Entropy	Right hand: Time (in ms)
MMSE	0.064	0.006*	0.006*	0.002*	0.003*	0.013*
ACE total	0.026*	<0.001*	<0.001*	<0.001*	<0.001*	0.003*
RAVLT sum-5	0.037*	0.001*	0.002*	0.007*	0.004*	0.121
RAVLT7	0.097	0.002*	0.025*	0.002*	0.002*	0.037*
TMT-A (in sec) [a]	0.009*	<0.001*	<0.001*	0.129	0.009*	0.203
TMT-B (in sec) [a]	0.022*	<0.001*	<0.001*	0.225	0.046*	0.139
CDR [a]	<0.001*	<0.001*	<0.001*	<0.001*	<0.001*	<0.001*
BDI [a]	0.074	0.784	0.984	0.319	0.081	0.544
STAI-S [a]	0.321	0.373	0.047	0.217	0.370	0.227
STAI-T [a]	0.280	0.241	0.356	0.719	0.437	0.842

DOI:10.14753/SE.2024.3035

	ACE total	*MMSE*	*RAVLT sum-5*	*RAVLT7*	*CDR*	*TMT-A*	*TMT-B*	*BDI*	*STAI-S*	*STAI-T*	*Distance (average) left hand*	*Entropy (average) left hand*	*Time (average) left hand*	*Distance (average) right hand*	*Entropy (average) right hand*	*Time (average) right hand*
ACE total	1	0.777	0.769	0.649	-0.626	-0.616	-0.634	-0.127	-0.122	0.084	-0.277	-0.490	-0.503	-0.456	-0.473	-0.362
MMSE	0.777	1	0.615	0.567	-0.543	-0.546	-0.532	-0.251	-0.101	0.040	-0.231	-0.338	-0.348	-0.377	-0.358	-0.310
RAVLT sum-5	0.769	0.615	1	0.831	-0.572	-0.667	-0.633	-0.114	-0.152	0.080	-0.259	-0.414	-0.391	-0.325	-0.348	-0.196
RAVLT7	0.649	0.567	0.831	1	-0.473	-0.483	-0.407	-0.103	-0.180	0.011	-0.208	-0.380	-0.289	-0.364	-0.368	-0.261
CDR	-0.626	-0.543	-0.572	-0.473	1	0.576	0.594	0.115	0.199	-0.051	0.428	0.454	0.609	0.402	0.405	0.135
TMT-A	-0.616	-0.546	-0.667	-0.483	0.576	1	0.870	0.132	0.120	0.015	0.323	0.535	0.615	0.189	0.316	0.163
TMT-B	-0.634	-0.532	-0.633	-0.407	0.594	0.870	1	0.031	0.216	0.033	0.290	0.492	0.654	0.154	0.248	0.188
BDI	-0.127	-0.251	-0.114	-0.103	0.115	0.132	0.031	1	0.340	0.668	0.223	0.034	0.003	0.124	0.213	0.077
STAI-S	-0.122	-0.101	-0.152	-0.180	0.199	0.120	0.216	0.340	1	0.584	0.128	0.114	0.264	0.156	0.113	0.157
STAI-T	0.084	0.040	0.080	0.011	-0.051	0.015	0.033	0.668	0.584	1	0.141	0.151	0.125	0.046	0.099	-0.026
Distance (average) left hand	-0.277	-0.231	-0.259	-0.208	0.428	0.323	0.290	0.223	0.128	0.141	1	0.696	0.712	0.493	0.415	0.361
Entropy (average) left hand	-0.490	-0.338	-0.414	-0.380	0.454	0.535	0.492	0.034	0.114	0.151	0.696	1	0.837	0.493	0.585	0.550
Time (average) left hand	-0.503	-0.348	-0.391	-0.289	0.609	0.615	0.654	0.003	0.264	0.125	0.712	0.837	1	0.447	0.548	0.501
Distance (average) right hand	-0.456	-0.377	-0.325	-0.364	0.402	0.189	0.154	0.124	0.156	0.046	0.493	0.493	0.447	1	0.774	0.800
Entropy (average) right hand	-0.473	-0.358	-0.348	-0.368	0.405	0.316	0.248	0.213	0.113	0.099	0.415	0.585	0.548	0.774	1	0.907
Time (average) right hand	-0.362	-0.310	-0.196	-0.261	0.135	0.163	0.188	0.077	0.157	-0.026	0.361	0.550	0.501	0.800	0.907	1

Figure 11. Correlation matrix displaying correlations (r values) between neuropsychological test results and computer mouse movement parameters. The colour green signifies negative correlations, while the colour burgundy signals positive correlations. Participants with worse cognitive performance indicated by lower scores in ACE, MMSE, RAVLT tests had higher values of movement parameter values, resulting in negative r values. In addition, the tests where higher scores indicate worse cognitive performance, such as CDR and TMT, correlated positively with computer mouse movements. To conclude, higher values of movement parameters signal impaired cognitive functioning.

Note: ACE: Addenbrooke's Cognitive Examination, MMSE: Mini-Mental State Examination, RAVLT: Rey Auditory Verbal Learning Test, CDR: Clinical Dementia Rating scale, TMT: Trail-Making Test, STAI-T: Spielberger State-Trait Anxiety Inventory Trait Score, STAI-S: Spielberger State-Trait Anxiety Inventory State Score, BDI: Beck Depression Inventory.

DOI:10.14753/SE.2024.3035

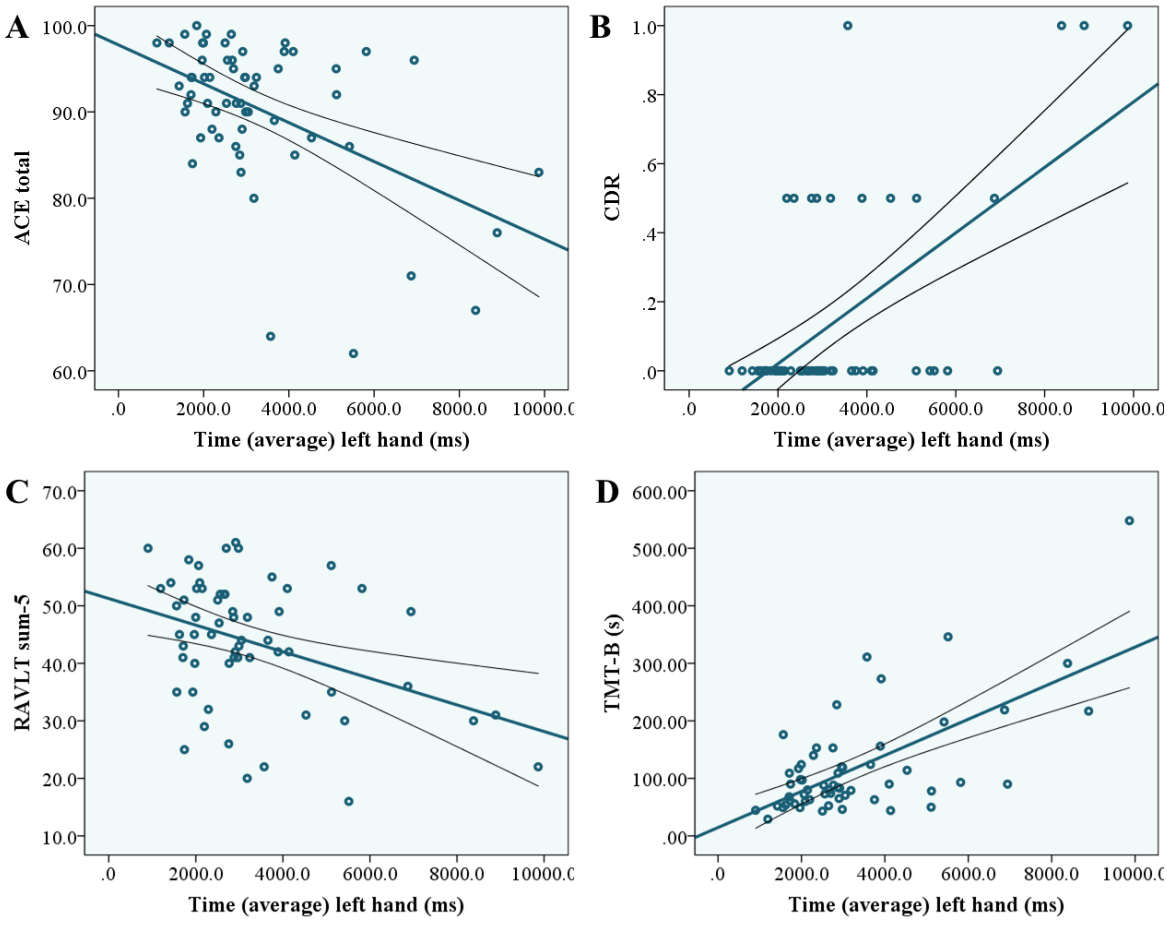

Figure 12. Pearson correlation analysis between neuropsychological test scores and average time of left-handed computer mouse movements (in ms). Negative correlation was found between required time for task completion and neuropsychological test scores in which lower scores indicate worse cognitive functioning such as ACE: Addenbrooke's Cognitive Examination and RAVLT sum-5: Rey Auditory Verbal Learning Test summary of first 5 trials (part A and C). On the contrary, positive correlation was present between time required for task completion and neuropsychological test scores where higher scores indicate worse cognitive functioning such as CDR: Clinical Dementia Rating Scale and TMT-B: Trail-Making Test part B (part B and D).

Note: ms: millisecond; s: second.

DOI:10.14753/SE.2024.3035

V. Discussion

Around 50% of dementia patients remain undiagnosed notwithstanding the major efforts aimed at a timely diagnosis (29). There are a number of possible causes contributing to difficulty with diagnosis: first, the limited availability of screening: only 16 percent of older adults aged 65 years and over receive routine cognitive evaluation (30). Furthermore, there is a limit in the availability of trained medical personnel and the geographical constraint of these examinations also has a role since the patient must be physically present at the examination. Additionally, neuropsychological tests struggle to identify early-stage cognitive impairment due to a lack of sensitivity (31).

