VILNIUS UNIVERSITY

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The significance of cognitive and demographic indicators in predicting the response of Alzheimer's disease to treatment

SUMMARY OF DOCTORAL DISSERTATION

Medicine and Health Sciences,
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The dissertation was written between 2016 and 2020 in Vilnius University Faculty of Medicine Neurology and Neurosurgery Clinics.

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This doctoral dissertation will be defended at a public meeting of the Dissertation Defence Board:

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VILNIAUS UNIVERSITETAS

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Kognityvinių ir demografinių rodiklių reikšmė prognozuojant Alzheimerio ligos atsaką į gydymą

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ABBREVIATIONS

AD	Alzheimer's disease
AChE	Acetylcholinesterase
ADAS-cog	Alzheimer Disease Assessment Scale - cognitive subscale
CANTAB	Cambridge Neuropsychological Test Automated Battery
CCT	Computerized cognitive tests
CG	Control group
ChE	Cholinesterase
GDS	Geriatric Depression Scale
CRT	Choice Reaction Time
CRT_MeanCorLat	Choice Reaction Time "Mean Correct latency"
CRT_TotCorTRL	Choice Reaction Time "Total Correct Trials"
CRT_TotIncTRL	Choice Reaction Time "Total Incorrect Trials"
HIS	Hachinski Ischemic Score
MMSE	Mini Mental State Examination
ns	Not significant
PAL_FTMS	Paired Associates Learning "First trial memory score"
PAL_FTMS RS2-1	PAL test "First trial memory score" change between 1st and 2nd testing
PAL_FTMS RS3-1	PAL test measure "First trial memory score" change between 1st and 3rd testing
PAL_METS	Paired Associates Learning "Mean errors to success"
PAL_MTTS	Paired Associates Learning "Mean trials to success"
PAL_MTTS RS1	PAL test measure "Mean trials to success" baseline score at 1st testing

PAL_MTTS RS2-1	PAL test measure “Mean trials to success” change between 1st and 2nd testing
PAL_MTTS RS3-1	PAL test measure “Mean trials to success” change between 1st and 3rd testing
PAL_SC	Paired Associates Learning “Stages completed”
PAL_SC RS1	PAL test measure “Stages completed” baseline score at 1st testing
PAL_SC RS2-1	PAL test measure “Stages completed” change between 1st and 2nd testing
PAL_SC RS3-1	PAL test measure “Stages completed” change between 1st and 3rd testing
PAL_SC1T	Paired Associates Learning “Stages completed on the first trial”
PAL_TEA	Paired Associates Learning “Total errors adjusted”
PAL_TEA RS2-1	PAL test measure “Total errors adjusted” change between 1st and 2nd testing
PAL_TEA RS3-1	PAL test measure “Total errors adjusted” change between 1st and 3rd testing
PAL_TE1/2/3/6/8ad	Paired Associates Learning “Total errors adjusted” at different pattern stages (1, 2, 3, 6 and 8 shapes)
PAL_TE6ad RS1	PAL test measure “Total errors adjusted 6 shapes” baseline score at 1st testing
PAL_TE6ad RS2-1	PAL test measure “Total errors adjusted 6 shapes” change between 1st and 2nd testing
PAL_TE6ad RS3-1	PAL test measure “Total errors adjusted 6 shapes” change between 1st and 3rd testing
PAL_TE8ad RS1	PAL test measure “Total errors adjusted 8 shapes” baseline score at 1st testing
PAL_TE8ad RS3-1	PAL test measure “Total errors adjusted 8 shapes” change between 1st and 3rd testing
PAL_TT	Paired Associates Learning “Total trials”

PAL_TT1/2/3/6/8	Paired Associates Learning “Total trials” at different pattern stages (1, 2, 3, 6 and 8 shapes)
PAL_TTad	Paired Associates Learning “Total trials adjusted”
PAL_TTad RS2-1	PAL test measure “Total trials” change between 1st and 2nd testing
PAL_TTad RS3-1	PAL test measure “Total trials” change between 1st and 3rd testing
PRMi	Pattern Recognition Memory immediate
PRMi_NC	Pattern Recognition Memory immediate “Number correct”
PRMd	Pattern Recognition Memory delayed
PRMd_NC RS1	PRMd test measure “Number correct” baseline score at 1st testing
PRMd_NC RS2-1	PRMd test measure “Number correct” change between 1st and 2nd testing
PRMd_NC RS3-1	PRMd test measure “Number correct” change between 1st and 3rd testing
PRMd_NC	Pattern Recognition Memory delayed “Number correct”
R^2	The coefficient of determination
SD	Standard deviation
SOC	Stockings of Cambridge
SOC_MM2	Stockings of Cambridge “Mean Moves (2 moves)”
SOC_MM3	Stockings of Cambridge “Mean Moves (3 moves)”
SOC_MM4	Stockings of Cambridge “Mean Moves (4 moves)”
SOC_MM5	Stockings of Cambridge “Mean Moves (5 moves)”
SOC_PSMM2	Stockings of Cambridge “Problems solved in minimum moves (2 moves)”

SOC_PSMM3	Stockings of Cambridge “Problems solved in minimum moves (3 moves)”
SOC_PSMM4	Stockings of Cambridge Problems solved in minimum moves (4 moves)”
SOC_PSMM5	Stockings of Cambridge “Problems solved in minimum moves (5 moves)”
SWM	Spatial Working Memory
SWM_BE	Spatial Working Memory “Total Between errors”
SWM_BE4/6/8	Spatial Working Memory “Between errors” at different pattern stages (4, 6 and 8 boxes)
SWM_WE	Spatial Working Memory “Total Within errors”
SWM_WE4/6/8	Spatial Working Memory “Within errors” at different pattern stages (4, 6 and 8 boxes)
SWM_DE	Spatial Working Memory “Double Errors”
SWM_DE4/6/8	Spatial Working Memory “Double Errors” at different pattern stages (4, 6 and 8 boxes)
SWM_TE4/6/8	Spatial Working Memory “Total errors” at different pattern stages (4, 6 and 8 boxes)

1. INTRODUCTION

Alzheimer's disease (AD) is a chronic progressive neurodegenerative disease accompanied by decline of cognitive functions and difficulty with daily living activities. Alzheimer's disease is most common in people over the age of 60-65. It is the most frequent cause of dementia syndrome in old age, which is becoming an increasingly important medical and social issue.

From the age of 65, the incidence of AD increases exponentially 2 times every 5 years. Early diagnosis of AD provides an opportunity to apply symptomatic or in the future, disease-modifying treatment and slow down cognitive decline, prolong the period of independent living, propose psychosocial measures, and reduce the cost of care for the sick.

Although the pathogenesis of AD is not fully understood, it is established that the degeneration of cholinergic system and the deficiency of acetylcholine in the brain has a major role in occurrence of cognitive symptoms in AD. Symptomatic therapies are the only treatment approved for AD. Cholinesterase (ChE) inhibitors diminish the degradation of acetylcholine and increase its accumulation in the synapse, thereby enhancing cholinergic neurotransmission and cognitive functions in AD patients. Donepezil is the most commonly used cholinergic medication in AD treatment.

In clinical practice, the diagnosis of AD is based on cognitive impairment, which is determined by cognitive tests. Usual paper-pencil tests such as Mini Mental State Examination (MMSE) are suitable for diagnosis while Alzheimer Disease Assessment Scale - cognitive subscale (ADAS-cog) - is suitable for assessing disease progression; however, these tests are not sensitive and detailed enough to reflect episodic declarative memory, which disorders can already be found in the prodromal and early stages of AD. In recent years, efforts have been made to develop and improve cognitive research methodologies that allow more reliable detection of cognitive impairment in the very early stages of the disease and to assess and

compare small changes in cognitive functions as early as possible with medical treatment.

Computerized cognitive tests (CCT) and their batteries are more extensive and reliable, more accurate and more sensitive while assessing visual / semantic / verbal memory and learning, working memory, executive functions and decision making, alertness, reaction time; when the treatment is administered and continued, CCTs allow for a faster detection and assessment of the change in cognitive status which would predict the patient's response to the prescribed medication. The tests by Cambridge Neuropsychological Test Automated Battery (CANTAB), especially the PAL test, have many advantages in AD studies. The PAL test is sensitive for the diagnosis of early Alzheimer's disease and amnestic mild cognitive impairment. Groton Maze Learning Test (GMLT), a part of the computerized CogState battery, has been shown to detect changes in cognitive functions after a single dose of donepezil even though this test is used for evaluating executive functions, working memory, and speed of information processed, that is, cognitive domain which impairment in AD occurs later than episodic memory, learning, and recall impairment. To the best of our knowledge, CANTAB tests have not been studied to determine the efficacy of a single dose of donepezil.

The dissertation presents a work that has been dealing with the CCTs which are reliable for AD diagnosis and suitable for monitoring the dynamics of cognitive functions in patients with AD after initiation of treatment, and which allow to predict the efficacy of a long-term treatment with donepezil.

1.1. The Aim of the Study

To determine which CANTAB test scores reliably differentiate Alzheimer's patients from controls and are suitable for monitoring the dynamics of cognitive functions; to analyse if the results of computerized cognitive tests before treatment and test results after a single dose of donepezil in combination with clinical and demographic

indicators of Alzheimer's disease, can predict the efficacy of a long-term treatment with donepezil.

1.2. The Objectives of the Study

1. To determine which CANTAB test measures reliably differentiate Alzheimer's patients from controls.
2. To evaluate the similarity of cognitive dysfunction profiles of the subjects *de novo*-diagnosed with Alzheimer's disease who have not yet started treatment and who have been receiving donepezil for more than four months.
3. To identify which CANTAB test measures reliably and strongly correlate with the overall dementia severity as evaluated by MMSE scores.
4. To assess which CANTAB test measures reliably evaluate the change in cognitive functions after a single dose of donepezil.
5. To determine which changes in CANTAB test measures after a single dose of donepezil, alongside the baseline scores and demographic and clinical factors reliably predict the efficacy of a long-term Alzheimer’s disease treatment with donepezil.
6. To determine the correlation of changes in CANTAB test measures with changes in MMSE scores over a four-month period of Alzheimer's disease treatment with donepezil.

1.3. Significance and Scientific Novelty of the Study

The ability of paper-pencil tests to reliably detect changes in cognitive functions after treatment with donepezil periods of various durations has been extensively studied worldwide and in Lithuania. In many clinical trials, non-computerized tests can reliably detect improvements in cognitive functions only after 2-4 weeks of the treatment. For the first time, a detailed analysis was carried out in the

paper to evaluate a change in cognitive functions after a single dose of the acetylcholinesterase (AChE) inhibitor, donepezil, using the CANTAB computerized test battery.

There are no comprehensive studies performed in Lithuania and worldwide to examine changes in the cognitive status of AD patients after a single dose of the drug. Taking into account that some of the patients respond poorly to symptomatic treatment or that symptomatic treatment has no effect at all in some cases, there is a particular need for cognitive tests to reliably determine the efficacy or lack of efficacy of the drug as early as possible. Using computerized cognitive tests and demographic and clinical data applied in the paper, it would be possible to have an early prediction of the patient's likely response to symptomatic medication and optimize individual treatment for specific patients. In this way, the maximum possible efficacy of the drug could be achieved with minimal side effects.

2. MATERIALS AND METHODS

The research and data analysis were performed in the Center of Neurology of Vilnius University Hospital Santaros Klinikos in 2010-2020.

The study Protocol was approved by the Vilnius Regional Biomedical Research Ethics Committee (approval No. 158200-12-128-36).

2.1. Selection and Grouping of Participants

102 subjects were enrolled in the study: 72 patients with Alzheimer's disease at Neurology Center of Vilnius University Hospital Santaros Klinikos and 30 healthy persons (CG) selected by age, gender and duration of education. The patients' group includes 62 *de novo*-diagnosed, treatment-naïve AD patients and 10 patients receiving the stable donepezil dose of 10 mg/day for at least 3 months. *De novo*-diagnosed, treatment-naïve AD patients were randomly assigned to

AD+ or AD- groups by the online Research Randomizer at http: //www.randomizer.org/. AD+ includes patients, who have been diagnosed with clinically probable AD, have not received treatment with specific anti-AD medications and who have taken the first dose of the prescribed drug, donepezil, after CCT Session 1. AD- is a group of patients, who have been diagnosed with clinically probable AD, have not received treatment with specific anti-AD medications and who have taken the first dose of the prescribed drug, donepezil, on the same day but after CCT Session 2. The third group consists of patients who have been diagnosed with clinically probable AD, have already been treated and have received a stable dose of donepezil for at least 3 months but have not taken the medicine in between CCT sessions.

Inclusion criteria for AD patients are the following:

1. The patients who are diagnosed with clinically probable AD based on NINCDS - ADRDA criteria, according to the order No. 382 by Minister of Health of Lithuania;
2. The patients who are diagnosed with mild or mild-to-moderate dementia: Mini - Mental State Examination (MMSE) score of at least 18, and no greater than 23;
3. The diagnosis of pure Alzheimer's disease without vascular comorbidity, confirmed by estimating the ischemia index on the basis of the Hachinski scale, where Hachinski ischemic score is equal to or less than 4;
4. The patients without diabetes mellitus;
5. The patients who have had a CT or MRI in less than 12 recent months, and their findings are consistent with the diagnosis of probable AD;
6. The patients over 65 years old;
7. The patients who have a newly diagnosed AD and are treatment-naïve or have been treated with the stable daily donepezil dose for 3 months or more prior to assessment; the treatment was initiated at the discretion of the treating physician based on standard clinical practice. Conventional

clinical practice treatment has not been modified in any way due to the conduct of this study;

8. The patients whose neurological examination does not show focal neurological signs except for cognitive symptoms which belong to AD characteristics;
9. The patients whose education is equal to or more than 4 years;
10. The patients are proficient in the Lithuanian language;
11. The patients' eyesight and hearing are sufficient for compliance with the study assessment.

