ABSTRACT

Title of Document: CARDIOVASCULAR FITNESS MODIFIES THE RELATIONSHIP BETWEEN GENOTYPE AND NEUROCOGNITIVE FUNCTION DURING EXECUTIVE CHALLENGE IN LATE ADOLESCENCE

Minjung Woo, Doctor of Philosophy, 2008

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Cardiovascular fitness and physical activity have been positively associated with executive cognitive functioning (i.e., planning, scheduling, coordinating, response inhibition, and working memory), which rely on the frontal region of the brain. Recent studies suggest that the benefit is particularly strong in middle-aged individuals who carry the Apolipoprotein (ApoE) e4 allele, a known genetic risk factor for Alzheimer's disease (AD). However, there have been no studies to determine this interactive relationship in adolescents. Therefore, the present study examined if cardiovascular fitness mediates the relationship between genotype and cerebral cortical responses in college-age males during a frontally-mediated executive challenge. Twenty nine e4 carriers (N=29; 15 high-fit, 14 low-fit) and thirty non-carriers (N=30; 15 high-fit, 15 low-fit) were stratified by cardiovascular fitness. Cognitive function was assessed by neuroelectric response, event-related potentials (ERPs) recorded at 11 sites (F3, Fz, F4,

C3, Cz, C4, P3, Pz, P4, O1 and O2) to both an auditory Go-nogo executive task (ECF) and a non-executive Oddball task (non-ECF). The P300 amplitude, which is indicative of the recruitment of attentional resources, exhibited by the high-fit e4 carriers was higher relative to that observed in the low-fit e4 carriers during both the ECF and non-ECF tasks. Importantly, the high-fit e4 carriers were also undifferentiated from both groups of the non-carriers. Furthermore, high-fit individuals, regardless of genotype, exhibited shorter P300 latency than did the low-fit individuals at sites Fz, Cz and Pz during ECF task and site Pz during non-ECF task. The current findings revealed genetic specificity in the relationship between cardiovascular fitness and the brain processes indexed by P300 amplitude function during late adolescence in response to both ECF and non-ECF challenge, with greater benefit incurred for the ECF task. The results suggest that cardiovascular fitness in e4 carriers is protective against the susceptibility to the liabilities (i.e., hypometabolism and cortical thinning) associated with this allele.

CARDIOVASCULAR FITNESS MODIFIES THE RELATIONSHIP BETWEEN GENOTYPE AND NEUROCOGNITIVE FUNCTION DURING EXECUTIVE CHALLENGE IN LATE ADOLESCENCE

By

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Dedication

This dissertation is dedicated to my Mother (Kyungsook Yoon) and Father (Dr. Sungkoo Woo). Without their continuous encouragement, the pursuit of this degree would not have been possible.

Mom and Dad, this is for you.

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My son, Jihoon Woo Ahn, you are the reason why I live.

Table of Contents

List of Tables

List of Figures

INTRODUCTION & REVIEW OF LIFERATURE (Chapter 1 & 2)

MANUSCRIPT (Chapter 3)

CHAPTER 1: INTRODUCTION

Continued brain development after puberty

The second decade of life is marked by major cerebral cortical and neuro-cognitive reorganization. In particular, the prefrontal cortex has been shown to continue to myelinate through the early adult years (i.e. until 25 years of age) (Giedd et al., 1999; Huttenlocher, 1990). Additionally, synaptogenesis in prefrontal cortex during childhood and at puberty was followed by a plateau phase and subsequent elimination and reorganization of prefrontal synaptic connections is accelerated between the early 20s and up to the age of 30 (Sowell et al., 2003). This synaptic pruning reflects CNS developmental processes such that non-essential connections are eliminated while essential ones are strengthened (Pfefferbaum et al., 1994). Accumulated neuroimaging studies have supported that the prefrontal cortex is the last brain region to mature in the course of development (Sowell, Thompson, Tessner, & Toga, 2001; Sowell et al., 2003). Complex interconnectivity of prefrontal cortex with a number of brain structures result in late development in prefrontal cortex, which is responsible for highly integrative cognitive functions, such as executive control function (ECF) (Fuster, 2001). Executive function, which heavily relies on prefrontal lobe has been shown to continues to develop until early adulthood (Luna et al., 2004, Mackinlay, Charman, & Karmiloff-Smith, 2003). Given the protracted period of plasticity in the frontal lobe, as compared to other brain regions, the impact of environmental stimuli on this brain region is greater during this critical period relative to after maturation of brain is completed. Accordingly, the development of adult-like executive cognitive function, heavily dependent on the frontal

lobe, is expected to be strongly influenced by environmental factors such as physical activity, nutrition, and family size while brain continues to develop.

Physical activity and frontal lobe

Regular participation in physical activity is associated with a variety of mental health benefits. Exercise increases brain-derived neurotrophic factor (BDNF) and nerve growth factor (NGF) that mediate short and long-term enhancement of synaptic strength and reduce cell death in the hippocampus (Cotman & Berchtold, 2002). Furthermore, aerobic exercise increases cerebral blood flow, the number of capillaries and capillary density in brain regions (Black, Isaacs, Anderson, Alcantara, & Greenough, 1990; Rogers, Meyer, & Mortel, 1990; Swain et al., 2003). Allan et al. (2001) found a significant positive correlation between cell proliferation and the distance run in rats. These neurotrophic, angiogenic and neurogenic effects of exercise on the brain may play a role as a protector and enhancer of cognitive function and central nervous system (CNS) integrity. Indeed, Colcombe and colleagues (2003) demonstrated that cardiovascular fitness ameliorated age-related brain tissue loss in the prefrontal, superior, parietal and temporal cortices, which play central roles in successful everyday functioning. In addition, increased cardiovascular fitness has been shown to improve plasticity of the aging human brain. For example, study participants who are highly fit or aerobically trained showed increased activity in the regions of cerebral cortex thought to be necessary for successful attentional processes. Such change is remarkable after only 6months of exercise training (Colcombe et al., 2004).

Along with structural and functional anatomical changes in the brain, exercise has been shown to improve cognitive function and protect against age-related cognitive decline. Exercise training is particularly beneficial in the improvement of the executive control function (ECF) which is mediated by frontal lobe, relative to speed tasks (RT), visuospatial tasks, or controlled processing tasks in normal aging (Colcombe & Kramer, 2003). The positive relationship between physical activity and a variety of cognitive functions, including perceptual skills, executive function, IQ, academic achievement, math and verbal tests has been exhibited in childhood (Buck, Hillman & Castelli, 2007; Davis et al., 2007; Sibley & Etnier, 2003; Tomporowski et al., 2007). Importantly, the benefit of physical activity was greatest on executive function performance (Buck et al; Davis et al; Tomporowski et al).

However, the benefit of physical activity on neurocognitive function is less clear in the young population, since there have been fewer studies of this population. Although the result of these studies generally revealed no difference in cognitive function between high and low active young adults, Hansen, Johnsen, Sollers III, Stenvik and Thayer (2004) did observed better performance in executive control function (ECF) task in an exercise group with a mean age of 19 years without difference between groups in non-ECF task. This finding clearly showed the importance of the task characteristics with which study participants are challenged. Exclusive benefit of exercise on executive function in children and early young adult is speculated to result from greater plasticity in underdeveloped frontal lobe which executive function is heavily dependent on. The changes at the cortical level in young and healthy populations may not be observable with

behavioral measurement. Therefore, the use of neuroimaging techniques may allow detection of change due to exercise not readily apparent at the behavioral level.

Physical activity and executive function in the developing brain – the psychophysiological approach

Electroencephalography (EEG) is a procedure that measure electrical activity of the brain through the skull and scalp. Event-related potential (ERP) can be reliably measured using the EEG. As the EEG reflects thousands of simultaneous ongoing brain processes, the brain response to a certain stimulus or event of interest is usually not visibly in the EEG. By averaging many dozens or hundred of individual trials, the waveform/pattern of the ERP is able to be distinguished from the background EEG activity. One ERP component that has been widely investigated by psychologists is the P300. The P300 waveform is a positive deflection that peaks in amplitude around 300 ms or more after the presentation of a stimulus. The component can be characterized by both amplitude and latency components. P300 Amplitude is defined as the voltage difference between a prestimulus baseline and the largest positive-going peak of the ERP waveform within a latency range (e.g., 270-650 ms). P300 latency (ms) is defined as the time from stimulus onset to the point of maximum positive amplitude within the latency window (Polich, 1996). The P300 amplitude is sensitive to the allocation of attentional resources during task processing since P300 amplitude is larger when subjects devote more effort to a task leading to the proposal (Wickens, Kramer, Vanasse, & Donchin, 1983). The P300 latency indexes stimulus classification speed, with longer latencies reflecting increased

processing time (Duncan-Johnson, 1981, Kutas, McCathy, & Donchin, 1997, McCarthy & Donchin, 1981).

The use of ERPs has been employed to examine the effects of exercise on brain functions. Consistent with behavioral studies described above, the benefit of regular participation in physical activity has been observed in P300 component with increase in P300 amplitude and reduction in P300 latency in old (Dustman et al., 1990; Hillman, Belopolsky, Snook, Kramer, & McAuley, 2004; Hillman, Kramer, Belopolsky, & Smith, 2006; Hillman, Weiss, Hagberg, & Hatfield, 2002;,McDowell, Kerick, Santa Maria & Hatfield, 2003), young adults (Hillman et al., 2006; Polich & Lardon, 1997), and children (Hillman, Castelli, & Buck, 2005). More specifically, greater P300 amplitude (Bashore, 1989; McDowell et al.; Hillman et al., 2004; 2006) and faster P300 latency (Dustman et al. ; Hillman et al., 2002; 2004; 2006) were observed for high active old adults compared to low active old adults both during executive function task and non-executive functions. In addition, the positive relationship between aerobic fitness and P300 amplitude was also shown in children during non-executive task (Hillman, Castelli, & Buck, 2005). However, the impact of physical fitness on P300 components is controversial in young adults. Young individuals who engage in high amounts of aerobic exercise (> 5 h/week, mean age of 30 years) showed greater amplitude during non-executive function task, relative to individuals who engage in low amounts of exercise (< 5h/week, mean age of 34.7 years) but no difference found on P300 latency (Polich & Lardon, 1997). In contrary, other studies failed to find any relationship between P300 components and physical activity or fitness in young age group during non-executive function task (Bashore, 1989, Dustman et al., 1990, Hillman et al., 2002). The non-significant effect of

physical fitness in young adult is speculated to result from task type employed in the studies. Accordingly, Hillman et al. (2006) examined the impact of physical activity on P300 components in a task-switching paradigm consisting of tasks, which require different levels of executive function (from less to more ECF requirement) in young (mean age of 19.3 years) and old (64.8 years). These authors found that higher physical activity was associated with greater P300 amplitude regardless of task type, but the association between physical activity and P300 latency was found on more challenged task. The P3 latency measure indicated that this processing speed benefit was specific to more challenging task. These effects were statistically equivalent for younger and older adults. Consistent with behavior result described above, this result indicates that the effects of physical activity on P300 components were more pronounced during an ECF task in this young age group. Given that early young adults exhibit greater plasticity in the frontal lobe, the fact that the benefit of physical activity on P300 components was greater during ECF compared to non-ECF task in early adulthood seems to be reasonable. Contrary to a study of Hillman et al. (2006), Scisco, Leynes and Kang (2008) failed to find any interaction between cardiovascular fitness and P300 components during executive control task. This result may be due to measurement specificity (i.e., physical activity in Hillman et al.; cardiovascular fitness in Scisco et al.). The other possible reason may be the age range of participants recruited. Highest age in Scisco et al.'s study was 28. Since brain maturation is completed by late 20s, influence of cardiovascular fitness on matured brain is likely to be less than one on underdeveloped brain.

Collectively, the results of these studies suggest that physical activity enhances neuronal and cortical functioning through various biological benefits (BDNF &

angiogeniesis) thereby enabling increased (improved) allocation of attentional resources and reducing cognitive processing speed during development (childhood and young adulthood). This benefit extends to both non-ECF and ECF domains, while the benefit is particularly pronounced during ECF tasks in young adults. This may be due to greater plasticity in frontal region associated with ECF during early adulthood and stronger benefit of physical activity on executive function. As shown in both behavioral and neuro-cortical studies, being physically active during early adulthood plays an important role for healthy brain development.

Healthy brain development and neurocognitive integrity in later life

The importance of healthy brain development in young adults is underscored by the findings on cognitive integrity, as measured by idea density, in young women (mean age 22 years) who were entering the religious orders as reported by Riley, Snowdon, Desrosiers and Markesbery (2005). More specifically, low linguistic ability in early-life (i.e., mean age of 22 years) has been associated with greater cognitive impairment, lower brain weight, and higher degree of cerebral atrophy in late-life (80 years and beyond). Furthermore, the childhood and adolescent environment is associated with the risk of Alzheimer's disease (AD) (Moceri, Kukull, Emanuel, van Belle, & Larson, 2000). That is, the exposure to enriched environment in young adulthood resulting in healthy brain development appears to reduce the risk of cognitive impairment and neurogenerative disease such as AD in late-life. Such a linkage supports the essential need to promote healthy brain and cognitive development during this critical period of young adulthood. As such, physical fitness may contribute to cognitive reserve at this developmental stage

resulting in delay of cognitive decline in later life. Moreover, the positive effects of physical activity may also help to attenuate the effects of known genetic factors which predispose individuals to hypoactivation of the brain and impairment in cognitive function.

Apolipoprotein (ApoE) and brain development

Apolipoprotein (ApoE) is a gene that has been associated with cognitive impairments in late adulthood. ApoE is expressed in terms of three alleles –e2, e3 and e4. It plays an important role in the uptake of lipids generated after neuronal degeneration and their redistribution to cells requiring lipids for proliferation, membrane repair, or remyelination of new axons. For this reason, neurons synthesize and secrete ApoE in response to injury. ApoE e3 and ApoE e2 are more effective in the normal maintenance and repair of cells by increasing the level of cholesterol. On the other hand, ApoE e4 does not appear to be less as effective and therefore causes the deficiency in cholesterol transport, necessary in the repair of brain injury and age-related degeneration in the brain. Inheritance of one e4 allele has been associated with susceptibility to late onset of AD. Strittmatter et al. (1993) observed that 52% of late-onset AD patients carry at least one e4 allele, while only 16% of age-matched controls carry this allele. The age of onset of the disease was dramatically reduced in e4 carriers (Corder et al., 1993).

Importantly, brain imaging studies have demonstrated abnormality in individuals who carry the e4 allele. Studies employed ERP measurement were found significant difference in P300 components between healthy middle-aged either group carrying e4 allele or positive family history of AD (FH+) and non-e4 carriers or individuals who do

not have family history of AD (FH-). Specifically, Green and Levey (1999) initially demonstrated that abnormal prolongation in the latency of P300 in Pz and Cz during Oddball paradigm in e4 carriers who have FH, while no abnormality found in P300 amplitude. Recently, similar result was observed such that smaller P300 amplitudes and prolonged P300 latency during Oddball paradigm were observed in healthy middle aged FH+ group (Ally, Jones, Cole & Budson, 2006). Both findings revealed that groups at increased risk for developing AD show ERP changes consistent with those observed in patients diagnosed as having AD. These changes were observed in the absence of neuropsychological deficits. However, a study to examine the effect of risk genetic factor of AD on P300 component in young female adults aged 19-21 years failed to find significant difference between e4 carriers and non-e4 carriers (Yu, Lin, Chen, Hong, & Tsai, 2000).

The relationship between APOE and brain activity was examined with fMRI. Specifically, middle-aged non-demented healthy e4 carriers exhibited hypoactivation in the hippocampus, temporal, parietal and prefrontal regions, compared to age-matched counterparts (Reiman et al., 1998). Additionally, the reduction in glucose metabolism in those brain regions over 2 years of period was greater in e4 carriers (Reiman et al., 2001). These results raised the possibility that deficits in brain activity in e4 carriers may occur several years before onset of AD. Thus, Reiman et al. (2004) investigated brain activation in young adults (20-39 years) who carry the e4 allele and found that degraded functional brain begins several decades before the possible onset of dementia. More recently, Shaw and colleagues (2007) observed that the e4 allele is associated with distinct neuroanatomic signatures, identifiable in children mean age of 11 years. Their study

showed that a thinner cortex in the entorhinal, medial temporal and posterior-medial orbitofrontal areas in children who carried e4 allele, relative to non-carriers.

In contrast to previous studies examining neural activity during resting state, greater and more widespread activation of the left hippocampal, prefrontal and parietal regions was observed during a memory task in middle-age e4 carriers (Bookheimer et al., 2000). The increased level and volume of activation in the e4 carriers was interpreted as a compensatory response in which additional brain regions are recruited to perform a cognitive operation. This is consistent with notion of reduction in regional specificity and neural efficacy in e4 carriers. This hyperactivation shown in e4 carriers during memory task was also observed in young age adult. Brain imaging comparing brain activation in e4 carriers and non-carriers during non-verbal memory in college age adult and young adults also supports this compensatory mechanism with e4 carriers exhibiting significantly higher activation in frontal areas and cingulated gyri than non-carriers during memory task (Filbey, Slack, Sunderland & Cohen, 2006; Scarmeas et al., 2005).This evidence suggests that the presence of the APOE e4 allele has physiological consequences before age-related decline that may contribute to risk for Alzheimer's disease. In addition, the brain does not experience E4 allele-related decline passively, but compensates for decline in areas of the brain no longer functioning efficiently by engaging other regionss of the brain (Bruckner, 2004). This finding underscores the importance of the exposure to an enriched environment in childhood and early adulthood while the brain continues to develop, particularly for those with the e4 allele.

Physical activity attenuates the effects of ApoE

Participation in physical activity has been shown to compensate for the harmful effects of carrying the genetic risk of AD in older adults. Schuit, Feskens, Launer and Kromhount (2001) revealed that physical activity modified the link between genetic factors and brain functions in non-demented, healthy middle-aged adults. Specifically, the risk of cognitive decline in physically-inactive e4 carriers was nearly 4 times greater than active carriers, whereas no difference was found in non-e4 carriers. This result implies that e4 carriers are particularly vulnerable to a sedentary life style, and regular participation in physical activity considerably reduces the risk of cognitive decline. Most recently, Deeny (2008) also found greater benefit of physical activity in middle age e4 carriers who showed increased activation in task-relevant brain region, relative to non-e4 carriers during a cognitively challenging task. That is, exposure to an enriched environment such as regular participation in exercise would compensate for cognitive decline and hypoactivation accelerated by this genetic risk factor. However, it is not clear if physical fitness during early adulthood also reduces any cortical abnormality associated with e4.

Statement of the Problem

Physical activity is a major contributor to developing, enhancing, maintaining, and protecting cognitive function and even appears to compensate for harmful genetic risk factors. Physical fitness holds a greater effect on the performance of executive control function (ECF) tasks requiring response inhibition compared to other types of cognitive tasks. Given that the development of the frontal lobe continues until early or young adulthood, the benefit of physical fitness on ECF is expected to be significant in

younger college age adults. Moreover, it is anticipated that even those at genetic risk of AD (i.e. ApoE e4 carriers) may benefit from physical fitness, especially on ECF performance.

Moreover, recent advances in neuroelectrical measure and genetics allow researchers to examine subtle changes in brain processes several decades prior to any potential clinical decline in populations genetically at risk for dementia. Previous investigations have examined the effect of physical activity on brain and cognitive function in non-demented older individuals who are at genetic risk for dementia (carriers of ApoE4). However, no research to date has been conducted on the role of the physical fitness in a young population in mediating the relationship between ApoE and brain function. Therefore, the purpose of the study is to examine if the relationship between ApoE genotype and neuro-cognitive function (cognitive function and cortical activation) is modified by cardiovascular fitness in young healthy adults.

Hypotheses

Hypothesis 1. P300 Amplitude

1.1. High fit e4 carriers will show higher P300 amplitude compared to low fit e4 carriers during cognitive challenge and will be undifferentiated from non-carriers.

1.2. This pattern of amplitude differences will be specific to the frontal region of the brain.

1.3. The pattern of differences outlined in 1.1 will be observed for the executive function task (Go-nogo), but not for non-executive function task (Oddball). These predictions are illustrated in Figure 1.

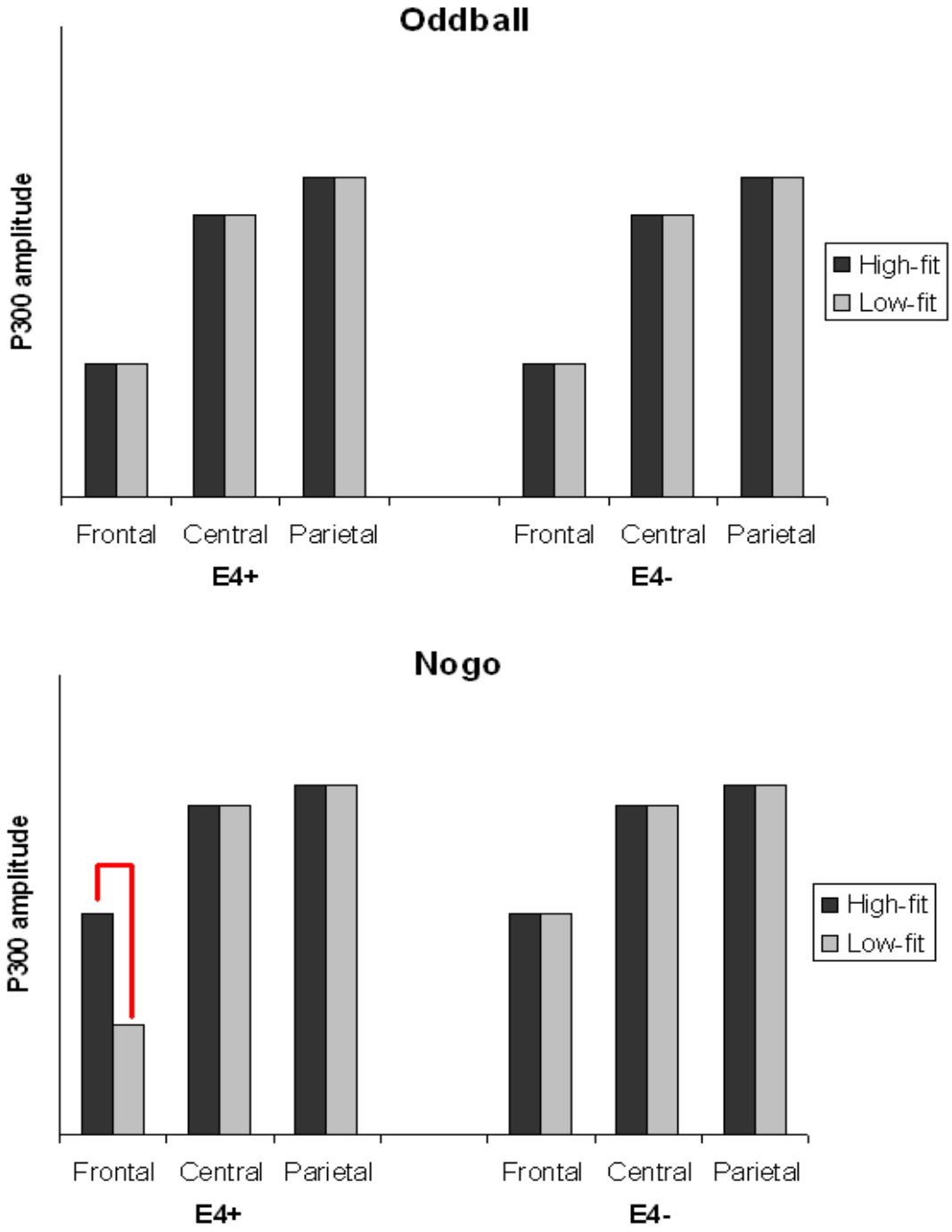


Figure 1. Histograms representing the predicted interactive relationship between cardiovascular fitness and genotype to P300 amplitude in Oddball and Go-nogo tasks

1.4. Cardiovascular fitness will be positively related to P300 amplitude in e4 carriers during executive challenge while no such relationship will be seen in non-carriers.