Most neuropsychological evaluations of dementia patients are heavily focused on memory tasks and less so on other cognitive domains (the proportion of memory scores from the total scores are 35/70 in ADAS-Cog, 35/100 in ACE, 5/30 in MoCA, while that of visuospatial abilities are 0/70 in ADAS-Cog, 5/100 in ACE, and 4/30 in MoCA). However, visuospatial abilities have been found to carry significant diagnostic and prognostic capabilities (40). A recent study by Wasserman et al. proposed that the assessment of visuospatial abilities could be an accurate method to detect multiple domain MCI (90). In the first phase of our research, we aimed to investigate the impairment of different cognitive domains and their contribution to the global cognitive deficit of AD patients by using and analysing a comprehensive neuropsychological test battery.

In our study of 110 clinically defined AD patients (Group 1-3) and 45 cognitively healthy individuals (Group 0), we found that visuospatial abilities demonstrated the most precipitous deterioration with increased disease duration, thus, it could be a promising candidate for observing the advancement of the disease stages.

Regarding the most affected cognitive domain early on the disease continuum, we found verbal fluency to be the most defective in the beginning of the disease course, similar to the level of memory deficiency in early AD. Regarding global cognition, we found that ACE total score decreases with increased disease duration supported by correlation analysis and intergroup comparisons. This finding is in accordance with previous literature stating that the

impairment of global cognition in AD follows a linear trajectory (91, 92), and literature supporting the capacity of ACE in indicating the patient's stage on the disease continuum (93). When breaking down ACE total score to subscores according to cognitive domains, orientation, attention, memory, verbal fluency, language, and visuospatial skills, all show decreased subscores with later disease stages. However, outstanding differences in the level of impairment of the cognitive domains appear at different disease stages, which is in line with previous literature discussing the relevance of assessing a set of cognitive domains in identifying diverse disease courses (92). While memory impairment is a distinct feature of AD, especially involving the episodic memory, the associated disease stage is debated. Literature is present supporting the concept that decline in episodic memory is more pronounced later in the disease continuum (69, 94). On the contrary, other studies point to the presence of episodic memory impairment in the early disease stages (95, 96). Our results may provide more insight into the above-mentioned discrepancy in the literature: we found that memory is already heavily impaired in the early stage of the disease because its normalized score (0.78) was the third most affected subscore after verbal fluency (0.64) and attention (0.77). However, the ensuing decline in the 2-3 years following the diagnosis was inconspicuous, as the memory subscores are not significantly different between Group1 and Group 2. These results question the rasion d'être of the consecutive use of memory tests for monitoring disease progression and cognitive impairment. Nevertheless, 4 years after the disease onset, memory functions deteriorate severely again. This reinforces the findings of de Boer et al. that the deterioration of memory is also apparent further on in the AD continuum (69). This finding draws attention to the variable contribution of different cognitive domains to the constant deterioration of global cognition: the significance of loss of episodic memory function decreases whereas the contribution of the other cognitive domains enlarges. These results point to the importance of assessing various cognitive domains for disease monitoring and evaluating the effectiveness of disease modifying interventions as opposed to the sole assessment of memory.

Our results showed that verbal fluency was the most severely affected cognitive domain in the early phase of the disease, even more so than memory (normalized score of 0.64 vs 0.78, respectively). Literature suggests that the impairment of verbal fluency appears as early as

the preclinical phase of the disease (97) and is also noticeable in the subsequent phase in MCI, even in its amnestic type (98). However, there is a debate in the literature concerning which category of verbal fluency is affected more. Some studies emphasize the pre-eminence of testing letter/phonemic fluency (98), while other studies accentuate the assessment of semantic/category fluency (99-101). Further studies are needed to clarify this question so that the most appropriate types of verbal fluency tests may be used in dementia screening. Our results underline the relevance of verbal fluency tests in the evaluation of early-stage cognitive impairment and might contribute to the advancement of innovative diagnostic methods. Not only memory and verbal fluency, but all six subscores of the ACE test demonstrated a significant negative correlation with disease duration. Of special note is the correlation between disease duration and the visuospatial subscore, which demonstrated a notably robust negative value, the highest among the six subscores ($r= -0.85$). The visuospatial subscore of the ACE test is composed of three tasks: 1) copying two overlapping pentagons, 2) copying a cube, 3) drawing a clock face with the 12 numbers indicated and the placement of the hands indicating a given time. Our results point to the possible advantage of visuospatial function assessment in the follow-up of patients to appraise whether the cognitive deficit is progressing, and if so, at what rate it is advancing.

The importance of the impairment of visuospatial function was reinforced by our study of 78 individuals, 32 patients with multiple domain a-MCI and 46 healthy control participants. The characterization of the a-MCI group is described in our study in detail (70). In this study, we completed a thorough statistical analysis of different neuropsychological test results, including ACE. We calculated intergroup comparisons for ACE subscores which showed interesting results: even though the MCI patients belonged to the multiple domain amnestic subtype reinforced by tests such as MMSE and TMT, the visuospatial subscore was the only subscore that was significantly different between the two groups (F-value=8.32, corrected $p<0.001$). Even though the MCI group was significantly older, significant modifying effect of age and sex was ruled out by ANCOVA analysis ($p>0.05$). It is important to emphasize that, contrary to our previous study involving AD patients, this study was composed of MCI patients, an earlier phase on the disease continuum, yet the impairment of visuospatial function was already present in this patient group. While the literature is more extensive on

the evaluation of visuospatial function in AD, the studies focusing on visuospatial skills in MCI imply interesting findings: a study by Mapstone et al. found defective visual motion perception in one-third of MCI patients. The extent of deficit was linked to impairment in spatial navigation assessed by the Money Road Map test. However, no association was found between the deficit in visual motion perception and memory, neither visual nor verbal.(102) Furthermore, it was found that impaired visuospatial function could signal the initial manifestation of neurodegeneration (103).

The limitations of the study are 1) the lack of cerebrospinal fluid analysis, PET acquisition and genetic testing. 2) Exact disease duration may differ among participants, as cognitive impairment could start years before AD diagnosis is established. Based on the description of relatives and caregivers, we only included AD patients with a brief history of cognitive impairment preceding the patients' AD diagnosis. Nonetheless, family members' and caregivers' reports might not be precise due to various reasons. On the contrary, we applied a comprehensive diagnostic approach combined with meticulous participant selection, which give the strength of this study.

In this study of 78 participants, we also assiduously analysed the structural integration and functional connectivity of the visuospatial network using structural and functional MRI data. Regarding structural MRI data, we found slightly reduced cortical thickness in numerous cortical areas in the a-MCI group. We corrected for multiple comparisons, and this way, only two cortical regions remained significantly different: the right superior temporal gyrus and the left temporal pole ($p<0.05$). We also analysed the modifying effect of age and sex, as even healthy aging is associated with reduced cortical thickness. Sex did not have a significant modifying effect ($p>0.05$). However, decreased cortical thickness of the frontal and temporal cortices in both the left and right hemispheres was linked to older age ($p<0.05$). Howbeit, we found no significant modifying effect of age once Benjamini-Hochberg correction was applied ($p>0.05$). Our findings regarding the two significantly different cortical regions are in accordance with the literature: a study by Choi et al. analysed the Alzheimer's Disease Neuroimaging Initiative (ADNI) database and found that impaired visuospatial function not only signals a-MCI appropriately but also its conversion to

Alzheimer's disease (104). They also found that it's primarily the reduced cortical thickness of the temporal lobe that marks a-MCI patients. Other studies likewise identified the distinguished role of the lateral and medial temporal cortices in the identification of a-MCI (62, 105, 106). Furthermore, the pre-eminence of the lateral temporal lobe has been proposed in the literature (107). Even though the temporal pole and the superior temporal gyrus are key components of the visuospatial network, there are several more regions that are essential (e.g., caudal part of the middle frontal gyrus, insula, precentral gyrus, angular gyrus, supramarginal gyrus, posterior part of the superior parietal lobule, calcarine cortex, inferior temporal gyrus and occipitotemporal gyrus) (72), yet their cortical thickness was not significantly reduced in the a-MCI group in our study. These findings hardly explain the outstanding differences in visuospatial function between healthy controls and a-MCI patients, and they point to the importance of functional analysis. Notably, when analysing the ADNI 2 database, the most extensive neuroimaging repository, Choi et al. found that impaired visuospatial functioning was primarily linked to decreased cortical thickness of the superior temporal gyrus (104). This result fortifies our findings and also signifies the relevance of the analysis of functional MRI data.

All 78 participants in this study had resting-state functional MRI recordings available. In this study, we specifically analysed the resting state functional connectivity of the visuospatial network, which shed light on increased functional connectivity of the left frontotemporal network in the a-MCI group. Simultaneously, functional connectivity was decreased between the right temporal regions and the left frontal and temporal regions. Therefore, increased connectivity and decreased connectivity are present concurrently, but involve different regions of the brain. This contrasts with the results of Bonanni et al., who propose that these two phenomena are consecutive (108). However, our study solely analysed the visuospatial network, so whole brain connectivity cannot be judged based on our results. Differences in connectivity in the a-MCI group could indicate the altered functioning of the visuospatial network: the lengthy commissural connections of the visuospatial network's organizers in the subdominant/right hemisphere are lost, and associative short connections of the left hemisphere become preeminent as a compensatory mechanism for functional impairment. Our findings are in accordance with the literature pointing to the decreased commissural

connections in early MCI detected by diffusion tensor imaging (109, 110). The uniqueness of our study lies in the combination of the a-MCI patient population and the resting-state functional MRI method. At the time of our study, plenty of studies identified disturbed visuospatial function in AD (111-113), but there were merely 3 studies evaluating the visuospatial network in MCI (114-116). Furthermore, these studies used task-based paradigms, and no study analysed the functional connectivity of the visuospatial network on resting-state functional MRI data. The three studies using task-based fMRI protocols found that activation heightened in the left frontal regions in the group of MCI patients. Due to the proposed preeminent role of the right hemisphere in visuospatial function (84, 85), these three studies suggested a compensatory mechanism for the functional deficit in MCI behind the decreased right hemispheric activity and increased engagement of the left hemisphere at the time of the visuospatial paradigm (114-116). In accordance with the conclusion of these studies, our proposal also lies within a compensatory mechanism. Our study added novelty to the literature by introducing distinct changes of the visuospatial network in MCI with resting-state MRI, which might serve as a possible new biomarker for the early detection of cognitive impairment. However, more studies are needed to verify our results and to assess whether they can be used for the detection of the earliest phases of cognitive decline: people with subjective memory complaints or those at high risk of dementia.