Exclusion criteria for AD patients are the following:

1. The patients who do not meet at least one inclusion criterion;
2. The patients with other type of dementia;
3. The patients with other neurodegenerative diseases, other primary nervous system or systemic medical diseases which are likely to affect cognitive functions;
4. The patients who have had a stroke;
5. The patients with a clinically severe depression (rated ≥ 20 by the Geriatric Depression Scale);
6. The patients who are diagnosed with renal or hepatic insufficiency, thyroid hypofunction, vitamin B12 deficiency;
7. The patients who suffer from mental illnesses;
8. The patients receiving a stable dose of donepezil for less than 3 months;
9. The patients who are currently taking any other cognition-enhancing medication or food supplements (e.g. memantine or over-the-counter medicines).

A written Informed Consent Form was obtained from all the subjects prior to any study related procedures.

2.2. Study Methods

2.2.1. The Schedule of Study Assessments

All subjects were examined 3 times. At the first visit, general clinical and neurological examination was performed on each subject. Demographic data of the subjects (age, gender, duration of education) were recorded, medical and AD disease history was collected, vital signs, haematological and biochemical tests' results, and concomitant medications were assessed. Vascular risk factors were assessed using the Hachinski Ischemic Score (HIS), and depressive symptoms were assessed using the Geriatric Depression Scale (GDS). At the first visit, assessment of cognitive functions was performed twice with a 4-hour break; the third assessment of cognitive functions was performed at the second visit 4 months later (Table 1).

Table 1. Flowchart of Study Specific Assessments

Visit	**Visit 1**		**Visit 2**
Assessment	**Test 1**	**Test 2** (4 hours after the Test 1)	**Test 3** (4 months after the Visit 1)
Informed consent	+		
Demographics	+		
Medical history, AD history	+		
Vital signs	+	+	+
Inclusion/exclusion criteria	+		
MMSE	+ MMSE 1		+ MMSE 2
GDS	+		+
Hachinski Ischemic Score	+		
Concomitant medication	+		+
Adverse events	+	+	+
CANTAB battery tests (CRT, SOC, PAL, PRM immediate, SWM, PRM delayed)	+ CANTAB 1	+ CANTAB 2	+ CANTAB 3

2.2.2. Examination of Cognitive Functions

MMSE test and computerized system CANTAB eclipse 3.0.0 were used for cognitive assessment. All tests for all participants were performed by the same investigator.

The standard MMSE paper-pencil test was used to evaluate the overall level of cognitive functioning (global severity of dementia) for all the patients. The MMSE test was performed at the first and second visits, i.e. in four months following the first visit.

After the MMSE test, cognitive functions were assessed using the computerized Cambridge Neuropsychological Test Battery CANTAB eclipse 3.0.0. A laptop with a touch screen and a press pad with two buttons were used for the tests. Performing a task, the subject sat in front of the screen to be able to comfortably touch the screen or press buttons on the press pad. The researcher sitting next to the subject explained how to perform the task before each test. The CANTAB test battery was performed twice at the first visit. Following the CANTAB testing session 1, the AD+ patients' group received the first prescribed dose of 5mg donepezil hydrochloride. All the subjects had a 4 hour interval after the first CANTAB testing session. The second CANTAB testing session took place after the interval. Following the CANTAB testing session 2, the patients from the group AD- received their first doze of 5mg donepezil hydrochloride, while the patients with AD who had been taking donepezil hydrochloride for longer than 3 months, received their regular daily dose of the drug. The third CANTAB testing session took place in 4 months following the first visit. The tests were repeated in the same sequence throughout the study. In order to avoid the learning effect, parallel test versions of the CANTAB battery were used for the repeated testing. To assess the possible practice effect, a group of controls was examined according to the same study protocol.

The cognitive test battery consisted of 6 tests most appropriate for AD, which were selected from 22 tests possible to perform by CANTAB eclipse 3.0.0 battery. The tests assess alertness, spatial

planning and spatial working memory, learning, short-term memory, recall, recognition, and working memory capacity.

Choice Reaction Time (CRT) test shows the subject's alertness and speed of psychomotor reactions. The subject uses a press pad with two buttons during the test. An arrow pointing to the right or left is displayed on the right or left side of the screen. The subject shall press the right button as quickly as possible if an arrow pointing to the right is displayed on the screen or the left button if an arrow pointing to the left is displayed on the screen (Figure 1). The test consists of three stages: a learning stage of 24 trials and two assessment stages, each including 50 trials.

The outcome measures for CRT test are the following:

• Choice Reaction Time Mean Correct latency, CRT_MeanCorLat;
• Choice Reaction Time Total Correct Trials, CRT_TotCorTRL;
• Choice Reaction Time Total Incorrect Trials, CRT_TotIncTRL.

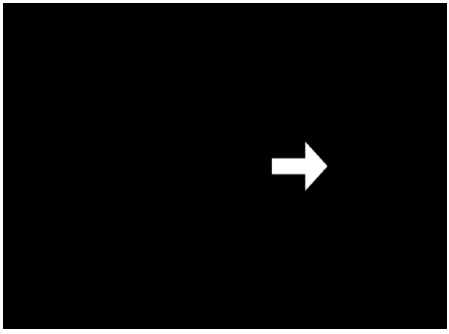


Figure 1. The Example of Choice Reaction Time Test

Stockings of Cambridge (SOC) test demonstrates the subject's spatial planning and problem-solving abilities, which reflect frontal executive functions. SOC test is composed of two types of task. Task Type 1 is for the subject to place red, green, and blue balls in socks of different lengths at the bottom of the screen, with a minimum number of moves and as quickly as possible, according to the example at the

top of the screen. At first, the subject only has to move one ball, then the tasks get harder and the number of moves required increases to 5. A procedure controlling motor performance is inserted in between the tasks: in the upper part of the screen, the balls move themselves and change their position, then, the subject shall copy moves made by the computer and put the same ball to its new position in the lower part of the screen, respectively. The moves are arranged mimicking the moves the participant made when originally solving the problem. Task Type 2 evaluates how much time the subject has spent on solving the task (cognitive functions) and how much on the transfer action itself (motor function) (Figure 2).

The outcome measures for SOC test are the following:

• Stockings of Cambridge Mean Moves (2 moves), SOC_MM2;
• Stockings of Cambridge Mean Moves (3 moves), SOC_MM3;
• Stockings of Cambridge Mean Moves (4 moves), SOC_MM4;
• Stockings of Cambridge Mean Moves (5 moves), SOC_MM5;
•Stockings of Cambridge Problems solved in minimum moves (2 moves), SOC_PSMM2;
• Stockings of Cambridge Problems solved in minimum moves (3 moves), SOC_PSMM3);
• Stockings of Cambridge Problems solved in minimum moves (4 moves), SOC_PSMM4;
• Stockings of Cambridge Problems solved in minimum moves (5 moves), SOC_PSMM5.

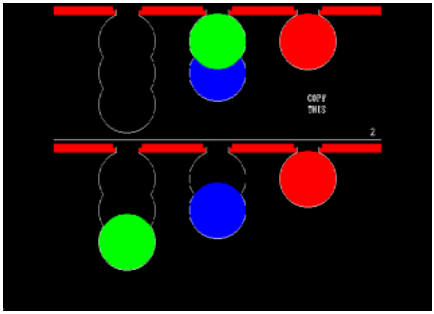


Figure 2. The Example of Stockings of Cambridge Test

Paired Associates Learning (PAL) test assesses the subject's visual memory and new learning. The screen displays six (eight in the last stage) white boxes which are arranged in a circle and are randomly opened every 3 seconds. At the first stage, only one box shows an abstract geometric figure, and during each subsequent stage, the number of boxes showing different abstract geometric figures is increased, i. e. two, three, six, and eight boxes each shows a different shape. The subject shall remember which figure was displayed in which box. When all the boxes are opened on the screen and all the shapes are displayed, the shapes appear randomly one after another in the middle of the screen, and the subject shall touch the box where the displayed shape has appeared. If the subject indicates the location of all the figures correctly, the program proceeds to the next more complex stage of the test with 2, 3, 6, or 8 figures. The stages of the two and three figures are repeated twice. If the subject makes a mistake, the program reopens the boxes randomly, but the location of the shapes remains same as the first time. At each stage, the subject can perform 10 trials to learn how to position all the figures correctly. The PAL test is terminated if the subject fails to indicate the correct location of all the figures throughout 10 trials (Figure 3).

The outcome measures for PAL test are the following:

• Paired Associates Learning First trial memory score, PAL_FTMS;
• Paired Associates Learning Mean errors to success, PAL_METS;
• Paired Associates Learning Mean trials to success, PAL_MTTS;
• Paired Associates Learning Stages completed, PAL_SC;
• Paired Associates Learning Stages completed on the first trial, PAL_SC1T;
• Paired Associates Learning Total errors adjusted, PAL_TEA and total errors adjusted at different pattern stages (1, 2, 3, 6 and 8 shapes): PAL_TE1ad, PAL_TE2ad, PAL_TE3ad, PAL_TE6ad and PAL_TE8ad;
• Paired Associates Learning Total trials, PAL_TT and total trials at different pattern stages (1, 2, 3, 6 and 8 shapes): PAL_TT1, PAL_TT2, PAL_TT3, PAL_TT6 and PAL TT8;

• Paired Associates Learning Total trials (adjusted), PAL_TTad.

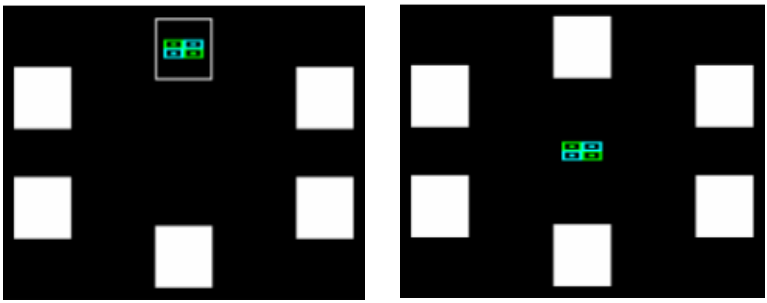


Figure 3. The Example of Paired Associates Learning Test

Pattern Recognition Memory immediate (PRMi) tests the subject's visual recognition memory. 12 coloured abstract geometric shapes appear on the screen one after another. When all the 12 figures are demonstrated to the subject, 2 figures are displayed on the screen: 1 is already seen by the participant and 1 is new. The subject shall select the one which has already been seen (Figure 4).

The outcome measure for PRMi test is:

• Pattern Recognition Memory immediate Number correct, RMi_NC.

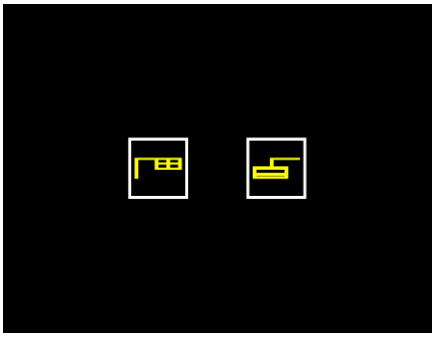


Figure 4. Pattern Recognition Memory Test

Spatial Working Memory (SWM) test is concerned with working memory. The test assesses the subject's ability to retain and

manipulate visual-spatial information in working memory. Colored boxes are displayed on the screen; at each stage, the quantity of the boxes is increased from three to eight, the color and position is altered as well. The participant shall select each box on the screen and open it to check if there is a blue square. The square shall be placed in the column at the edge of the screen. At each stage, once a square is found, it will no longer appear in the same box and shall be found in all remaining boxes, both untouched and those which have previously been empty, but the box in which the square has already been found shall not be selected. This eliminates the number of boxes in which the blue square can be found (Figure 5).

The outcome measures for SWM test are the following:

• Spatial Working Memory Total Between errors, SWM_BE. Intermediate errors revisiting boxes which have already been found to contain a blue square;

• Spatial Working Memory Between errors at different pattern stages (4, 6 and 8 boxes): SWM_BE4, SWM_BE6 and SWM_BE8;

• Spatial Working Memory Total Within errors, SWM_WE. Internal errors selecting boxes which have already been found to be empty;

• Spatial Working Memory Within errors at different pattern stages (4, 6 and 8 boxes): SWM_WE4, SWM_WE6 and SWM_WE8;

• Spatial Working Memory Double Errors, SWM_DE. These are occasions where the subject has committed an error that can be categorised as both a within and a between error. This is calculated for trials of four or more boxes only.

• Spatial Working Memory Double Errors at different pattern stages (4, 6 and 8 boxes): SWM_DE4, SWM_DE6 and SWM_DE8;

• Spatial Working Memory Total errors, SWM_TE. This is the number of times a box is selected that is certain not to contain a blue square and therefore should not have been visited by the subject, i.e. between errors + within errors - double errors;

• Spatial Working Memory Total errors at different pattern stages (4, 6 and 8 boxes): SWM_TE4, SWM_TE6 and SWM_TE8.

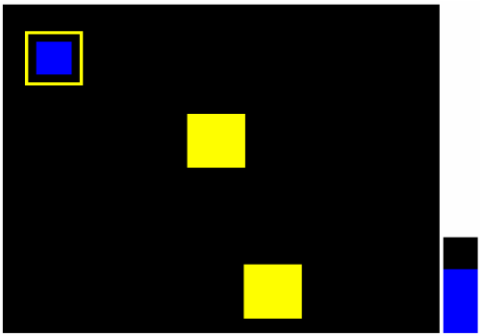


Figure 5. Spatial Working Memory Test

Pattern Recognition Memory delayed (PRMd). The test deals with the subject's visual recognition memory (delayed recognition). The stimuli presentation stage is performed immediately after the PRMi test and there should be a 30 minute interval between the display of stimuli and the recognition stage. The SWM test is performed after the presentation of PRMd stimuli and the recognition of PRMd stimuli is assessed after the SWM test is finished. At the recognition stage, the screen displays two coloured abstract geometric shapes at a time: one of which has already been seen while showing shapes one by one immediately after the completion of the PRMi test; another figure has not been seen. The subject shall touch the figure which has already been seen.

