**Background**

This report summarizes a longitudinal cohort study following healthy, community dwelling, and cognitively intact elders. Participants were followed up annually and investigators collected information such as gender, age, Hollingshead scale for SES, cognitive assessments, and Clinical Dementia Rating scale to determine Mild Cognitive Impairment (MCI). Short-term this study aimed to define noticeable rates of decline using memory tests and compare to the normal aging process. Long-term the goals are to develop biomarkers for the rate of memory decline. The primary hypothesis for this report were that rate of memory decline will be different for participants with a MCI diagnosis compared to healthy, aging controls. The secondary hypothesis is the rate of decline accelerates four years prior to MCI diagnosis. The statistical hypotheses are as follows: 1) The animal category fluency scores will decline significantly different for MCI cases compared to aging controls. 2) Four years prior to the MCI diagnosis the rate of decline splines and significantly decreases for MCI cases.

**Methods**

There was minimal data management for this report. Univariate descriptive statistics were used to examine variables for outliers, extreme values, and percentage of missing data. Visit count and memory test totals were counted to assist in determining average visits and length of time in the study. Participants that had CDR’s greater than 0.05 two consecutive visits were given an MCI diagnosis and considered an MCI case. Memory rates were determined using scores from the Animal Category Fluency test. Participants that had less than three animal scores were removed from the analysis.

A new age variable was created to assist with interpretability of results. The minimum age of MCI diagnosis was 67 and was used. A change point was created to assess if the rate of memory decline splines four years prior to diagnosis for the MCI group. The *max* function was used to select when the age is greater than zero, and allow for memory rates to spline when MCI cases were at four years before their diagnosis.

Bivariate descriptive statistics were performed to determine demographic differences between MCI groups at first visit and ten years using chi-squared (gender) and independent t-tests (age, SES, average years in study). Spaghetti plots were created to examine the rate of animal test decline per participant as they age by MCI group.

Mixed linear regression analysis was used to differentiate the rates of memory decline by MCI status. The primary explanatory variables in the analysis were MCI status (demind) and age. Gender and SES status were added to the model as covariates and were decided a-priori based on investigator clinical knowledge. An interaction term was to assess the significance between aging and case status. *Proc Mixed* was used to account for multiple observations on each participant.

Examination of spaghetti plots for the animals’ outcome suggested that there were unique intercepts and slopes for each participant. This mixed model was designed with a *random* statement to allow for the variation among subjects and get a more precise standard error without using up degrees of freedom. Restricted maximum likelihood estimates (REML) were used to account for the estimation of the fixed effects. Three models were tested to determine which random effects components were the best fit for the data. Likelihood ratio tests were performed between random intercept, random slope of age, or both to determine the final model. Although REML was used for log likelihood estimates, all three models were nested, had the same fixed effects, and the same data was used allowing for a valid likelihood ratio test. Pearson’s r correlations were used to examine relationships between explanatory variables and covariates. An alpha of <0.05 was considered significant. SAS University Edition Software was used for this analysis.

**Results**

There were 216 participants in the cohort completing 3,385 total observations. 187 participants completed at least 3 animal fluency tests. Table 1 has demographic differences between these participants at first visit and ten years (visits) later. There were 68 participants with two consecutive CDRs greater than 0.05 and categorized as a MCI case. Cases entered the study at an older age (84 yrs [5.94]) compared to controls (77 yrs [8.82]). There were more females (68%) than males in the cases compared to the controls (49%). There were no demographic changes by year five, but by year 10 there were 31 controls that dropped out. At year 10 the controls were younger (82 [7.86]). Graph 2 in the appendix shows that controls were in the study shorter (16.42 [8.27]) and dropped out earlier compared to controls (19.36 [5.54]).