It is important to emphasize that this study was directed at multiple domain a-MCI patients that form a high dementia risk but heterogeneous patient group. This subtype of MCI has the highest conversion rate to dementia of any kind (117). Furthermore, AD cases are not fully homogenous either: 25% of the cases present as atypical. In these cases, the cognitive symptoms as well as the location of neurodegeneration are dissimilar to typical AD. The corticobasal variant, the behavioural variant, the logopenic primary progressive aphasia, and the posterior cortical atrophy constitute the atypical types of AD (118). It might be argued that the atypical forms of AD could influence our results due to the lack of a final outcome for the patients. However, posterior cortical atrophy is of special interest, as the visuospatial functions are greatly affected early in the disease continuum. Therefore, the association between visuospatial impairment in the prodromal phase and the risk of conversion to dementia needs to be studied further with regular follow-ups.

The limitations of our study are 1) MCI patients' cognitive impairment belongs to the multiple domain type, which might be relatively heterogenous. Nevertheless, multiple domain MCI patients have the highest risk of conversion to dementia. 2) The site was not included as a confounding factor in the statistical analysis, thus, site-specific factors could influence our results. 3) Within-group sample sizes are not large, which could affect the reliability of between-group differences in neuroimaging data. 4) The MCI and healthy control groups significantly differed in age and sex. Although we used corrections and analysed the effect of sex and age separately as well, our understanding of the results might still be altered by the differences. In addition, age and sex are correlated with the group factor, thus, separating their effect in the model might not be fully possible. Though the two study groups differed significantly in the visuospatial subscore of ACE, we do not have a more detailed characterisation of visuospatial functions of the study groups. On the contrary, the strengths of our study include the rigorous patient selection, the involvement of two centres, and the meticulous statistical analysis inclusive of reporting effect sizes and multiple comparisons corrections.

In the study of 68 participants composed of 46 healthy controls and 22 patients with multiple domain a-MCI, we introduced the pilot phase of the Precognize paradigm, a custom-made digital diagnostic system. In this study, we focused on the applicability of this system in distinguishing MCI patients from cognitively healthy elderly people. The Precognize system assesses visuospatial and visuomotor functions based on a fine movement task and is automated; thus, it is designed for independent use. It is important to emphasize that the MCI patients in our study belonged to the multiple domain amnestic subtype based on the results of the neuropsychological evaluation. Furthermore, neuroimaging also confirmed the neurodegenerative background of our MCI patients: the patients' total grey matter volume and thickness of the entorhinal cortex were significantly reduced. Due to our extensive exclusion criteria, cognitive impairment due to reversible causes has been discarded in this study. To control for the impact of selection bias, further studies are needed on a representative sample of the whole MCI patient group, not only those with likely neurodegenerative aetiology.

In this study, we asked right-handed daily internet user elderly individuals to complete the paradigm, clicking on Arabic numerals from 1 to 9 in ascending order using a regular computer mouse, with both their right and left hands. We obtained six different movement parameters: distance, duration, velocity, speed, and entropy of computer mouse movements and the number of clicks or tries. The intergroup comparisons of these parameters between the a-MCI and healthy control group showed interesting results: the duration of task solving was significantly longer in the MCI group coupled with higher levels of movement entropy. Of the two hands, the subdominant left hand seemed superior in distinguishing the two groups based on greater effect size. The effect sizes were rather large for movement parameters in general (>0.8), which point to the powerful discriminative capacity of the movement characteristics. Anxiety level, sex, and age did not have a significant modifying effect on movement parameters.

To assess the interrelation between cognition and movement characteristics, we applied correlation analysis between movement parameters and neuropsychological test results. Reduced movement entropy and shorter task execution time were associated with better memory performance (RAVLT scores) as well as better global cognitive functioning (MMSE and ACE scores). On the basis of the r values, these interrelations were mild or moderate. Of particular attention is the ACE total score, which resulted in moderate r values throughout. The importance of the latter is that the ACE test is commonly used in the diagnostic workup of cognitive impairment; thus, its steady association with the movement parameters reinforces the possible diagnostic potential of our digital tool. Furthermore, we found higher levels of movement entropy and task completion time in participants who had decreased flexibility and speed of cognitive processing (larger scores on the TMT test) as well as poorer daily functioning indicated by a higher score on the CDR scale. These interrelations were moderate. The correlation analysis also pointed to the supremacy of the left-hand data with greater r values. Importantly, mood and anxiety measures (BDI and STAI tests, respectively) did not show significant correlations with movement parameters. In consonance with our findings concerning longer task completion time, psychomotor slowing and reduced speed of motor function are described in the literature (119).

In this study, movement entropy appeared to have the greatest potential in distinguishing between healthy individuals and those with cognitive impairment based on the intergroup comparisons' effect sizes. MCI patients had increased levels of entropy in their computer mouse movements, which means their movements were more unruly, diverting from the anticipated path (120). Despite that, in the field of neurocognitive research, Shannon's entropy of movements is scarcely studied (121). On the other hand, different fields of research have focused on the application of entropy analysis in eye movements, rapid aimed movements, and polyrhythmic hand movements (122-124). Their results propose that increased entropy levels signal disrupted movement coordination. Furthermore, other reports suggested that patients with Parkinson's disease and essential tremor could be distinguished from healthy individuals based on entropy measurements of pentagon copying and Archimedes spiral drawing tasks (125, 126). However, movement entropy analysis appears to be less commonly used in the AD continuum. Nonetheless, two studies have shown that MCI patients differ from healthy controls in entropy measures of gross motor function (127, 128). Taken all these into account, the analysis of fine movement entropy seems a promising novel frontier in the field of MCI diagnostics.

Several electronic devices are being developed with various cognitive tasks for identifying MCI patients (129). However, the use of conventional cognitive tests with electronic diagnostic systems poses some challenges: certain pre-evaluation conditions, such as higher levels of anxiety, mental tiredness impact the outcome of the tests and the administration takes a considerable time, on average 30 minutes per person (129). However, visuomotor paradigms have been used successfully for identifying MCI patients with motion sensors and eye-trackers (130, 131). Another study applied a computerized visuomotor paradigm and found that AD patients needed longer time to respond than healthy controls (52). Furthermore, research on kinematic handwriting analysis of patients with cognitive impairment has also been published (132, 133). However, a substantial modifying effect of age and anxiety has previously been described on the movement features of handwriting (134, 135). To summarize, the above-described studies of electronic screening tools for cognitive impairment share some common limitations: these tools can be costly, age and anxiety can have a strong modifying effect, and the arrangement of the diagnostic apparatus

DOI:10.14753/SE.2024.3035

may be too difficult to handle for some people. By comparison, the acquisition time with our tool is less than 5 minutes; the applied hardware and software are easy to handle and inexpensive. Furthermore, age, sex, mood, and anxiety did not have a modifying effect on movement parameters measured by our diagnostic tool.

Similarly to other protocols analysing movement data in the MCI diagnostic process, the limitations of this study are the lack of validation on separate datasets and the small size of the sample. To conquer these problems, we plan to run an extended acquisition protocol, including a diverse subset of the population.

DOI:10.14753/SE.2024.3035

VI. Conclusions

1. Rigorous neuropsychological assessment contributes to early detection of cognitive decline.
2. Verbal fluency seems to be the most affected cognitive domain in early AD; thus, its evaluation could have a central role in early diagnosis of AD.
3. The decline of visuospatial abilities follows a linear trajectory over the AD continuum; thus, VS abilities could have a possible role in tracking the advancement of cognitive impairment.
4. Due to the linear trajectory of VS abilities' decline, they could have a role in validating drug trials.
5. VS abilities were the most impaired cognitive domain in our cohort of multiple domain a-MCI patients.
6. We demonstrated—after rigorous statistical correction—that reduced cortical thickness of the superior temporal gyrus and temporal pole are distinctive structural features of a-MCI.
7. The fMRI-based analysis of the visuospatial network pointed to reduced functional connectivity between the left and right frontal areas, whereas functional connectivity was increased between the left frontotemporal regions.
8. Simultaneously increased and decreased levels of functional connectivity of different brain regions might represent a compensatory mechanism of the VS network.
9. With the possibility of automatized data analysis, alteration of VS abilities detectable by resting state fMRI could be a sensitive and non-invasive early biomarker for cognitive impairment with low need of man-hour.
10. Fine motor control is impaired early in MCI.
11. Characterizing fine movement entropy could be applied for the early detection of cognitive impairment.
12. Considering the opportunity for self-administration, automatization and the potential use of artificial intelligence, fine movement analysis could serve as a population-wide cognitive screening tool for older adults.

DOI:10.14753/SE.2024.3035

VII. Summary

Alzheimer's disease (AD) is the principal cause of major neurodegenerative disorders in older adults, and it poses a tremendous socioeconomic burden on society. To this day, AD is incurable, treatment is aimed at slowing the progression of cognitive decline. The sooner the treatment is initiated the better; however, adequate screening methods for the disease are yet to be developed. Previous studies pointed to the impairment of visuospatial and visuomotor functions in AD, which could have screening potential contributing to timely diagnosis. We examined 110 AD patients with different disease duration and 45 healthy controls (HC) with an extensive neuropsychological test battery and found that visuospatial subscore of the Addenbrooke's Cognitive Examination (ACE) was impaired already in the early phase of the disease. Furthermore, it showed a linear trajectory of decline over the disease course, which pointed to its potential in the diagnosis and follow-up of patients. Since intervention has better outcomes if started already in mild cognitive impairment (MCI), the prodromal phase of AD, we investigated 46 HC and 32 multiple domain amnestic MCI patients with detailed neuropsychological examination and structural and functional MRI acquisition. We found that the visuospatial abilities were the most impaired subscore of the ACE test. We also found reduced cortical thickness of the temporal pole and superior temporal gyrus. Furthermore, upon analysing the visuospatial network with functional MRI, we found decreased functional connectivity between left and right frontal areas, whereas functional connectivity was increased between left frontotemporal regions. To assess the diagnostic potential of visuospatial and visuomotor functions, we applied the Trail-making test based computerized Precognize paradigm on 22 MCI patients and 46 HCs. Upon analysing the computer mouse movement characteristics of the two groups derived from Precognize, we found impaired fine motor control in the MCI group. From the movement characteristics, entropy of hand movements showed the highest discriminative potential between cognitively healthy older adults and MCI patients. Based on our results, visuospatial and visuomotor abilities are impaired already in the MCI stage of the AD continuum. The Precognize paradigm pointed to the promising potential of fine movement analysis in the development of population-wide automated screening tool for cognitive impairment in older adults.