The outcome measure for PRMd test is:

• Pattern Recognition Memory delayed Number correct, PRMd_NC.

2.3. Methods of Statistical Analysis

The statistical package SPSS 20.0 (*Windows* version) was used for data analysis. Qualitative variables are presented in absolute numbers and percentages. Quantitative variables are presented in mean and standard deviation (SD) if distribution is normal. Normal distribution of data was verified using the Shapiro-Wilk test. The Student's t-test or Analysis of Variance (ANOVA) was applied comparing groups

with respect to a quantitative variable. A chi-square ($\chi 2$) test was used to compare qualitative variables. The one - way analysis of variance (ANOVA) with intergroup comparison using the Bonferroni *post-hoc* test was performed to compare the test scores of the first CANTAB session in more than two groups of the subjects. The Levene test was used to assess the homogeneity of variances across participant groups.

The Pearson correlation coefficient r was used to assess the data correlation for normally distributed samples.

Repeated-measures ANOVA was applied to assess the change in CANTAB test scores in more than two study groups between the first and second test sessions. The Bonferroni *post hoc* test was used for intergroup comparison. The sphericity of the variances of the differences between all possible pairs of groups was checked using the Mauchly sphericity test, when the number of repeated measurements was greater than two.

A cluster analysis was applied to evaluate the differences of cognitive impairment profiles in different AD groups.

A Power analysis was used to determine the sample size.

In order to determine the cognitive, demographic, and clinical variables, which might be prognostic factors in the effectiveness of a long-term treatment, the multiple General Linear Models (GLM) were designed.

The significance level was considered to be $p < 0.05$.

3. RESULTS

3.1. Demographic, Clinical and Cognitive Characteristics of the Participants

102 subjects were enrolled in the study: 62 participants *de novo*-diagnosed with AD and who have taken the first dose of the prescribed drug, donepezil, after CCT Session 1 or CCT Session 2; 10 AD patients taking the stable donepezil dose of 10 mg/day for at least 3 months; and 30 controls (CG). All 4 groups were matched on the demographic characteristics. MMSE did not differ significantly

among the three AD study groups, but it was significantly higher in the control group. Demographic characteristics, depression level by GDS and MMSE scores of all the subjects in the study are provided in table 2.

Table 2. Demographic Characteristics in Participant Groups.

Variables	AD+ group	AD- group	DZP treated AD group	Control group	Statistical methods
Number of subjects, N	30	32	10	30	
Age (Years) Mean ± SD	77.30±5.11	77.03±5.28	76.50±4.43	76.43±6.36	One-way ANOVA F(3,98)=0.148; p=0.931, ns
Gender Women/ Men, N	17/13	17/15	4/6	17/13	Pearson Chi square=0.970; p=0.809, ns
Education (Years), Mean ± SD	13.17±4.79	13.47±4.02	11.10±3.73	13.20±3.61	One-way ANOVA F(3,98)=0.875; p=0.457, ns
Depression (GDS scale) Mean ± SD	7.670±4.93	6.84±3.91	7.50±4.33	6.77±4.34	One-way ANOVA F(3,98)=0.286; p=0.835, ns
MMSE test Mean ± SD	21.57±1.57	21.25±1.48	21.80±1.14	29.47±0.57	One-way ANOVA F(3,98)=277.1; p<0.001 Bonferroni post-hoc: AD+=AD-= DZP treated AD (p=1.0); CG>AD+, AD-, DZP treated AD (p<0.001)

SD – standard deviation, ns – not significant.

3.2. A Assessment of Differences of Cognitive Impairment Profiles in AD Groups

Before proceeding to the evaluation of prognostic factors it was necessary to determine and assure that cognitive profiles in all three groups of AD patients were not significantly different (Table 1).

Cluster analysis was performed to evaluate the difference of cognitive impairment profiles in AD groups. The groups were assigned into clusters based on the results of all the variables examined (Figure 6). AD+ and AD- groups, both being *de novo*, is highly similar in cognitive profiles. The third AD patients' group is far from AD+ and AD- and in terms of its linkage distance, is in the intermediate position between *de novo* AD subjects and controls.

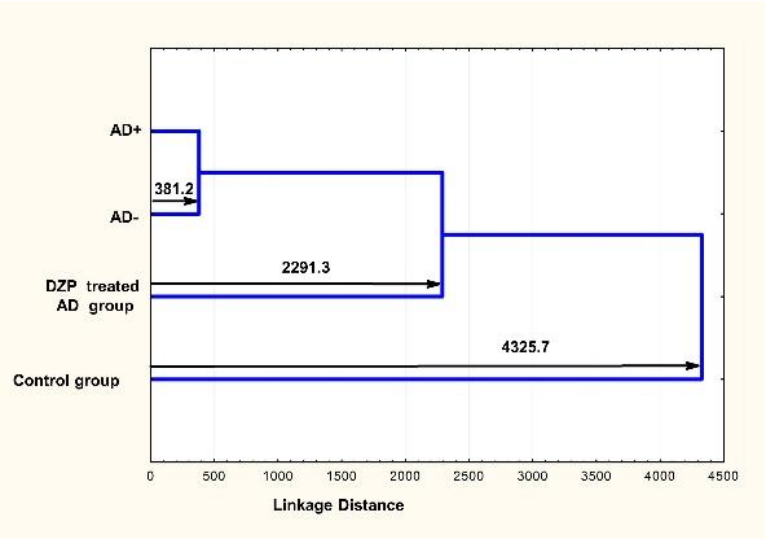


Figure 6. Dendrogram of the Linkage Distance of the Cognitive Profiles in the Four Study Groups.

Euclidean distance (Table 3) compared to linkage distance provides additional information on cognitive heterogeneity of the research groups. Euclidean distance scores show the study groups

arranged not on the same axis and the distance between treated AD group and CG is even greater than *de novo*-diagnosed AD and CG. It would indicate that the overall cognitive profile of the donepezil treated AD patients' group is even further away from the control group than from any of the *de novo*-diagnosed AD groups.

Table 3. Euclidean Distance in Participant Groups.

Group	AD+	AD-	Treated DZP AD group	CG
AD+	0	381	2671	4326
AD-	381	0	2291	4654
Treated DZP AD group	2671	2291	0	6676
CG	4326	4654	6676	0

Due to the significant difference in the profile of cognitive impairment between untreated and treated AD subjects, the AD treated subjects' group is not suitable for the analysis of the ability of CANTAB test measures to distinguish between AD and CG.

3.3. Comparison of CANTAB Test Measures in Participant Groups at a Baseline (1st testing session)

Before proceeding to the main purpose of this study, it was necessary to establish which CANTAB test measures were able to reliably distinguish AD patients from the Control group; hence, CANTAB test scores were compared among AD+, AD- and CG at a baseline. All scores of PAL test measures, PRMi_NC, PRMd_NC measures, SWM_TE, SWM_TE4, SWM_TE6, SWM_TE8, SWM_BE6 and SWM_BE8 had a statistically significant worse results in AD patients (both AD+ and AD-) than CG. A CANTAB CRT baseline score did not significantly differ between AD and CG. Only two measures of SOC test (SOC_MM3 and SOC_PSMM3) showed significant difference between both AD groups (AD+ and AD-) and CG. CANTAB testing measures for alertness and reaction time (CRT),

executive function measures (SOC) are shown in Table 4; Table 5 presents measures for learning and episodic recall memory (PAL); episodic immediate and delayed pattern recognition memory (PRMi and PRMd), and working memory measures are provided in Table 7.

Table 4. Comparison of Choice Reaction Time (CRT) Test and Stockings of Cambridge (SOC) Test Measures in Participant Groups.

Measure	AD+ group	AD- group	Control group	One-way ANOVA	Bonferroni post-hoc
CRT_ MeanCorLat, ms	573.0±192.9	548.0±166.8	536.6±230.9	F=0.266; p=0.767, ns	CG=AD+=AD-
CRT_ TotCorTRL, N	98.97±1.40	98.72±2.14	98.50±2.01	F=0.459; p=0.633, ns	CG=AD+=AD-
CRT_ TotIncTRL, N	0.87±1.20	1.00±1.81	1.20±1.64	F=0.338; p=0.714, ns	CG=AD+=AD-
SOC_MM2	2.15±0.35	2.28±0.51	2.03±1.83	F=3.394*; p<0.05	Comparison non valid
SOC_PSMM2	1.83±0.38	1.69±0.59	1.97±0.18	F=3.361; p<0.05*	Comparison non valid
SOC_MM3	3.75±0.73	3.55±0.65	3.16±0.37	F=7.164; p=0.001	CG<AD+, AD-AD+=AD-
SOC_ PSMM3	1.23±0.63	1.34±0.70	1.80±0.41	F=7.714; p=0.001	CG>AD+, AD-AD+=AD-
SOC_MM4	5.11±0.98	5.15±1.04	4.90±0.93	F=0.576; p=0.564, ns	CG=AD+=AD-
SOC_PSMM4	2.60±0.93	2.38±1.09	2.54±1.25	F=0.343; p=0.711, ns	CG=AD+=AD-
SOC_MM5	6.60±1.65	7.03±1.38	6.61±1.51	F=0.820; p=0.444, ns	CG=AD+=AD-
SOC_PSMM5	1.37±0.99	1.45±1.14	1.83±1.23	F=1.427; p=0.246, ns	CG=AD+=AD-

Measures of CANTAB tests provided as Mean ± SD;
* Levene test significant (p<0.05), ANOVA invalidated;
ns – not significant.

Table 5. Comparison of Paired Associates Learning (PAL) Test Measures in Participant Groups.

Measure	AD+ group	AD- group	Control group	One-way ANOVA	Bonferroni post-hoc
PAL_FTMS	9.50±3.59	8.78±3.79	17.83±4.68	F=47.08; p<0.001	CG>AD+, AD-AD+=AD-
PAL_METS	8.47±3.31	8.48±2.63	2.64±2.86	F=39.88; p<0.001	CG<AD+, AD-AD+=AD-
PAL_MTTS	3.67±0.68	3.74±0.76	1.85±0.71	F=68.14; p<0.001	CG<AD+, AD-AD+=AD-
PAL_SC	6.30±1.12	6.00±0.88	7.93±0.25	F=47.27; p<0.001	CG>AD+, AD-AD+=AD-
PAL_SC1T	3.60±1.28	3.50±1.19	5.40±1.22	F=23.01; p<0.001	CG>AD+, AD-AD+=AD-
PAL_TEA	107.3±36.6	119.2±32.2	20.83±22.3	F=91.42; p<0.001	CG<AD+, AD-AD+=AD-
PAL_TE1ad	0.10±0.40	0.34±0,83	0.00±0.00	F=3.349*; p<0.05	Comparison non valid
PAL_TE2ad	2.10±2.39	2.15±3.07	0.33±1.02	F=5.930*; p<0.05	Comparison non valid
PAL_TE3ad	12.63±12.5	13.15±12.2	1.20±1.39	F=13.43*; p<0.001	Comparison non valid
PAL_TE6ad	33.00±15.2	38.71±13.8	6.07±6.87	F=59.09; p<0.001	CG<AD+, AD-AL+=AL-
PAL_TE8ad	59.50±16.9	64.84±14.5	13.23 ± 14.1	F=105.9; p<0.001	CG<AD+, AD-AD+=AD-
PAL_TTad	31.67±7.16	32.75±7.27	14.57±5.08	F=72.59; p<0.001	CG<AD+, AD-AD+=AD-
PAL_TT1	2.10±0.40	2.34±0.82	2.00±0.00	F=3.349*; p<0.05	Comparison non valid
PAL_TT2	3.23 ± 1.36	3.46±2.09	2.20± 0.61	F=6.186*; p<0.05	Comparison non valid
PAL_TT3	7.10±3.99	7.28±3.99	2.73±0.86	F=18.31; p<0.001	CG<AD+, AD-AD+=AD-
PAL_TT6	6.57±3.79	7.37±4.11	3.10±1.94	F=13.36; p<0.001	CG<AD+, AD-AD+=AD-
PAL_TT8	3.97±4.88	1.66±3.60	4.50±2.42	F=5.039*# p<0.05	Comparison non valid

Measures of CANTAB tests provided as Mean ± SD;

* Levene test significant (p<0.05), ANOVA invalidated;

The mean includes participants, who did not reach the stage of 8 shapes (mostly AD patients), whose result for this measure is automatically entered as 0. Due to this, the mean is irrelevant.

Table 6. Comparison of Pattern Recognition Memory Immediate (PRMi) Test and Pattern Recognition Memory Delayed (PRMd) Test Measures in Participant Groups.

Measure	AD+ group	AD- group	Control group	One-way ANOVA	Bonferroni post-hoc
PRMi_NC	8.03±1.47	7.44±1.65	9.93±1.39	F=22.84; p<0.001	CG>AD+, AD- AD+=AD-
PRMd_NC	6.30±1.84	6.43±1.39	9.40±1.69	F=34.25; p<0.001	CG>AD+, AD- AD+=AD-

Measures of CANTAB tests provided as Mean ± SD;

Table 7. Comparison of Spatial Working Memory (SWM) Test Measures in Participant Groups.