**Table 1:** MCI Group Demographics at first visit and five years

|  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- |
|  | **No MCI** | | **MCI/Dementia** | |  |
| **n=119** | | **n=68** | |
| Variable | Mean | STD | Mean | STD | Pr > |t| |
| Average Visits | 10.74 | 7.18 | 10.96 | 6.76 | 0.4071 |
| Time Followed | 16.42 | 8.27 | 19.36 | 5.54 | 0.0095 |
| Age Onset | N/A | N/A | 90.54 | 4.87 |  |
|  | **First Visit** | | | |  |
| Age | 76.85 | 8.82 | 84.47 | 5.94 | <0.0001 |
| SES | 49.66 | 10.86 | 48.74 | 13.07 | 0.6057 |
| Gender- Male | 59 | 49.58% | 23 | 33.82% | 0.0367 |
| Animal Score | 18.8 | 4.91 | 16.56 | 4.68 | 0.0101 |
|  | **Ten Years** | | | |  |
|  | **n=88** | | **n=68** | |  |
| Age | 82.38 | 7.86 | 89.02 | 5.9 | <0.0001 |
| SES | 48.34 | 11.21 | 48.73 | 13.06 | 0.8397 |
| Gender- Male | 43 | 48.86% | 23 | 33.82% | 0.0594 |
| Animal Score | 18.86 | 4.89 | 14.25 | 6.19 | <0.0001 |

Graph 1 shows the spaghetti plot of each individual’s animal memory score as they age. The graph identifies a downward trend with age, but the MCI group (1-blue) has average lower scores compared to controls (0-red). Additionally, the graph shows that MCI cases entered the study at an older age.