VIII. References

1. Rasmussen J, Langerman H. Alzheimer's Disease - Why We Need Early Diagnosis. Degener Neurol Neuromuscul Dis. 2019;9:123-130.

2. Brookmeyer R, Johnson E, Ziegler-Graham K, Arrighi HM. Forecasting the global burden of Alzheimer's disease. Alzheimers Dement. 2007;3(3):186-191.

3. Prince M, Ali GC, Guerchet M, Prina AM, Albanese E, Wu YT. Recent global trends in the prevalence and incidence of dementia, and survival with dementia. Alzheimers Res Ther. 2016;8(1):23.

4. Varatharajah Y, Ramanan VK, Iyer R, Vemuri P, Weiner MW, Aisen P, Petersen R, Jack CR, Saykin AJ, Jagust W, Trojanowki JQ, Toga AW, Beckett L, Green RC, Morris J, Shaw LM, Khachaturian Z, Sorensen G, Carrillo M, Kuller L, Raichle M, Paul S, Davies P, Fillit H, Hefti F, Holtzman D, Mesulam MM, Potter W, Snyder P, Schwartz A, Montine T, Thomas RG, Donohue M, Walter S, Gessert D, Sather T, Jiminez G, Balasubramanian AB, Mason J, Sim I, Harvey D, Bernstein M, Fox N, Thompson P, Schuff N, DeCarli C, Borowski B, Gunter J, Senjem M, Jones D, Kantarci K, Ward C, Koeppe RA, Foster N, Reiman EM, Chen K, Mathis C, Landau S, Cairns NJ, Franklin E, Taylor-Reinwald L, Lee V, Korecka M, Figurski M, Crawford K, Neu S, Foroud TM, Potkin S, Faber K, Kim S, Nho K, Thal L, Buckholtz N, Albert M, Frank R, Hsiao J, Kaye J, Quinn J, Silbert L, Lind B, Carter R, Dolen S, Schneider LS, Pawluczyk S, Beccera M, Teodoro L, Spann BM, Brewer J, Vanderswag H, Fleisher A, Heidebrink JL, Lord JL, Mason SS, Albers CS, Knopman D, Johnson K, Doody RS, Villanueva-Meyer J, Pavlik V, Shibley V, Chowdhury M, Rountree S, Dang M, Stern Y, Honig LS, Bell KL, Ances B, Carroll M, Creech ML, Franklin E, Mintun MA, Schneider S, Oliver A, Marson D, Geldmacher D, Love MN, Griffith R, Clark D, Brockington J, Roberson E, Grossman H, Mitsis E, Shah RC, deToledo-Morrell L, Duara R, Greig-Custo MT, Barker W, Onyike C, D'Agostino D, Kielb S, Sadowski M, Sheikh MO, Ulysse A, Gaikwad M, Doraiswamy PM, Petrella JR, Borges-Neto S, Wong TZ, Coleman E, Arnold SE, Karlawish JH, Wolk DA, Clark CM, Smith CD, Jicha G, Hardy P, Sinha P, Oates E, Conrad G, Lopez OL, Oakley MA, Simpson DM, Porsteinsson AP, Goldstein BS, Martin K, Makino KM, Ismail MS, Brand C, Preda A, Nguyen D, Womack K, Mathews D, Quiceno

M, Levey AI, Lah JJ, Cellar JS, Burns JM, Swerdlow RH, Brooks WM, Apostolova L, Tingus K, Woo E, Silverman DHS, Lu PH, Bartzokis G, Graff-Radford NR, Parfitt F, Poki-Walker K, Farlow MR, Hake AM, Matthews BR, Brosch JR, Herring S, van Dyck CH, Carson RE, MacAvoy MG, Varma P, Chertkow H, Bergman H, Hosein C, Black S, Stefanovic B, Caldwell C, Hsiung G-YR, Mudge B, Sossi V, Feldman H, Assaly M, Finger E, Pasternack S, Rachisky I, Rogers J, Trost D, Kertesz A, Bernick C, Munic D, Rogalski E, Lipowski K, Weintraub S, Bonakdarpour B, Kerwin D, Wu C-K, Johnson N, Sadowsky C, Villena T, Turner RS, Johnson K, Reynolds B, Sperling RA, Johnson KA, Marshall G, Yesavage J, Taylor JL, Lane B, Rosen A, Tinklenberg J, Sabbagh MN, Belden CM, Jacobson SA, Sirrel SA, Kowall N, Killiany R, Budson AE, Norbash A, Johnson PL, Obisesan TO, Wolday S, Allard J, Lerner A, Ogrocki P, Tatsuoka C, Fatica P, Fletcher E, Maillard P, Olichney J, DeCarli C, Carmichael O, Kittur S, Borrie M, Lee TY, Bartha R, Johnson S, Asthana S, Carlsson CM, Tariot P, Burke A, Milliken AM, Trncic N, Fleisher A, Reeder S, Bates V, Capote H, Rainka M, Scharre DW, Kataki M, Kelly B, Zimmerman EA, Celmins D, Brown AD, Pearlson GD, Blank K, Anderson K, Flashman LA, Seltzer M, Hynes ML, Santulli RB, Sink KM, Gordineer L, Williamson JD, Garg P, Watkins F, Ott BR, Tremont G, Daiello LA, Salloway S, Malloy P, Correia S, Rosen HJ, Miller BL, Perry D, Mintzer J, Spicer K, Bachman D, Pomara N, Hernando R, Sarrael A, Schultz SK, Smith KE, Koleva H, For the Alzheimer’s Disease Neuroimaging I. Predicting Short-term MCI-to-AD Progression Using Imaging, CSF, Genetic Factors, Cognitive Resilience, and Demographics. Sci Rep. 2019;9(1):2235.

5. Wilson RS, Leurgans SE, Boyle PA, Bennett DA. Cognitive decline in prodromal Alzheimer disease and mild cognitive impairment. Arch Neurol. 2011;68(3):351-356.

6. Sperling RA, Aisen PS, Beckett LA, Bennett DA, Craft S, Fagan AM, Iwatsubo T, Jack CR, Jr., Kaye J, Montine TJ, Park DC, Reiman EM, Rowe CC, Siemers E, Stern Y, Yaffe K, Carrillo MC, Thies B, Morrison-Bogorad M, Wagster MV, Phelps CH. Toward defining the preclinical stages of Alzheimer's disease: recommendations from the National Institute on Aging-Alzheimer's Association workgroups on diagnostic guidelines for Alzheimer's disease. Alzheimers Dement. 2011;7(3):280-292.

7. Maurer K, Volk S, Gerbaldo H. Auguste D and Alzheimer's disease. Lancet. 1997;349(9064):1546-1549.

8. Alzheimer A. Uber eigenartige Erkrankung der Hirnrinde. All Z Psychiatr. 1907;64:146-148.

9. Cipriani G, Dolciotti C, Picchi L, Bonuccelli U. Alzheimer and his disease: a brief history. Neurol Sci. 2011;32(2):275-279.

10. Guo T, Zhang D, Zeng Y, Huang TY, Xu H, Zhao Y. Molecular and cellular mechanisms underlying the pathogenesis of Alzheimer's disease. Mol Neurodegeneration. 2020;15(1):40.

11. Braak H, Braak E. Neuropathological stageing of Alzheimer-related changes. Acta Neuropathol. 1991;82(4):239-259.

12. Reitz C, Brayne C, Mayeux R. Epidemiology of Alzheimer disease. Nat Rev Neurol. 2011;7(3):137-152.

13. Livingston G, Sommerlad A, Orgeta V, Costafreda SG, Huntley J, Ames D, Ballard C, Banerjee S, Burns A, Cohen-Mansfield J, Cooper C, Fox N, Gitlin LN, Howard R, Kales HC, Larson EB, Ritchie K, Rockwood K, Sampson EL, Samus Q, Schneider LS, Selbæk G, Teri L, Mukadam N. Dementia prevention, intervention, and care. Lancet. 2017;390(10113):2673-2734.

14. Kivipelto M, Solomon A, Ahtiluoto S, Ngandu T, Lehtisalo J, Antikainen R, Bäckman L, Hänninen T, Jula A, Laatikainen T, Lindström J, Mangialasche F, Nissinen A, Paajanen T, Pajala S, Peltonen M, Rauramaa R, Stigsdotter-Neely A, Strandberg T, Tuomilehto J, Soininen H. The Finnish Geriatric Intervention Study to Prevent Cognitive Impairment and Disability (FINGER): study design and progress. Alzheimers Dement. 2013;9(6):657-665.

15. Ngandu T, Lehtisalo J, Solomon A, Levälahti E, Ahtiluoto S, Antikainen R, Bäckman L, Hänninen T, Jula A, Laatikainen T, Lindström J, Mangialasche F, Paajanen T, Pajala S, Peltonen M, Rauramaa R, Stigsdotter-Neely A, Strandberg T, Tuomilehto J, Soininen H, Kivipelto M. A 2 year multidomain intervention of diet, exercise, cognitive training, and vascular risk monitoring versus control to prevent cognitive decline in at-risk elderly people (FINGER): a randomised controlled trial. Lancet. 2015;385(9984):2255-2263.

16. Barnes DE, Yaffe K. The projected effect of risk factor reduction on Alzheimer's disease prevalence. Lancet Neurol. 2011;10(9):819-828.

17. Aisen PS, Cummings J, Jack CR, Morris JC, Sperling R, Frölich L, Jones RW, Dowsett SA, Matthews BR, Raskin J, Scheltens P, Dubois B. On the path to 2025: understanding the Alzheimer's disease continuum. Alzheimer's Res Ther. 2017;9(1):60.