Measure	AD+ group	AD- group	Control group	One-way ANOVA	Bonferroni post-hoc
SWM_BE	58.83±13.95	62.12±10.54	34.73±16.85	F=35.0*; p<0.001	Comparison non valid
SWM_BE4	4.50±3.26	5.69±3.21	1.70±1.78	F=15,19*; p<0.001	Comparison non valid
SWM_BE6	19.77±6.33	19.34±6.48	10.73±7.75	F=16.66; p<0.001	CG<AD-, AD+ AD+=AD-
SWM_BE8	34.57±7.69	37.09±6.64	22.30±8.99	F=31.37; p<0.001	CG<AD+, AD- AL+=AL-
SWM_TE	60.3±14.5	64.1±10.5	36.2±17.2	F=34.29; p<0.001	CG<AD+, AD- AD+=AD-
SWM_TE4	5.03±3.58	6.12±3.35	2.37±2.31	F=11.66; p<0.001	CG<AD+, AD- AL+=AL-
SWM_TE6	20.0±6.53	19.7±6.58	11.0±7.89	F=16.17; p<0.001	CG<AD+, AD- AD+=AD-
SWM_TE8	35.2±7.86	38.2±6.67	22.8±9.01	F=32.78; p<0.001	CG<AD+, AD- AD+=AD-
SWM_DE	1.67±1.71	2.09±2.53	1.03±1.30	F=2.35; p=0,101, ns	CG=AD+=AD-
SWM_DE4	0.13±0.35	0.13±0.55	0.10±0.40	F=0.05; p=0.949, ns	CG=AD+=AD-
SWM_DE6	0.33±0.76	0.56±0.88	0.37±1.03	F=0.59; p=0.553, ns	CG=AD+=AD-
SWM_DE8	1.20±1.40	1.41±2.18	0.57±0.77	F=2.35; p=0.101, ns	CG=AD+=AD-
SWM_WE	3.10±2.56	4.06±3.35	2.50±2.66	F=2.31 p=0.105, ns	CG=AD+=AD-
SWM_WE4	0.67±0.99	0.56±1.01	0.77±1.33	F=0.26; p=0.774, ns	CG=AD+=AD-
SWM_WE6	0.60±1.13	0.94±1.19	0.63±1.33	F=0.729; p=0,485, ns	CG=AD+=AD-
SWM_WE8	1.83±1.74	2.58±2.84	1.10±1.06	F=4.073*; p=0.02	Comparison non valid

Measures of CANTAB tests provided as Mean ±SD; ns – not significant;

* Levene test significant (p<0.05), ANOVA invalidated.

3.4. Correlation between CANTAB Test Measuresand MMSE

A correlation between CANTAB test measures and MMSE (as a measure of global dementia severity) was established to evaluate whether change in CANTAB test measures may be treated as a clinically relevant cognitive change. Only those test measures able to distinguish donepezil treatment-naïve *de novo*-diagnosed AD groups from the Control Group were selected for correlation analysis. Eight measures of PAL test and one measure of PRMd test showed statistically significant and strong correlation (r>0.7) with MMSE. Table 8 presents the findings.

Table 8. Correlation of CANTAB PAL, SOC, PRMi, PRMd, SWM Test Results with MMSE (Global Measure of Dementia Severity)

Measure	Pearson r	p	Selected for further analysis
PAL_FTMS	**0.763**	p<0.001	**Yes**
PAL_METS	-0.651	p<0.001	No
PAL_MTTS	**-0.803**	p<0.001	**Yes**
PAL_SC	**0.791**	p<0.001	**Yes**
PAL_SC1T	0.623	p<0.001	No
PAL_TEA	**-0.870**	p<0.001	**Yes**
PAL_TE6ad	**-0.807**	p<0.001	**Yes**
PAL_TE8ad	**-0.849**	p<0.001	**Yes**
PAL_TTad	**-0.847**	p<0.001	**Yes**
PAL_TT3	-0.632	p<0.001	No
PAL_TT6	-0.365	p<0.001	No
PRMi_NC	0.509	p<0.001	No
PRMd_NC	**0.702**	p<0.001	**Yes**
SOC_MM3	-0.411	p<0.001	No
SOC_ PSMM3	0.433	p<0.001	No
SWM_TE	-0.632	p<0.001	No
SWM_TE4	-0.403	p<0.001	No
SWM_TE6	-0.501	p<0.001	No
SWM_TE8	-0.624	p<0.001	No
SWM_BE6	-0,501	p<0.001	No
SWM_BE8	-0.613	p<0.001	No

3.5. Assessment of Cognitive Change due to a Single Dose of Donepezil Based on CANTAB Test Measures in the Three Participant Groups (Change between 1st and 2nd CANTAB Testing Sessions)

For those CANTAB test measures which were able to reliably distinguish AD and Controls and demonstrated the statistically significant and strong correlation ($r>0.7$) with MMSE (as a measure of global dementia severity), repeated-measures ANOVA was used to assess which CANTAB tests and measures are able to detect significant cognitive change due to a single dose of donepezil.

Significant difference of change in findings between 1st (Testing 1) and 2nd (Testing 2) CANTAB testing sessions in AD+ and AD- groups (interaction effect "Testing session" * "Group") was established for seven PAL test measures: PAL_FTMS, PAL_MTTS, PAL_SC, PAL_TEA, PAL_TE6ad, PAL_TE8ad and PAL_TTad. For all 7 PAL test measures Bonferroni *post-hoc* revealed that the difference is significant in the results of Testing 2 between AD+ and AD– groups (between-group effect), while Testing 1 showed insignificant differences between AD+ and AD– groups. 4 PAL measures, PAL_MTTS, PAL_TEA, PAL_TE6ad, PAL_TTad, and PRMd for test measure PRMd_NC demonstrated statistically significant difference based on results of CANTAB Testing 1 and Testing 2 in the AD+ group where subjects received a single dose of donepezil between first and second testing (within treatment group effect). All three evaluation criteria were fulfilled for the effect of donepezil first single dose in 4 PAL test measures: PAL_MTTS, PAL_TEA, PAL_TE6ad and PAL_TTad. The efficacy of a single dose of donepezil was expressed in PRMd_NC only in terms of significant change in the AD+ group between Testing 1 and Testing 2. Three testing measures: PAL_FTMS, PAL_SC and PAL_TE8ad, showed the efficacy of a single dose of donepezil in relation to significant difference of change in testing results in AD+ and AD- between CANTAB Testing 1 and Testing 2 as well as significant

difference of testing results in AD+ and AD- at Testing 2. Table 9 indicates the findings of the analysis of CANTAB repeated-measures in participant groups based on Testing Session 1 and Testing Session 2.

Table 9. Results of Repeated-Measures ANOVA with Bonferroni *Post-Hoc* in Participant Groups Based on the First and Second Testing Sessions.

Measure	Difference of changes in AD+ and AD- groups between 1st and 2nd testing (interaction effect)	p	Difference of AD+ and AD- groups based on the results of the second testing (between group effect)	p	Difference of first and second testing results in AD+ group (within group effect)	p
PAL_FTMS	3.39	0.0016	4.11	<0.001	1.80	0.130, ns
PAL_MTTS	-0.59	0.0014	-0.62	0.003	-0.51	<0.001
PAL_SC	0.95	<0.0001	1.25	<0.001	0.23	1.00, ns
PAL_TEA	-35.4	<0.0001	-47.3	<0.001	-18.3	0.005
PAL_TE6ad	-11.7	0.0002	-17.5	<0.001	-7.03	0.008
PAL_TE8ad	-8.89	0.0447	-14.2	0.005	-4.67	1.00, ns
PAL_TTad	-8.65	<0.0001	-9.73	<0.001	-4.40	0.002
PRMd_NC	0.61	0.240, ns	0.47	1.00, ns	1.20	0.003

ns – not significant

3.6. Calculation of the Study Sample Size

Before the start of our study there were no data of CANTAB testing to calculate a sample size for a study of relevant design, hence, the sample size assessment was performed after obtaining the necessary baseline data, but before proceeding to the main purpose of the study – the evaluation of treatment effectiveness predictors, therefore, the sample size determination in this study should be treated as “a priori”, but not as “post hoc”. The sample size was calculated when it was already clear which CANTAB tests were able to reliably distinguish both AD groups from CG individuals and which testing measures reliably correlated with MMSE and detected cognitive change after a

single dose of donepezil. The sample size was estimated with regards to 4 CANTAB testing measures which were informative and reliably fulfilled all three criteria (Table 9). Sample size calculations are given in Table 10.

Table 10. Sample Size, Power, and Effect size in CANTAB Testing Measures

Measure	Mean of test measure change between 1st and 2nd testing in AD+ group (N=30)	Mean of test measure change between 1st and 2nd testing in AD- group (N=32)	Standard deviation of both AD+ and AD- groups sample (N=62)	Power, 1−β	Type I error rate, α	Sample size	Effect size, Cohen's d
PAL MTTS	-0.510	0.077	0.791	0.80	0.05	29	0.742 (medium-large)
PAL TEA	-18.367	17.063	36.599	0.95	0.05	28	0.968 (large-very large)
PAL TE6ad	-7.033	4.718	14.107	0.85	0.05	26	0.833 (large)
PAL TTad	-4.400	4.250	8.426	0.95	0.05	25	1.027 (large-very large)

3.7. Evaluation of Treatment Effectiveness Predictors

For CANTAB testing measures as dependent variables multiple General Linear Models (GLM) were built to determine which independent demographic variables, cognitive testing baseline scores, and changes in cognitive functions after a single dose of donepezil were able to predict the efficacy of a long-term, 4-month treatment.

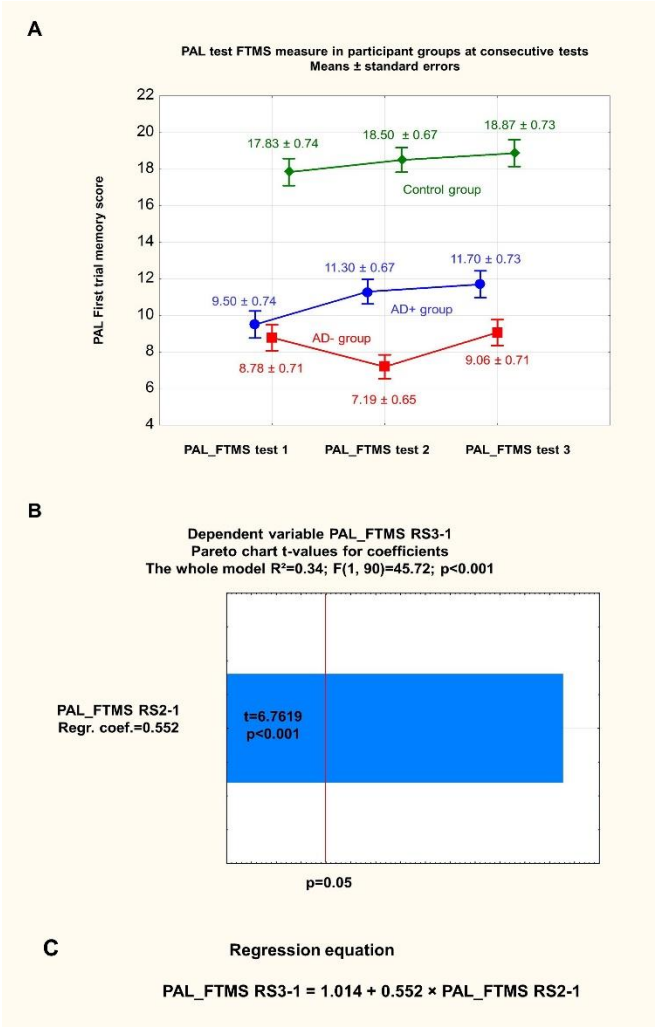
The GLM models were designed and calculated for those CANTAB testing measures which were able to differentiate the subjects from AD and CG, showed statistically significant and strong

correlation ($r>0.7$) with MMSE and were able to identify the efficacy of a single dose of donepezil (Table 8).

The best predictor variables were determined using a stepwise backward removal method. The regression model was considered suitable for the prediction if the whole model was statistically significant (coefficient of determination R^2 was significant at the level of $p<0.05$) and all coefficients in the regression equation of the final model were statistically significant ($p<0.05$).

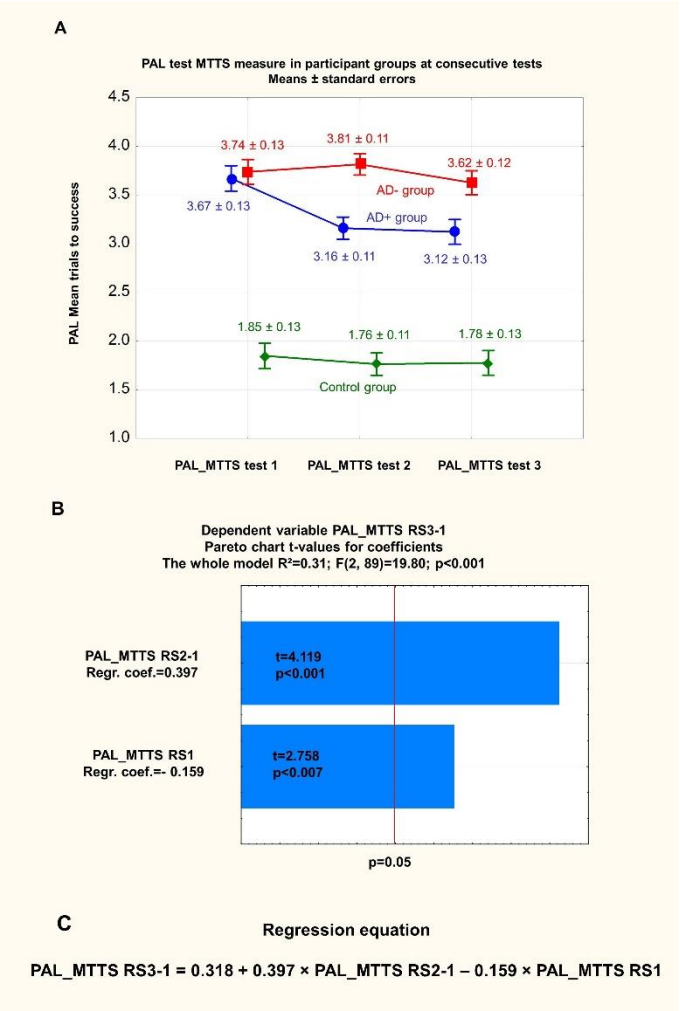
The differences of scores on the CANTAB at Testing 3 and Testing 1 were entered in a GLMs as a dependent variables. As independent continuous predictors in the GLM models were entered age, education, baseline MMSE score (i.e. MMSE score at Testing 1), depression (by GDS), results of corresponding CANTAB tests' at a baseline (Testing 1) and the change of scores between Testing 1 and Testing 2; As independent categorical predictors in the GLM models were entered gender and study group (AD+, AD- or CG).