**Graph 1:** Spaghetti Plot of Animal Memory Scores with Age and MCI status

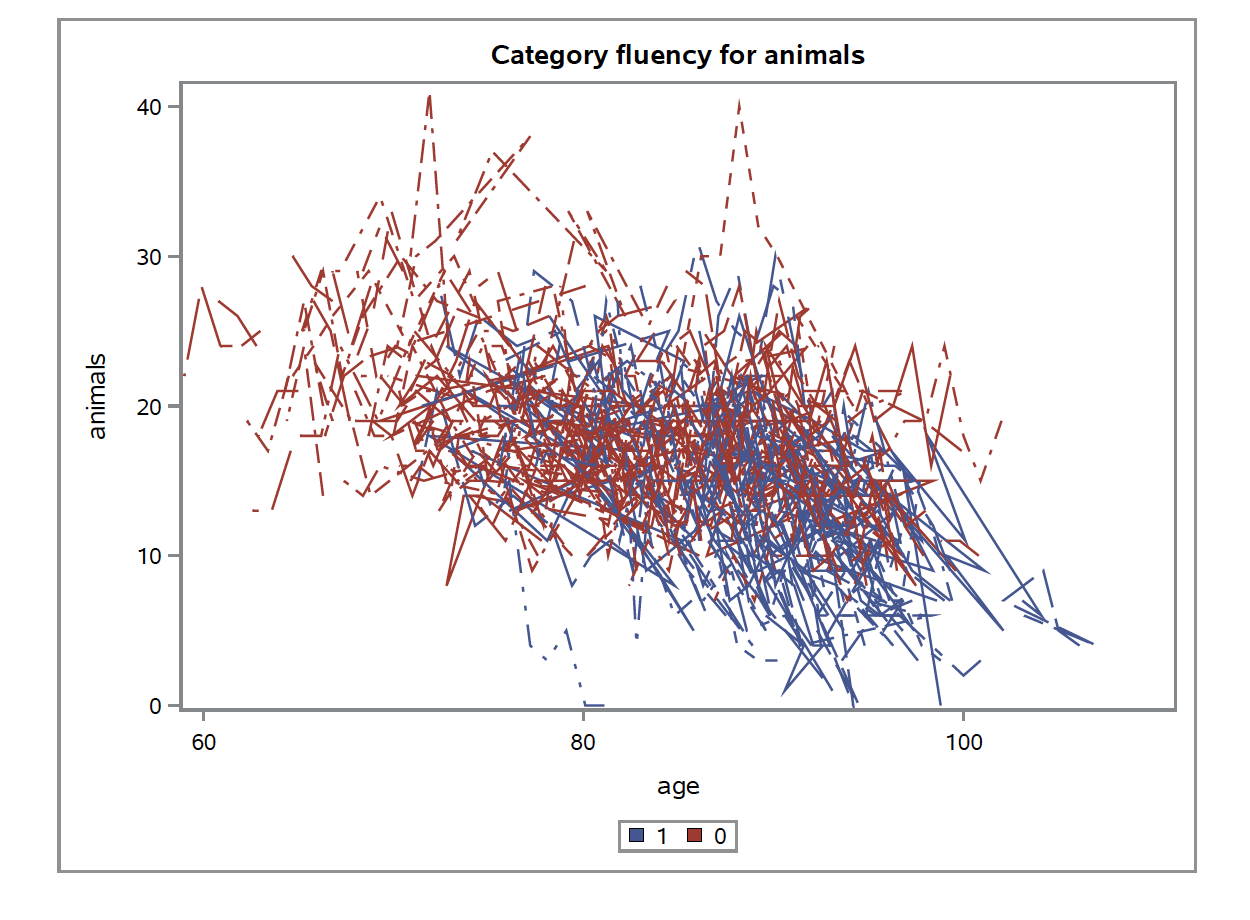
****

Table 2 presents the fixed effects from the analysis. The intercept is interpreted as: the average animal fluency score for male controls at age 67 is 19.12 (1.42; p=<0.0001). As participants aged (every unit of age) there was an average 0.18 (0.03) decline in animal scores (p=<0.0001). There was no significant effect of gender (p=0.3325), but for every unit increase in SES status there was a 0.05 (0.02) increase in scores (p=0.0462).

MCI cases scored 1.16 (1.42) less than controls when holding other characteristics constant, but this difference between groups was not significant (p=0.4552). Furthermore, there was not a significant interaction between aging and case status (p= 0.7201). There was a significant acceleration in decreasing scores for MCI cases at four years prior to diagnosis (p= <0.0001). For every unit of age starting four years before age of onset scores decreased an additional 0.96 (0.1027). Pearson’s r correlations showed MCI case status is strongly correlated with the four-year change point (r=0.64; p=<0.0001). Log likelihood ratio tests found a full model with random intercept and slope to be more significant than a random intercept only model (p=<0.0014).