18. Dubois B, Hampel H, Feldman HH, Scheltens P, Aisen P, Andrieu S, Bakardjian H, Benali H, Bertram L, Blennow K, Broich K, Cavedo E, Crutch S, Dartigues JF, Duyckaerts C, Epelbaum S, Frisoni GB, Gauthier S, Genthon R, Gouw AA, Habert MO, Holtzman DM, Kivipelto M, Lista S, Molinuevo JL, O'Bryant SE, Rabinovici GD, Rowe C, Salloway S, Schneider LS, Sperling R, Teichmann M, Carrillo MC, Cummings J, Jack CR, Jr. Preclinical Alzheimer's disease: Definition, natural history, and diagnostic criteria. Alzheimers Dement. 2016;12(3):292-323.

19. Petersen RC, Roberts RO, Knopman DS, Boeve BF, Geda YE, Ivnik RJ, Smith GE, Jack CR, Jr. Mild cognitive impairment: ten years later. Arch Neurol. 2009;66(12):1447-1455.

20. Calzà L, Beltrami D, Gagliardi G, Ghidoni E, Marcello N, Rossini-Favretti R, Tamburini F. Should we screen for cognitive decline and dementia? Maturitas. 2015;82(1):28-35.

21. Breijyeh Z, Karaman R. Comprehensive Review on Alzheimer's Disease: Causes and Treatment. Molecules. 2020;25(24):5789.

22. McKhann G, Drachman D, Folstein M, Katzman R, Price D, Stadlan EM. Clinical diagnosis of Alzheimer's disease: report of the NINCDS-ADRDA Work Group under the auspices of Department of Health and Human Services Task Force on Alzheimer's Disease. Neurology. 1984;34(7):939-944.

23. McKhann GM, Knopman DS, Chertkow H, Hyman BT, Jack CR, Jr., Kawas CH, Klunk WE, Koroshetz WJ, Manly JJ, Mayeux R, Mohs RC, Morris JC, Rossor MN, Scheltens P, Carrillo MC, Thies B, Weintraub S, Phelps CH. The diagnosis of dementia due to Alzheimer's disease: recommendations from the National Institute on Aging-Alzheimer's Association workgroups on diagnostic guidelines for Alzheimer's disease. Alzheimers Dement. 2011;7(3):263-269.

24. Weller J, Budson A. Current understanding of Alzheimer’s disease diagnosis and treatment. F1000Res. 2018;7:1161.

25. Constantinides VC, Paraskevas GP, Boufidou F, Bourbouli M, Pyrgelis E-S, Stefanis L, Kapaki E. CSF Aβ42 and Aβ42/Aβ40 Ratio in Alzheimer's Disease and Frontotemporal Dementias. Diagnostics (Basel, Switzerland) [Internet]. 2023 2023/02//; 13(4):[783 p.].

26. Dubois B, Villain N, Frisoni GB, Rabinovici GD, Sabbagh M, Cappa S, Bejanin A, Bombois S, Epelbaum S, Teichmann M, Habert MO, Nordberg A, Blennow K, Galasko D, Stern Y, Rowe CC, Salloway S, Schneider LS, Cummings JL, Feldman HH. Clinical diagnosis of Alzheimer's disease: recommendations of the International Working Group. Lancet Neurol. 2021;20(6):484-496.

27. Frisoni GB, Fox NC, Jack CR, Jr., Scheltens P, Thompson PM. The clinical use of structural MRI in Alzheimer disease. Nat Rev Neurol. 2010;6(2):67-77.

28. De Jager CA, Hogervorst E, Combrinck M, Budge MM. Sensitivity and specificity of neuropsychological tests for mild cognitive impairment, vascular cognitive impairment and Alzheimer's disease. Psychol Med. 2003;33(6):1039-1050.

29. Amjad H, Roth DL, Sheehan OC, Lyketsos CG, Wolff JL, Samus QM. Underdiagnosis of Dementia: an Observational Study of Patterns in Diagnosis and Awareness in US Older Adults. J Gen Intern Med. 2018;33(7):1131-1138.

30. Livingston G, Huntley J, Sommerlad A, Ames D, Ballard C, Banerjee S, Brayne C, Burns A, Cohen-Mansfield J, Cooper C, Costafreda SG, Dias A, Fox N, Gitlin LN, Howard R, Kales HC, Kivimäki M, Larson EB, Ogunniyi A, Orgeta V, Ritchie K, Rockwood K, Sampson EL, Samus Q, Schneider LS, Selbæk G, Teri L, Mukadam N. Dementia prevention, intervention, and care: 2020 report of the Lancet Commission. Lancet. 2020;396(10248):413-446.

31. Albert MS, DeKosky ST, Dickson D, Dubois B, Feldman HH, Fox NC, Gamst A, Holtzman DM, Jagust WJ, Petersen RC, Snyder PJ, Carrillo MC, Thies B, Phelps CH. The diagnosis of mild cognitive impairment due to Alzheimer's disease: recommendations from the National Institute on Aging-Alzheimer's Association workgroups on diagnostic guidelines for Alzheimer's disease. Alzheimers Dement. 2011;7(3):270-279.

32. Meskó B, Drobni Z, Bényei É, Gergely B, Győrffy Z. Digital health is a cultural transformation of traditional healthcare. MHealth. 2017;3:38.

33. Maruff P, Lim YY, Darby D, Ellis KA, Pietrzak RH, Snyder PJ, Bush AI, Szoeke C, Schembri A, Ames D, Masters CL. Clinical utility of the cogstate brief battery in identifying cognitive impairment in mild cognitive impairment and Alzheimer's disease. BMC Psychol. 2013;1(1):30.

34. Berg JL, Durant J, Léger GC, Cummings JL, Nasreddine Z, Miller JB. Comparing the Electronic and Standard Versions of the Montreal Cognitive Assessment in an Outpatient Memory Disorders Clinic: A Validation Study. J Alzheimers Dis. 2018;62(1):93-97.

35. Wild K, Howieson D, Webbe F, Seelye A, Kaye J. Status of computerized cognitive testing in aging: a systematic review. Alzheimers Dement. 2008;4(6):428-437.

36. Ramsey AT, Wetherell JL, Depp C, Dixon D, Lenze E. Feasibility and Acceptability of Smartphone Assessment in Older Adults with Cognitive and Emotional Difficulties. J Technol Hum Serv. 2016;34(2):209-223.

37. Yiannopoulou KG, Papageorgiou SG. Current and Future Treatments in Alzheimer Disease: An Update. J Cent Nerv Syst Dis. 2020;12:1179573520907397.

38. Hoy SM. Lecanemab: First Approval. Drugs. 2023;83(4):359-365.

39. Shi M, Chu F, Zhu F, Zhu J. Impact of Anti-amyloid-β Monoclonal Antibodies on the Pathology and Clinical Profile of Alzheimer's Disease: A Focus on Aducanumab and Lecanemab. Front Aging Neurosci. 2022;14:870517.

40. Salimi S, Irish M, Foxe D, Hodges JR, Piguet O, Burrell JR. Can visuospatial measures improve the diagnosis of Alzheimer's disease? Alzheimers Dement (Amst). 2018;10:66-74.

41. Muffato V, Meneghetti C, De Beni R. Not all is lost in older adults' route learning: The role of visuo-spatial abilities and type of task. J Environ Psychol. 2016;47:230-241.

42. Quental NB, Brucki SM, Bueno OF. Visuospatial function in early Alzheimer's disease--the use of the Visual Object and Space Perception (VOSP) battery. PLoS One. 2013;8(7):e68398.

43. Kravitz D, Saleem K, Baker C, Mishkin M. A new neural network for visuospatial processing. Nat Rev Neurosci. 2011;12:217-230.

44. Trés ES, Brucki SMD. Visuospatial processing: A review from basic to current concepts. Dement Neuropsychol. 2014;8(2):175-181.

45. Beretta L, Caminiti SP, Santangelo R, Magnani G, Ferrari-Pellegrini F, Caffarra P, Perani D. Two distinct pathological substrates associated with MMSE-pentagons item deficit in DLB and AD. Neuropsychologia. 2019;133:107174.

46. Thiyagesh SN, Farrow TFD, Parks RW, Accosta-Mesa H, Young C, Wilkinson ID, Hunter MD, Woodruff PWR. The neural basis of visuospatial perception in Alzheimer's disease and healthy elderly comparison subjects: An fMRI study. Psychiatry Res Neuroimaging. 2009;172(2):109-116.

47. Shin S, Crapse TB, Mayo JP, Sommer MA. Visuomotor Integration. In: Binder MD, Hirokawa N, Windhorst U, editors. Encyclopedia of Neuroscience. Berlin, Heidelberg: Springer Berlin Heidelberg; 2009. p. 4354-4359.

48. Hosseini SMH, Bruno JL, Baker JM, Gundran A, Harbott LK, Gerdes JC, Reiss AL. Neural, physiological, and behavioral correlates of visuomotor cognitive load. Sci Rep. 2017;7(1):8866.

49. Tippett WJ, Sergio LE. Visuomotor integration is impaired in early stage Alzheimer's disease. Brain Res. 2006;1102(1):92-102.

50. Ilardi CR, Chieffi S, Iachini T, Iavarone A. Neuropsychology of posteromedial parietal cortex and conversion factors from Mild Cognitive Impairment to Alzheimer’s disease: systematic search and state-of-the-art review. Aging Clin Exp Res. 2022;34(2):289-307.

51. Hawkins KM, Sergio LE. Visuomotor impairments in older adults at increased Alzheimer's disease risk. J Alzheimers Dis. 2014;42(2):607-621.

52. Mollica MA, Navarra J, Fernández-Prieto I, Olives J, Tort A, Valech N, Coll-Padros N, Molinuevo JL, Rami L. Subtle visuomotor difficulties in preclinical Alzheimer's disease. J Neuropsychol. 2017;11:56–73.

53. Stachó L, Dudás R, Ivády R, Kothencz G, Janka Z. Addenbrooke’s Kognitív Vizsgálat: a magyar változat kifejlesztése. Psychiatr Hung. 2003;18(4):226-240.