GLM models were built for eight relevant dependent variables to evaluate the predictive value of specified above independent variables. The results of the General Linear Models are provided in Figures 7-14.



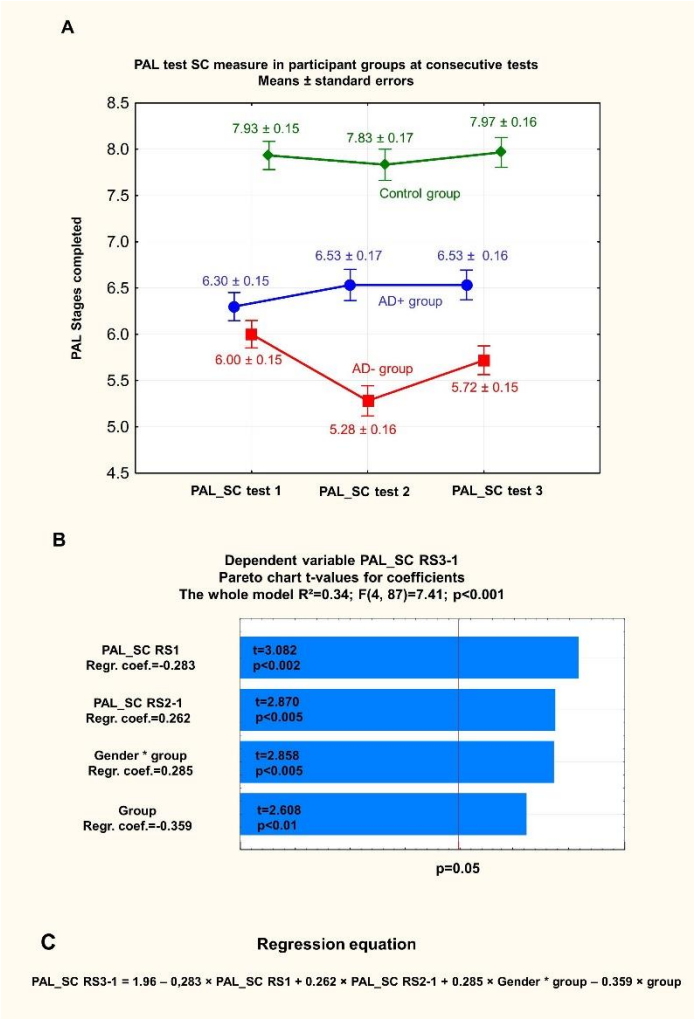
* Dependent variable PAL_FTMS RS3-1 is a PAL test measure "First trial memory score" change between 1st and 3rd testing;
** PAL_FTMS RS2-1 is a PAL test "First trial memory score" change between 1st and 2nd testing.

Figure 7. The results of the general linear regression model of PAL test FTMS measure: (A) Mean ±SD in participant groups at the three consistent tests. (B) Pareto chart of regression coefficients. (C) Regression equation.



* Dependent variable PAL_MTTS RS3-1 is a PAL test measure "Mean trials to success" change between 1st and 3rd testing;
** PAL_MTTS RS2-1 is a PAL test measure "Mean trials to success" change between 1st and 2nd testing.
*** PAL_MTTS RS1 is a PAL test measure "Mean trials to success" score at 1st testing.

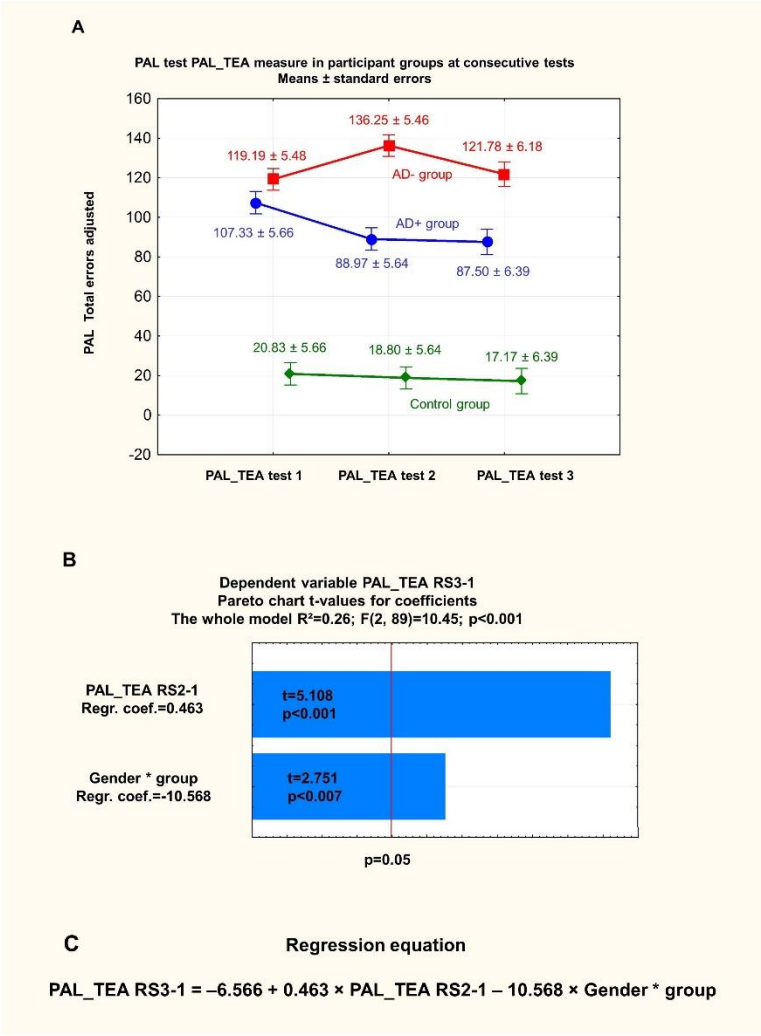
Figure 8. The results of the general linear regression model of PAL test MTTS measure: (A) Mean ±SD in participant groups at the three consistent tests. (B) Pareto chart of regression coefficients. (C) Regression equation.



Dependent variable PAL_SC RS3-1 is a PAL test measure "Stages completed" change between 1st and 3rd testing;

** PAL_SC RS1 is a PAL test measure "Stages completed" baseline score at 1st testing.
** PAL_SC RS2-1 is a PAL test measure "Stages completed" change between 1st and 2nd testing.
**** Gender * Participant group is an interaction between a gender and a participant group.

Figure 9. The results of the general linear regression model of PAL test SC measure: (A) Mean ±SD in participant groups at the three consistent tests. (B) Pareto chart of regression coefficients. (C) Regression equation.

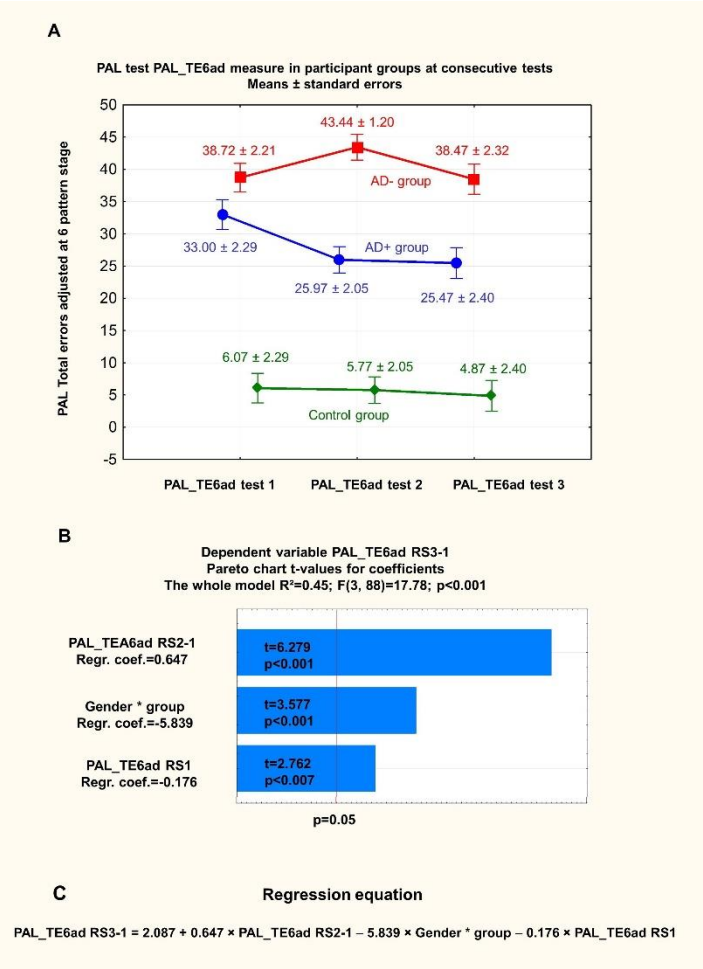


* Dependent variable PAL_TEA RS3-1 is a PAL test measure "Total errors adjusted" change between 1st and 3rd testing;

** PAL_TEA RS2-1 is a PAL test measure "Total errors adjusted" change between 1st and 2nd testing.

*** Gender * Participant group is an interaction between a gender and a participant group.

Figure 10. The results of the general linear regression model of PAL test TEA measure: (A) Mean ±SD in participant groups at the three consistent tests. (B) Pareto chart of regression coefficients. (C) Regression equation.



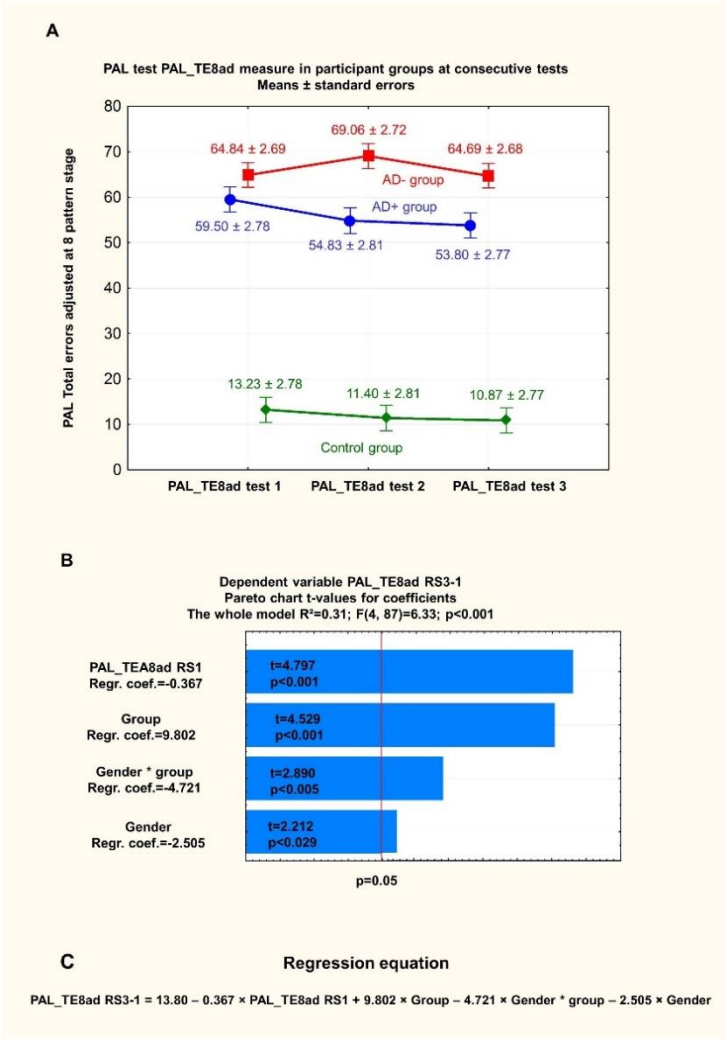
* Dependent variable PAL_TE6ad RS3-1 is a PAL test measure "Total errors adjusted 6 shapes" change between 1st and 3rd testing;

** PAL_TE6ad RS2-1 is a PAL test measure "Total errors adjusted 6 shapes" change between 1st and 2nd testing.

*** Gender * Participant group is an interaction between a gender and a participant group.

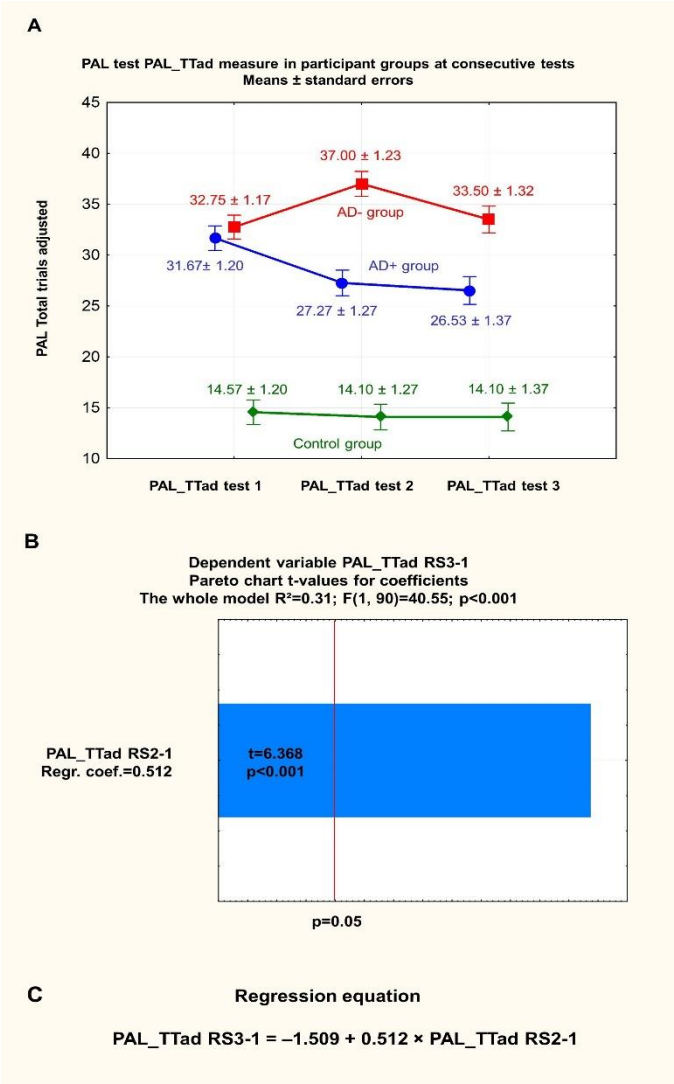
**** PAL_TE6ad RS1 is a PAL test measure "Total errors adjusted 6 shapes" baseline score at 1st testing.