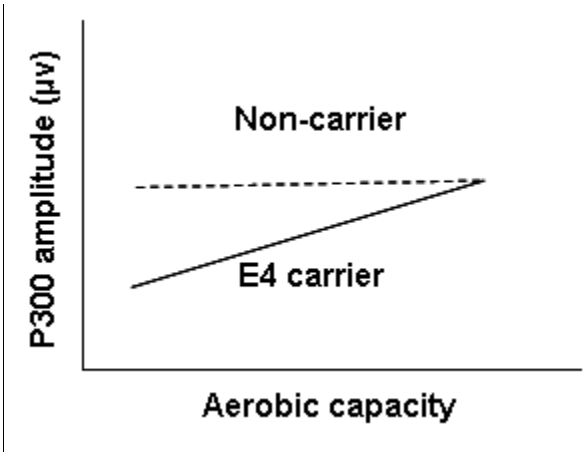


Figure 2. Plot representing the predicted relationship between cardiovascular fitness and P300 amplitude in e4 and non-carriers in response to ECF challenge

Hypothesis 2. P300 Latency

2.1. High-fit e4 carriers will exhibit shorter P300 latency compared to low-fit e4 carriers and will be undifferentiated from non-carriers.

2.2. This pattern of latency differences will be specific to the frontal region of the brain.

2.3. The pattern of differences outlined in number 1 will be observed only for the executive function task (Go-nogo). These predictions are illustrated in Figure 2.

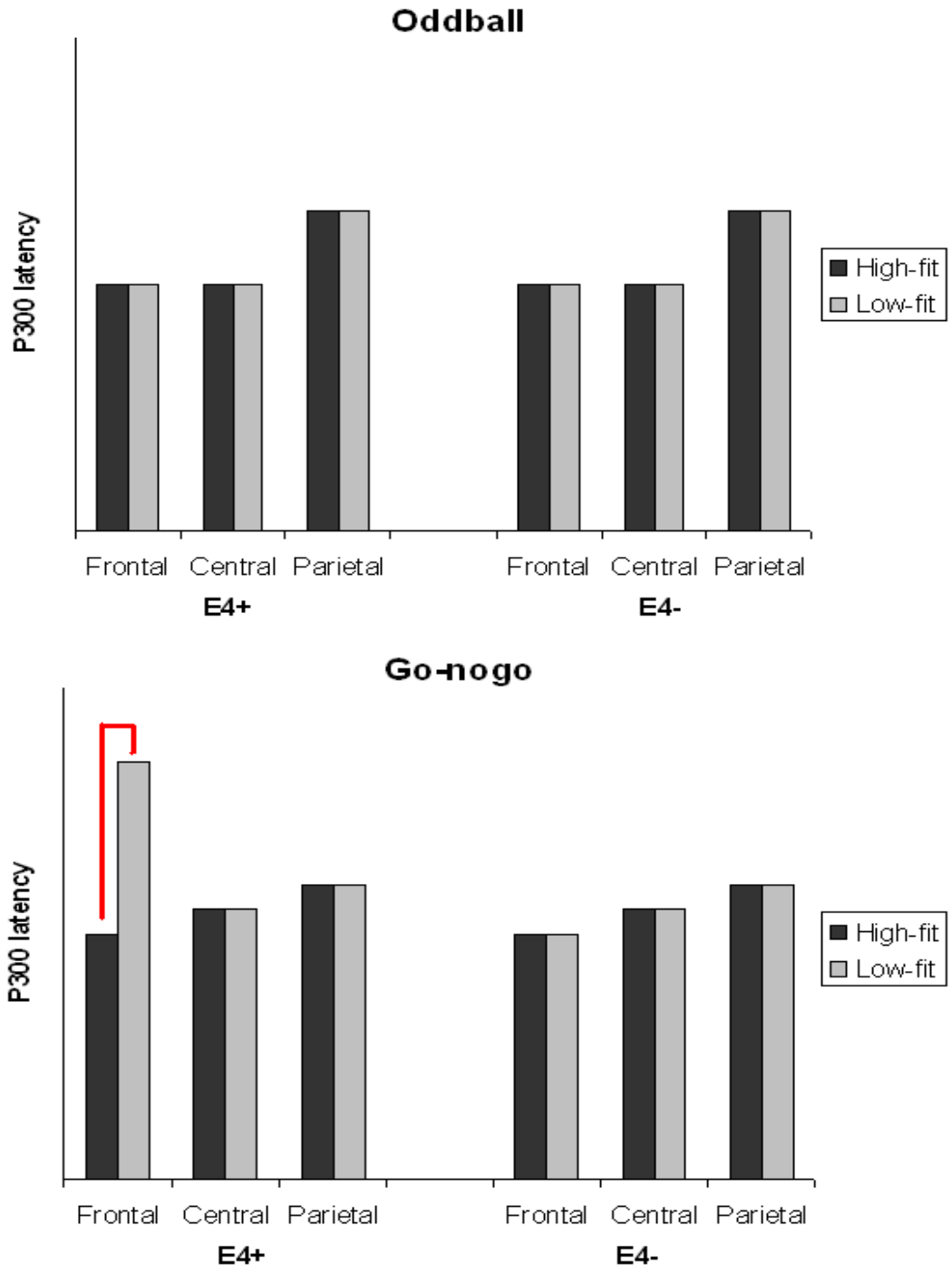


Figure 3. Histograms representing the predicted interactive relationship between cardiovascular fitness and genotype to P300 latency in the Oddball and Go-nogo tasks

2.4. Cardiovascular fitness will be negatively related to P300 latency during executive challenge while no such relationship will emerge in non-carriers

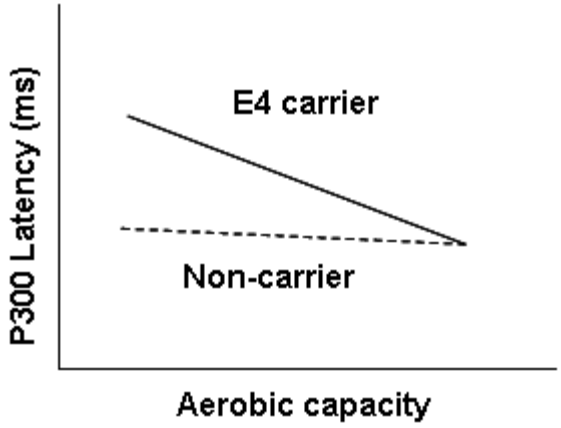


Figure 4. Plot of predicted relationship between aerobic capacity and P300 latency in e4 and non-carriers in response to ECF challenge

Hypothesis 3. Behavioral data

3.1. Percent correct response during executive and non-executive challenges

High-fit subjects will show superior performance relative to low-fit participants during executive challenge and the magnitude difference will be greater in e4 carriers. No differences are expected between groups during non-executive challenge.

3.2. Reaction time during non-executive challenge

There will be no difference in reaction time for non-executive function task between groups (high-fit e4 carriers, low-fit e4 carriers, high-fit non-carriers, low-fit non-carriers).

3.3. Stroop interference score

High-fit subjects will show superior performance relative to low-fit participants and the magnitude difference will be greater in e4 carriers.

CHAPTER 2: REVIEW OF LITERATURE

Brain development

The second decade of life is marked by major cortical and cognitive reorganization. Especially, during late childhood through adolescence, brain structure and functioning are fundamentally transformed. In particular, the development of the frontal lobe has been examined by psychologists, since the prefrontal cortex, which is connected to a variety of brain structure is responsible for higher-order cognitive function, and is the last area to reach maturation in human cerebrum.

Frontal lobe and executive function

The frontal lobe, the largest of the cerebral lobes, lies superior to the lateral sulcus and the anterior to ventral sulcus and consists of prefrontal and premotor areas. The premotor area is involved in voluntary movement, whereas the prefrontal lobe has been implicated in planning complex cognitive behaviors, personality expression, and moderating adaptive social behavior.

The prefrontal cortex lies anterior to the motor and premotor areas, and can be divided into orbitofrontal (OFC), ventormedial (vm-PFC) and dorsolateral cortex (DL). Especially, the dorsolateral prefrontal cortex (DL-PFC) is connected to numerous brain regions, which include the thalamus, parts of the basal ganglia (the dorsal caudate nucleus), the hippocampus, and primary and secondary association areas of neocortex, including posterior temporal, parietal, and occipital areas (Procyk & Goldman-Rakic, 2006). Thus, DL-PFC serves as the highest cortical area responsible for sensory and

mnemomic information, the regulation of intellectual function and executive function control. Executive function relates to abilities that reserve competing thoughts, determination of good and bad, discrimination of better and best, future consequences of current activities, working toward a defined goal, prediction of outcomes, expectation based on actions, and social "control" (the ability to suppress urges that, if not suppressed, could lead to socially-unacceptable outcomes). This is supported by a study showing dysfunction both in executive control and working memory in DLPFC-impaired individuals (Robertson, Tormos, Maeda, & Pascual-Leone, 2001). This complex connection of prefrontal cortex with a number of brain regions may be responsible for the late development of this brain region, especially dorsolateral prefrontal cortex (DLPFC).

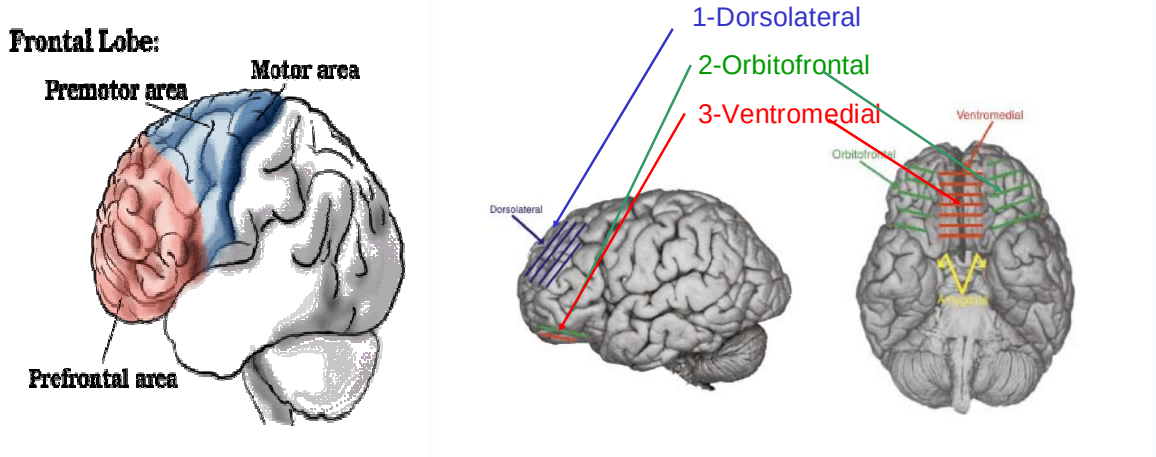


Figure 5. Anatomy of frontal cortex. The frontal cortex consists of prefrontal and premotor areas. The prefrontal cortex is further divided into orbitofrontal, ventromedial and dorsolateral cortex (www.thebrainlabs.com/brain.shtml)

Development of frontal lobe

Sensory and motor systems complete myelination quite early in development by the age of 7 years, but the intra- and inter-hemispheric association fibers and prefrontal

cortex continue to myelinate through the early adult years (Giedd et al., 1999; Pfefferbaum et al., 1994; Sowell, Thompson, Holmes, Jernigan & Tonga, 1999; Yakovlev & Lecours, 1967). Research by Sowell et al. (2003) revealed increased myelin in the frontal lobe white matter of young adults compared to that of teens. Given that the myelination is associated with speed of axonal conduction, the implication of this research is that the transmission velocity of neural information in the frontal cortex should increase throughout adolescence and early adulthood.

Concurrently, another change of the brain that occurs before or after puberty is synaptic density in the prefrontal cortex. Early in postnatal development, the brain begins to form new synapses, so that the synaptic density (the number of synapses per unit volume of brain tissue) greatly exceeds adult levels. Synaptogenesis, which is initially "overproduced" up to several months after birth, is subsequently pruned. That is, unused processes (synaptic connections) are eliminated while active ones are strengthened and their arborization further developed (Changeuz & Danchin, 1976, Purves, 1988). Interestingly, synaptogenesis and synaptic pruning in the prefrontal cortex have shown that there is a proliferation of synapses in the subgranular layers of the prefrontal cortex during childhood and at puberty, followed by a plateau phase and a subsequent elimination and reorganization of prefrontal synaptic connections after puberty (Huttenlocher, 1979; Woo, Pucak, Kye, Matus, & Lewis, 1997; Zecevic & Rakic, 2001).

A number of brain imaging studies have provided further evidence of the ongoing maturation of the frontal cortex into adolescence and even into adulthood. Several Magnetic Resonance Imaging (MRI) studies have been conducted to investigate the development of the structure of the brain during childhood and adolescence in humans

(Paus, 2005; Casey, Tottenham, Liston, & Durston, 2005). One of the most consistent findings from MRI studies is that there is a steady increase in white matter in frontal region during childhood and adolescence (Barnea-Goraly etal., 2005; Giedd et al., 1999; Paus et al., 1999) (Figure 6). This has consistently been interpreted as reflecting continued axonal myelination during childhood and adolescence.

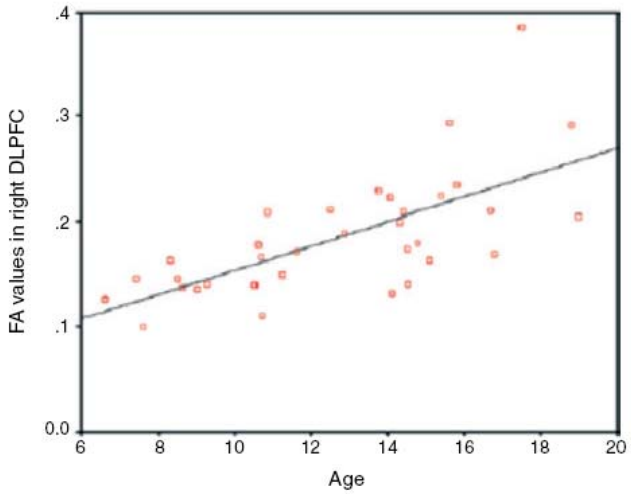


Figure 6. Linear development of white matter with increasing age in the right DLPFC. The graph shows the correlation between age and amount of white matter (Barnea-Goraly et al., 2005)

In contrast to the development of white matter with increasing age, the pattern of development of grey matter in frontal and parietal regions follows an inverted- U shape. A MRI study of 35 normally developing children (7-11 years), adolescents (12-16 years) and young adults (23-30 years) demonstrated a sharp acceleration in the loss of grey matter between childhood and adolescence in the dorsal prefrontal cortex and the parietal cortex (Sowell et al., 2001). In the frontal lobes, the decrease in grey matter density was even more pronounced between adolescence and adulthood. Furthermore, recent MRI studies indicate that the time at which the brain reaches maturity may be much later than the end of adolescence. One such study of participants aged between 7 and 30 years

revealed that the refinement of grey matter in the frontal cortex accelerated during adulthood between the early 20s and up to the age of 30 (Sowell et al., 2001; 2003). In summary, at or after puberty, grey matter volume in the frontal lobe reaches a peak, followed by a plateau until early 20s and then a decline continuing until third decade of life. The non-linear decrease in grey matter in the frontal region throughout early and young adulthood seems to result from increased myelination and synaptic pruning occurring at the onset of and after puberty (Bourgeois, Goldman-Rakic, & Rakic, 1994; Huttenlocher, 1979).

Likewise, accumulated neuroimaging studies suggest that the prefrontal cortex is the brain area to develop most and last in the course of individual development (Giedd et al., 1999; Jernigan & Tallal, 1990; Pfefferbaum et al., 1994; Reiss et al., 1996). Development in other indices of cortical maturation including the prolongation of axons and the arborization of dendrites in the frontal lobe seems also to lag chronologically behind that of other cortical areas (Huttenlocher, 1990; Mrzljak, Uylings, Van Eden, & Judas, 1990; Scheibel, 1990). *Such delayed development of the frontal region offers great opportunity for developmental enhancement from the neurobiological benefit of exercise.*

Development of Executive function

The term executive function is used to describe the capacity that allows one to control and coordinate thought and behavior (Luria, 1966; Shallice, 1982). These skills include selective attention, decision-making, voluntary response inhibition and working memory. Each of these executive functions has a role in cognitive control, for example filtering unimportant information, holding in mind a plan to carry out to execute in the future and inhibiting impulses. Functional imaging experiments suggest that such skills

rely heavily on the frontal lobes (Casey et al., 1997; Rubia, Smith, Brammer, & Taylor, 2003).

The prefrontal cortex has consistently been shown to undergo continued development until early adulthood. Given the continued structural changes in the region during adolescence or early adulthood, it is expected that executive function, cognitive abilities that rely on the functioning of this region and its complex interconnectivity with other regions should also change during this time period. Indeed, behavioral studies revealed that executive function continues to improve during adolescence (Luciana, Conklin, Cooper, & Yarger, 2005; Hooper, Luciana, Conklin, & Yarger, 2004) and until young adulthood (mean age 25) (Luna et al., 2004, Mackinlay, Charman, & Karmiloff-Smith, 2003). Based on these findings and the structural MRI studies discussed above, it was suggested that until pruning occurs after puberty, synaptic connections in the frontal cortex generate a low signal to noise ratio due to an excess of synapses, which renders the cognitive performance less efficient. Therefore, until nonessential proliferated synapses at puberty are pruned by fourth decade, non optimal executive function performance is expected to occur during this time period (Blakemore, & Choudhury, 2006).

Given the protracted period of plasticity in the frontal lobe, as compared to other brain regions, it is highly influenced by environmental stimuli. Accordingly, the development of adult-like executive cognitive function, heavily dependent on the frontal lobes, is strongly influenced by physical activity, nutrition, and family size during the late adolescence and early young age years.

Physical activity and brain

Physical activity and executive function

Regular participation in physical activity is associated with a variety of mental health benefits. The influence of physical activity on brain processes was initially studied in older adults with neuromuscular tasks such as simple reaction time (RT) and finger tapping, since protective effects on the brain would be predicted to be most pronounced in areas engaged during physical movement. Indeed, Spirduso (1980, 1983) provided evidence that physical activity improves RT. Subsequently, some researchers began to examine the effects of exercise on higher-level cognitive processes more susceptible to aging. Some found that aerobic fitness enhances performance in visuospatial tasks (Shay & Roth, 1992, Stones & Kozma, 1989), while others demonstrated that cognitive task type in which the beneficial effect of exercise is particularly strong are those requiring more effortful processing (Chodzko-Zajko & Moore, 1994). Lastly, Kramer, Hahn, and Gopher (1999) proposed that the tasks most protected and enhanced by aerobic fitness would be frontally mediated executive control function (ECF) tasks including planning, coordination, inhibition, and working memory, since ECF is a type of cognitive function on which normal aging individuals have shown the greatest decline (West, 1996). Although a number of studies revealed that exercise improves various types of cognitive functions, there had been no systematic study examining exercise effects on cognition. Accordingly, Colcombe and Kramer (2003) conducted a meta-analysis on exercise intervention studies over the previous 35 years. The results supported Kramer et al.'s hypothesis (1999) by showing greater effect sizes for improvements in cognitive function

on ECF tasks relative to speed tasks (RT), visuospatial tasks, or controlled processing tasks in normal aging (Colcombe & Kramer, 2003).