54. Dudas RB, Berrios GE, Hodges JR. The Addenbrooke's Cognitive Examination (ACE) in the Differential Diagnosis of Early Dementias Versus Affective Disorder. Am J Geriatr Psychiatry. 2005;13(3):218-226.

55. Strauss ESEMSSOSO. A compendium of neuropsychological tests : administration, norms, and commentary. Oxford; New York: Oxford University Press; 2006.166-168

56. Crawford S, Whitnall L, Robertson J, Evans JJ. A systematic review of the accuracy and clinical utility of the Addenbrooke's Cognitive Examination and the Addenbrooke's Cognitive Examination-Revised in the diagnosis of dementia. Int J Geriatr Psychiatry. 2012;27(7):659-669.

57. Mathuranath PS, Nestor PJ, Berrios GE, Rakowicz W, Hodges JR. A brief cognitive test battery to differentiate Alzheimer's disease and frontotemporal dementia. Neurology. 2000;55(11):1613-1620.

58. Kónya A, Verseghi A, Rey T. Rey- Emlékezeti vizsgálatok. Pszicho- Teszt Szerviz. 1995.

59. Petersen RC. Mild cognitive impairment as a diagnostic entity. J Intern Med. 2004;256(3):183-194.

60. Alladi S, Arnold R, Mitchell J, Nestor PJ, Hodges JR. Mild cognitive impairment: applicability of research criteria in a memory clinic and characterization of cognitive profile. Psychol Med. 2006;36(4):507-515.

61. Gomar JJ, Bobes-Bascaran MT, Conejero-Goldberg C, Davies P, Goldberg TE. Utility of combinations of biomarkers, cognitive markers, and risk factors to predict conversion from mild cognitive impairment to Alzheimer disease in patients in the Alzheimer's disease neuroimaging initiative. Arch Gen Psychiatry. 2011;68(9):961-969.

62. Csukly G, Sirály E, Fodor Z, Horváth A, Salacz P, Hidasi Z, Csibri É, Rudas G, Szabó Á. The Differentiation of Amnestic Type MCI from the Non-Amnestic Types by Structural MRI. Front Aging Neurosci. 2016;8:52.

63. Bowie CR, Harvey PD. Administration and interpretation of the Trail Making Test. Nat Protoc. 2006;1(5):2277-2281.

64. Kramer SI, Reifler BV. Depression, dementia, and reversible dementia. Clin Geriatr Med. 1992;8(2):289-297.

65. Seignourel PJ, Kunik ME, Snow L, Wilson N, Stanley M. Anxiety in dementia: a critical review. Clin Psychol Rev. 2008;28(7):1071-1082.

66. Sipos K, Sipos M. The development and validation of the Hungarian Form of the State-Trait Anxiety Inventory. Series in Clinical & Community Psychology: Stress & Anxiety. 1983;2:27-39.

67. Miklosi M, Martos T, Kocsis-Bogar K, Dora P. Psychometric properties of the Hungarian version of the Cognitive Emotion Regulation Questionnaire. Psychiatr Hung. 2011;26:102-111.

68. Lynch CA, Walsh C, Blanco A, Moran M, Coen RF, Walsh JB, Lawlor BA. The clinical dementia rating sum of box score in mild dementia. Dement Geriatr Cogn Disord. 2006;21(1):40-43.

69. de Boer C, Mattace-Raso F, van der Steen J, Pel JJ. Mini-Mental State Examination subscores indicate visuomotor deficits in Alzheimer's disease patients: A cross-sectional study in a Dutch population. Geriatr Gerontol Int. 2014;14(4):880-885.

70. Berente DB, Zsuffa J, Werber T, Kiss M, Drotos A, Kamondi A, Csukly G, Horvath AA. Alteration of Visuospatial System as an Early Marker of Cognitive Decline: A Double-Center Neuroimaging Study. Front Aging Neurosci. 2022;14:854368.

71. Whitfield-Gabrieli S, Nieto-Castanon A. Conn: a functional connectivity toolbox for correlated and anticorrelated brain networks. Brain Connect. 2012;2(3):125-141.

72. de Graaf TA, Roebroeck A, Goebel R, Sack AT. Brain network dynamics underlying visuospatial judgment: an FMRI connectivity study. J Cogn Neurosci. 2010;22(9):2012-2026.

73. Postle BR, Stern CE, Rosen BR, Corkin S. An fMRI investigation of cortical contributions to spatial and nonspatial visual working memory. Neuroimage. 2000;11(5 Pt 1):409-423.

74. Formisano E, Linden DE, Di Salle F, Trojano L, Esposito F, Sack AT, Grossi D, Zanella FE, Goebel R. Tracking the mind's image in the brain I: time-resolved fMRI during visuospatial mental imagery. Neuron. 2002;35(1):185-194.

75. Klingberg T, Forssberg H, Westerberg H. Increased brain activity in frontal and parietal cortex underlies the development of visuospatial working memory capacity during childhood. J Cogn Neurosci. 2002;14(1):1-10.

76. Culham JC, Cavina-Pratesi C, Singhal A. The role of parietal cortex in visuomotor control: what have we learned from neuroimaging? Neuropsychologia. 2006;44(13):2668-2684.

77. Scherf KS, Sweeney JA, Luna B. Brain basis of developmental change in visuospatial working memory. J Cogn Neurosci. 2006;18(7):1045-1058.

78. Trojano L, Conson M. Visuospatial and visuoconstructive deficits. Handb Clin Neurol. 2008;88:373-391.

79. Ghaem O, Mellet E, Crivello F, Tzourio N, Mazoyer B, Berthoz A, Denis M. Mental navigation along memorized routes activates the hippocampus, precuneus, and insula. Neuroreport. 1997;8(3):739-744.

80. Ricciardi E, Bonino D, Gentili C, Sani L, Pietrini P, Vecchi T. Neural correlates of spatial working memory in humans: a functional magnetic resonance imaging study comparing visual and tactile processes. Neuroscience. 2006;139(1):339-349.

81. Suchan B, Botko R, Gizewski E, Forsting M, Daum I. Neural substrates of manipulation in visuospatial working memory. Neuroscience. 2006;139(1):351-357.

82. Zimmer HD. Visual and spatial working memory: from boxes to networks. Neurosci Biobehav Rev. 2008;32(8):1373-1395.

83. Thakral PP, Slotnick SD. The role of parietal cortex during sustained visual spatial attention. Brain Res. 2009;1302:157-166.

84. Ng VW, Eslinger PJ, Williams SC, Brammer MJ, Bullmore ET, Andrew CM, Suckling J, Morris RG, Benton AL. Hemispheric preference in visuospatial processing: a complementary approach with fMRI and lesion studies. Hum Brain Mapp. 2000;10(2):80-86.

85. Corballis PM. Visuospatial processing and the right-hemisphere interpreter. Brain Cogn. 2003;53(2):171-176.

86. Sheline YI, Raichle ME. Resting state functional connectivity in preclinical Alzheimer's disease. Biol Psychiatry. 2013;74(5):340-347.

87. Worsley KJ, Marrett S, Neelin P, Vandal AC, Friston KJ, Evans AC. A unified statistical approach for determining significant signals in images of cerebral activation. Hum Brain Mapp. 1996;4(1):58-73.

88. Horvath AA, Berente DB, Vertes B, Farkas D, Csukly G, Werber T, Zsuffa JA, Kiss M, Kamondi A. Differentiation of patients with mild cognitive impairment and healthy controls based on computer assisted hand movement analysis: a proof-of-concept study. Sci Rep. 2022;12(1):19128.

89. Berente DB, Kamondi A, Horvath AA. The Assessment of Visuospatial Skills and Verbal Fluency in the Diagnosis of Alzheimer's Disease. Front Aging Neurosci. 2021;13:737104.

90. Wasserman V, Emrani S, Matusz EF, Peven J, Cleary S, Price CC, Ginsberg TB, Swenson R, Heilman KM, Lamar M, Libon DJ. Visuospatial performance in patients with statistically-defined mild cognitive impairment. J Clin Exp Neuropsychol. 2020;42(3):319-328.

91. Suh GH, Ju YS, Yeon BK, Shah A. A longitudinal study of Alzheimer's disease: rates of cognitive and functional decline. Int J Geriatr Psychiatry. 2004;19(9):817-824.

92. Wilkosz PA, Seltman HJ, Devlin B, Weamer EA, Lopez OL, DeKosky ST, Sweet RA. Trajectories of cognitive decline in Alzheimer's disease. Int Psychogeriatr. 2010;22(2):281-290.

93. Hodges JR, Larner AJ. Addenbrooke’s Cognitive Examinations: ACE, ACE-R, ACE-III, ACEapp, and M-ACE. In: Larner AJ, editor. Cognitive Screening Instruments: A Practical Approach. Cham: Springer International Publishing; 2017. p. 109-137.

94. Förstl H, Kurz A. Clinical features of Alzheimer’s disease. Eur Arch Psychiatry Clin Neurosci. 1999;249(6):288-290.

95. Baudic S, Barba GD, Thibaudet MC, Smagghe A, Remy P, Traykov L. Executive function deficits in early Alzheimer's disease and their relations with episodic memory. Arch Clin Neuropsychol. 2006;21(1):15-21.

96. Sperling RA, Dickerson BC, Pihlajamaki M, Vannini P, LaViolette PS, Vitolo OV, Hedden T, Becker JA, Rentz DM, Selkoe DJ, Johnson KA. Functional alterations in memory networks in early Alzheimer's disease. Neuromolecular Med. 2010;12(1):27-43.

97. Clark LJ, Gatz M, Zheng L, Chen YL, McCleary C, Mack WJ. Longitudinal verbal fluency in normal aging, preclinical, and prevalent Alzheimer's disease. Am J Alzheimers Dis Other Demen. 2009;24(6):461-468.

98. Murphy KJ, Rich JB, Troyer AK. Verbal fluency patterns in amnestic mild cognitive impairment are characteristic of Alzheimer's type dementia. J Int Neuropsychol Soc. 2006;12(4):570-574.

99. Monsch AU, Bondi MW, Butters N, Salmon DP, Katzman R, Thal LJ. Comparisons of verbal fluency tasks in the detection of dementia of the Alzheimer type. Arch Neurol. 1992;49(12):1253-1258.