Figure 11. The results of the general linear regression model of PAL test TE6ad measure: (A) Mean ±SD in participant groups at the three consistent tests. (B) Pareto chart of regression coefficients. (C) Regression equation.



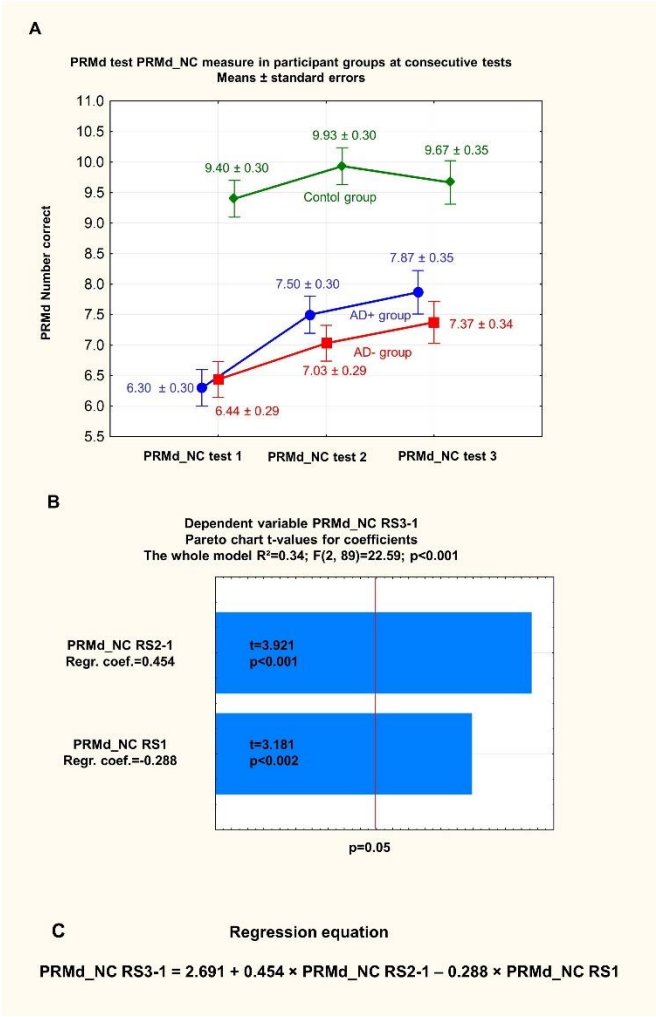
* Dependent variable PAL_TE8ad RS3-1 is a PAL test measure "Total errors adjusted 8 shapes" change between 1st and 3rd testing;
** PAL_TE8ad RS1 is a PAL test measure "Total errors adjusted 8 shapes" baseline score at 1st testing.
*** Gender * Participant group is an interaction between a gender and a participant group.

Figure 12. The results of the general linear regression model of PAL test TE8ad measure: (A); Mean ±SD in participant groups at the three consistent tests. (B) Pareto chart of regression coefficients. (C) Regression equation.



* Dependent variable PAL_TTad RS3-1 is a PAL test measure "Total trials" change between 1st and 3rd testing;
** PAL_TTad RS2-1 is a PAL test measure "Total trials" change between 1st and 2nd testing.

Figure 13. The results of the general linear regression model of PAL test TTad measure: (A) Mean ±SD in participant groups at the three consistent tests. (B) Pareto chart of regression coefficients. (C) Regression equation.



* Dependent variable PRMd_NC RS3-1 is a PRMd test measure "Number correct" change between 1st and 3rd testing;

** PRMd_NC RS2-1 is a PRMd test measure "Number correct" change between 1st and 2nd testing.

*** PRMd_NC RS1 is a PRMd test measure "Number correct" " baseline score at 1st testing.

Figure 14. The results of the general linear regression model of PRMd test NC measure: (A) Mean ±SD in participant groups at the three consistent tests. (B) Pareto chart of regression coefficients. (C) Regression equation.

3.8. Correlations between the Cognitive Changes in CANTAB Test Measures and MMSE over the 4-month Treatment Period

The change in cognitive functions over a 4-month period, that is, between CANTAB testing 1 and 3, reflects the dynamics in one cognitive domain. In order to assess the relationship between individual cognitive functions and overall dementia severity dynamics, the correlation between change in CANTAB tests' scores and change in MMSE was evaluated.

A statistically significant ($p<0.05$) correlation was found, with a Pearson correlation coefficient $r>0.5$, between a change in scores of PAL_FTMS, PAL_MTTS, PAL_SC, PAL_TEA, PAL_TE6ad, PALTE8ad and PAL_TTad at 1st and 3rd testing and a change in MMSE at 1st and 2nd testing. A statistically significant ($p<0.05$) correlation was detected, with a Pearson correlation coefficient $r> 0.3$, between a change in PRMd_NC measure at 1st and 3rd testing and a change in MMSE at 1st and 2nd testing.

4. CONCLUSIONS

1. The four CANTAB tests - PAL, PRM, SWM, and SOC - reliably differentiate Alzheimer's subjects from controls.
2. There is a difference in cognitive dysfunction profile between *de novo*-diagnosed treatment naïve AD patients and subjects taking donepezil for more than 4 months.
3. A significant and strong correlation with overall dementia severity, as assessed by MMSE score, is found for PAL_FTMS, PAL_MTTS, PAL_SC, PAL_TEA, PAL_TE6ad, PAL_TTad, PAL_TE8ad and PRMd_NC test measures.

4. Eight CANTAB test measures - PAL_FTMS, PAL_MTTS, PAL_SC, PAL_TEA, PAL_TE6ad, PAL_TE8ad, PAL_TTad, and PRMd_NC - are able to reliably determine the change in cognitive functions of Alzheimer's disease patients after a single dose of donepezil.
5. The change in PAL_FTMS, PAL_MTTS, PAL_SC, PAL_TEA, PAL_TE6ad, PAL_TTad and PRMd_NC test measures after a single dose of donepezil is reliable efficacy predictor of a long-term treatment with donepezil; The baseline estimates of PAL_MMTS, PAL_SC, PAL_TE6ad, PAL_TE8ad and PRMd_NC measures are reliable efficacy predictors of a long-term treatment with donepezil; A gender together with a PAL_TE8ad measure baseline estimate are reliable efficacy predictors of a long-term treatment with donepezil.
6. The changes in PAL_FTMS, PAL_MTTS, PAL_SC, PAL_TEA, PAL_TE6ad, PAL_TE8ad, and PAL_TTad measures significantly and moderately strongly correlate with the changes in MMSE score in a four month period of donepezil treatment; the change in PRMd_NC measure reliably but mildly correlates with the change in MMSE score in a four month period of donepezil treatment.

5. PUBLICATIONS

1. **Kuzmickienė J**, Kaubrys G. Cognitive Results of CANTAB Tests and Their Change Due to the First Dose of Donepezil May Predict Treatment Efficacy in Alzheimer Disease. Med Sci Monit 2015; 21: 3887-99. DOI: 10.12659/msm.896327. **"Clarivate Analytics Web of Science".**

2. **Kuzmickienė J**, Kaubrys G. Selective Ability of Some CANTAB Battery Test Measures to Detect Cognitive Response to a Single Dose of Donepezil in Alzheimer's Disease. Med Sci Monit 2015; 21: 2572-82. DOI: 10.12659/MSM.895381. **"Clarivate Analytics Web of Science".**

3. **Kuzmickienė J**, Kaubrys G. Specific Features of Executive Dysfunction in Alzheimer-Type Mild Dementia Based on Computerized Cambridge Neuropsychological Test Automated Battery (CANTAB) Test Results. Med Sci Monit. 2016; 22: 3605-13. DOI: 10.12659/msm.900992. **"Clarivate Analytics Web of Science".**

6. PRESENTATIONS

1. **Kuzmickienė J**, Kaubrys G. "Demographic, Clinical, and Cognitive Predictors of the Response of Alzheimer's Disease to Treatment with Donepezil". The 4th International Conference Evolutionary Medicine: Health and Diseases in Changing Environment. 5-10th Jun 2018. Vilnius, Lithuania.

2. **Kuzmickienė J**, Kaubrys G. "The Baseline Scores of the Cambridge Neuropsychological Test Automated Battery as Cognitive Predictors of Donepezil Treatment Efficacy in Alzheimer's disease". International Conference the 9th Baltic Congress of Neurology, BALCONE. 6-8th Sep 2018. Kaunas, Lithuania.

3. **Kuzmickienė J**, Kaubrys G. "Relationships between Alzheimer's disease progression and treatment effectiveness with cognitive phenotype and demographics". 7th Oct 2016. Vilnius, Lithuania.

SUMMARY IN LITHUANIAN

KOGNITYVINIŲ IR DEMOGRAFINIŲ RODIKLIŲ REIKŠMĖ PROGNOZUOJANT ALZHEIMERIO LIGOS ATSAKĄ Į GYDYMĄ

SANTRUMPOS

AL	Alzheimerio liga
ADAS	Alzheimerio ligos įvertinimo skalė (angl. *Alzheimer Disease Assesment Scale*
CANTAB	Kembridžio neuropsichologinių kompiuterinių testų rinkinys (angl. *Cambridge Neuropsychological Test Automated Battery*)
CRT	Pasirinkimo reakcijos laikas (angl. *Choise Reaction Time*)
GDS	Geriatrinė depresijos skalė
HII	Hačinskio išemijos indeksas
KKT	Kompiuterizuoti kognityviniai testai
KG	Kontrolinė grupė
MMSE	Protinės būklės mini tyrimas (angl. *Mini Mental State Examination)*
PAL	Porinių asociacijų išmokimas (angl. *Paired Associates Learning*)
PAL_FTMS	PAL testo rodiklis - pirmu bandymu teisingoje vietoje nurodytų figūrų skaičius (angl. *First trial memory score*)
PAL_FTMS RS2-1	PAL testo FTMS rodiklio pokytis tarp 1-o ir 2-o testavimų

PAL_FTMS RS3-1	PAL testo FTMS rodiklio pokytis tarp 1-o ir 3-io testavimų
PAL_MTTS	PAL testo rodiklis - vidutinis bandymų skaičius iki visiškai teisingo visų figūrų vietos nurodymo (angl. *Mean trials to success)*
PAL_MTTS RS1	PAL testo MTTS rodiklio pradinis įvertis
PAL_MTTS RS2-1	PAL testo MTTS rodiklio pokytis 1-o ir 2-o testavimų
PAL_MTTS RS3-1	PAL testo MTTS rodiklio pokytis tarp 1-o ir 3-io testavimų
PAL_SC	PAL testo rodiklis - sėkmingai įvykdytų stadijų skaičius (angl. *Stages completed)*
PAL_SC RS1	PAL testo SC rodiklio pradinis įvertis
PAL_SC RS2-1	PAL testo SC rodiklio pokytis tarp 1-o ir 2-o testavimų
PAL_SC RS3-1	PAL testo SC rodiklio pokytis tarp 1-o ir 3-io testavimų
PAL_TEA	PAL testo rodiklis - bendras koreguotas klaidų skaičius visose stadijose (angl. *Total errors adjusted)*
PAL_TEA RS2-1	PAL testo TEA rodiklio pokytis tarp 1-o ir 2-o testavimų
PAL_TEA RS3-1	PAL testo TEA rodiklio pokytis tarp 1-o ir 3-io testavimų
PAL_TE6ad	PAL testo rodiklis - bendras koreguotas klaidų skaičius 6 figūrų stadijos
PAL_TE6ad RS1	PAL testo TE6ad rodiklio pradinis įvertis
PAL_TE6ad RS2-1	PAL testo TE6ad rodiklio pokytis tarp 1-o ir 2-o testavimų
PAL_TE6ad RS3-1	PAL testoTE6ad rodiklio pokytis tarp 1-o ir 3-io testavimų
PAL_TE8ad	PAL testo rodiklis - bendras koreguotas klaidų skaičius 8 figūrų stadijos

PAL_TE8ad RS1	PAL testo TE8ad rodiklio pradinis įvertis
PAL_TE8ad RS3-1	PAL testoTE8ad rodiklio pokytis tarp 1-o ir 3-io testavimų
PAL_TTad	PAL testo rodiklis - bendras koreguotas atliktų bandymų skaičius (angl. *Total trials (adjusted)*
PAL_TTad RS2-1	PAL testo TTad rodiklio pokytis tarp 1-o ir 2-o testavimų
PAL_TTad RS3-1	PAL testo TTad rodiklio pokytis tarp 1-o ir 3-io testavimų
SOC_MM3	SOC testo rodiklis - vidutinis ėjimų skaičius, kurių reikėjo tiriamajam, kad atliktų užduotį, kurią galima atlikti minimaliu 3 ėjimų skaičiumi (angl. *SOC Mean Moves (3 moves))*
SOC_PSMM3	SOC testo rodiklis - minimaliu įmanomu ėjimų skaičiumi išspręstų užduočių skaičius atliekant 3 ėjimų užduotis (angl. *Problems solved in minimum moves (3 moves)*)
SWM_TE	SWM testo rodiklis - testo bendras klaidų skaičius (angl. *Total errors*)
SWM_TE4/6/8	SWM testo rodiklis - bendras klaidų skaičius 4/6/8 dėžučių stadijos

ĮVADAS

Alzheimerio liga (AL) – tai lėtinė progresuojanti neurodegeneracinė liga, pasireiškianti kognityvinių funkcijų blogėjimu ir kasdienės veiklos sutrikimu. Dažniau susergama virš 60–65 metų amžiaus. AL yra dažniausia demencijos sindromo priežastis vyresniame amžiuje ir tampa vis aktualesne medicinos ir socialine problema.