Table 2: Fixed Effects

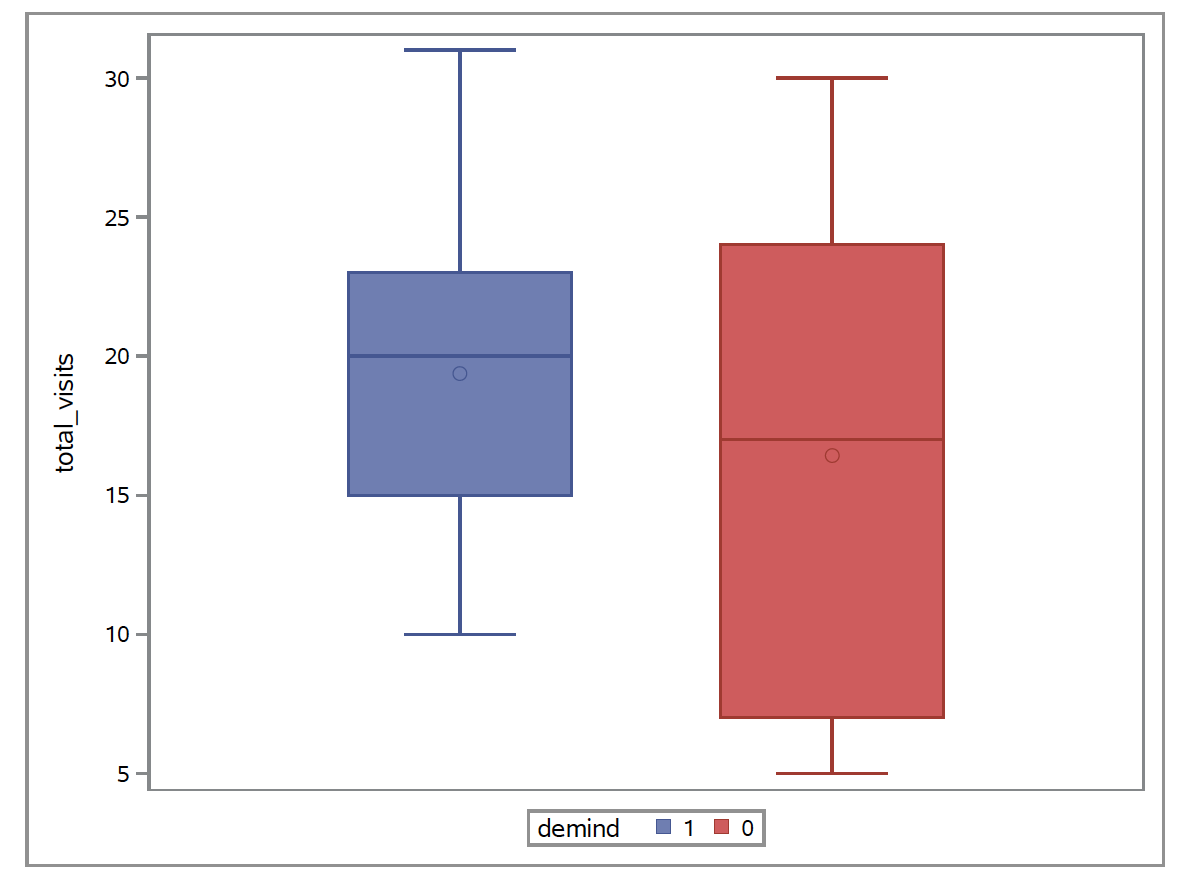
|  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- |
|  | Estimate | SE | Lower CI | Upper CI | P-value |
| Intercept | 19.12 | 1.42 | 16.31 | 21.92 | <0.0001 |
| Demind (Yes) | -1.17 | 1.56 | -4.23 | 1.90 | 0.4552 |
| Age | -0.18 | 0.03 | -0.24 | -0.12 | <0.0001 |
| Demind\*Age | 0.03 | 0.08 | -0.13 | 0.19 | 0.7201 |
| Change Point (4 yrs) | -0.96 | 0.10 | -1.16 | -0.76 | <0.0001 |
| SES | 0.05 | 0.02 | 0.00 | 0.09 | 0.0462 |
| Gender (Female) | -0.55 | 0.57 | -1.67 | 0.57 | 0.3325 |

**Discussion**

As participants age, the average rate of memory decline on animal fluency tests was 0.18 (0.03). There appeared to be a protective effect for participants with higher SES. There was no significant difference between MCI groups or an interaction between aging and case status suggesting that the rate of memory decline is the same for MCI cases and controls. However, there is a significant acceleration for MCI cases in which scores begin to decline an additional 0.96(0.10) four years prior to MCI onset. This is consistent with the secondary hypothesis that four years prior to onset there is a significant decrease in aging adults. There could be multicolinearity with the 4-year change point and MCI status because the two variables were strongly correlated. This would explain why the cases status variable and interaction was not significant. The relationship could be masked because the change point explains more of the relationship between MCI and animal scores.

Limitations in this analysis included 1816 missing animal outcomes. Although the analysis included only participants with three or more outcomes, some participants had only 6 measurements while some had up to 16 measurements. Another limitation was that healthy controls dropped out of the study sooner and were on average younger than the cases. The average age of onset was 90 years and it could be that controls dropped out of the study before onset of MCI. Further studies should attempt to recruit and maintain aging participants that meet an expected age range.

**Appendix**

**Graph 2: Total visits by MCI group**