100. Pasquier F, Lebert F, Grymonprez L, Petit H. Verbal fluency in dementia of frontal lobe type and dementia of Alzheimer type. J Neurol Neurosurg Psychiatry. 1995;58:81-84.

101. Gomez RG, White DA. Using verbal fluency to detect very mild dementia of the Alzheimer type. Arch Clin Neuropsychol. 2006;21(8):771-775.

102. Mapstone M, Steffenella TM, Duffy CJ. A visuospatial variant of mild cognitive impairment: getting lost between aging and AD. Neurology. 2003;60(5):802-808.

103. Iachini I, Iavarone A, Senese VP, Ruotolo F, Ruggiero G. Visuospatial memory in healthy elderly, AD and MCI: a review. Curr Aging Sci. 2009;2(1):43-59.

104. Choi SE, Mukherjee S, Gibbons LE, Sanders RE, Jones RN, Tommet D, Mez J, Trittschuh EH, Saykin A, Lamar M, Rabin L, Foldi NS, Sikkes S, Jutten RJ, Grandoit E, Mac Donald C, Risacher S, Groot C, Ossenkoppele R, Crane PK. Development and validation of language and visuospatial composite scores in ADNI. Alzheimers Dement (N Y). 2020;6(1):e12072.

105. Karas G, Sluimer J, Goekoop R, van der Flier W, Rombouts SA, Vrenken H, Scheltens P, Fox N, Barkhof F. Amnestic mild cognitive impairment: structural MR imaging findings predictive of conversion to Alzheimer disease. AJNR Am J Neuroradiol. 2008;29(5):944-949.

106. Xie Y, Cui Z, Zhang Z, Sun Y, Sheng C, Li K, Gong G, Han Y, Jia J. Identification of Amnestic Mild Cognitive Impairment Using Multi-Modal Brain Features: A Combined Structural MRI and Diffusion Tensor Imaging Study. J Alzheimers Dis. 2015;47(2):509-522.

107. Shen Q, Loewenstein DA, Potter E, Zhao W, Appel J, Greig MT, Raj A, Acevedo A, Schofield E, Barker W, Wu Y, Potter H, Duara R. Volumetric and visual rating of magnetic resonance imaging scans in the diagnosis of amnestic mild cognitive impairment and Alzheimer's disease. Alzheimers Dement. 2011;7(4):e101-108.

108. Bonanni L, Moretti D, Benussi A, Ferri L, Russo M, Carrarini C, Barbone F, Arnaldi D, Falasca NW, Koch G, Cagnin A, Nobili F, Babiloni C, Borroni B, Padovani A, Onofrj M, Franciotti R, group-SINDEM tFIs. Hyperconnectivity in Dementia Is Early and Focal and Wanes with Progression. Cerebral Cortex. 2020;31(1):97-105.

109. Zhuang L, Wen W, Zhu W, Trollor J, Kochan N, Crawford J, Reppermund S, Brodaty H, Sachdev P. White matter integrity in mild cognitive impairment: a tract-based spatial statistics study. Neuroimage. 2010;53(1):16-25.

110. Nir TM, Jahanshad N, Villalon-Reina JE, Toga AW, Jack CR, Weiner MW, Thompson PM. Effectiveness of regional DTI measures in distinguishing Alzheimer's disease, MCI, and normal aging. Neuroimage Clin. 2013;3:180-195.

111. Thulborn KR, Martin C, Voyvodic JT. Functional MR imaging using a visually guided saccade paradigm for comparing activation patterns in patients with probable Alzheimer's disease and in cognitively able elderly volunteers. AJNR Am J Neuroradiol. 2000;21(3):524-531.

112. Prvulovic D, Hubl D, Sack AT, Melillo L, Maurer K, Frölich L, Lanfermann H, Zanella FE, Goebel R, Linden DE, Dierks T. Functional imaging of visuospatial processing in Alzheimer's disease. Neuroimage. 2002;17(3):1403-1414.

113. Bokde ALW, Lopez-Bayo P, Born C, Ewers M, Meindl T, Teipel SJ, Faltraco F, Reiser MF, Möller H-J, Hampel H. Alzheimer Disease: Functional Abnormalities in the Dorsal Visual Pathway. Radiology. 2010;254(1):219-226.

114. Vannini P, Almkvist O, Dierks T, Lehmann C, Wahlund LO. Reduced neuronal efficacy in progressive mild cognitive impairment: a prospective fMRI study on visuospatial processing. Psychiatry Res. 2007;156(1):43-57.

115. Bokde AL, Lopez-Bayo P, Born C, Dong W, Meindl T, Leinsinger G, Teipel SJ, Faltraco F, Reiser M, Möller HJ, Hampel H. Functional abnormalities of the visual

processing system in subjects with mild cognitive impairment: an fMRI study. Psychiatry Res. 2008;163(3):248-259.

116. Alichniewicz K, Brunner F, Klünemann H, Greenlee M. Structural and functional neural correlates of visuospatial information processing in normal aging and amnestic mild cognitive impairment. Neurobiol Aging. 2012;33:2782-2797.

117. Petersen RC, Morris JC. Mild cognitive impairment as a clinical entity and treatment target. Arch Neurol. 2005;62(7):1160-1163; discussion 1167.

118. Frisoni GB, Altomare D, Thal DR, Ribaldi F, van der Kant R, Ossenkoppele R, Blennow K, Cummings J, van Duijn C, Nilsson PM, Dietrich PY, Scheltens P, Dubois B. The probabilistic model of Alzheimer disease: the amyloid hypothesis revised. Nat Rev Neurosci. 2022;23(1):53-66.

119. Chehrehnegar N, Nejati V, Shati M, Rashedi V, Lotfi M, Adelirad F, Foroughan M. Early detection of cognitive disturbances in mild cognitive impairment: a systematic review of observational studies. Psychogeriatrics. 2020;20(2):212-228.

120. Drotár P, Mekyska J, Rektorová I, Masarová L, Smékal Z, Faundez-Zanuy M. Decision support framework for Parkinson's disease based on novel handwriting markers. IEEE Trans Neural Syst Rehabil Eng. 2015;23(3):508-516.

121. Shannon CE. A Mathematical Theory of Communication. Bell Syst Tech J. 1948;27(3):379-423.

122. Engbert R, Scheffczyk C, Krampe RT, Rosenblum M, Kurths J, Kliegl R. Tempo-induced transitions in polyrhythmic hand movements. Phys Rev E 1997;56(5):5823-5833.

123. Soukoreff RW, Zhao J, Ren X. The entropy of a rapid aimed movement: Fitts' index of difficulty versus Shannon's entropy. Proceedings of the 13th IFIP TC 13 international conference on Human-computer interaction - Volume Part IV; Lisbon, Portugal: Springer-Verlag; 2011. p. 222–239.

124. Krejtz K, Szmidt T, Duchowski AT, Krejtz I. Entropy-based statistical analysis of eye movement transitions. Proceedings of the Symposium on Eye Tracking Research and Applications; Safety Harbor, Florida: Association for Computing Machinery; 2014. p. 159–166.

125. Lopez-de-Ipina K, Solé-Casals J, Faúndez-Zanuy M, Calvo PM, Sesa E, Roure J, Martinez-de-Lizarduy U, Beitia B, Fernández E, Iradi J, Garcia-Melero J, Bergareche A. Automatic Analysis of Archimedes' Spiral for Characterization of Genetic Essential Tremor Based on Shannon's Entropy and Fractal Dimension. Entropy (Basel). 2018;20(7):531.

126. Brabenec L, Klobusiakova P, Mekyska J, Rektorova I. Shannon entropy: A novel parameter for quantifying pentagon copying performance in non-demented Parkinson's disease patients. Parkinsonism Relat Disord. 2022;94:45-48.

127. Martín-Gonzalo J-A, Pulido-Valdeolivas I, Wang Y, Wang T, Chiclana-Actis G, Algarra-Lucas MdC, Palmí-Cortés I, Fernández Travieso J, Torrecillas-Narváez MD, Miralles-Martinez AA, Rausell E, Gómez-Andrés D, Zanin M. Permutation Entropy and Irreversibility in Gait Kinematic Time Series from Patients with Mild Cognitive Decline and Early Alzheimer's Dementia. Entropy. 2019;21(9):868.

128. Khan T, Jacobs PG. Prediction of Mild Cognitive Impairment Using Movement Complexity. IEEE J Biomed Health Inform. 2021;25(1):227-236.

129. Aslam RW, Bates V, Dundar Y, Hounsome J, Richardson M, Krishan A, Dickson R, Boland A, Fisher J, Robinson L, Sikdar S. A systematic review of the diagnostic accuracy of automated tests for cognitive impairment. Int J Geriatr Psychiatry. 2018;33(4):561-575.

130. Staal J, Mattace-Raso F, Daniels HAM, van der Steen J, Pel JJM. To Explore the Predictive Power of Visuomotor Network Dysfunctions in Mild Cognitive Impairment and Alzheimer's Disease. Front Neurosci. 2021;15:654003.

131. Lagun D, Manzanares C, Zola SM, Buffalo EA, Agichtein E. Detecting cognitive impairment by eye movement analysis using automatic classification algorithms. J Neurosci Methods. 2011;201(1):196-203.

132. Yan JH, Rountree S, Massman P, Doody RS, Li H. Alzheimer's disease and mild cognitive impairment deteriorate fine movement control. J Psychiatr Res. 2008;42(14):1203-1212.

133. Yu N-Y, Chang S-H. Characterization of the fine motor problems in patients with cognitive dysfunction – A computerized handwriting analysis. Hum Mov Sci. 2019;65:71-79.

134. Schröter A, Mergl R, Bürger K, Hampel H, Möller HJ, Hegerl U. Kinematic analysis of handwriting movements in patients with Alzheimer's disease, mild cognitive impairment, depression and healthy subjects. Dement Geriatr Cogn Disord. 2003;15(3):132-142.

135. Likforman-Sulem L, Esposito A, Faundez-Zanuy M, Clémençon S, Cordasco G. EMOTHAW: A Novel Database for Emotional State Recognition From Handwriting and Drawing. IEEE Trans Hum Mach Syst. 2017;47(2):273-284.