Nuo 65 metų amžiaus sergamumas AL didėja eksponentiškai du kartus kas 5 metai. Ankstyva AL diagnostika suteikia galimybę taikyti simptominį ar ateityje leis paskirti ligos eigą modifikuojantį (LEM) gydymą ir sulėtinti kognityvinį blogėjimą, prailginti nepriklausomo gyvenimo laikotarpį, skirti psichosocialines priemones, sumažinti išlaidas, skiriamas sergantiems prižiūrėti.

Klinikinėje praktikoje diagnozuojant AL remiamasi kognityvinių funkcijų sutrikimais, kurie nustatomi kognityviniais testais. Įprastiniai „popieriaus – pieštuko“ testai, tokie kaip Protinės būklės mini tyrimas (angl. *Mini Mental State Examination,* MMSE), tinkamas diagnozuoti, Alzheimerio ligos įvertinimo skalė (angl. *Alzheimer Disease Assesment Scale,* ADAS) – ligos progresavimui vertinti, yra nepakankamai jautrūs ir detalūs kad galėtų atspindėti epizodinę deklaratyviąją atmintį, kurios sutrikimus jau galima rasti prodrominėje ir ankstyvojoje AL stadijoje. Pastaraisiais metais stengiamasi sukurti ir tobulinti kognityvinio tyrimo metodikas, kurios leistų patikimiau nustatyti kognityvinius sutrikimus labai ankstyvose ligos stadijose ir įvertinti bei palyginti nedidelius pažinimo funkcijų pokyčius kuo anksčiau pradėjus gydymą vaistais.

Kompiuterizuoti kognityviniai testai (KKT) ir jų baterijos yra išsamesni ir patikimesni, tikslesni ir jautresni vertinant regimąją / semantinę / žodinę atmintį ir išmokimą, darbinę atmintį, vykdomąsias funkcijas ir sprendimų priėmimą, dėmesį, reakcijos laiką. KKT naudojami siekiant anksčiau ir tiksliau identifikuoti epizodinės deklaratyviosios atminties sutrikimus, o paskyrus gydymą ir jį tęsiant KKT leidžia greičiau pastebėti ir vertinti kognityvinės būklės pokytį,

kuris leistų prognozuoti paciento atsaką į skiriamą vaistą. Kembridžo neuropsichologinių kompiuterinių testų rinkinys (angl. *Cambridge Neuropsychological Test Automated Battery*), ypač PAL testas, turi daug privalumų AL tyrimuose. PAL testas yra jautrus ankstyvos Alzheimerio ligos ir amnestinio lengvo kognityvinio sutrikimo diagnostikai. Įrodyta, kad kompiuterizuotos CogState baterijos dalis Grotono labirinto išmokimo testas (angl. *Groton Maze Learning Test*, GMLT) gali aptikti kognityvinių funkcijų pokyčius jau po vienkartinės donepezilio dozės, tačiau šis testas vertina vykdomąsias funkcijas, darbinę atmintį ir apdorojamos informacijos greitį, t. y. kognityvinius domenus, kurių pažeidimas, sergant AL, atsiranda vėliau nei epizodinės atminties, mokymosi ir atsiminimo sutrikimai. Kiek mums žinoma, CANTAB testai nebuvo tirti siekiant nustatyti vienos donepezilio dozės efektyvumą.

Disertacijoje analizuojama ir aptariama, kokie KKT patikimai diagnozuoja AL, tinka sekti pacientų, sergančių AL, kognityvinių funkcijų dinamiką pradėjus gydymą bei leidžia prognozuoti ilgalaikio gydymo donepeziliu efektyvumą.

DARBO TIKSLAS

Nustatyti, kurie CANTAB testų rodikliai patikimai skiria sergančius Alzheimerio liga pacientus nuo kontrolinės grupės asmenų ir tinka stebėti kognityvinių funkcijų dinamiką bei ištirti ar kompiuterizuotų kognityvinių testų rezultatai prieš pradedant gydymą, testų rezultatai po pirmos donepezilio dozės vertinant kartu su Alzheimerio ligos klinikiniais ir demografiniais rodikliais, gali prognozuoti ilgalaikio gydymo donepeziliu maksimalų įmanomą efektyvumą.

DARBO UŽDAVINIAI

1. Nustatyti, kurių CANTAB testų rodikliai patikimai skiria Alzheimerio liga sergančius tiriamuosius nuo kontrolinės grupės asmenų.
2. Įvertinti naujai diagnozuotų dar nepradėjusių gydymo ir daugiau nei keturis mėnesius besigydančių donepeziliu Alzheimerio liga sergančių tiriamųjų kognityvinės disfunkcijos profilių vienodumą.
3. Nustatyti, kurie CANTAB testų rodikliai patikimai ir stipriai koreliuoja su bendru demencijos sunkumu, vertinamu MMSE balais.
4. Nustatyti, kurie CANTAB testų rodikliai patikimai nustato kognityvinių funkcijų pokytį po pirmosios donepezilio dozės.
5. Nustatyti, kurių CANTAB testų rodiklių pokyčiai po pirmos donepezilio dozės kartu su rodiklių įverčiais pradiniame taške bei demografiniais ir klinikiniais veiksniais patikimai prognozuoja ilgalaikio Alzheimerio ligos gydymo donepeziliu efektyvumą.
6. Nustatyti CANTAB testų rodiklių pokyčių koreliaciją su MMSE įverčio pokyčiu per keturių mėnesių trukmės Alzheimerio ligos gydymo donepeziliu laikotarpį.

PRAKTINĖ DARBO REIKŠMĖ IR NAUJUMAS

Darbe išsamiai kompiuterizuotu kognityvinių testų rinkiniu ištirtos pažintinės funkcijos sergančiųjų lengvo ir lengvo-vidutinio sunkumo Alzheimerio liga ir sveikų asmenų, vertinta jų dinamika po pirmosios vaisto dozės ir 4 mėnesių laikotarpyje bei joms įtaką darantys klinikiniai, demografiniai veiksniai.

Mūsų tyrimas skirtas CANTAB kompiuterizuotų kognityvinių testų rodikliams, kurie galėtų patikimai nustatyti kognityvinių

funkcijų pokyčius jau po vienkartinės donepezilo dozės, vertinti ir šių testų rodiklių prognozuojamai vertei ilgalaikio gydymo efektyvumui nustatyti. Tyrimo metu sudarytos kompiuterizuotos kognityvinės baterijos taikymas klinikinėje praktikoje leistų optimizuoti gydymą individualiam Alzheimerio liga sergančiam pacientui, taip pat galėtų būti jautria ir detalia kognityvinių funkcijų dinamikos vertinimo priemone naujų AL gydymo metodų klinikiniuose tyrimuose.

TIRIAMIEJI IR TYRIMO METODIKA

Tyrimas ir duomenų analizė atlikti Vilniaus universiteto Medicinos fakulteto Neurologijos ir neurochirurgijos klinikoje, Vilniaus universiteto ligoninės Santaros klinikų Nervų ligų skyriuje ir Konsultacijų poliklinikoje 2010–2020 m. Tyrimui gautas Vilniaus regioninio biomedicininių tyrimų etikos komiteto leidimas 2009-12-02 Nr. 158200-12-128-36.

Į tyrimą įtraukti 102 tiriamieji: 72 pacientai, sergantys Alzheimerio liga, ir 30 pagal amžių, lytį bei išsilavinimo trukmę atrinktų kontrolinių tiriamųjų, kurie nesirgo AL ar kito tipo demencija. Pacientų grupę sudarė 62 asmenys, kuriems buvo naujai diagnozuota AL ir pradėtas įprastinis gydymas donepeziliu, 10 asmenų, sergančių AL ir daugiau kaip 3 mėnesius gydomų stabilia 10 mg donepezilio doze. Pirmoji AL pacientų grupė buvo randomizuota į dvi grupes: AL+ ir AL–. Tiriamųjų randomizacijai panaudota internetinė randomizacijos sistema „Reasearch Randomizer“ http://www.randomizer.org/. AL+ yra pacientų grupė, kuriai diagnozuota kliniškai tikėtina AL, dar nepradėtas gydymas specifiniais vaistais nuo AL ir kuri pirmąją paskirto vaisto – donepezilio – dozę išgėrė po pirmosios KKT sesijos. AL– yra pacientų grupė, kuriai diagnozuota kliniškai tikėtina AL, dar nepradėtas gydymas specifiniais vaistais nuo AL ir kuri pirmąją paskirto vaisto – donepezilio – dozę išgėrė tą pačią dieną, bet po antrosios KKT sesijos.

Trečioji pacientų grupė, kuriai diagnozuota kliniškai tikėtina AL, jau buvo gydoma ir stabilią donepezilio dozę vartojo ne mažiau kaip 3 mėnesius, o tarp pirmosios ir antrosios KKT sesijų vaisto negėrė.

Visi tiriamieji buvo ištirti tris kartus. Pirmojo vizito metu kiekvienam tiriamajam buvo atliktas bendras klinikinis ir neurologinis ištyrimas. Įvertinti demografiniai tiriamųjų duomenys (amžius, lytis, išsilavinimas metais), surinkta medicininė ir AL ligos anamnezė, įvertintos gyvybinės funkcijos, įvertinti atlikti kraujo tyrimai (hematologinis ir biocheminis), vartojami medikamentai. Kraujagysliniai rizikos veiksniai vertinti Hačinskio išemijos indeksu (HII), depresijos reiškiniai vertinti naudojant Yesavage geriatrinę depresijos skalę (GDS). Pirmojo vizito metu kognityvinių funkcijų įvertinimas buvo atliktas du kartus su 4 valandų pertrauka, trečiasis kognityvinių funkcijų įvertinimas buvo atliktas antrojo vizito metu po 4 mėnesių. Kognityvinėms funkcijoms vertinti buvo naudojamas MMSE testas ir kompiuterizuota sistema CANTABeclipse 3.0.0. Visi testai atlikti vieno tyrėjo.

Atlikus MMSE testą, kognityvinės funkcijos vertintos kompiuterizuota Kembridžo neuropsichologinių testų baterija CANTABeclipse 3.0.0. Testams atlikti naudotas nešiojamasis kompiuteris su liesti jautriu ekranu ir pultelis. CANTAB testų baterija pirmojo vizito metu buvo atlikta du kartus. Atlikus CANTAB 1 testavimo sesiją, AL+ pacientų grupė suvartojo paskirtą pirmąją donepezilio hidrochlorido 5 mg dozę. Visi tiriamieji po pirmosios CANTAB testų sesijos turėjo 4 valandų pertrauką. Po šios pertraukos buvo atlikta CANTAB 2 testų sesija. Atlikus CANTAB 2 testų sesiją, pirmąją donepezilio hidrochlorido 5 mg dozę suvartojo AL– grupės tiriamieji, o sergantys AL ir daugiau nei 3 mėn. vartojantys donepezilio hidrocholoridą suvartojo įprastinę kasdienę šio vaisto dozę. CANTAB 3 testų sesija buvo atlikta praėjus 4 mėnesiams nuo pirmojo vizito. Visų tyrimų metu testai kartojosi ta pačia seka. Siekiant išvengti išmokimo efekto, kartotiniam testavimui buvo taikytos CANTAB rinkinio paralelinės sesijos. Siekiant įvertinti

galimą praktikos efektą, tirta kontrolinių asmenų grupė pagal tą patį ištyrimo protokolą.

Kognityvinių funkcijų tyrimo baterija buvo sudaryta iš 6 AL tinkamiausių testų, kurie atrinkti iš 22 testų, kuriuos įmanoma atlikti CANTABeclipse 3.0.0 baterija. Pasirinkti testai, vertinantys dėmesį, erdvinį planavimą ir erdvinę darbinę atmintį, išmokimą, trumpalaikę atmintį, įsiminimą ir darbinės atminties talpą:

• pasirinkimo reakcijos laikas (CRT, angl. *Choise Reaction Time*);
• Kembridžo kojinės (SOC, angl. *Stockings of Cambridge*);
• porinių asociacijų išmokimas (PAL, angl. *Paired Associates Learning*);
• figūrų atpažinimo testas, betarpiškas atpažinimas (PRMi, angl. *Pattern Recognition Memory immediate*);
• erdvinės darbinės atminties testas (SWM, angl. *Spatial Working Memory*);
• figūrų atpažinimo testas, uždelstas atpažinimas (PRMd, angl. *Pattern Recognition Memory delayed*).

REZULTATAI

Tyrime dalyvavo 102 asmenys: 62 asmenys, kuriems buvo naujai diagnozuota AL ir pradėtas įprastinis gydymas donepeziliu, 10 asmenų, sergančių AL ir daugiau kaip 3 mėnesius gydomų stabilia 10 mg donepezilio doze, ir 30 – sveiki asmenys (KG). Tiriamųjų grupės patikimai nesiskyrė pagal amžių ($p = 0,931$), išsilavinimą metais ($p = 0,457$), lytį ($p = 0,809$), depresijos lygį, GDS skalę balais ($p = 0,835$). MMSE patikimai nesiskyrė tarp trijų AL tiriamųjų grupių, tačiau statistiškai reikšmingai buvo didesnis kontrolinės grupės tiriamųjų.