This relationship between physical activity and cognition was also found in children with meta-analytic reviews of research studies conducted by Sibley and Etnier (2003) in which they identified 44 studies that yielded 125 comparisons for analysis. The overall effect size of 0.32 indicated that physical activity was significantly related to improved cognition in children. The effect of physical activity on cognition was task dependent. Effect sizes were largest for tests of perceptual skills (S=.49), followed by IQ (ES=.34), achievement (ES=.30) and then math test (ES=.20) and verbal tests (ES=.17). *Most recently, clear evidence for a selective facilitation effect of aerobic exercise on children's executive function was obtained in a randomized clinical trial experiment conducted by Davis et al. (2007).* The children were randomly assigned to one of three experimental conditions; no exercise control, 20-min exercise, or 40-min exercise condition. Children participated in physical training games 5 days/week after school. The program consisted of games (e.g., running game, jump rope, soccer) designed to maintain average heart rates of above 150 bpm and to exert a vigorous physical challenge on children. A standardized test of cognitive function, the Cognitive Assessment System (CAS) (Naglieri & Das, 1997), was administered to each child before and after the intervention period. The CAS provides four scales of cognitive functioning: Planning (which assesses executive function; i.e., cognitive control, utilization of processes and knowledge, intentionality and self-regulation), Attention (which assesses focus, selective cognitive activity and resistance to distraction), Simultaneous (which assesses focused, selective cognitive activity and resistance to distraction), Successive (which assesses

processing of sequential information). Analysis of covariance performed on post-test scores revealed that exercise influenced the Planning scale. Children in the high-dose exercise group improved their Planning scale scores significantly more than did children in the control group (ES=0.30). No effects of the exercise intervention were observed on the remaining CAS scales. There were no differences in the CAS performance of children who performed 20 min of daily exercise and those children in the control condition, suggesting that positive effects may accrue only with a large amount of vigorous physical activity. The results suggest that childrens' performance improves exclusively on tests that involve executive function following aerobic exercise training (Tomporowski et al., 2007). In addition, Buck, Hillman and Castelli (2007), examining the relation of aerobic fitness to executive function performance in children between 7 and 12 year of age, also demonstrated that greater aerobic fitness is associated with better inhibitory control. These studies clearly show particular benefit of physical activity and physical fitness on executive function in children.

However, the benefit from physical activity has been less clear in the young adult population since there have been far fewer studies involving young people. In most of studies examining exercise effect, the young adult was used as the purpose of comparison with old adult. The results of most studies failed to show exercise benefit in cognitive function. However, one recent study examining the relationship between aerobic exercise and cognitive function in late adolescence with a mean of 19 years showed better performance in executive function in the exercise group than in the non-exercise group but this enhancement was not observed in the performance of a non-executive function task (Hansen et al., 2004). The result is consistent with previous study exhibiting

improvement of executive function following exercise in children (Davis et al., 2007). Exclusive benefit of exercise on executive function in children and early young adults is speculated to result from greater plasticity in underdeveloped frontal lobe which executive function is heavily dependent on. In contrast, the greatest benefit in ECF shown in old adults may be because frontal lobe is the first to undergo involution with aging, so that there are more rooms to be repaired by exercise. Taken together, exercise is particularly beneficial on development and maintenance of executive function relative to other cognitive functions due to greater plasticity in frontal area during developmental and degenerated period.

Mechanism underlying benefit of physical activity on cognition

Several biological mechanisms underlying protective effect of exercise on brain functioning have been proposed.

Oxygenation and Angiogenesis hypothesis

The level of blood flow in brain regions is associated with the functioning of the brain. Especially, decreases in cerebral blood flow (CBF) to the frontal lobe and hippocampus have been connected to impairment in executive functioning and memory loss with advancing age. This relation has also found in healthy young adults with supplemental oxygen administration significantly enhancing memory function (Scholey, Moss, Neave, & Wesnes, 1999).

Exercise plays a role to increase cerebral blood flow in the brain to enhance cognitive function and protect against age-related decline. This oxygenation hypothesis was initially proposed by Spirduso (1980). Spirduso suggested that increased blood and oxygen supply in brain areas engaged during exercise may contribute to maintenance and

enhancement of cognitive function. The notion of maintained blood flow and oxygenation in the brain was supported in animal studies, which showed increased in blood flow in the motor cortex of rat during exercise, and angiogenesis in the motor cortex and cerebellum of rats following voluntary wheel running (Black et al., 1990, Churchill et al., 2002; Swain et al., 2003).

The oxygenation hypothesis was supported by Rogers et al. (1990) who examined changes in cognition and cerebral blood flow in older adults classified into three discrete groups: 1) currently working; 2) retired, high activity; and 3) retired, low activity over four-year interval. As a result, retirees who elected to become physically inactive exhibited significant declines in cerebral blood flow and cognitive function, while high active group sustained more constant CBF levels and scored better on cognitive testing after the fourth year of follow-up compared to inactive retirees. In conclusion, being active seems helpful to maintain cognitive function by sustaining cerebral blood flow.

Neurotrophic hypothesis (BDNF and NGF level)

This hypothesis suggests that exercise may facilitate production of molecules such as neurotrophic factors that protect neurons, increase neuronal plasticity, enhance learning, and assist in the overall maintenance of the brain. Animal studies by Cotman and colleagues (Cotman & Berchtold, 2002; Cotman & Engessar-Cesar, 2002) supported this hypothesis by showing that physical activity, in the form of voluntary wheel running results in upregulation of brain-derived neurotrophic factor (BDNF), a member of a neurotrophic family that supports the health and functioning of glutamatergic neurons and is known to maintain longevity of cells, protect cells against insults, and facilitate long-term potentiation (LTP) as well as neuro- and synaptogenesis in the brain. The

amount of increase in BDNF was proportionate to the amount of exercise (More exercise, more BDNF). Although the earliest and most sustained increase in BDNF following exercise has been shown in hippocampus, the frontal cortex has also exhibited increase in BDNF. Thus, exercise results in upregulated BDNF levels, which lead to less cortical atrophy in hippocampus and frontal region (Cotman & Berchtold, 2002).

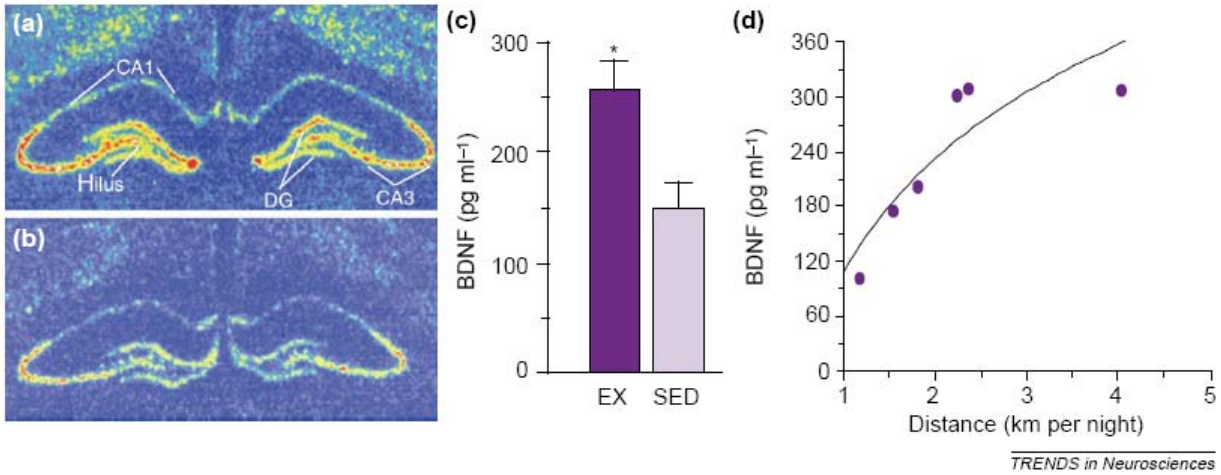


Figure 7. Effects of exercise on hippocampal brain-derived neurotrophic factor (BDNF) mRNA and protein levels. (a) In situ hybridization shows that expression of BDNF mRNA in the rat dentate gyrus (DG), hilus, CA1–CA3 regions and cortex is greater following exercise (seven days of voluntary wheel-running) than in (b) sedentary animals. (c) ELISA quantification of hippocampal BDNF protein levels in the hippocampus in sedentary (SED) and exercising (EX) animals, after five days of wheel-running (*P <0.05). (d) BDNF protein levels correlate with running distance (average over 14 days running; R2 = 0.771) (Cotman & Berchtold, 2002)

Neurogenesis and synaptogenesis hypothesis

Up until quite recently, it was assumed that neurogenesis, or the production of new neurons, occurs only during development and never in the adult organism. However, after initial work showing that the number of new cells in the adult hippocampus can be regulated using a paradigm of environmental enrichment (Kempermann, Kuhn, & Gage, 1997), a number of studies demonstrated that environmental enrichment including increased opportunity for learning, socialization and physical activity facilitates

synaptogenesis. Black et al. (1990) revealed that motor-skill learning such as acrobatic exercise increases the number of synapse in cerebellar cortex. Allan et al. (2001) investigated whether the amount of running influences the number of new cells produced. As the result, there was a significant positive correlation between cell proliferation and distance run. That is, animals that ran the most had the greatest increase in neurogenesis. Exercise-induced neurogenesis and cell proliferation was only found in hippocampus area. Given that the hippocampus is important for learning and memory, elevated neurogenesis at this area may lead to improve cognitive function and delay functional loss which is associated with AD.

Neural efficiency hypothesis

As described above, physical activity and exercise enhance or maintain cognitive function, particularly executive function in all age group (i.e., children, young and old adults). Initially, Dustman et al. (1990) proposed the neural efficiency hypothesis that improved performance accompanying increased aerobic fitness may reflect alteration of basic neurobiological processes. To test this hypothesis, EEG and ERP of young and older groups in poor-fair and excellent-superior fitness categories were examined. The results suggested a positive relationship between neurophysiological performance and aerobic fitness. Increased EEG regional specificity for high-fit subjects was associated with better cognitive and visual function, rapid processing of attention and stimuli detection, and enhanced inhibition control for the high-fit subjects. This result reflects that physical fitness results in more efficient functioning of central nervous system. This exercise effect was shown in both young and old adults group, but the benefit was much stronger in the older group than the younger group. The result raised the possibility that

greater physical fitness facilitate healthy brain development until early adulthood by increasing neural efficiency.

Recently, a brain imaging study examining the impact of physical activity on cortical plasticity in young and old adults also supported that exercise increased cortical efficiency (Colcombe et al., 2004). Specifically, high-active older adults demonstrated significantly greater activation in several cortical regions associated with effective attentional control, than did low-active old adults and showed reduced activation in anterior cingulated cortex (ACC), and area that responsible for error detection. Furthermore, after 6 month intervention of aerobic exercise, exercise group increased activation in task-relevant brain region and reduced task-irrelevant activity with better performance on executive function task. Collectively, these studies suggest that increased cardiovascular fitness enhances cognitive function by improving plasticity of the young and aging human brain.

Healthy young brain and cognitive function of later-life

Physical activity, exercise, and physical fitness have been shown to be a critical environmental factors which are positively associated with cognitive function (i.e., executive function) in all age groups. As described above, physical activity is particularly beneficial while the prefrontal region continues to develop. Healthy development in this critical period has been shown to protect or delay age-related decline and genetically-determined decline in brain by demonstrating the association of early-life factors with later-life development of AD. Specifically, the area of residence before age 18 years and number of siblings were associated with subsequent development of AD. This result implies that the early-life childhood and adolescent environment is associated with the

risk of AD (Moceri et al., 2000). A more recent study examining the relationships between early life variables, cognitive function and neuropathology of the Religious Orders (nuns and priests) supported an importance of healthy development of the young brain. The result was that low early-life linguistic ability at a mean age of 22 years was associated with greater cognitive impairment, lower brain weight, higher degree of cerebral atrophy, more severe neurofibrillary pathology in late-life (Riley et al., 2005). These evidences suggest that healthy brain development appears to be critical to protect against age-related decline in brain in later-life. Therefore, exposure to enriched environment such as exercise during adolescence and early adulthood is likely to reduce risk of development of AD and reduce impacts of an AD-related genetic factor (i.e., APOE e4 allele) on neurocognitive function.

Genetic influence on brain development

The phenotype is determined by multifactorial combination of genetic traits and environmental factors. There is a gene which has been related to cognitive impairment and AD in later-life.

Apolipoprotein (ApoE) and Alzhiemer's disease (AD)

Human Apolipoprotein E (ApoE) is a polymorphic protein 299 amino acids long. Its gene, on chromosome 19, encodes three alleles that differ only at two positions, 112 and 158. The molecular basis for the ApoE polymorphism has been elucidated by amino acid sequence analysis. The major alleles are called e2 which has cynsteines at both positions, e3 having cysteine at 112 and arginine at 158, and e4 having arginine at both positions. These single amino acid sequence changes cause profound difference in the

properties of the isoform. The ApoE gene is inherited as one of three alleles – e2, e3 and e4- with mean frequencies in the general population of about 8%, 78%, and 14%, respectively.

The role of ApoE is to take up lipids generated after neuronal degeneration and redistribute them to cells requiring lipids for proliferation, membrane repair, or remyelinaton of new axons. For this reason, neurons synthesize and secrete ApoE in response to injury. For example, ApoE levels increase 250 to 350 fold in response to peripheral nerve injury in a rat model. However, ApoE e2, e3 and e4 have markedly different effects on neurite extension. ApoE e3 and ApoE e2 are more effective in the normal maintenance and repair of cells by increasing level of cholesterol, but ApoE e4 seems plays an opposite role. ApoE e4 causes deficiency of transport cholesterol, which is known to involve in repairing brain injury and age-related degeneration in the brain. Thus, inheritance of one e4 allele is associated with increased vulnerability of neurons to stress; this may underlie the increased susceptibility of e4 carriers to Alzheimer's disease.

ApolipoproteinE (ApoE) gene has been shown to be associated with susceptibility to late onset AD with 52% of late-onset AD patients carrying ApoE e4 allele while only 16% age-matched controls carrying this allele (Strittmatter et al., 1993). The robust nature of this association was rapidly and widely confirmed in laboratories across the world (Anwar, Lovestone, & Cheetham, 1993, Borgaonkar et al., 1993, Houlden, Crook & Duff, 1993, Noguchi, Murakami & Yamada, 1993, Poirier et al., 1993, Rebeck, Reiter, Strickland & Hyman, 1993). If two e4 alleles are inherited, the risk is increased further and age of onset of AD lowers. In contrast, the e2 allele appears to confer a protective effect with regard to AD risk. ApoE e4 allele has a gene-dose effect on the risk and age

of onset of AD. As the number of ApoE4 alleles increase from 0 to 1 to 2, the risk of developing late-onset familial AD increases from 20% to 90%, and the mean age of onset decreases from 84 to 68 yr (Corder et al., 1993). Therefore, a variation in the ApoE gene has been shown to be a risk factor for AD.

ApoE e4 and brain

The e4 allele of the ApoE gene has emerged as the most robust genetic risk factor for the cognitive decline and development of AD. Early studies to examine the genetic influence on brain have focused on non-demented old individuals who are at risk for AD, as identification of early markers of dementia may facilitate research on interventions to slow the progression of the pathology before onset of any clinical symptoms.

ApoE and brain during resting condition

Initially, Reiman and colleagues (1996) examined whether non-demented ApoE e4 carriers show similar changes in brain to AD patients by observing glucose metabolism via PET in middle- aged (50-65 years) individuals. The result was that non-demented e4 carriers showed hypoactivation in hippocampus, temporal, parietal, and prefrontal regions in which AD patients showed abnormality. Afterwards, Reiman et al. (2001) examined whether there is a significant difference in rate of decline in the brain activity for approximately 2 years between non-demented e4 carriers and non-carriers who are 50-60 years of age with a family history of AD (Reiman et al., 2001). As a result, e4 heterozygote carriers showed a faster decline in glucose metabolism in temporal, posterior cingulated, and prefrontal cortex, basal forebrain and thalamus after 2 years than do e4 non-carriers. The result implies that carrying the e4 allele seems to cause not only abnormality in brain regions, but also accelerates brain degeneration that is related

to AD in middle age individuals. Both studies suggested that brain abnormalities progress several years before onset of AD. However, how early the brain dysfunction occurs was unclear.

Reiman et al. (2004) also examined brain activity in young e4 carriers who are 20-30 years of age. Interestingly, young e4 carriers had abnormally low CMRgl in the posterior cingulated, parietal, temporal, and prefrontal cortex as observed in middle age e4 carriers. The result implies that carrying the e4 allele causes neural alteration several decades before AD related behavioral signs occur. The rate of decline in CMRgl from young adulthood to late middle age was faster in the e4 carriers than in the controls. Taken together, brain degeneration in e4 carriers which began in young adulthood appears to progress gradually with aging (Reiman et al., 2004) but the rate of decline become accelerated after middle age (Reiman et al., 2001). Additionally, these results raised possibility that functional brain abnormalities in e4 carriers may begin earlier than young adulthood.

Most recently, to address this possibility, a study was conducted to examine whether possession of an e4 allele would confer children with a neural substrate that might render them at risk for AD. The result was that e4 carriers exhibited thinner cortex in the entorhinal, medial temporal and prefrontal area than did non-carriers. The rate of change in thickness in the entorhinal cortex between ages 8 to 20 years was not different by genotype. The result implies that the thinner cortex in e4 carriers could represent a genetically determined neuroanatomic property (Shaw et al., 2007). That is, possession of an e4 allele could be associated with a cortical endophenotype, characterized by a thinner

cortex, which seems to be cognitively silent in childhood, but could render individuals more prone to the later development of Alzheimer's disease.

ApoE and brain activation during task performance

In contrast to previous studies, Bookheimer and colleagues (2000) observed greater and more widespread activation of the left hippocampal, prefrontal and parietal regions during a memory task in older e4 carriers. Bookheimer et al. interpreted the increased level and volume of activation in the e4 carriers as a compensatory response in which additional brain regions are recruited to perform a cognitive operation. This is consistent with the notion of reduced neuro efficiency e4 carriers. This hyperactivation shown in e4 carriers during the episodic memory task was also observed in young adults. Brain imaging study comparing brain activation in e4 carriers and non-carriers during non-verbal memory in college age adult also supports this compensatory mechanism (Scarmeas et al., 2005) with e4 carriers exhibiting significantly lower or higher activation than non-carriers during memory task depending upon brain region assessed. A study examining difference in neural activity between young healthy e4 carriers (age=26 ± 3 years) and non-carriers (age=25 ± 4 years) with functional magnetic resonance imaging and magnetoencephalography also showed greater activity in frontal areas and cingulated gyri while performing a visual working memory task (Filbey, Slack, Sunderland, & Cohen, 2006). These evidences suggest that the presence of the APOE e4 allele has physiological consequences before aging that may contribute to risk for Alzheimer's disease. In addition, the brain does not receive E4 allele-related decline passively, but compensates for decline in areas of the brain no longer functioning efficiently by engaging other areas of the brain (Bruckner, 2004). As described earlier, given that the

phenotype is determined by a multifactorial combination of genetic traits and environmental factors, exposure to enriched environment in early adulthood during which highest cortical areas continue to develop is expected to slow or protect negative impact of genetic factor.

Physical activity and genetic risk of AD

Angiogenesis, neurogenesis, synaptogenesis and increased oxygenation and neurotrophic factors resulting from exercise is expected to ameliorate brain abnormalities and cognitive decline which is characterized in group who carry ApoE e4 allele. Schuit et al (2001) examined whether the association between physical activity and cognitive decline differs between individuals at high risk and low risk of AD. A study conducted in the Netherlands tested basic cognitive function of 60 to 70 year old people over the course of 3 years with Mini Mental State Examination (MMSE). As a result, cognitive impairment was significantly higher in low-active e4 carriers than high–active e4 carriers and non-carriers regardless of the level of physical activity. Furthermore, the risk of cognitive decline in inactive carriers of the ApoE e4 allele was nearly 4 times bigger relative to active carriers. However, there was no significant difference between inactive non-carriers and active non-carriers. This result implies that the negative impact of low activity is particularly strong in individuals carrying the e4 allele, but the risk of cognitive decline was considerably reduced in ApoE e4 carriers who are active more than 1 hour a day. That is, exercise would compensate for cognitive decline that is accelerated by carrying a genetic risk factor.

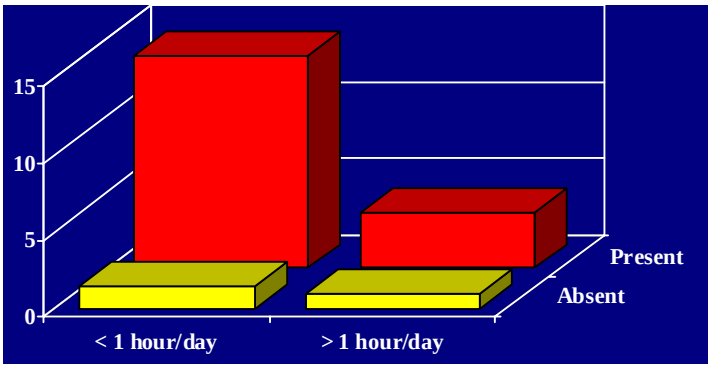


Figure 8. Risk of cognitive decline in physically active and inactive group in ApoE e4 carriers and non-carriers (Red bar indicates e4 carriers and Yellow bar indicates e4 non-carriers). The risk of cognitive decline is considerably reduced in e4 carriers who are physically active (Schuit et al., 2001).

Most recently, Deeny (2005) and Deeny et al. (2008) examining this relationship with magnetoencephalographic (MEG) measures of cortical activation during executive function task and memory task found that highly physically active e4 carriers and non-carriers who are 50-70 year of age and free from dementia exhibited greater cortical activation in task-relevant regions both during ECF and memory tasks relative to their low-active counterparts. On working memory task, high-active e4 carriers exhibited greater activation than low-active e4 carriers in the right temporal region, while being undifferentiated from both the high active and low active non e4 carriers. All groups performed similarly on accuracy and reaction time in both tasks. The findings support the notion that populations genetically at risk for dementia who remain sedentary may be at greater risk for decline in brain function relative to those who are physically active.

Although Schuit et al. (2001) and Deeny (2005) found a strong relationship between physical activity and brain in individuals at high risk of AD, Podewils et al. (2005) failed to demonstrate this relationship in e4 carriers. Podewils et al. (2005) examined the relation of physical activity (PA) and dementia by using a large,

community-based cohort of non-demented 65 years or older adults who carry e4 or not with MRI and cognition assessment. As the results, the inverse association of energy expenditure and activity index with dementia risk was limited to ApoE e4 non-carriers. They interpreted this result as any potential protective effect associated with physical activity is not enough to overcome the effect of ApoE e4 alleles, or physical activity and dementia are simply unrelated in persons with e4 genotypes

Event Related Potential (ERP)

Neuroelectric measures can provide direct imaging of central nervous system (CNS) function. Indeed, electroencephalography (EEG) and event-related potentials (ERP) have been used clinically to assess cortical and sensory function. Especially, ERP provides information regarding neural resources allocation, and topographical and temporal aspects of neural processing in response to a behavioral challenge. ERP components can be broadly divided into exogenous component reflecting presentation of the stimuli and endogenous component representing psychological reaction of the stimuli.