IX. Bibliography of the candidate's publications

IX.1. Publications related to the thesis:

1. Horvath AA, **Berente DB**, Vertes B, Farkas D, Csukly G, Werber T, Zsuffa JA, Kiss M, Kamondi A. Differentiation of patients with mild cognitive impairment and healthy controls based on computer assisted hand movement analysis: a proof-of-concept study. Sci Rep. 2022;12(1):19128. (**IF:4.6**)
2. **Berente DB**, Zsuffa J, Werber T, Kiss M, Drotos A, Kamondi A, Csukly G, Horvath AA. Alteration of Visuospatial System as an Early Marker of Cognitive Decline: A Double-Center Neuroimaging Study. Front Aging Neurosci. 2022;14:854368. (**IF:4.8**)
3. **Berente DB**, Kamondi A, Horvath AA. The Assessment of Visuospatial Skills and Verbal Fluency in the Diagnosis of Alzheimer's Disease. Front Aging Neurosci. 2021;13:737104. (**IF:4.8**)

Cumulated impact factor of publications related to the thesis: **14.2**

IX.2 Publications not related to the thesis:

1. Csukly G, Tombor L, Hidasi Z, Csibri E, Fullajtár M, Huszár Z, Koszovácz V, Lányi O, Vass E, Koleszár B, Kóbor I, Farkas K, Rosenfeld V, **Berente DB**, Bolla G, Kiss M, Kamondi A, Horvath AA. Low Functional network integrity in cognitively unimpaired and MCI subjects with depressive symptoms: results from a multi-center fMRI study. Translational Psychiatry. 2024;14(1):179. (**IF:6.8**)**
2. Zsuffa JA, Katz S, Koszovacz V, **Berente DB**, Kamondi A, Csukly G, Mangialasche F, Rocha ASL, Kivipelto M, Horvath AA. Lifestyle and behavioural changes in older adults during the Covid-19 pandemic are associated with subjective cognitive complaints. Sci Rep. 2024;14(1):2502. (**IF:4.6**)**

3. Bolla G, **Berente D**, Andrássy A, Zsuffa J, Hidasi Z, Csibri E, Csukly G, Kamondi A, Kiss M, Horváth A. Comparison of the diagnostic accuracy of resting-state fMRI driven machine learning algorithms in the detection of mild cognitive impairment. Sci Rep. 2023;13. (**IF:4.6*)**
4. Zsuffa J, Koszovácz V, **Berente D**, Bálint Z, Katz S, Kamondi A, Csukly G, Horváth A. A COVID-19-pandémia harmadik hullámának hatása a 60 év feletti magyar lakosság életmódjára, mentális és fizikai egészségére. Orvosi hetilap. 2022;163:1215-1223. (**IF:0.6)**

Cumulated impact factor of publications not related to the thesis: **16.6**

**,* Expected IF

DOI:10.14753/SE.2024.3035

X. Acknowledgements

I would like to express my gratitude to everyone who has contributed to this thesis.

I am profoundly grateful to my mentor, Dr. András Attila Horváth, for the unwavering support and guidance he has given over the years. His enthusiasm and dedication to research motivated me to pursue my PhD in neuroscience. He provides me with great opportunities to learn and participate in research projects for which I am immensely grateful.

I would like to express my deepest gratitude to Professor Dr. Anita Kamondi. Her guidance, kind support and extraordinary knowledge have encouraged and motivated me in every phase of my scientific research.

I wish to express my gratitude to Dr. Gábor Csukly for his support. His knowledge and approach to research has inspired me to explore new ideas.

I am grateful to Anita Andrássy, Máté Kiss, Gergő Levente Bolla, Tom Werber and Dr. János Zsuffa; their knowledge and help have been invaluable in the neuroimaging studies.

I am also grateful to Anna Csilla Kegyes-Brassai for her kindness and unwavering support from the beginning of our scientific journeys.

Thanks should also go to the members of the MTA-OMIII Lendület 'Momentum' Neurocognitive Research Center for providing a nourishing and inspiring atmosphere.

I would like to extend my sincere thanks to Professor Cornelis Jan Stam and Dr. Arjan Hillebrand for providing me the opportunity to complete my internship in Amsterdam. Their knowledge and guidance have been instrumental for the beginning of my journey in studying EEG connectivity.

I am also thankful to every participant of these studies without whom our research, the basis of this thesis, would not have been possible.

I'd like to acknowledge the members of the Neurology Department of the National Institute of Mental Health, Neurology and Neurosurgery for their support.

I could not have undertaken this journey without the support and encouragement of my friends and family, especially my husband, Márk Kerestély, for which I am forever grateful.

DOI:10.14753/SE.2024.3035

Appendix

Intergroup differences in cortical thickness of healthy control participants and amnestic MCI patients.

Note: HC: healthy control, a-MCI: amnestic mild cognitive impairment, a: mean followed by standard deviation, b: nominal p-value, c: reported in Cohen's d, *signals significantly different results after Benjamini-Hochberg correction ($p<0.05$).

Anatomical region	Left hemisphere (in mm)				Right hemisphere (in mm)			
	HC	a-MCI	p [b]	Effect size [c]	HC	a-MCI	p [b]	Effect size [c]
cortex around superior temporal sulcus [a]	2.26±0.2	2.19±0.2	0.51	0.396	2.36±0.2	2.43±0.4	0.212	0.230
caudal anterior cingulate gyrus [a]	2.61±0.3	2.63±0.3	0.64	0.074	2.5±0.2	2.42±0.3	0.004	0.353
caudal middle frontal gyrus [a]	2.43±0.1	2.28±0.3	0.003	0.664	2.42±0.2	2.38±0.2	0.017	0.241
cuneus [a]	1.75±0.1	1.81±0.2	0.566	0.345	1.81±0.1	1.8±0.1	0.190	0.077
entorhinal cortex [a]	3.35±0.3	3.15±0.5	0.003	0.515	3.52±0.4	3.36±0.4	0.002	0.385
fusiform gyrus [a]	2.66±0.1	2.52±0.2	0.004	0.830	2.7±0.1	2.59±0.2	0.006	0.667
inferior parietal lobule [a]	2.3±0.11	2.22±0.2	0.005	0.559	2.31±0.1	2.3±0.2	0.180	0.062
inferior temporal gyrus [a]	2.65±0.1	2.56±0.2	0.005	0.588	2.73±0.1	2.64±0.2	0.005	0.566
isthmus of cingulate gyrus [a]	2.21±0.2	2.15±0.2	0.052	0.333	2.23±0.2	2.17±0.2	0.003	0.291
lateral occipital cortex [a]	2.11±0.1	2.07±0.2	0.046	0.274	2.14±0.1	2.07±0.2	0.115	0.424

lateral orbitofrontal cortex [a]	2.54±0.1	2.48±0.2	0.002	0.339	2.57±0.2	2.49±0.2	0.012	0.483
lingual gyrus [a]	1.92±0.1	1.85±0.1	0.095	0.595	1.97±0.1	1.88±0.2	0.009	0.663
medial orbitofrontal cortex [a]	2.33±0.1	2.25±0.2	0.005	0.608	2.34±0.2	2.31±0.2	0.004	0.187
middle temporal gyrus [a]	2.66±0.1	2.55±0.2	0.08	0.609	2.73±0.1	2.72±0.2	0.014	0.067
para-hippocampal gyrus [a]	2.7±0.3	2.61±0.2	0.011	0.379	2.63±0.3	2.57±0.3	0.005	0.222
paracentral lobule [a]	2.27±0.1	2.2±0.2	0.019	0.430	2.35±0.2	2.25±0.2	0.05	0.500
inferior frontal gyrus pars opercularis [a]	2.41±0.1	2.31±0.2	0.004	0.527	2.41±0.1	2.41±0.1	0.165	0
inferior frontal gyrus pars orbitalis [a]	2.53±0.2	2.48±0.4	0.006	0.170	2.64±0.2	2.6±0.2	0.023	0.202
inferior frontal gyrus pars triangularis [a]	2.26±0.1	2.18±0.3	0.197	0.395	2.31±0.1	2.32±0.2	0.171	0.072
pericalcarine cortex [a]	1.52±0.1	1.58±0.2	0.118	0.404	1.55±0.1	1.59±0.2	0.537	0.229
postcentral gyrus [a]	1.91±0.1	1.89±0.1	0.066	0.166	1.91±0.1	1.89±0.2	0.005	0.123
posterior cingulate gyrus [a]	2.4±0.1	2.15±0.6	0.014	0.626	2.39±0.2	2.31±0.2	0.36	0.453
precentral gyrus [a]	2.43±0.2	2.35±0.2	0.173	0.442	2.41±0.1	2.33±0.2	0.002	0.420
precuneus [a]	2.2±0.1	2.15±0.2	0.035	0.327	2.24±0.1	2.21±0.1	0.006	0.230
rostral anterior cingulate gyrus [a]	2.74±0.2	2.61±0.2	0.071	0.616	2.83±0.2	2.73±0.2	0.355	0.465

rostral middle frontal gyrus [a]	2.25±0.1	2.22±0.2	0.242	0.219	2.24±0.1	2.27±0.2	0.031	0.235
superior frontal gyrus [a]	2.6±0.1	2.49±0.2	0.002	0.656	2.58±0.1	2.51±0.2	0.004	0.463
superior parietal lobule [a]	2.06±0.1	2.01±0.2	0.018	0.18	2.02±0.1	2±0.2	0.011	0.135
superior temporal gyrus [a]	2.57±0.2	2.46±0.3	0.003	0.429	2.62±0.2	2.42±0.2	<0.001*	1
supra-marginal gyrus [a]	2.34±0.1	2.27±0.4	0.079	0.235	2.37±0.1	2.38±0.3	0.217	0.048
frontal pole [a]	2.58±0.2	2.59±0.3	0.886	0.041	2.56±0.3	2.6±0.2	0.702	0.159
temporal pole [a]	3.48±0.2	3.32±0.5	0.001*	0.414	3.6±0.4	3.55±0.3	0.004	0.145
transverse temporal cortex [a]	2.08±0.2	2.01±0.3	0.016	0.286	2.13±0.2	2.11±0.3	0.013	0.068
insular cortex [a]	2.88±0.2	2.74±0.4	0.222	0.471	2.89±0.2	2.81±0.3	0.130	0.367