Kognityvinių sutrikimų profilių skirtingose AL grupėse vienodumui įvertinti taikyta klasterinė analizė, grupių paskirstymui į klasterius naudoti visų tirtų rodiklių rezultatai. Remiantis rezultatais, dėl žymaus kokybinio kognityvinių sutrikimų profilio skirtumo tarp

negydomų ir gydomų AL tiriamųjų, gydomų AL tiriamųjų grupė į tolimesnę analizę netraukta.

Siekiant pagrindinio tyrimo tikslo, pradžioje reikėjo išsiaiškinti, kurie CANTAB testų rodikliai gali patikimai atskirti AL pacientus nuo kontrolinės grupės asmenų. Buvo palyginti AL+, AL– ir KG asmenų CANTAB testų rezultatai pirmojo testavimo metu. PAL testo visų rodiklių, PRMi_NC, PRMd_NC rodiklių, SWM_TE, SWM_TE4, SWM_TE6, SWM_TE8, SWM_BE6, SWM_BE8 rodiklių reikšmės buvo statistiškai patikimai blogesnės AL ligonių (tiek AL+, tiek AL–) nei KG asmenų. CANTAB CRT testo rezultatai pirmojo testavimo metu tarp AL ir KG asmenų statsitiškai reikšmingai nesiskyrė. SOC testo tik dviems rodikliams: SOC_MM3 ir SOC_PSMM3 gautas statistiškai reikšmingas skirtumas tarp abiejų AL grupių (AL+, AL–) ir KG asmenų.

Siekiant įvertinti ar CANTAB testų pokytis gali būti laikomas kliniškai reikšmingu kognityvinių funkcijų pokyčiu, buvo įvertinta koreliacija tarp CANTAB testų rezultatų ir MMSE balų kaip visuminio demencijos sunkumo rodiklio. Aštuoniems PAL testo rodikliams: PAL_FTMS, PAL_MTTS, PAL_SC, PAL_TEA, PAL_TE6ad, PAL_TE8ad, PAL_TTad, ir PRMd testo PRMd_NC rodikliui gautos statistiškai reikšmingos ir stiprios koreliacijos ($r>0.7$) su MMSE.

Tiems CANTAB testų rodikliams, kurie patikimai skiria AL ir KG tiriamuosius ir kuriems buvo gautos statistiškai patikimos ir stiprios koreliacijos ($r > 0,7$) su MMSE buvo atlikta kartotinių bandymų dispersinė analizė (angl. *Repeated-measures ANOVA*). Siekta įvertinti, kurie CANTAB testai ir jų rodikliai gali nustatyti reikšmingą kognityvinį pokytį jau po pirmos donepezilio dozės.

Septyniems PAL testo rodikliams: PAL_FTMS, PAL_MTTS, PAL_SC, PAL_TEA, PAL_TE6ad, PAL_TE8ad, PAL_TTad – nustatytas reikšmingas testų rezultatų pokyčių skirtumas tarp 1-osios ir 2-osios CANTAB testavimo sesijų AL+ ir AL– grupėse (sąveikos efektas „Testavimo sesija“*„Grupė“). Atlikus Bonferinio *post-hoc* testą nustatyta, kad statistiškai reikšmingas rezultatų skirtumas gautas

visiems septyniems PAL testo rodikliams antrosios CANTAB testavimo sesijos metu tarp AL+ ir AL– grupių (tarpgrupinis efektas). Reikšmingo skirtumo tarp AL+ ir AL– grupių pirmosios CANTAB testavimo sesijos metu nebuvo. Keturiems PAL testo rodikliams: PAL_MTTS, PAL_TEA, PAL_TE6ad, PAL_TTad – ir PRMd testo rodikliui PRMd_NC gautas statistiškai reikšmingas skirtumas tarp pirmosios ir antrosios CANTAB testavimo sesijos rezultatų AL+ grupėje, kuriai priskirti tiriamieji išgėrė pirmąją donepezilio dozę tarp pirmo ir antro testavimo (vidinis grupės efektas). Keturi PAL testo rodikliai: PAL_MTTS, PAL_TEA, PAL_TE6ad, PAL_TTad – rodo donepezilio dozės poveikį pagal visus tris vertinimo kriterijus. PRMd_NC rodo vienos donepezilio dozės efektyvumą tik remiantis reikšmingu pokyčiu AL+ grupėje tarp 1-ojo ir 2-ojo testavimo. Trys PAL testo rodikliai: PAL_FTMS, PAL_SC, PAL_TE8ad – rodo vienos donepezilio dozės efektyvumą pagal reikšmingą testų rezultatų pokyčių skirtumą tarp 1-osios ir 2-osios CANTAB testavimo sesijų AL+ ir AL– grupėse bei reikšmingą rezultatų skirtumą antrosios testavimo sesijos metu tarp AL+ ir AL– grupių.

Norėdami nustatyti, kurie demografiniai, klinikiniai, kognityvinių testų pradinio taško (angl. *Baseline*) ir kognityvinių funkcijų pokyčio po pirmos vaisto dozės rodikliai galėtų prognozuoti ilgalaikio (4 mėnesių trukmės) gydymo efektyvumą, remiantis CANTAB ir MMSE testo rodikliais, konstravome bendruosius tiesinės regresijos (angl. *General Linear Models,* GLM) modelius. Tam tikro CANTAB testo rodiklio pokytis tarp 1-o ir 3-io testų į modelius įtrauktas kaip priklausomas kintamasis. Nepriklausomais kintamaisiais įtraukti amžius, išsilavinimas, pradinio taško MMSE balas (t. y. MMSE balas 1-o testavimo metu), depresiškumas (GDS skalės balai), atitinkamo CANTAB testo rodiklio pradinio taško rezultatas ir šio CANTAB testo rodiklio pokytis tarp 1-o ir 2-o testavimų, nepriklausomais kategoriniais kintamaisiais – lytis ir tiriamųjų grupė (AL+, AL– ar KG). Pažingsninio pašalinimo procedūra atmetė kaip nereikšmingus amžių, lytį, išsilavinimą, pradinio taško MMSE balą, depresiškumą visuose regresijos modeliuose, sudarytuose analizuojamiems

CANTAB testų (PAL ir PRMd) rodikliams. Iš viso analizuoti aštuoni rodikliaiGauti rezultatai pateikiami 1 lentelėje.

1 lentelė. 7 PAL testo rodiklių ir PRMd_NC rodiklio regresijos lygtys, viso modelio determinacijos koeficientas R^2, p – regresijos modelio patikimumas.

Priklausomas kintamasis	Regresijos lygtis	Modelio p	Modelio R^2
PAL_FTMS RS3-1	1,014 + 0,552 × PAL_FTMS RS2-1	0,001	0,34
PAL_MTTS RS3-1	0,318 + 0,397 × PAL_MTTS RS2-1 – 0,159 × PAL_MTTS RS1	0,001	0,31
PAL_SC RS3-1	1,96 – 0,283 × PAL_SC RS1 + 0,262 × PAL_SC RS2-1 + 0,285 × Lytis * tiriamųjų grupė – 0,359 × tiriamųjų grupė	0,001	0,34
PAL_TEA RS3-1	–6,566 + 0,463 × PAL_TEA RS2-1 – 10,568 × Lytis * tiriamųjų grupė	0,001	0,26
PAL_TE6ad RS3-1	2,087 + 0,647 × PAL_TE6ad RS2-1 – 5,839 × Lytis * tiriamųjų grupė – 0,176 × PAL_TE6ad RS1	0,001	0,45
PAL_TE8ad RS3-1	13,80 – 0,367 × PAL_TE8ad RS1 + 9,802 × Tiriamųjų grupė – 4,721 x Lytis * tiriamųjų grupė – 2,505 × lytis	0,001	0,31
PAL_TTad RS3-1	–1,509 + 0,512 × PAL_TTad RS2-1	0,001	0,31
PRMd_NC RS3-1	2,691 + 0,454 × PRMd_NC RS2-1 – 0,288 × PRMd_NC RS1	0,001	0,34

Kognityvinių funkcijų pokytis per 4 mėnesių laikotarpį, t. y. tarp 1-o ir 3-io CANTAB testavimo, atspindi dinamiką viename kognityviniame domene. Norėdami įvertinti atskirų kognityvinių funkcijų ir bendro demencijos sunkumo dinamikos ryšį, įvertinome koreliaciją tarp CANTAB testų rodiklių pokyčio ir MMSE pokyčio.

Nustatyta statistiškai patikima ($p < 0,05$) koreliacija, kurios Pirsono koreliacijos koeficientas $r > 0,5$ tarp PAL_FTMS, PAL_MTTS, PAL_SC, PAL_TEA, PAL_TE6ad, PALTE8ad, PAL_TTad rodiklių pokyčio tarp 1-o ir 3-io testavimų ir MMSE pokyčio tarp 1-o ir 2-o testavimų. Nustatyta statistiškai patikima ($p < 0,05$) koreliacija, kurios Pirsono koreliacijos koeficientas $r > 0,3$ tarp PRMd_NC rodiklio pokyčio tarp 1-o ir 3-io testavimų ir MMSE pokyčio tarp 1-o ir 2-o testavimų.

IŠVADOS

1. Keturių CANTAB testų – PAL, PRM, SWM ir SOC rodikliai – patikimai skiria tiriamuosius, sergančius Alzheimerio liga, nuo kontrolinės grupės asmenų.
2. Naujai diagnozuotų dar nepradėjusių gydymo ir daugiau nei keturis mėnesius besigydančių donepeziliu Alzheimerio liga sergančių tiriamųjų kognityvinės disfunkcijos profilis yra skirtingas.
3. PAL_FTMS, PAL_MTTS, PAL_SC, PAL_TEA, PAL_TE6ad, PAL_TTad, PAL_TE8ad ir PRMd_NC patikimai ir stipriai koreliuoja su bendru demencijos sunkumu, vertinamu MMSE balais.
4. Aštuoni CANTAB testų rodikliai: PAL_FTMS, PAL_MTTS, PAL_SC, PAL_TEA, PAL_TE6ad, PAL_TE8ad, PAL_TTad ir PRMd_NC – patikimai nustato kognityvinių funkcijų pokytį jau po pirmosios donepezilio dozės.
5. PAL_FTMS, PAL_MTTS, PAL_SC, PAL_TEA, PAL_TE6ad, PAL_TTad ir PRMd_NC testų rodiklių pokyčiai, suvartojus pirmą donepezilio dozę, yra patikimi ilgalaikio gydymo donepeziliu efektyvumo prediktoriai; PAL_MMTS, PAL_SC, PAL_TE6ad, PAL_TE8ad ir PRMd_NC rodiklių pradiniai įverčiai yra patikimi ilgalaikio gydymo donepeziliu efektyvumo prediktoriai; lytis kartu su PAL_TE8ad rodiklio pradiniu įverčiu yra patikimi ilgalaikio gydymo donepeziliu efektyvumo prediktoriai.
6. PAL_FTMS, PAL_MTTS, PAL_SC, PAL_TEA, PAL_TE6ad, PAL_TE8ad, PAL_TTad rodiklių pokyčiai patikimai ir vidutiniškai stipriai koreliuoja su MMSE įverčio pokyčiu per keturių mėnesių trukmės gydymo donepeziliu laikotarpį; PRMd_NC rodiklio pokytis patikimai, bet silpnai koreliuoja su MMSE įverčio pokyčiu per keturių mėnesių trukmės gydymo donepeziliu laikotarpį.

PRAKTINĖS REKOMENDACIJOS

1. Detalesniam kognityvinių funkcijų ištyrimui, sergant Alzheimerio liga, rekomenduojama naudoti kompiuterizuotų CANTAB kognityvinių testų bateriją.
2. CANTAB testų baterijos CRT ir SOC testai gali padėti įvertinti dėmesio, psichomotorinių reakcijų greičio ir vykdomųjų funkcijų sutrikimus, kurių įvertinimas MMSE testu yra ribotas.
3. Diagnozavus lengvą ir vidutinio sunkumo Alzheimerio ligos demenciją, skiriamas gydymas AChE inhibitoriais, dažniausiai donepeziliu. Remiantis didelių daugiacentrių AL tyrimų duomenimis, žinoma, kad tik maždaug pusei AL pacientų gaunamas reikšmingas kognityvinių funkcijų pagerėjimas gydant donepeziliu. Kadangi šiuo metu egzistuoja tik simptominis Alzheimerio ligos gydymas, nepriklausomai nuo išankstinių prognozių šis gydymas skiriamas visais atvejais nustačius lengvos ir vidutinės AL diagnozę. Tačiau AChEI vartojimas gali sukelti daug šalutinių reakcijų, todėl, esant nepalankiems simptominio vaisto efektyvumo rodikliams, rekomenduojama dažniau vertinti ligonio kognityvinės būklės dinamiką bei atidžiau stebėti dėl nepageidaujamų reakcijų ir, esant neefektyviam gydymui, rekomenduotina anksčiau spręsti klausimą dėl alternatyvaus simptominio gydymo.
4. Rekomenduojama ambulatoriniame darbe naudoti sutrumpintą baterijos versiją, sudarytą iš PAL ir PRMd testų, nes jų rodikliai yra informatyviausi sergant AL.
5. Naudoti CANTAB bateriją klinikinėje praktikoje rekomenduotina parenkant individualų gydymą konkrečiam pacientui, atsižvelgiant į paciento kognityvinius, klinikinius nekognityvinius ir demografinius ypatumus, t. y. taikyti personalizuotos medicinos principus.

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