P300

The P300 component out of endogenous components has been widely applied to study neurologic and psychiatric cognitive impairment. The P300 waveform is a positive deflection that peaks 300 ms or more (up to 900 ms) after stimulus. The component can be characterized by both amplitude and latency. P300 amplitude is defined as the voltage difference between a prestimulus baseline and the largest positive-going peak of the ERP waveform within a latency range (e.g., 270-650 ms). P300 latency (ms) is defined as the

time from stimulus onset to the point of maximum positive amplitude within the latency window (Polich, 1996).

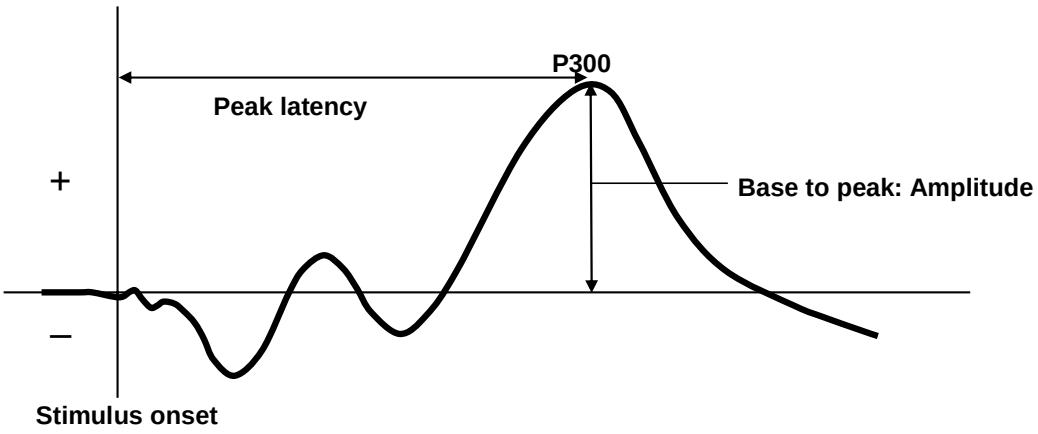


Figure 9. An example of ERP waveform with the component of P300. P300 is a positive deflection that peak around 300 ms after stimlus. P300 latency is the time from stimulus onset to the point of maximum positive amplitude within the latency window**.**

P300 amplitude has been linked with memory processes but is perhaps more sensitive to the allocation of attentional resources during task processing (Wickens et al., 1983). P300 latency indexes stimulus classification speed (Duncan-Johnson, 1981, Kutas et al., 1997, McCarthy & Donchin, 1981). Shorter latencies are associated with superior cognitive performance from neuropsychological test of attention and immediate memory (Polich, 1992, Reinvang, 1999), with increased latency found for normal aging (Fjell & Walhovd, 2001, Polich, 1997). Traditionally, the P300 has been elicited in stimulus discrimination paradigms that manipulate probability such as the "Oddball" task, in which rare target stimuli have to be detected within a train of frequent nontarget stimuli that do not require a response and "go-nogo" task, in which rare target stimuli have to be inhibited to respond within a train of frequent nontarget stimuli requiring responses. Go-

nogo task is executive function task which requires inhibitory control and is heavily dependent on frontal lobe function, whereas Oddball task is non-executive function task that are depend on other areas of the brain rather than frontal lobe.

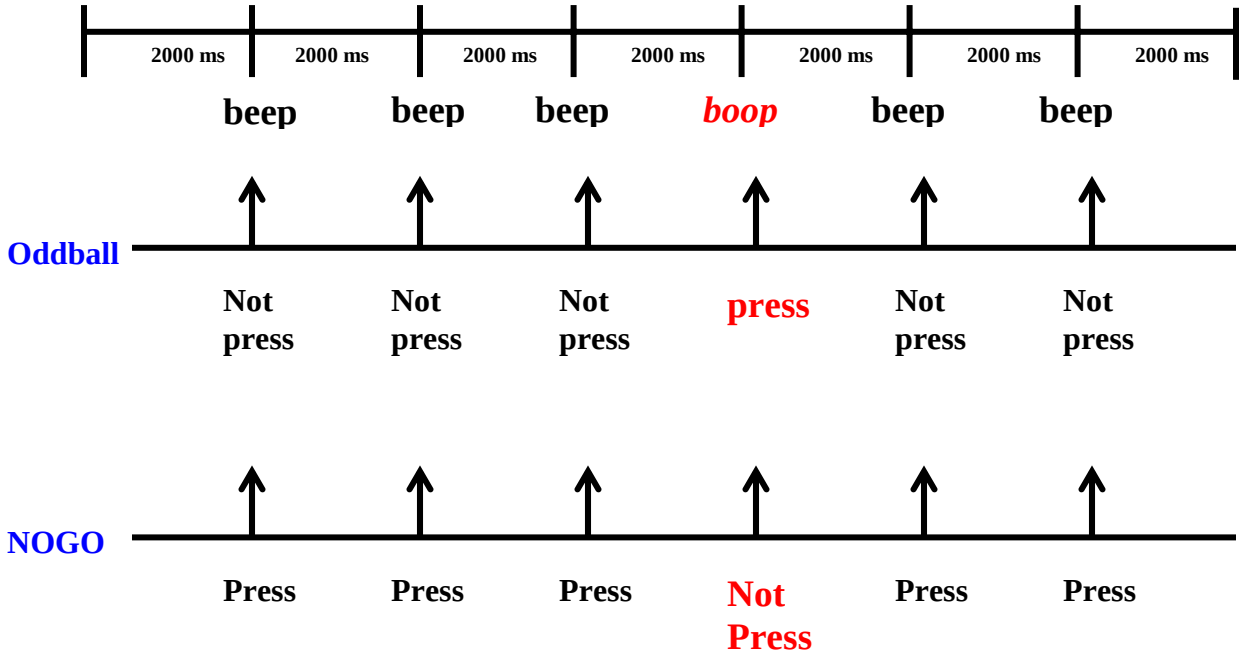


Figure 10. Protocol of Oddball and Go-nogo tasks

These paradigms reliably yield P300 responses with a parietocentral scalp distribution to target compared to standard stimuli irrespective of stimulus or response in young adult. The "hilliness" of the topographic P300 in central and parietal region shown in young adult has been shown to be changed with aging or disease-related brain degeneration. In these protocols, the amplitude and latency of the P300 component indicate the integrity of cortical response to executive challenge and non-executive challenge.

P300 and development

Goodin, Squires, Henderson, and Starr (1978) were the first to suggest that ERPs might be a useful tool for investigating changes in cognitive function and found that the P300 component provides a sensitive neuroelectric index of age-related change. Afterwards, the age-related change in P300 component has been studied extensively across the lifespan in order to evaluate the neurophysiological basis of the changes in cognition that occur from development to aging.

ERP is employed to examine the developmental process of brain and subsequent cognitive processes in preadolescent children exhibited significantly longer P3 latency than adults during Oddball tasks, indicating slower cognitive processing speed due to immaturation of brain (Curry & Polich, 1992; Hillman, Castelli & Buck, 2005). Howard and Polich's study (1985) examining change in P300 latency across age demonstrated that P300 latency gradually decreases until early adulthood. Given that the developmental change in P300 latency is negatively associated with myelination in the brain, longer P300 latency in preadolescent children relative to adults may well reflect that neuro-cognitive function is at a dynamic developmental stage in this age group. Johnstone, Barry, Anderson and Coyle (1996) also examined age-related changes in children's and adolescents' (8 to 18 years of age) event-related potential components in an auditory Oddball task. The result was that age was negatively related to P300 latency at Fz and Cz.

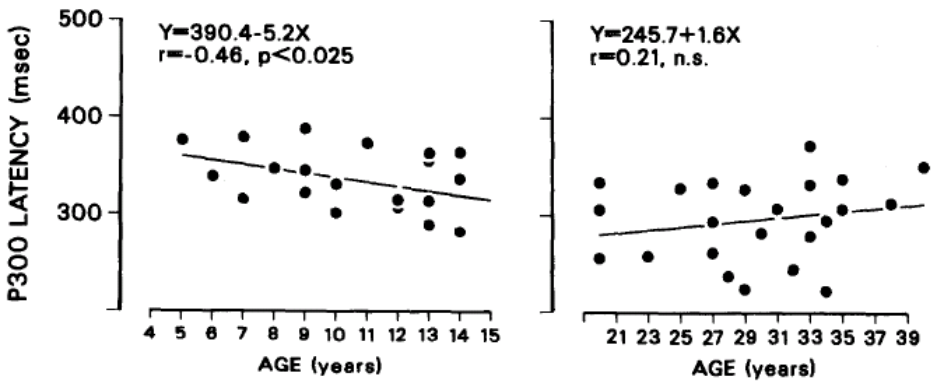


Figure 11. Scattergrams and regression analyses of P300 latency and age for children and adult subjects. The negative directional relationship between age and P300 latency has been found until preadolescence but the direction of this relationship become positive after 21 years of age. This figure is taken from Howard & Polich (1985).

However, the findings regarding P3 amplitude are less clear, though with results indicating larger (Batty & Taylor, 2002), smaller (Johnstone, Barry, Anderson & Coyle, 1996) or no difference (Curry & Polich, 1992) of amplitude in children when compared with adults. Accordingly, the relationship between development and P3 amplitude in response to cognitive tasks remains unclear. These controversial results may be related to differences in maturation, characteristics of the study sample (population specificity), or the cognitive task employed (task specificity). Despite this lack of consensus regarding P3 amplitude, robust behavioral differences have been observed, with preadolescents exhibiting longer reaction time and decreased response accuracy compared with adults.

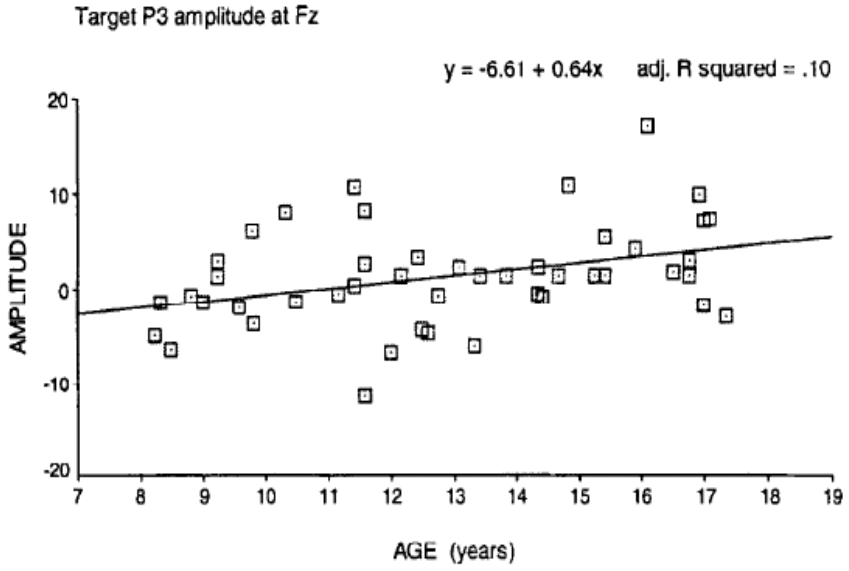


Figure 12. Scattergram of P300 amplitude at Fz elicited by target stimuli as a function of age (Johnstone, Barry, Anderson, & Coyle, 1996)

P300 and aging brain

A number of researchers examining the relationship between P3 components and aging demonstrated robust age-related differences for P3, with older adults exhibiting decreased amplitude and increased latency (Dustman et al., 1990, Hillman et al., 2004; Polich, 1997). Although there is an apparent discrepancy in the literature regarding rate of increase in P300 latency with aging, the overall agreement is a support of age-related latency increases (Beck, Swanson & Dustman, 1980, Brown, Marsh & LaRue, 1983). Additionally, the hilliness of the topographic P300 in central and parietal region which is characterized by young adults is reduced with advancing age, resulting in a more frontally oriented scalp distribution during task performance (Dustman et al., 1990; Hillman, Weiss, Hagberg, & Hatfield, 2002; Hillman et al., 2004; Polich, 1997,).

Decreased amplitude appears to imply reduced neuronal activity resulting from age-related neurogeneration or neuron death, rather than reduced amount of attentional resources. Slower neuronal communication throughout reduced myelination may be responsible for increased latency. Reduced regional specificity (greater equipotentiality of the scalp distribution) may be a compensatory mechanism to make up reduced neuronal activity for completion of task. Genetic factors which have been associated with cognitive impairment such as ApoE e4 allele may accelerate the age-related change in P300 components and scalp distribution.

P300 and ApoE

P300 component has also been associated with heredity. Green and Levey (1999) initially used ERP measurement to examine influence of genetic risk factor of AD on brain in non-demented, middle-aged individuals. The result was that group carrying e4 allele and positive family history of AD showed abnormal prolongation in the latency of P300 in Pz and Cz, while no abnormality found in P300 amplitude. A study examined P300 in AD patients and biological children of patients of Alzheimer's disease (AD). The result was that as predicted, amplitude in AD patients was significantly smaller compared to age- and gender-matched control group. Both amplitude and latency in offspring of AD patients (FH+) were significantly impaired when compared to its age- and gender-matched control group. Interestingly, FH+ group demonstrated similar P300 amplitudes to the older control group. That is, P300 amplitude in participants with a family history of AD and a mean age of 54 is comparable to participants with a mean age of 75 and no family history of the disease. This similarity in P300 amplitude may lead one to draw the hypothesis that participants with a family history of AD are prematurely aging (Ally,

Jones, Cole, & Budson, 2006). Both findings indicate that participants with a family history of AD demonstrate possible preclinical phase of the disease more than 20 years prior to its clinical presentation at the electrophysiological level. The findings raise the possibility that assessment of ERPs may contribute to early detection of AD.

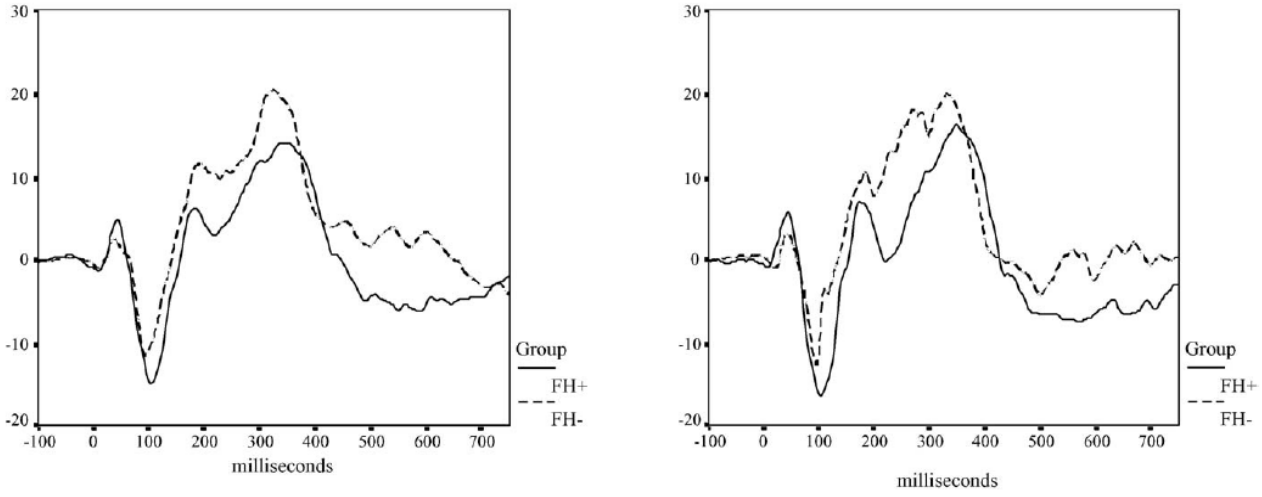


Figure 13. ERP waveforms at Fz (left) and Cz (right) for the group of family history of AD (FH+) vs FH- (Ally, Jones, Cole, & Budson, 2006)

However, a study to examine the effect of genetic risk factor of AD on P300 component in young female adults aged 19-21 years failed to find significant difference between e4 carriers and non-e4 carriers (Yu, Lin, Chen, Hong, & Tsai, 2000). This may be because brain abnormality shown in young e4 carriers may be too little to be observed with ERP assessment. The brain abnormality is outcome of interaction between environmental factors and genetic factors. Thus, consideration of environmental factors affecting brain function in study design may increase sensitivity to detect brain abnormality in young e4 carriers with neuroelectric measurement.

P300 and Physical activity

Neuroelectric function has been known to be influenced by external factors. For example, higher intelligence has been related to faster P3 latency, and children with deficit/hyperactivity disorder have shown greater equipotentiality of P300 amplitude across scalp-placed recording sites. Physical activity has also been associated with P300 components.

Aging brain

Early studies examined the impact of physical fitness on brain function with older population. Bashore (1989) found age-related slowing in P300 latency and reduced P300 amplitude in physically low-fit old adults. Dustman et al. (1990) observed that physically active older men who are 50 -60 years of age exhibited shorter ERP latencies, stronger central inhibition, better neurocognitive performance, and better visual sensitivity compared to low fit old men. Hillman et al. (2002) also replicated the results of previous studies by showing that older sedentary individuals exhibite

d the longest latency relative to older-fit and young adults. The effect of physical activity on P300 components was most pronounced during executive function tasks (Hillman et al., 2004; 2006). During executive function task, fitness was positively related to P300 amplitude and negatively associated with P300 latency in old adult. Specifically, high and moderate active older adults exhibited increased P300 amplitude during executive control at the frontal region, compared to young adult (Hillman et al., 2004). Sedentary individuals demonstrated slower P300 latencies and smaller amplitude at central and parietal scalp than high active participants (Hillman et al., 2004; 2006). Additional recruitment in task-relevant brain regions shown in active old adults is to

compensate for age-related deficits. That is, active old adults seem to be able to compensate age-related deficit but low active old adults could not utilize compensatory mechanism. Reduced amplitude and slower latency in sedentary individuals may be related to age-related degeneration in brain structures.

Young adults

The benefit of physical activity on cognitive function is not limited to old adults. As describe earlier, a positive relationship between physical activity and cognition was observed in children and adolescents (Buck et al., 2007; Davis et al., 2007; Sibley & Etnier, 2003; Tomporowski et al., 2007), but the benefit of physical activity was particularly strong in executive function tasks (Buck et al.; Davis et al.; Tomporowski et al.) Accordingly, Hillman et al. (2005) initially examined the relationship between age, aerobic fitness and cognitive function during non-executive function tasks by comparing high and low-fit preadolescent children and adults. As a result, physical fitness was positively associated with P300 amplitude in children, but not in young adult. The increase in P300 amplitude for higher fit children suggests greater allocation of attention and working memory resources related to stimulus processing. Greater neural resource allocation in high-fit children relative to low-fit children and low- and high-fit young adult may be related to mechanism to compensate immature cognitive function resulting from underdeveloped brain structure.

However, the results examining effect of physical fitness on P300 component in a young population are controversial. Young individuals who engage in high amounts of aerobic exercise (> 5 h/week, mean age of 30 years) showed greater amplitude during non-executive function task, relative to individuals who engage in low amounts of

exercise (< 5h/week, mean age of 34.7 years) but no difference found on P300 latency (Polish & Lardon, 1997). Other studies failed to find any difference in P300 amplitude and latency between high active or fit and low active or fit in young age group during non-executive function task (Bashore, 1989, Dustman et al., 1990, Hillman et al., 2002). The non-significant effect of physical fitness in young adult is speculated to result from the task type employed in studies. Accordingly, Hillman et al. (2006) examined the impact of physical activity on P300 components in a task-switching paradigm including executive function (ECF) and non-ECF tasks in young (mean age of 19.3 years) and old (64.8 years). These authors found that higher physical activity was associated with greater P300 amplitude in both task types, but shorter P3 latency with greater physical activity was only observed in executive function task. The P300 latency result indicated that this processing speed benefit was specific to the ECF task. The effects of physical activity on P300 measures were statistically equivalent for younger and older adults. However, consistent with behavior result described above, this result indicates that the effects of physical activity on P300 components were more pronounced during an ECF task in this young age group. Given that early young adults exhibit greater plasticity in the frontal lobe, the fact that the benefit of physical activity on P300 components was greater during ECF compared to non-ECF task in early adulthood seems to be reasonable.

In contrary to the study of Hillman et al. (2006), Scisco, Leynes and Kang (2008) examined the relationship between cardiovascular fitness and executive control during task-switching task in young adult (18 to 28 years of age). They failed to find any significant relationship between cardiovascular fitness and P300 components during executive control task. This result may be due to measurement specificity (i.e., physical

activity in Hillman et al.; cardiovascular fitness in Scisco et al.). That is, physical activity appears to be more sensitive to detect neuroelectrical changes in early young adult relative to cardiovascular fitness. The other possible reason may be age range of participants recruited. Highest age in Scisco et al.'s study was 28. Since brain maturation is completed by late 20s, influence of cardiovascular fitness on matured brain is likely to less than one on underdeveloped brain.

Collectively, the results of these studies suggest that physical activity enhances neuronal and cortical functioning through various biological benefits (BDNF & angiogeniesis) thereby enabling increased (improved) allocation of attentional resources and reducing cognitive processing speed during development (childhood and young adulthood). This benefit extends to both nonECF and ECF domains, while the benefit is particularly pronounced during ECF tasks in young adults. The result suggests that physical activity is also quite effective during young adulthood especially in performance of frontally-mediated ECF, since greater plasticity in underdeveloped frontal lobe may maximize the effect of physical activity. As shown in both behavioral and neuro-cortical studies, participation of physical activity during early adulthood plays an important role for healthy brain development. To date, the genetic specificity in the relationship between exercise and brain function has not been examined in young populations. Therefore, in the present study, we will determine whether the benefits of physical fitness on the brain are more prominent in those who are at risk of dementia (ApoE e4 carriers) during executive control function task in young age.

CHAPTER 3: Manuscript to be submitted for publication

ABSTRACT

Cardiovascular fitness and physical activity have been positively associated with executive cognitive functioning (i.e., planning, scheduling, coordinating, response inhibition, and working memory), which rely on the frontal region of the brain. Recent studies suggest that the benefit is particularly strong in middle-aged individuals who carry the Apolipoprotein (ApoE) e4 allele, a known genetic risk factor for Alzheimer's disease (AD). However, there have been no studies to determine this interactive relationship in adolescents. Therefore, the present study examined if cardiovascular fitness mediates the relationship between genotype and cerebral cortical responses in college-age males during a frontally-mediated executive challenge. Twenty nine e4 carriers (N=29; 15 high-fit, 14 low-fit) and thirty non-carriers (N=30; 15 high-fit, 15 low-fit) were stratified by cardiovascular fitness. Cognitive function was assessed by neuroelectric response, event-related potentials (ERPs) recorded at 11 sites (F3, Fz, F4, C3, Cz, C4, P3, Pz, P4, O1 and O2) to both an auditory Go-nogo executive task (ECF) and a non-executive Oddball task (non-ECF). The P300 amplitude, which is indicative of the recruitment of attentional resources, exhibited by the high-fit e4 carriers was higher relative to that observed in the low-fit e4 carriers during both the ECF and non-ECF tasks. Importantly, the high-fit e4 carriers were also undifferentiated from both groups of the non-carriers. Furthermore, high-fit individuals, regardless of genotype, exhibited shorter P300 latency than did the low-fit individuals at sites Fz, Cz and Pz during ECF task and site Pz during non-ECF task. The current findings revealed genetic specificity in the relationship between cardiovascular fitness and the brain processes indexed by P300

amplitude function during late adolescence in response to both ECF and non-ECF challenge, with greater benefit incurred for the ECF task. The results suggest that cardiovascular fitness in e4 carriers is protective against the susceptibility to the liabilities (i.e., hypometabolism and cortical thinning) associated with this allele.

INTRODUCTION

The second decade of life is marked by major cerebral cortical and neuro-cognitive reorganization. In particular, the prefrontal cortex has been shown to continue to myelinate through the early adult years (i.e. until 25 years of age) (Giedd et al., 1999; Huttenlocher, 1990), and synaptic pruning such that non-essential connections are eliminated while essential ones are strengthened still occurs during the third decades (Pfefferbaum et al., 1994; Sowell et al., 2003). Accumulated neuroimaging studies have supported that the prefrontal cortex is the last brain region to mature in the course of development (Chugani, Phelps, & Mzssiottad, 1987; Hudspeth & Pribram, 1992; Sowell, Thompson, Holmes, Jernigan & Toga, 1999). Connection of prefrontal cortex to a variety of brain structure is responsible for highly integrative cognitive function such as ECF (Fuster, 2001). Given the protracted period of plasticity in the frontal lobe, as compared to other brain regions, the development of adult-like executive cognitive function, heavily dependent on the frontal lobe, is strongly influenced by environmental factors such as physical activity, nutrition, and family size during the late adolescence and early adulthood (Tomporowski, Davis, Miller, & Naglieri, 2007).

Regular participation in physical activity is associated with a variety of mental health benefits. Exercise increases brain-derived neurotrophic factor (BDNF), nerve growth factor (NGF) in the hippocampus (Cotman, & Berchtold, 2002), cerebral blood flow, the number of capillaries, capillary density (Black, Issacs, Anderson, Alcantar, & Greenough, 1990; Rogers, Meyer, & Mortel, 1990; Swain et al., 2003) and cell proliferation in brain regions (Allan et al., 2001). These neurotrophic, angiogenic and neurogenic effects of exercise on the brain may play a role as a protector and enhancer of

cognitive function and central nervous system (CNS) integrity. Indeed, Colcombe and colleagues (2003) demonstrated that cardiovascular fitness ameliorated age-related brain tissue loss in the prefrontal, superior, parietal and temporal cortices, which play a central role in successful everyday functioning.

Along with structural and functional anatomical changes in the brain, exercise has been shown to improve cognitive function and to protect against age-related cognitive decline. A meta-analysis on exercise intervention conducted by Colcombe and Kramer (2003) demonstrated that effect sizes for improvements in cognitive function on ECF tasks relative to speed tasks (RT), visuospatial tasks, or controlled processing tasks in normal aging. The benefit of physical activity has also been shown to play an important role in developing cognitive function during childhood in ECF and non-ECF tasks (Sibley, & Etnier, 2003, Buck, Hillman, & Castelli, 2007, Tomporowski et al., 2007). However, the benefit of physical activity on neurocognitive function is less clear in the young population, since there have been few studies with this age group. Although the results of these studies generally revealed no difference in cognitive function between high and low active young adults, Hansen, Johnsen, Sollers III, Stenvik and Thayer (2004) observed better performance in ECF in an exercise group with a mean age of 19 years. This finding clearly showed the importance of the task characteristics with which study participants are challenged. Based on this discussion, it seems that executive challenges are most likely to reveal the benefit of exercise on the developing brain. However, the manifestation of behavioral improvement may occur long after changes at the cortical level or may not be obvious in young, healthy populations (i.e. ceiling

effects). Therefore, the use of psychophysiological techniques may allow detection of change due to exercise not readily apparent at the behavioral level.

The use of ERPs has been employed to examine the effects of exercise on brain functions. Consistent with behavioral studies described above, the benefit of regular participation in physical activity has been observed in P300 component with increase in P300 amplitude and reduction in P300 latency in old (Dustman et al., 1990; Hillman, Weiss, Hagberg, & Hatfield, 2002; Hillman, Belopolsky, Snook, Kramer, & McAuley, 2004; Hillman, Kramer, Belopolsky, & Smith, 2006; McDowell, Kerick, Santa Maria, & Hatfield, 2003), late adolescents and early young adults (Hillman et al., 2006; Polich & Lardon, 1997), and children (Hillman, Castelli, & Buck, 2005). More specifically, P300 amplitude was greater (Bashore, 1989, McDowell et al., 2003, Hillman et al., 2004; 2006) and P300 latency was faster (Dustman et al., 1990; Hillman et al., 2002; 2004; 2006) for high active old adults compared to low active old adults both during executive function task and non-executive functions. In addition, the positive relationship between aerobic fitness and P300 amplitude was observed in children during non-executive task (Hillman et al., 2005). However, the impact of physical fitness on P300 components is controversial in young adults. Young individuals who engage in high amounts of aerobic exercise (> 5 h/week, mean age of 30 years) showed greater amplitude during non-executive function task, relative to individuals who engage in low amounts of exercise (< 5h/week, mean age of 34.7 years) but no difference found on P300 latency (Polish & Lardon, 1997). Other studies failed to find any difference in P300 amplitude and latency between high active or fit and low active or fit in young age group during non-executive function task (Bashore, 1989, Dustman et al., 1990, Hillman et al., 2002). The non-

significant effect of physical fitness in young adult is speculated to result from task type employed in the studies. Accordingly, Hillman et al. (2006) examined the impact of physical activity on P300 components in a task-switching paradigm consisting of tasks, which require different level of executive function (from less to more ECF requirement) in late adolescents (mean age of 19.3 years) and old (64.8 years). These authors found that higher physical fitness was associated with greater P300 amplitude in all task types, but the association between physical activity and P300 latency was found on more challenged task. Importantly, P3 latency measure indicated that this processing speed benefit was specific to more challenged task. These effects were statistically equivalent for younger and older adults. Consistent with behavior result described above, this result indicates that the effects of physical activity on P300 components were more pronounced during an ECF task in this young age group. Given that early young adults exhibit greater plasticity in the frontal lobe, the fact that the benefit of physical activity on P300 components was greater during ECF compared to non-ECF task in early adulthood seems to be reasonable. In contrary to a study of Hillman et al. (2006), Scisco, Leynes and Kang (2008) failed to find any interaction between cardiovascular fitness and P300 components during executive control task. This result may be due to measurement specificity (i.e., physical activity in Hillman et al.; cardiovascular fitness in Scisco et al.). The other possible reason may be age range of participants recruited. Highest age in Scisco et al.'s study was 28. Since brain maturation is completed by late 20s, influence of cardiovascular fitness on matured brain is likely to less than one on underdeveloped brain.

Collectively, the results of these studies suggest that physical activity enhances neuronal and cortical functioning through various biological benefits (BDNF & angiogeniesis) thereby enabling improved allocation of attentional resources and reducing cognitive processing speed during development (childhood and young adulthood). This benefit extends to both non-ECF and ECF domains, while the benefit is particularly pronounced during ECF tasks in young adults. This may be due to greater plasticity in frontal region associated with ECF during early adulthood and stronger apparent benefit of physical activity on executive function. As shown in both behavioral and neuro-cortical studies, participation of physical activity during adolescence plays an important role for healthy brain development.

The importance of healthy brain development in young adults is underscored by the findings on cognitive integrity, as measured by idea density, in young women (mean age 22 years) who were entering the religious orders as reported by Riley, Snowdon, Desrosiers, & Markesbery (2005). More specifically, a remarkable relationship between idea density and the incidence of dementia in later life was revealed such that those with high cognitive ability were less likely to suffer these diseases. Such a linkage supports the essential need to promote healthy brain and cognitive development during this critical period of young adulthood. As such, physical fitness may contribute to cognitive reserve at this developmental stage resulting in delay of cognitive decline in later life.

Moreover, the positive effects of physical activity may also help to attenuate the effects of known genetic factors which predispose individuals to hypoactivation of the brain and impairment in cognitive function. Apolipoprotein (ApoE) is a gene that has been associated with cognitive impairments and late onset of the Alzheimer's disease

(AD) in late adulthood. ApoE e4 causes the deficiency in cholesterol transport, necessary in the repair of brain injury and age-related degeneration in the brain. Thus, inheritance of one e4 allele has been associated with susceptibility to late onset of AD (Corder et al., 1993; Strittmatter et al.,1993). Importantly, brain imaging studies have demonstrated abnormality in individuals who carry the e4 allele. More specifically, middle-age non-demented healthy e4 carriers exhibited hypoactivation in the hippocampus, temporal, parietal and prefrontal regions, compared to age-matched counterparts (Reiman, Uecker, Caselli et al., 1998). Furthermore, the degraded functional brain was also observed in young adults (20-39 years) who carry the e4 allele (Reiman et al., 2004). More recently, Shaw and colleagues (2007) observed that the e4 allele is associated with distinct neuroanatomic signatures, identifiable in children with mean age of 11. This study showed a thinner cortex in the entorhinal, medial temporal and posterior-medial orbitofrontal areas in children who carried e4 allele, relative to non-carriers. These findings indicate that brain abnormality (i.e., deficit in brain activity and shrinkage of cortex) typically observed in old adults who carry e4 allele would occur in childhood and early adulthood.

In contrast to previous studies examining neural activity during resting state, greater and more widespread activation of the left hippocampal, prefrontal and parietal regions was observed during a memory task in middle-age e4 carriers (Bookheimer et al., 2000). The increased level and volume of activation in the e4 carriers was interpreted as a compensatory response in which additional brain regions are recruited to perform a cognitive operation. This is consistent with the notion of reduction in regional specificity and neural efficiency in e4 carriers. This hyperactivation shown in e4 carriers during a

memory task was also observed in young age adults. Brain imaging study comparing brain activation in e4 carriers and non-carriers during non-verbal memory in college age adult and young adults also supports this compensatory mechanism with e4 carriers exhibiting significantly higher activation in frontal areas and cingulated gyri than non-carriers during memory task (Filbey, Slack, Sunderland, & Cohen, 2006; Scarmeas et al., 2005).These evidences suggest that the presence of the APOE e4 allele has physiological consequences before aging that may contribute to risk for Alzheimer's disease.

Studies employed ERP measurement were also found significant difference in P300 components between healthy middle-aged either group carrying e4 allele or positive family history of AD (FH+) and non-e4 carriers or individuals who do not have family history of AD (FH-). Specifically, Green and Levey (1999) initially demonstrated that abnormal prolongation in the latency of P300 in Pz and Cz during Oddball paradigm in e4 carriers who have FH, while no abnormality found in P300 amplitude. Recently, similar result was observed such that smaller P300 amplitudes and prolonged P300 latency during Oddball paradigm were observed in healthy middle aged FH+ group (Ally, Jones, Cole, & Budson, 2006). Both findings revealed that groups at increased risk for developing AD show ERP changes consistent with those observed in patients diagnosed as having AD. These changes were observed in the absence of neuropsyhchological deficits. However, a study to examine the effect of risk genetic factor of AD on P300 component in young female adults aged 19-21 years failed to find significant difference between e4 carriers and non-e4 carriers (Yu, Lin, Chen, Hong, & Tsai, 2000).

There are few studies to examine the relationship between participation in physical activity or physical fitness and the e4 allele on brain function in middle aged

adults. Schuit, Feskens, Launer and Kromhount (2001) revealed that physical activity modified the link between genetic factors and brain functions in non-demented, healthy middle-aged adults. Specifically, the risk of cognitive decline in physically-inactive e4 carriers was nearly 4 times greater than active carriers, whereas no difference was found in non-e4 carriers. This result implies that e4 carriers are particularly vulnerable to a sedentary life style, and regular participation in physical activity considerably reduces the risk of cognitive decline. Most recently, Deeny and colleagues (2008) also found a greater benefit of physical fitness in middle age e4 carriers by showing greater activation at task-relevant brain region in high-fit e4 carriers, relative to low fit e4 carriers during a cognitive challenging task. That is, exposure to an enriched environment through participation in physical activity would compensate for cognitive decline and hypoactivation accelerated by this genetic risk factor. However, it is not clear if physical fitness during early adulthood also reduces cortical abnormality associated with e4.

Therefore, the purpose of the study is to examine if the relationship between ApoE genotype and neuro-cognitive function (cognitive function and cortical activation) is modified by cardiovascular fitness in young healthy adults. Another purpose is to confirm the previous findings that the benefit of cardiovascular fitness is greater during frontally-mediated executive function task relative to non-ECF in late adolescents. In this regard, our prediction was that high-fit e4 carriers will show higher P300 amplitude and faster P300 latency, compared to low-fit carriers, but will be undifferentiated from non-carriers regardless of fitness. This pattern of differences in P300 amplitude and latency will be shown only in ECF task. This pattern will be specific to the frontal region.

METHOD

Participants

Young healthy males between the ages of 18 and 23 were recruited from University of Maryland. Thirty non-e4 carriers (5 of e2/e3 and 26 of e3/e3) and twenty nine e4 carriers (24 of e3/e4, 5 of e4/e4) were selected for current study. APOE genotype was determined based on presence (E4+) or absence (E4-) of at least one E4 allele. Each e4 group (n=29) and non-carrier group (n=30) was equally divided into high physical fitness and low physical fitness groups according to aerobic capacity (VO_2max). All participants provided written informed consent, in accordance with the institutional review board at the University of Maryland. Participants were free of neurological disorder, cardiovascular disease, any medications that influence central nervous system function. Participants' characteristics are shown in Table 1.

Table 1 about here

Fitness measures

Participants' fitness was assessed using YMCA cycle ergometry protocol, which predicts maximal oxygen consumption (VO_2max) based on the steady-state heart rate (HR) of a person exercising at a submaximal power level. The YMCA protocol uses three to four, 3-minute stages of continuous exercise. The test is designed to raise the steady state HR of the subject to between 110 beats.min^{-1} and 85% of the age-predicted maximal HR for at least two consecutive stages. Each work rate is performed for 3 minutes, and heart rates are recorded during the final 15 to 30 seconds of each stage. The heart rate measured during the last minute of each stage is plotted against work rate. The line

generated from the plotted points is then extrapolated to the age-predicted maximal heart rate (220-age), and a perpendicular line is dropped to the x-axis to estimate the work rate that would have been achieved if the person had worked to maximum. Then estimated VO_2max can be estimated using formula (Balady et al., 2000). Estimated VO_2max = 1.8 (work rate) · M^{-1} + 7, where M is the subject's body mass in kg. A study, which conducted cross-validation of this protocol to predict VO_2max with heterogenous study population, revealed that there was no statistical difference between the YMCA predicted VO_2max and the real treadmill VO_2max. This result suggests that the YMCA test is adequate for predicting VO_2max determined on a treadmill (Beekely et al., 2004).

Genotyping

Participants provided samples for DNA from a buccal cell sample collected using a mouthwash rinse. Genomic DNA was isolated using a Puregene DNA Purification kit (Gentra). Genotyping of the APOE e2, e3, and e4 alleles was performed using a modification of methods first described by Qiagen Inc. PCR was performed using typical primers [F-5' ACT GAC CCC GGT GGC GGA GGA GAC GCG GGC-3'; R-5' TGT TCC ACC AGG GGC AGG CGC TCG CGG 3'] at a final concentration of 300mM. The thermal cycling profile consisted of incubation at 95°C for 5 minutes followed by 30 cycles of 95°C for 10 seconds, 65°C for 30 seconds, and 72°C for 30 seconds and then incubated at 72°C for 5 minutes. The PCR reaction generated 50uL of amplification product, with a product size of 318 bases in length. Two restriction digests containing 10uL each of PCR product and either 3.75 U of AflIII or 4.0 U of HaeII were incubated at 37°C overnight and analyzed separately on a 3% agarose gel. Genotypes were

determined for each sample according the fragment sizes observed from each of the digests (Table 2). Genotype accuracy was determined by the use of DNA sequence-verified controls run with each genotyping reaction.

Table 2 about here

Kaufman Brief Intelligence Test (K-BIT*)*

The K-BIT was created to test intelligence of individuals aged 4-90 year. This test screens two cognitive functions, verbal and nonverbal intelligence. The verbal test containing expressive vocabulary and definitions measures crystallized thinking – knowledge of words and their meanings. The nonverbal test measures fluid thinking – the ability to solve new problems through perceiving relationships and completing analogies. All nonverbal test items contain pictures and abstract designs rather than words so nonverbal ability can be assessed even when language skills are limited. And if there is a significant disparity between verbal and nonverbal scores, K-Bit provides valuable in sight. For example, a low score on Verbal and a high score on nonverbal might suggest a language problem rather than low intelligence. The K-BIT has been scaled and normalized for easy comparison with several other more comprehensive batteries such as the Wechsler adult Intelligence Scale-Revised (WAIS-R) (Kaufman, Kaufmen, 1990 28). Reliability testing has revealed that internal consistency reliabilities are average .94 for the overall K-BIT IQ composite, .93 for the vocabulary subtest, and .99 for the matrices subtest (http:// ages, pearsonasessments.com). Estimate of construct validity in adult groups range from r = 0.61 to 0.75 when compared with the WAIS-R, and estimates of

concurrent validity in adult groups have ranged from r = 0.37 to 0.50 when compared with the Slosson Intelligence Test (Kaufman, Kaufman, 1990 28).

A study examining differences in ERP parameters related to intelligence demonstrated significant correlation between intelligence and ERP parameters with longer P300 latencies and reduced amplitudes in less intelligent young adults (18-21 years of age) (Jaušovec & Jaušovec, 2000). In addition, Bixby and colleagues (2007) also found the significant correlation between IQ and executive challenge score in old adults (Bixby et al., 2007). Therefore, it appears to be necessary to consider IQ in an examination of relationship between the cardiovascular fitness and P300 components during executive function task in the current study.

Seven-day physical activity recall test (PAR)

The PAR is one of the most widely used physical activity assessments in exercise science and epidemiological research. The PAR provides detail regarding the duration, intensity, and volume (energy expenditure) of physical activity and can therefore be used for a variety of applications. Participants is asked to estimate the number of hours spent during the last seven days in sleep, moderate, hard and very hard physical activity. The remaining time represents light activity. Work intensity is frequently described in multiples of resting metabolic rate. This index is calculated as the work metabolic rate / resting metabolic rate ratio (MET). It is well established that, at least for physical activities in which the body weight is moved, there is a direct association between work intensity and energy expenditure via the body's metabolic processes (ACSM, 1980). Caloric expenditure is estimated on the basis of 1 MET = 1 kcal $\cdot$ kg^{-1} $\cdot$ $hour^{-1}$. Sleep is

estimated as 1 MET, light activity as 1.5 METs, moderate activity as 4 METS, hard activity as 6 METs, and very hard activity as 10 METs. MET values are multiplied by the hours spent in each of the five categories, and the products are summed to give daily energy expenditure (kilocalories per kilogram per day). Construct validity of the PAR has been established through relationships with changes in objective measures of physical fitness such as VO_2max and percent body fat (Blair et al., 1985, Dishman & Steinhardt, 1988, Jacobs, Ainsworth, Hartman, & Leon, 1993). These studies have generally supported the validity and reliability of the PAR as a measure of physical activity in adults. Studies have also shown it to be reasonably valid among adolescents (Sallis, Buono, Roby, Micale, & Nelson, 1993).

Cognitive challenge

Stroop Color and word test

The Stroop test is selected as a measure of inhibition, a key executive function. The Stroop test requires suppression of one's response to a dominant stimulus pattern (printed words) while attending and responding to a secondary stimulus characteristic (i.e., ink color). There are three phases of the test, each 45s in duration. The first phase assesses the number of words that can be read (i.e., names of colors – red, green, and blue appearing in black-colored ink), and the second phase assesses the number of colored stimuli (i.e., a string of X's printed in one of the three respective colors) that can be correctly identified during the 45-s interval. The third phase assesses the participant's ability to identify the color of link while suppressing the response to identify text, which appears in an incongruent color (e.g., the word "**RED**" printed in blue ink). The Stroop

test yields four scores: one each for the word and color test, a color-word scores, and the interference score, which is derived from performance on the three phases described above (Golden, 1978). The raw word, color and color-word scores simply represent the total number of correct responses during each of the respective tasks. An age-based correction factor was applied to each of the word (w), color (c), and color-word (cw) scores. A predicted color-word score (cw'), which served to adjust for individual differences in the speed of naming words and colors, was then computed by dividing the product of the word and color scores by the sum of the word and color scores: (cw' = w × c/w + c). Finally, the interference (I) score is calculated by subtracting the predicted color-word score from the obtained color-word score (I = cw – cw') to assess resistance to cognitive interference (i.e., cognitive inhibition). It served as the primary index of executive function (Golden, 1978). A higher score represents more resistance to cognitive interference and therefore, better executive function.

Oddball and Go-nogo tasks

Participants engaged in two types of binaural auditory tasks: an Oddball and Go-nogo task, which have been used to elicit the ERP components. In both Oddball and Go-nogo task, participants subjected to three blocks of 100 tones (80 common, 20 rare in each block). In Oddball task, participants were asked to press a button when they heard the rare (target) tones and to count the number of rare tones they heard in each of the trials. However, they were asked to inhibit the button press to the rare tones and were not asked to count the number of tones in Go-nogo task. The Go-nogo task requires response inhibition so it has been used as a measure for executive function. Common tones were

1000Hz and rare tones were 2000 Hz. Intensity of tones employed in both tasks was 80 dB. Tones were presented in the ear canal via a soft earplug insert. The interstimulus interval was set a 2.00 seconds.

EEG

EEG was recorded from eleven sites of the scalp corresponding to Fz, F3, F4, Cz, C3, C4, Pz, P3, P4, O1 and O2 of the International 10-20 electrode placement system (Jasper, 1958), which were referenced to the right mastoid (A2) while AFz served as the ground. All electrode impedances were below 10 kΩ. EEG was acquired at a sampling rate of 512 Hz and amplified 20,000 times, while the eye channels was amplified 5,000 times using Grass model 12A5 Neurodata Acquisition amplifiers with band-pass filter settings of 0.1-100 Hz (96-db/octave). Continuous data were collected with Neuroscan Scan 4.2 software. Stimuli were generated using Neuroscan Stim software, which sent a trigger indicating the condition of each trial for offline sorting, reduction and analysis of EEG and behavioral data. EEG signal processing was conducted off-line with Neuroscan software (Neuroscan Labs, Neurosoft, Inc., version 4.3, Sterling, VA). EEG was re-referenced by linear derivation to average mastoids. Ocular correction was performed and eye blinks were corrected using the eye correction algorithm (Semlitsh, Anderer, Schuster, & Presslich, 1986). The time series data were epoched into 1000 msec segments, and baseline corrected based on a 100 msec prestimulus interval. Any epoch containing amplitudes greater than ± 75 μV was removed with artifact rejection procedures. To exclude trials that are contaminated by artifact, each epoch was visually inspected and removed manually. The rare epochs for the Oddball and Go-nogo tasks were sorted according to trial type. The Oddball rare and Nogo rare trials were then

averaged in the time domain to yield two averaged time series with event-related potentials and then filtered with low-pass filter settings of 10 Hz (24-db/octave). Waveforms were analyzed by peak picking with a latency window of 200 – 450 ms then the P300 amplitude and latency were saved as data file for further statistical analysis.

Testing procedure

The study entailed two visits to the lab. All participants were asked to refrain from alcohol, caffeine, and nicotine for at least 24 hours and from food for at least 75 minutes before psychophysiological testing was to begin. On day 1, after providing their informed consent, participants completed an interview on health history information and a non-invasive mouthwash procedure as a method of obtaining a DNA sample. Participants who meet the criteria of this project were invited for Day 2 testing procedure. On the second testing day, all participants completed the Kaufman Brief Intelligence Test (K-BIT), the Stroop color-word test (executive control function test) and seven day physical activity recall test (PAR). After providing a brief overview of the testing procedures, participants were seated in a comfortable chair in front of a computer screen. The participants were then prepared for electrocortical measurement in accordance with the Society for Psychophysiological Research guidelines (Putnam, Johnson, & Roth, 1992). A lycra electrode cap (Electro-Cap International, Eaton, OH) was fitted to the participant's head and 11 electrode sites were prepared using Omni-prep and electrode gel. EEG data collection was conducted in sound-attenuated room. During both task conditions, subjects were asked to open eye and inhibit vertical and horizontal eye movement by focusing fixation point (black cross with white background). After an

acceptable EEG signal was observed, the participant was given the task instructions and baseline EEG during eye open and eye close was recorded each for 2 minutes. Participants were given the opportunity to ask questions and 10 practice trials were provided to ensure that the subject understands the task requirements. Each Oddball and Go-nogo task was administered in three blocks each with a brief rest period between blocks. Total six blocks were counter-balanced. After completion of EEG data collection, physical fitness was assessed with the YMCA cycle ergometry protocol to yield submaximal aerobic capacity

Statistical analysis

Behavioral data

Percent correct and reaction time (RT) were separately analyzed in a 2 (Genotype) × 2 (Physical fitness) × 2 (Task) ANCOVAs with repeated measures on task. A 2 (Genotype) × 2 (Physical fitness) ANCOVAs was conducted on Stroop interference score to examine interactive relationship between genotype and physical fitness on behavioral performance on executive function task. Significant omnibus tests were followed up with Tukey HSD post-hoc comparisons. The alpha level was p=.05 for all analyses.

Event-related potential

ANCOVA approach

In order to control for the effect of intelligence on the dependent variables (P300 amplitude and latency), ANCOVA was employed with K-BIT scores used as a covariate.

Separate 2 (Genotype) × 2 (Physical fitness) × 2 (Task) × 3 (Region) × 3 (Sites) ANCOVAs with repeated measures on task, region and sites were conducted on P300

amplitude and P300 latency. The between-groups factors were genotype (E4+ vs E4-) and fitness (high vs low). The levels of within-groups factors were task (Oddball vs Go-nogo), region (frontal, central, parietal), and sites (mid, left, right). Additional 4-way ANCOVAs (2 (Genotype) × 2 (Physical fitness) × 2 (Task) × 3 (Region: Fz, Cz, Pz)) on midline sites were conducted separately on P300 amplitude and P300 latency to examine interaction among independent variables.

Regression approach

Hierarchical regression analyses were employed to examine the following dependent variables of interest: (1) the P300 amplitude during the Oddball task, (2) the P300 amplitude during the Go-nogo task, (3) the P300 latency during the Oddball task, and, finally, (4) the P300 latency during the Go-nogo task. Those dependent variables were run separately to the following predictors (1) genotype, (2) aerobic capacity, and the (3) interaction of genotype and aerobic capacity in order to determine the unique variance accounted for by each predictor as well as the nature of the relationship (i.e, direction and magnitude of the slope). The regression analyses applied to the P300 amplitudes and latencies of the Go-nogo and Oddball tasks were conducted for each of the 11 electrode sites. Furthermore, if the interaction term between genotype and aerobic capacity was statistically significant, then the P300 amplitude or latency (for each electrode) was regressed on aerobic capacity separately for e4 carriers and non-carriers. If correlations between all dependent variables and IQ are significant, the IQ variable was included in the regression equation (i.e., an empirical justification) that was applied to the dependent variable with which IQ was correlated. (Note: There were significant correlations between intelligence and P300 latency during the Oddball. Thus, intelligence was

included for empirical justification in the regression equation for P300 latency during Oddball task. A criterion alpha of 0.05 (two-tail test) was applied to all tests of significance.

RESULTS

Behavioral measures

A significant Genotype × Task effect emerged in percent correct ($F(1, 55) = 8.17$, $p=.006$, $\eta 2=.87$) with e4 carriers exhibiting lower percent correct in Go-nogo task (Mean=95.893, SE=.235), relative to Oddball task (Mean=99.323, SE=.118) (Figure 1, Table 3). However, this difference was not observed in non-carriers. No significant main effect and interaction effect was emerged for reaction time. In addition, no main and interaction effects exhibited for Stroop interference scores. Behavioral data are shown in Table 3.

Table 3 and Figure 1 are about here

ERP analysis

Grand averaged ERP waveforms and topographical voltage maps of each group (high-fit e4 carriers, low-fit e4 carriers, high-fit non-carriers, low-fit non-carriers) at each electrode site during non-ECF and ECF tasks are shown in Figure 2, Figure 3, Figure 4, and Figure 5, respectively. For the purpose of comparison, topographical voltage maps of six subjects located in extreme end of each cardiovascular physical fitness type (high-fit e4 carriers, low-fit e4 carriers, high-fit non-carriers, low-fit non-carriers) at each electrode site during non-ECF and ECF tasks are shown in Figure 6 and Figure 7.

Figure 2, 3, 4, 5, 6 and Figure 7 about here

P300 Amplitude

5-factor ANCOVA on midline and lateral sites

A significant Genotype × Fitness × Task × Region interaction effect emerged ($F(2, 53) = 3.28$, $p = .045$, $\eta 2 = .89$). Post-hoc testing revealed that during the Go-nogo test, the low-fit e4 carriers exhibited lower amplitude at frontal and central regions, relative to all other groups (high-fit carriers, high-fit non-carriers, low-fit non-carriers). *Importantly, as predicted, P300 amplitudes in high-fit e4 carriers were higher than low-fit e4 carriers, and undifferentiated from non-carriers* (Figure 8).

Figure 8 is about here

During Oddball task, high-fit e4 carriers exhibited higher amplitude at frontal and central regions, relative to low-fit e4 carriers. Such a difference was not observed in non-carriers. P300 amplitudes at frontal sites in high-fit e4 carriers were greater than those in high-fit non-carriers.

During Go-nogo test, low-fit e4 carriers exhibited smaller amplitude at frontal and central regions, relative to high-fit e4 carriers, high-fit non-carriers, and low-fit non-carriers. P300 amplitudes in high-fit e4 carriers were not differentiated from those in high- and low-fit non-carriers. There was no difference on P300 amplitude between high and low-fit non-carriers.

Comparison between Go-nogo and Oddball tasks revealed that P300 amplitude at frontal region in high-fit e4 carriers during Go-nogo task was greater than that during Oddball task. The same pattern was observed in both high and low-fit non-carriers with

amplitude during Go-nogo task being greater at frontal and central regions, relative to Oddball task.

4-factor ANCOVA on midline sites

A significant Genotype × Fitness × Region ($F(2, 53) = 5.76$, $p = .005$, $\eta^2 = .82$) effect emerged. These findings are essentially the same as described as above for the 5-factor ANCOVA but are collapsed across tasks. There were no main and interactive effects with task. Low-fit e4 carriers exhibited smaller amplitude at Fz, relative to high-fit e4 carriers, and low-fit non-carriers. Amplitude at Cz in low-fit e4 carriers was lower than those in all other groups, and high-fit e4 carriers were not differentiated from those in high- and low-fit non-carriers. Difference between high-fit and low-fit in P300 amplitude was not observed in non-carriers (Figure 9).

Figure 9 is about here

A Genotype × Task × Region ($F(2, 53) = 3.29$, $p = .045$, $\eta^2 = .89$) interaction effect was also significant. Post-hoc comparison revealed that during the Go-nogo task, e4-carriers exhibited lower amplitude at Fz, Cz, and Pz, relative to amplitudes at corresponding sites in non-carriers. In contrary, the amplitude at Fz during the Oddball task was greater in e4 carriers, relative to non-carriers. Comparison between Oddball and Go-nogo tasks revealed that non-e4 carriers during Go-nogo task exhibited greater P300 amplitude at Fz and Cz relative to those at corresponding sites in e4 and non-e4 carriers during the Oddball task. P300 amplitude at Fz in e4 carriers was greater than those at Fz in e4 and non-carriers during Oddball task. P300 amplitude at Cz in e4 carriers was also greater than that at Cz in e4 carriers during Oddball task (Figure 10).

Figure 10 is about here

P300 Latency

4 factor ANCOVA on midline site

A Fitness × Task × Region interaction effect was significant ((F (2, 53) =3.31, p=.044, η^2 =.89). Post-hoc comparisons revealed that P300 latencies at Fz and Cz during Go-nogo task were shorter in high-fit individuals, relative to those in low-fit during Go-nogo task and both low and high-fit individuals during Oddball task. However, latency at Cz in low-fit individuals during Go-nogo task was longer than one during oddball task. Similarly, latency at Pz during Go-nogo task in high-fit was shorter than those of low-fit individuals both during Go-nogo and Oddball tasks. The shorter latency shown in high-fit relative to low-fit was also observed at Pz during Oddball task (Figure 11).

Figure 11 is about here

Regression

Regression analysis was employed to investigate the relationship between aerobic capacity and P300 components.

Explained variance accounted for by the interaction between e4 and aerobic capacity

In Go-nogo task, the relationship between VO_2max and P300 amplitude was different depending upon genotype at sites Fz ($\Delta R^2 = 0.064$, p = .05), and F4 ($\Delta R^2 = 0.067$, p = .044) (Appendix J.1). Separate regressions to determine the difference in the VO_2max - P300 amplitude relationship between e4 carriers and non-carriers revealed that VO_2max was positively related to P300 amplitude at sites Fz ($R^2 = 0.19$, p = .018), and F4 ($\Delta R^2 = 0.175$, p = .024) in e4 carriers, not in non-carriers at sites Fz ($R^2 = 0.007$, p = .668), and F4 ($\Delta R^2 = 0.014$, p = .539) (Appendix J.2, Figure 12).

Figure 12 is about here

In Oddball task, the relationships between VO_2max and P300 amplitude at site C3 was significantly different depending on genotype ($\Delta R^2 = 0.076$, $p = .023$) (Appendix J.3). Separate regression analyses to determine the difference in the VO_2max - P300 amplitude relationship between e4 carriers and non-carriers showed the trend that VO_2max was positively related to P300 amplitude at C3 in e4 carriers ($\Delta R^2 = 0.26$, $p = .005$), but not in non-carriers (Appendix J.4, Figure 13). No significant interaction between the genotype and VO_2max was revealed for P300 latency (Appendix J.6.

Figure 13 is about here

Explained variance accounted for by e4 allele

No relationship was found between genotype and the dependent variables (P300 amplitude and P300 latency) both during Oddball and Go-nogo tasks.

Explained variance accounted for by aerobic capacity

The interaction between cardiovascular fitness and P300 amplitude was observed at F3 during Go-nogo task with greater VO_2max being associated with greater amplitude at F3 ($\Delta R^2 = 0.082$, $p = .036$) (Figure 14, Appendix J.1. The same interaction was also found at C3 during Oddball task ($\Delta R^2 = 0.084$, $p = .021$) (Figure 15, Appendix J.3). A significant relationship between cardiovascular fitness and P300 latency was observed at F4 ($\Delta R^2 = 0.096$, $p = .025$) and P3 ($\Delta R^2 = 0.082$, $p = .038$) with VO_2max being negatively related to P300 latency during Go-nogo task (Figure 16, Appendix J.5) The relationship was also observed at O2 during Oddball task ($\Delta R^2 = 0.143$, $p = .003$) (Figure 17, Appendix J.6)

Figure, 14, 15, 16, and 17 are about here

DISCUSSION

The purpose of the present study was to investigate the relationship between cardiovascular fitness and the integrity of cerebral cortical responses to executive cognitive challenge during the period of adolescence when brain maturation is ongoing. Accordingly, Blakemore & Choudhury (2006) reported significant gray matter refinement and white matter development in the cerebral cortex during this period and up to age 25 years. More specifically, the rationale for the investigation was based on two primary points – 1) the established neurobiological benefits of exercise and 2) the deficits in brain development and metabolism noted in young carriers of the APOE e4 allele. As such, it was hypothesized that this period of brain development enables a temporal window of opportunity in which to influence brain maturation in a positive manner, particularly in those vulnerable to developmental deficiency (i.e., carriers of e4). In regard to the latter issue, lower levels of cerebral glucose metabolism and relative thinning of the cortex have been reported in young e4 carriers relative to non-carriers of the allele (Reiman et al., 2004; Scarmeas et al., 2005; Shaw et al., 2007). In regard to the former point, the benefits of cardiovascular fitness on brain processes, as described by Hillman, Erickson and Kramer (2008) may be prominent particularly in those who are genetically at risk of compromised development. In this manner, cardiovascular fitness may induce benefits in the brain that counterbalance the liabilities associated with the e4 allele. The present results are supportive of such population specificity in regard to the amplitude findings for both the executive and non-executive challenges.

Furthermore, it is well established that the frontal lobe of the brain, which is essential to the mediation of executive processes (i.e., planning, scheduling, inhibition, working memory), is the slowest region of the forebrain to reach maturity. It is also the first region to suffer age-related decline. Kramer, Hahn and Gopher (1999) earlier reported a remarkable influence of aerobic exercise training on these cognitive functions in older men and women who developmentally are at a stage of decline of frontal integrity. As such, it is logical to assume that the neurobiological benefits of exercise are particularly salient in this brain region in light of accelerated decline relative to other brain regions that are less susceptible to age-related decline. *However, it is not clear at the present time whether cardiovascular fitness would enhance frontal brain development in the young who have not yet reached maturity, particularly in those who would logically derive great benefit (i.e., e4 carriers). The present findings are supportive of such influence as the amplitude responses in high-fit e4 carriers was undifferentiated from those at lower genetic risk of dementia (non-carriers) while the low-fit carriers showed an attenuated amplitude response compared to all other groups (high-fit e4 carriers, high-fit non-carriers, and low-fit non-carriers).*

Historically, the earliest studies that examined physical fitness and cognitive benefit employed rather crude measures of reactive capacity (i.e., reaction time RT) to simple auditory and visual stimuli (Spirduso, 1980). However, neuroimaging technology is now available to assess critical brain processes with instruments that are more sensitive in terms of temporal and spatial resolution. ERPs are ideal in their ability to assess neural responses to cognitive challenge with a high degree of temporal sensitivity and, thereby, index the speed and amplitude of cerebral cortical response in a precise and objective

manner. In addition, much of the literature on exercise / fitness and cognitive function to date has reported studies involving older participants. However, it is now clear that integrity of brain development during youth has significant implications for cognitive functioning and health of the nervous system later in life (Moceri, Kukull, Emanuel, van Belle, & Larson, 2000; Riley et al., 2005). As such, there is a need to employ highly sensitive measurements in investigations of physical fitness and neurocognitive function in the developing brain.

Importantly, the ERP studies that have reported findings on the relationship between cardiovascular fitness and neurocognitive function in adolescents are equivocal or mixed in their conclusions (Hillman et al., 2006; Scisco et al., 2008). For example, Hillman et al. (2002) observed that high-fit and low-fit adolescents were undifferentiated in regard to P300 amplitude and latency. However, these investigators employed the Oddball task and failed to examine brain responses to executive challenge, which more fully engage the frontal brain processes that would likely be more sensitive to exercise-induced benefits. However, other investigators have employed executive challenges while assessing ERP responses, but the findings are inconsistent across studies. Hillman et al. observed a positive relationship between cardiovascular responses and P300 amplitudes during executive challenge, but a more recent study by Scisco et al. did not reveal any such relationship between these variables. Similar discrepancies exist in the literature in regard to fitness and latency of the P300 component (Hillman et al.; Scisco et al.). As such, the investigations of cardiovascular fitness and neurocognitive integrity in adolescents are not compelling and they are surprising in light of the thoughtful employment of tasks (i.e., executive) that would be sensitive to the still-developing

frontal brain regions. Therefore, the consideration of genetic influence seems prudent in light of the individual differences in exercise-induced phenotypes based on genotype. This issue seems particularly important in terms of the present study in light of the arrested development of the frontal lobes in e4 carriers and the likelihood of greater derived benefit from cardiovascular fitness.

In the present study, the two groups of non-carriers of APOE e4 exhibited similar P300 amplitudes at all regions (i.e., frontal, central, and parietal) of the cortex when contrasted within each of the task challenges. As expected, the two groups elicited a classic pattern of ERP scalp topography or waveform morphology in response to the Oddball task characterized by a maximum parietal amplitude with progressive reductions in amplitude proceeding anterior to the frontal sites. The amplitudes were well within normal ranges for such an age group and both the high-fit and low-fit groups also exhibited higher amplitudes at the frontal and central sites during the executive challenge compared to those elicited during the non-executive task. The parietal amplitudes were undifferentiated across the tasks in both the high-fit and low-fit non-carrier groups. In this regard, the confirmation of expected characteristrics in relation to the overall topography of ERP waveforms, and the task-specific differences in amplitude observed in these non-carrier groups, provides confidence in their role as a useful and valid reference group with which to assess the impact of cardiovascular fitness on neurocognitive response in e4 carriers.

In comparison to the lack of differences noted in the two groups of non-carriers the high-fit e4 carriers exhibited higher amplitudes of the P300 component, as recorded at the anterior or frontal region of the brain (i.e., frontal and central sites), relative to those

observed in these same regions in the lower-fit e4 carriers. Furthermore, the P300 amplitudes of the high-fit e4 carriers were undifferentiated from those observed in both groups of the non-carriers. That is, the amplitudes generated in response to the tasks in the high-fit e4 carriers were similar to those evidenced in age-matched young males who are at lower genetic risk for dementia. The similarity in cortical response between the high-fit carriers and non-carriers was observed for both the executive and non-executive tasks. It is also important to note that the lower-fit e4 carriers showed significantly lower amplitudes at the frontal and central sites, not only in comparison to the high-fit e4 carriers, but in contrast to the two groups of non-carriers, as well. As such, high-fit e4 carriers appear to derive a benefit of cardiovascular fitness in that their neurocognitive response to challenge was similar to those at lower genetic risk of dementia while the lower-fit e4 carriers exhibited a neurocognitive response that was significantly reduced compared to that observed in all of the other groups of young men. The lower amplitudes in these regions exhibited by the lower-fit e4 carriers holds particular importance for the health of the brain as lower amplitudes of the P300 component are typically noted in older individuals compared to young. That is, ERP amplitudes decrease with aging and are indicative of neurodegenerative processes (Polich, 1997).

For example, a study was conducted by Ally, Jones, Cole and Budson (2006) in which the P300 component was examined in AD patients and biological children of patients with Alzheimer's disease (AD). As predicted, the amplitude in AD patients was significantly smaller compared to an age- and gender-matched control group. In addition, both amplitude and latency in offspring of AD patients (FH+) were significantly impaired when compared to its age- and gender- matched control group. Interestingly, the FH+

group demonstrated similar P300 amplitudes to the older control group. That is, the P300 amplitude in participants with a family history of AD and a mean age of 54 was comparable to participants with a mean age of 75 and no family history of the disease. This similarity in P300 amplitude may lead one to draw the hypothesis that participants with a family history of AD are prematurely aging. Both findings indicate that participants with a family history of AD demonstrate possible preclinical phase of the disease more than 20 years prior to its clinical presentation at the electrophysiological level. The findings raise the possibility that assessment of ERPs may contribute to early detection of AD. *As such, the present findings are suggestive of heightened integrity of cortical functioning in adolescent e4 carriers who are characterized by superior cardiovascular fitness relative to age matched men who carry the APOE e4 allele and characterized by lower cardiovascular fitness.*

On consideration of the full scalp topography (as assessed by the Gene x Fitness x Task x Region x Sites ANOVA), it is noteworthy that the magnitude of difference between the high-fit and lower-fit e4 carriers was greater in response to the executive challenge (Go-nogo task) during the response inhibition trials as compared to the group differences seen in response to the non-executive challenge (Oddball task). Such a difference was expected in light of the impact of APOE e4 on frontal brain development, the region that is essential to the mediation of executive processes, and the more salient effects of exercise programs that result in improved cardiovascular fitness on executive cognitive functioning. In essence, the deleterious effects of carrying the APOE e4 allele on frontal development would allow for more apparent benefit to neurocognitive processes that relay heavily on the frontal brain regions. The task difference in

magnitude of group difference disappeared on consideration of only the midline sites and was likely due to the decrease in the number of observations in each region, but the overall pattern of findings was otherwise essentially unchanged from that which emerged from the analysis based on the more comprehensive scalp topography

The relative benefit to frontally mediated cognitive processes was revealed previously by Colcombe & Kramer (2003) in their meta-analytic review of exercise and cognitive function in older men and women. In addition, Colcombe et al. (2004) reported a positive relationship between cardiovascular fitness, as indexed by aerobic capacity, and cerebral cortical tissue density in older men and women. *However, it is remarkable that such fitness-related differences were noted in the present study in young men.* As such, young carriers of the APOE e4 allele may derive particular benefit from the neurotrophic, angiogenic, and dopaminergic effects of exercise training as reported by Black et al. (1990), Cotman and Engessar-Cesar (2002), Rogers et al. (1990), and Spirduso (1983). Furthermore, the present pattern of amplitude findings show remarkable similarity to those found in older men and women (carriers and non-carriers of APOE e4 aged 50- 69 years) who were stratified on physical activity history (high vs low) during executive challenge (working memory) (Deeny et al., 2008). Based on magnetoencephalographic (MEG) responses to a letter recognition task, the high-active e4 carriers were undifferentiated from both high- and low-active non-carriers in amplitude of an M170 component, while the low-active e4 carriers exhibited a lower amplitude than all other comparison groups. Collectively, the results reported in the present study and by Deeny et al. support genetic specificity in the relationship between

cardiovascular fitness (and physical activity) with cerebral cortical response with studies employing different aged participants, neuroimaging technology, and cognitive tasks.

In contrast to the findings for amplitude, the assessment of P300 latency, indicative of speed of processing, revealed shorter processing periods in high-fit young males relative to lower-fit males, regardless of genotype. The group differences noted in latencies, as related to cardiovascular fitness, differed for the two tasks such that shorter latencies were observed for the appearance of the peak amplitudes in the parietal region during both tasks, but such an advantage extended to the central and frontal regions only for the executive task. These results are consistent with those of Hillman et al. (2006), but differ substantively from those reported recently by Scisco et al. (2008). The basis of the discrepancy with the latter findings is unclear at this time. However, it is important to note that the present findings are in agreement with those of Scisco et al. in terms of P300 amplitude as we noted no differences between the high-fit and low-fit non-carrier groups and well as between these same groups and the high-fit e4 carriers.

Collectively, the results for P300 amplitudes and latencies suggest that cardiovascular fitness bestows a robust benefit to both carriers and non-carriers of APOE e4 in terms of enhanced neural processing speed and, importantly, that low-fit e4 carriers are not at any disadvantage relative to low-fit non-carriers in terms of this specific dimension of neural processing. However, the findings for P300 amplitude are very different in that cardiovascular fitness apparently bestows a particular benefit to e4 carriers, enabling recruitment of neural resources to a level similar to those at reduced genetic risk of dementia, while a lower cardiovascular fitness is associated with a particular deficit in such recruitment of resources in e4 carriers. That is, they are less

capable of neural recruitment in response to challenge than low-fit non-carriers. As such, it appears that both genotypes benefit from cardiovascular fitness in regard to the neural processes indexed by P300 latency, but APOE e4 carriers derive particular benefit from fitness in terms of amplitude. The fact that the non-carriers who varied in cardiovascular fitness showed no differences in amplitudes across the tasks implies that they may have been optimized (i.e., ceiling effect) in terms of brain development at such a young age when the brain is healthy and intact. From this perspective there is no logical basis to reason that cardiovascular fitness would contribute benefit to the neural processes indexed by the P300 amplitude beyond that engendered by a healthy lifestyle and low genetic risk of brain pathology. It may be that other populations of adolescent non-carriers characterized by poor diet and cognitive stimulation would show benefit of cardiovascular fitness in terms of ERP components.

The general lack of behavioral findings as revealed by the lack of group differences on the Stroop task and the RTs observed during the Oddball and Go Nogo tasks) demonstrates the importance of multi-level of analysis to the neurocognitive benefits of cardiovascular fitness. It may be that common or undifferentiated behavioral outcomes may mask any differences in brain integrity due to compensatory strategies. For example, it may be that the Stroop performance in the low-fit e4 carriers required more cortical activation and recruitment of additional neural networks, but such a differential cost in neural resources would be "invisible' in the absence of neuroimaging. Longer duration of executive challenge may, in fact, result in behavioral differences between groups based on genotype and fitness similar in nature to those revealed by the ERP assessment.

Finally, the present results are meaningful in that healthy development of the brain at a young age is particularly important in terms of the long-term health of the individual (Riley et al., 2005). The Religious Order studies reveal that healthy cognitive development as revealed by ideational density and linguistic ability at a young age is remarkably protective against dementia in later life. Based on the present findings it would seem plausible that cardiovascular fitness also confers a benefit on brain development that bestows protection against undue age-related decline and dementia in later life. Such a possible benefit could be termed an investment hypothesis and would be particularly important in those at genetic risk for such disorders. The validity of such a notion will require scientific investigation with thoughtful consideration given to the level of measurement specificity conjoined with task specificity and genetic influence.

TABLES

Table 1. Mean and standard deviations for Age, Weight, VO_2max, Energy expenditure and intelligence score of participants.

	High fit E4- carriers	Low fit E4-carriers	High fit Non-carriers	Low fit Non-carriers
Number of subjects	15	14	15	15
Age	20.87 (1.30)	20.29 (1.20)	20.67 (1.05)	20.73 (1.71)
Weight	181.87 (35.16)	171.67 (31.03)	175.33 (26.50)	176.73 (26.28)
VO_2max	52.72 (7.48)	36.622 (5.94)	54.11 (3.91)	40.73 (5.60)
Energy Expenditure	40.19 (7.24)	40.72 (9.42)	43.60 (5.26)	40.42 (5.37)
K-BIT Composite	108.93 (9.22)	107.21 (8.63)	104.2 (9.20)	106.53 (9.81)

Table 2. APOE PCR fragment sizes after restriction enzyme digestion for use in APOE genotyping.

APOE genotype	Product size after HaeII digest (bases in length)	Product size after AflIII digest (bases in length)
e2/e2	267	231
e2/e3	267, 232	231
e2/e4	267, 232	295, 231
e3/e3	232	231
e3/e4	232	295, 231
e4/e4	232	295

Table 3. Mean and standard deviations for Reaction time (RT) and Response accuracy on non-executive Oddball task and executive Go-nogo task.

		Reaction Time (ms)		Response accuracy (%)	
		Oddball (SD)	Go-nogo (SD)	Oddball (SD)	Go-nogo (SD)
E4+		396.54 (75.54)	349.76 (70.90)	99.32 (0.52)	95.86 (1.35)
	High-Fit	381.50 (72.62)	338.11 (64.57)	99.29 (0.56)	95.67 (1.21)
	Low-Fit	412.66 (77.91)	362.24 (77.54)	99.36 (49.72)	96.12 (1.51)
E4-		403.32 (92.24)	366.58 (47.83)	99.11 (0.74)	96.61 (1.19)
	High-Fit	391.99 (85.64)	374.43 (48.86)	98.93 (0.79)	96.36 (1.14)
	Low-Fit	414.65 (100.07)	358.73 (47.12)	99.29 (0.67)	96.87 (1.23)

FIGURE CAPTIONS

Figure 1. Genotype × Task interaction on percent correct (%)

Figure 2. Grand averaged ERP waveforms by each group at each electrode site in Oddball task

Figure 3. Grand averaged ERP waveforms by group at each electrode sites in Go-nogo task

Figure 4. Topographical voltage map by group at each electrode site in Oddball task

Figure 5. Topographical voltage map by group at each electrode site in Go-nogo task

Figure 6. Topographical voltage map of six subjects located in extreme end of each physical activity type ((a) high fit e4 carriers, (b) low fit e4 carriers, (c) high fit non-carriers, (d) low fit non-carriers) at each electrode site in Oddball task

Figure 7. Topographical voltage map of six subjects located in extreme end of each physical activity type ((a) high fit e4 carriers, (b) low fit e4 carriers, (c) high fit non-carriers, (d) low fit non-carriers) at each electrode site in Go-nogo task

Figure 8. Genotype × Physical fitness × Task × Region interaction on the P300 amplitude. The line represents a significant difference ($p<.05$).

Figure 9. Genotype × Physical fitness × Region interaction on the P300 amplitude.

Figure 10. Genotype × Task × Region interaction on the P300 amplitude

Figure 1.

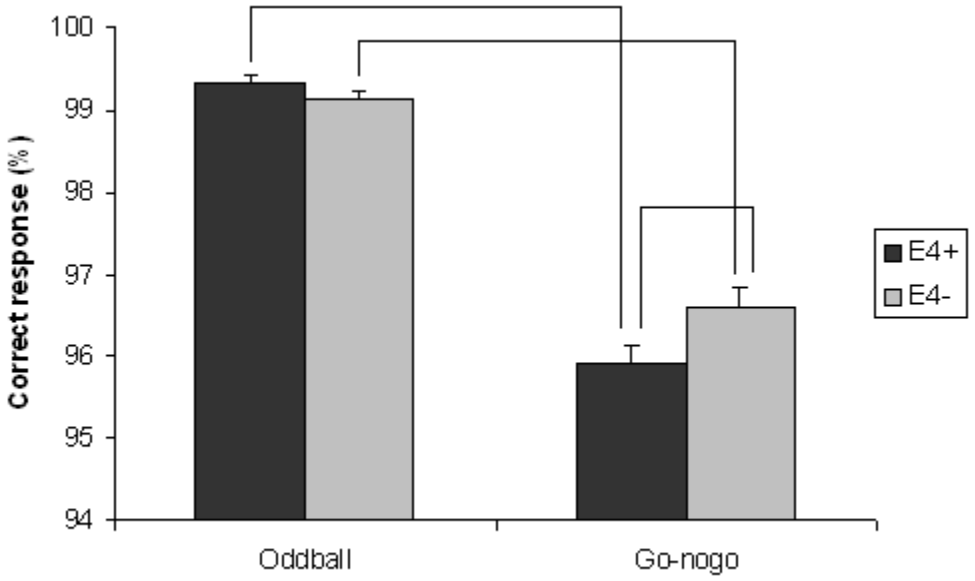


Figure 2.

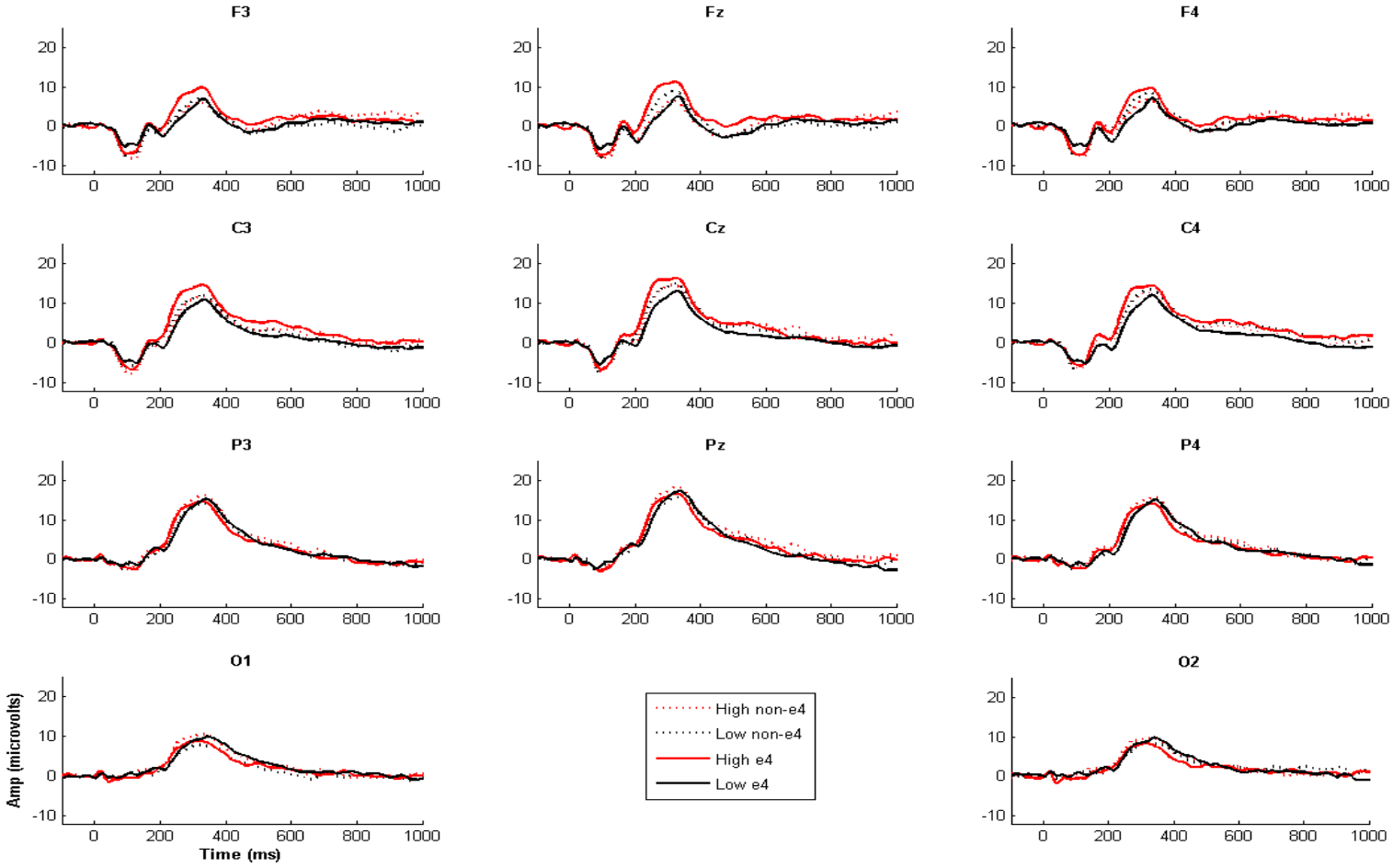

F3
Fz
F4
C3
Cz
C4
P3
Pz
P4
O1
O2
20
10
0
-10
0
200
400
600
800
1000
Amp (microvolts)
Time (ms)
High non-e4
Low non-e4
High e4
Low e4

Figure 3.

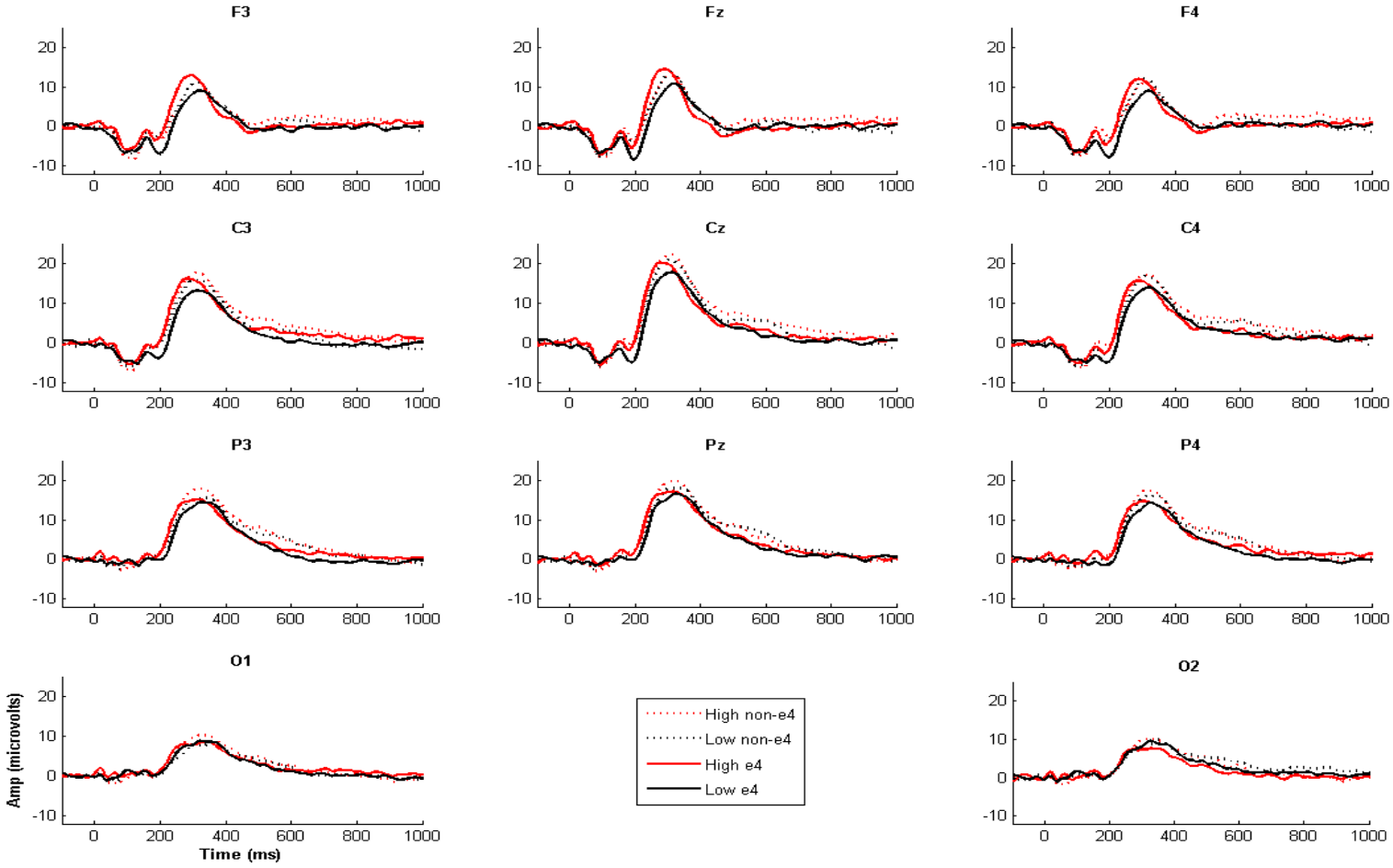

F3
Fz
F4
C3
Cz
C4
P3
Pz
P4
O1
O2
Amp (microvolts)
Time (ms)
High non-e4
Low non-e4
High e4
Low e4

Figure 4.

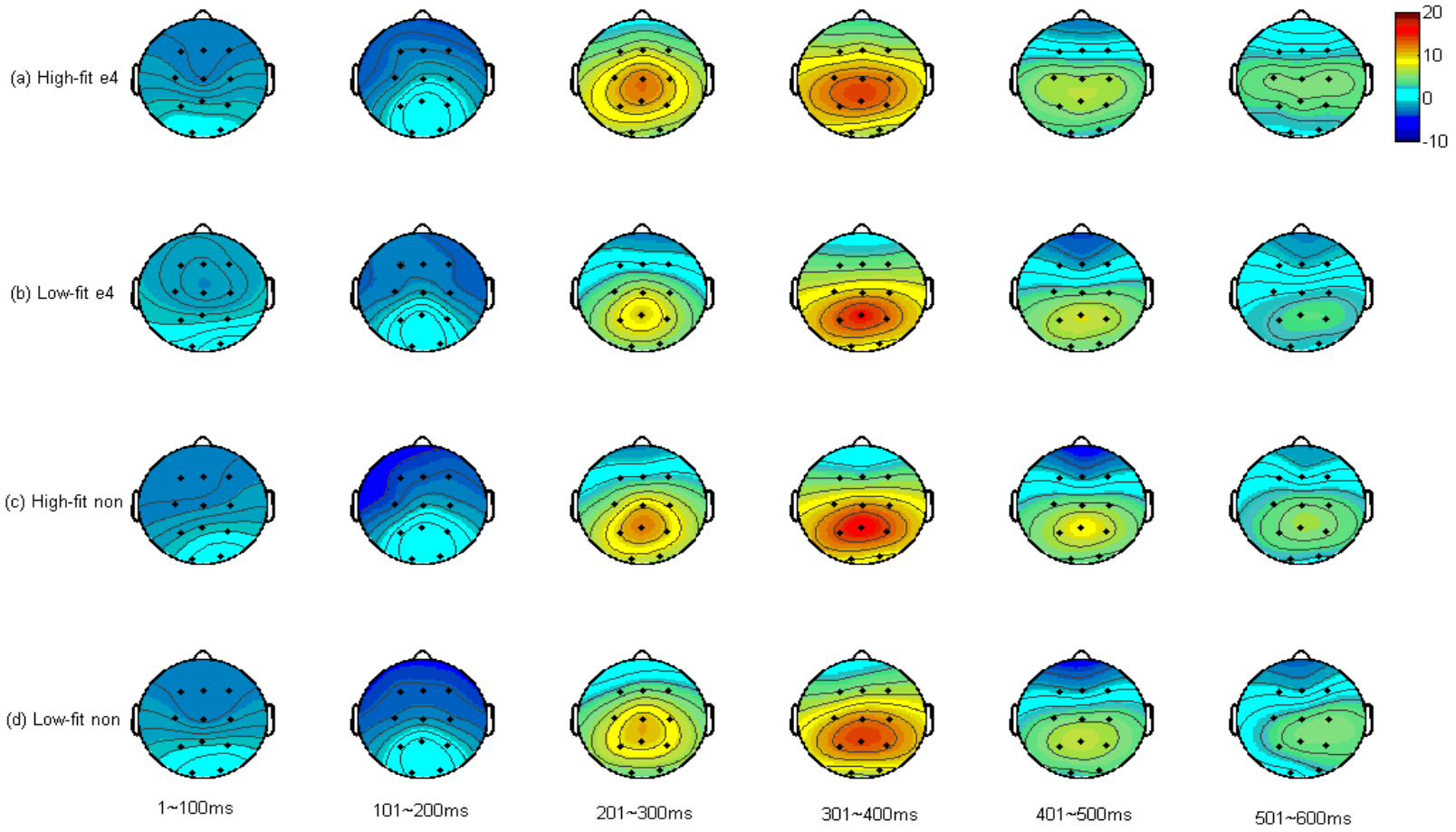

(a) High-fit e4
(b) Low-fit e4
(c) High-fit non
(d) Low-fit non
20
10
0
-10
1~100ms
101~200ms
201~300ms
301~400ms
401~500ms
501~600ms

Figure 5.

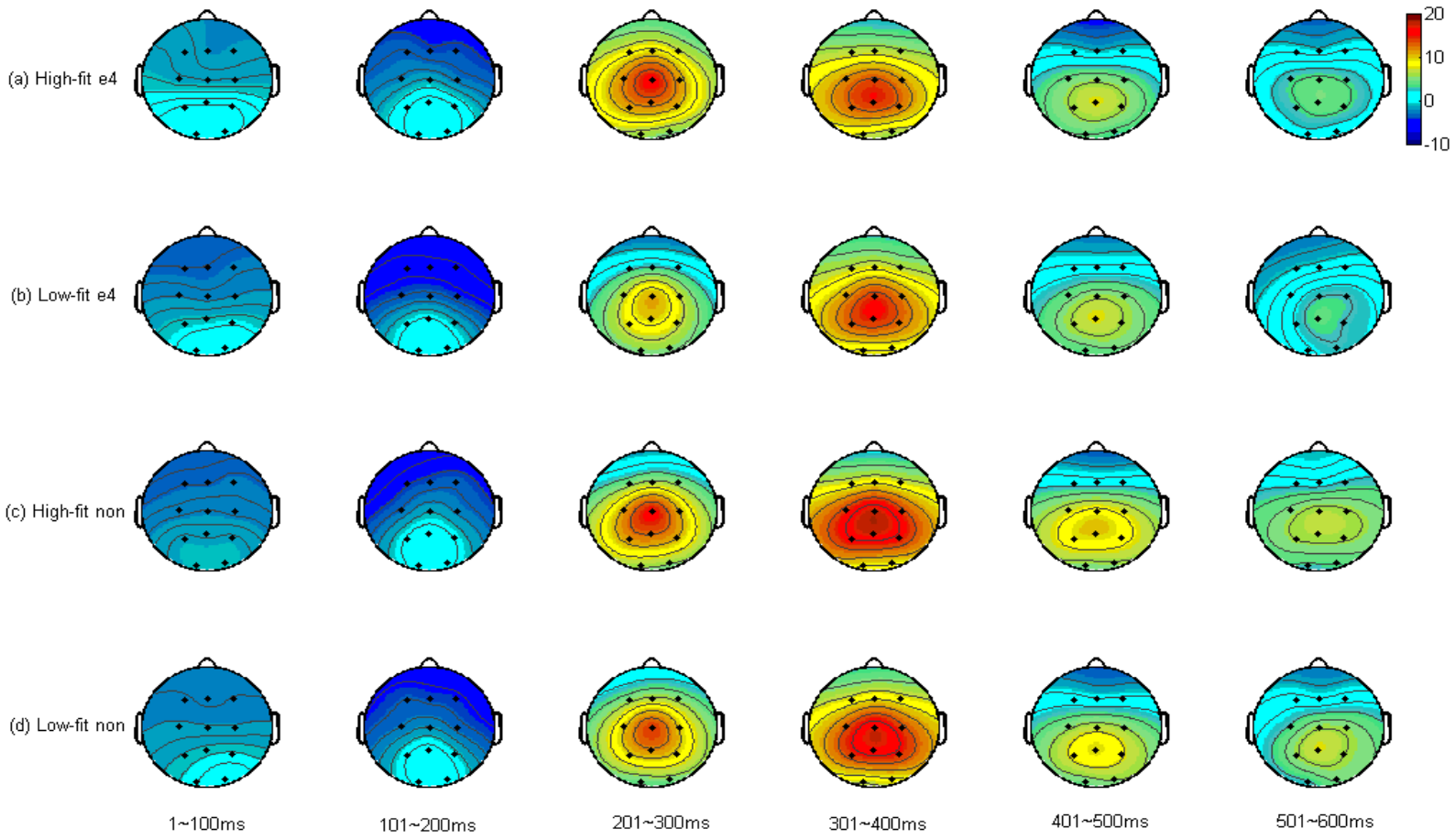

(a) High-fit e4
(b) Low-fit e4
(c) High-fit non
(d) Low-fit non
20
10
0
-10
1~100ms
101~200ms
201~300ms
301~400ms
401~500ms
501~600ms

Figure 6.

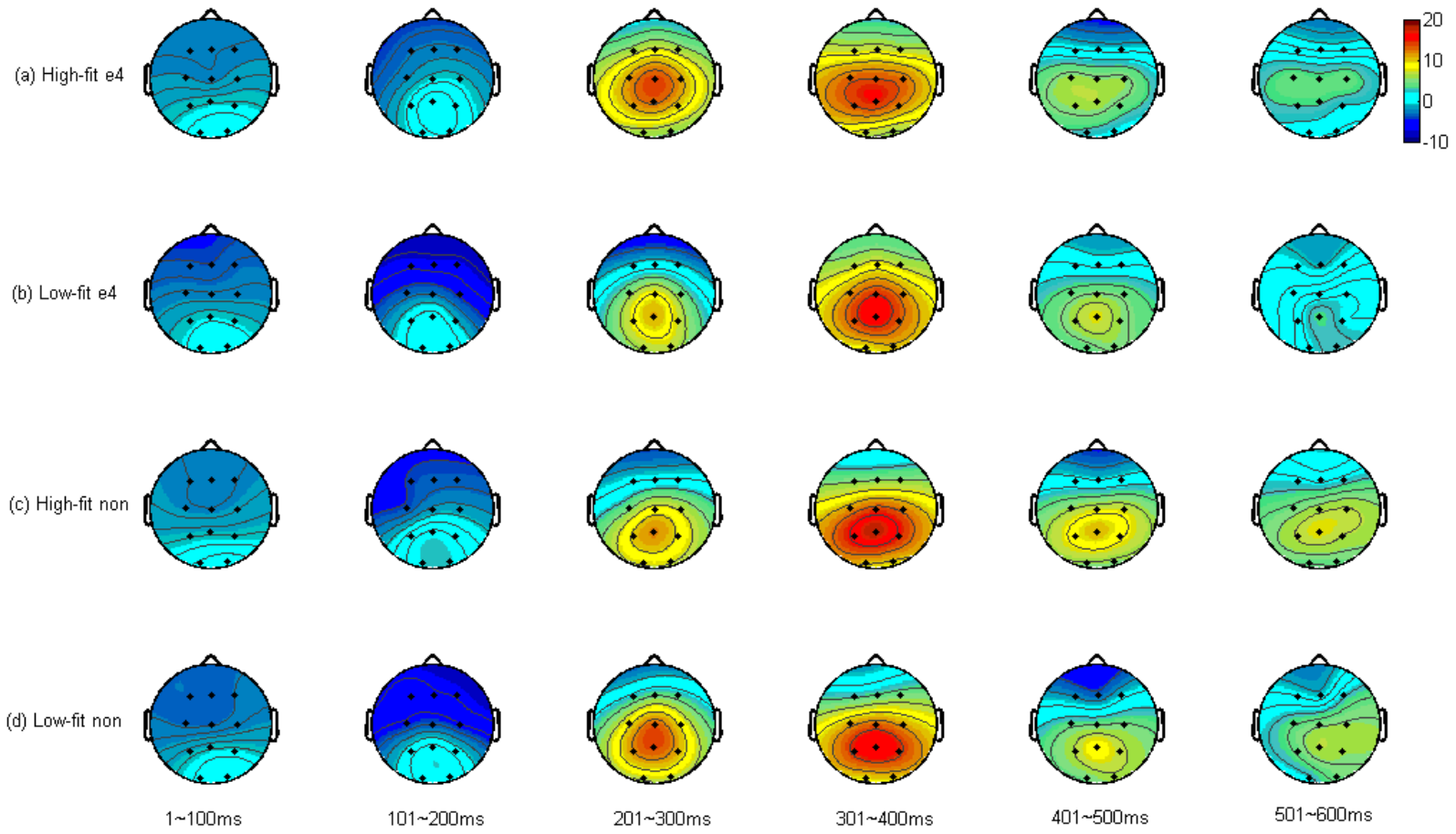

(a) High-fit e4
(b) Low-fit e4
(c) High-fit non
(d) Low-fit non
20
10
0
-10
1~100ms
101~200ms
201~300ms
301~400ms
401~500ms
501~600ms

Figure 7.

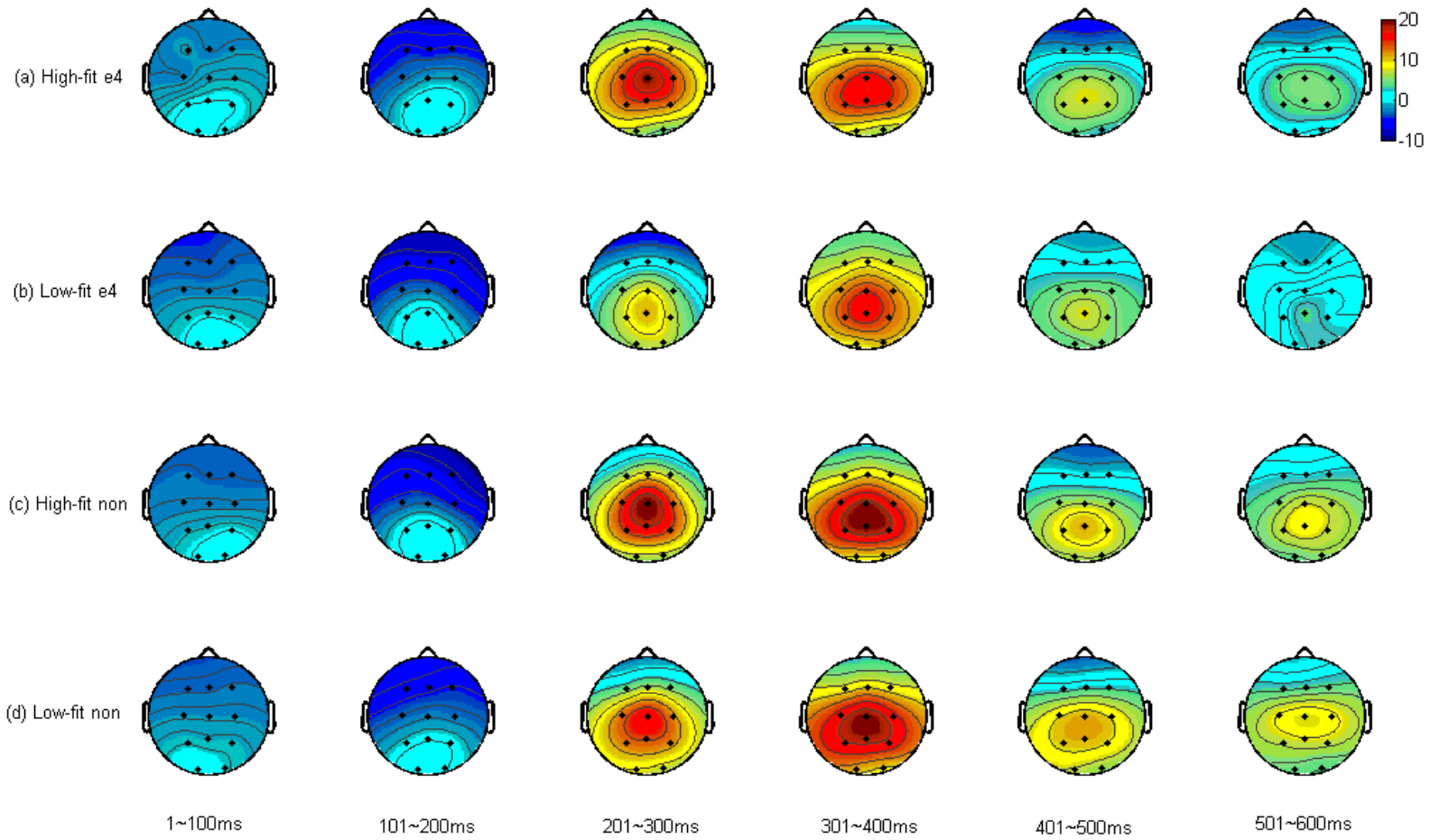

(a) High-fit e4
(b) Low-fit e4
(c) High-fit non
(d) Low-fit non
20
10
0
-10
1~100ms
101~200ms
201~300ms
301~400ms
401~500ms
501~600ms

Figure 8.

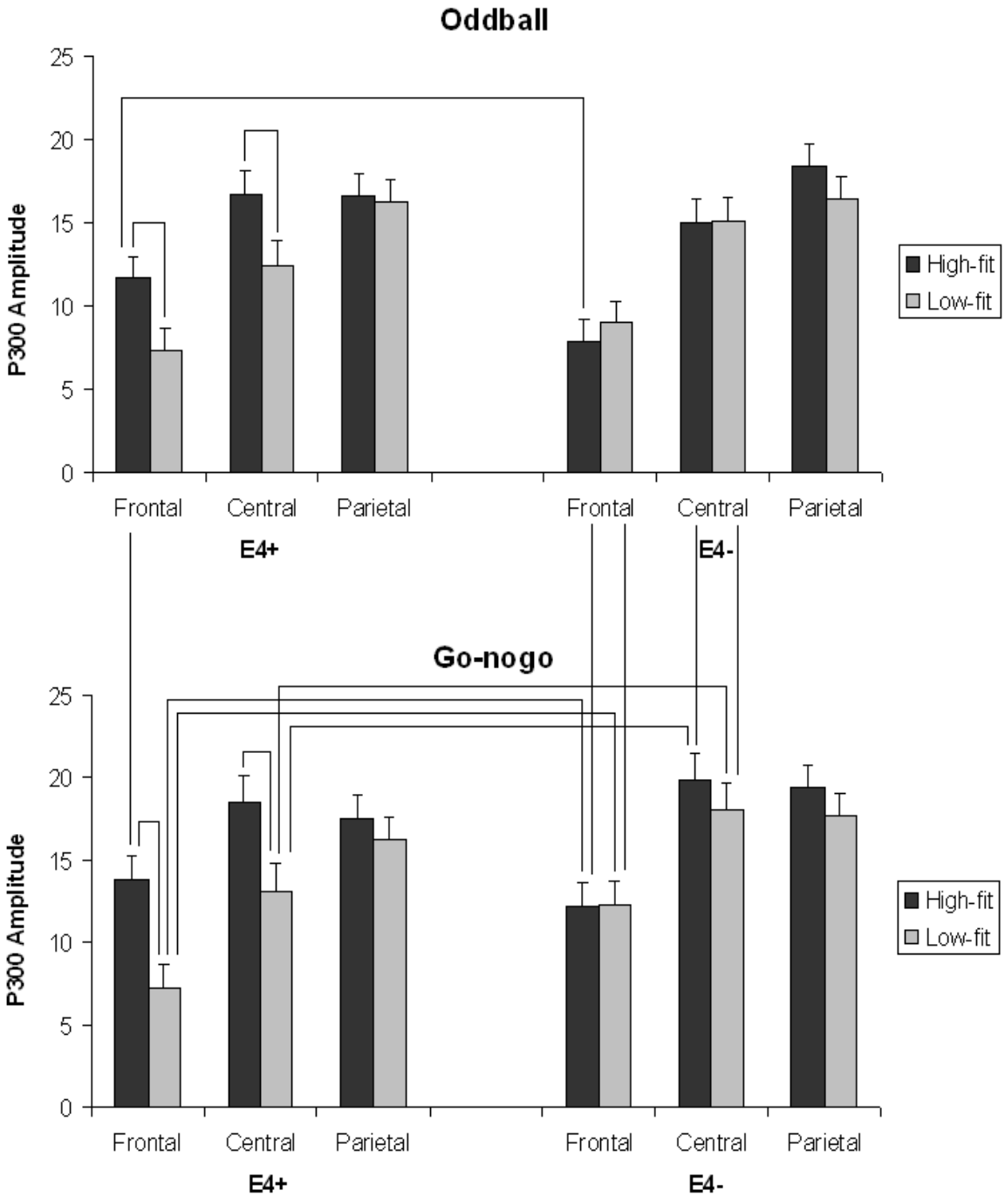

Oddball
P300 Amplitude
25
20
15
10
5
0
Frontal
Central
Parietal
Frontal
Central
Parietal
E4+
E4-
High-fit
Low-fit
Go-nogo
P300 Amplitude
25
20
15
10
5
0
Frontal
Central
Parietal
Frontal
Central
Parietal
E4+
E4-
High-fit
Low-fit

Figure 9.

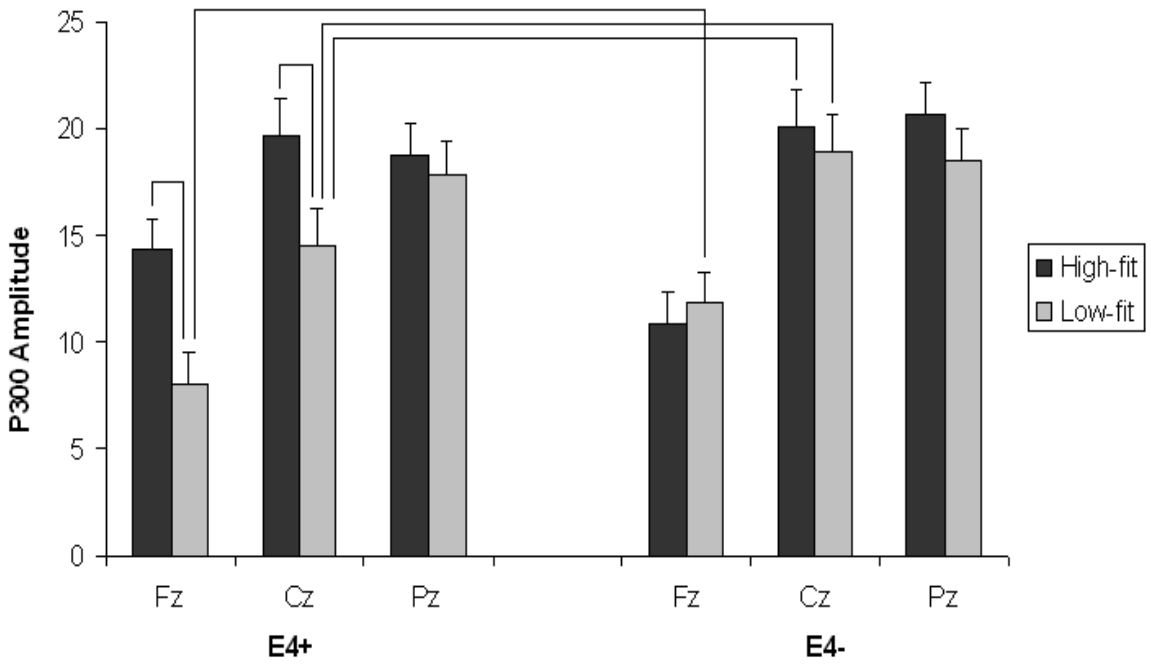

P300 Amplitude
25
20
15
10
5
0
Fz
Cz
Pz
Fz
Cz
Pz
E4+
E4-
High-fit
Low-fit

Figure 10.

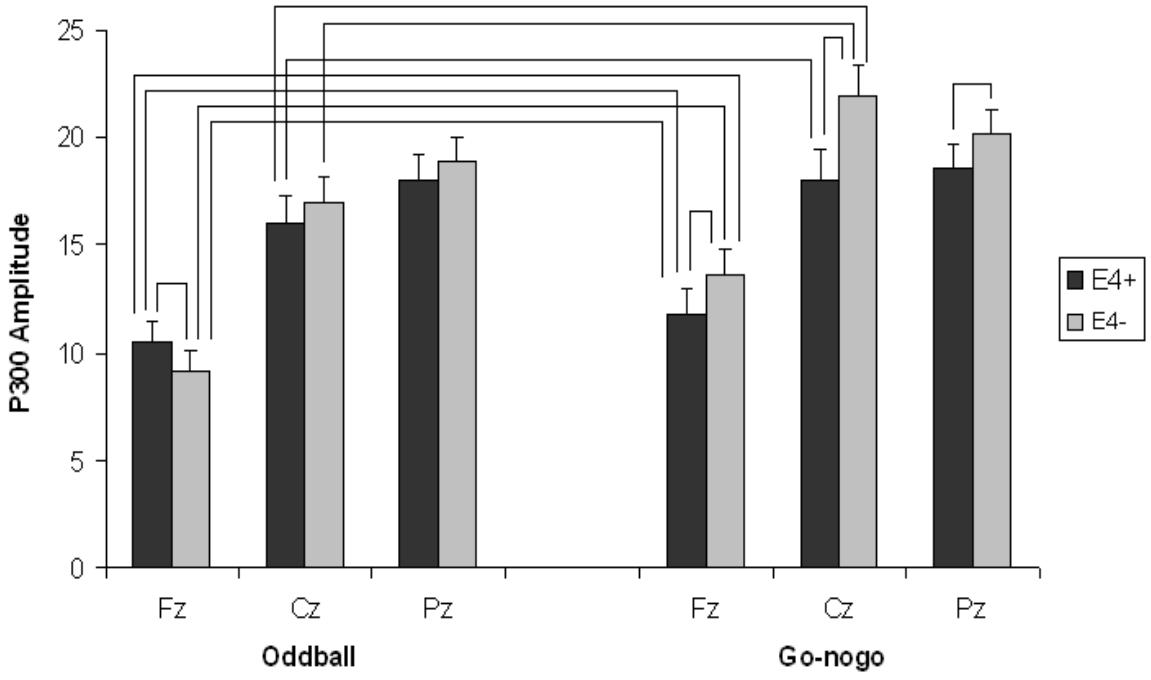

P300 Amplitude
25
20
15
10
5
0
Fz
Cz
Pz
Fz
Cz
Pz
Oddball
Go-nogo
E4+
E4-

Figure 11.

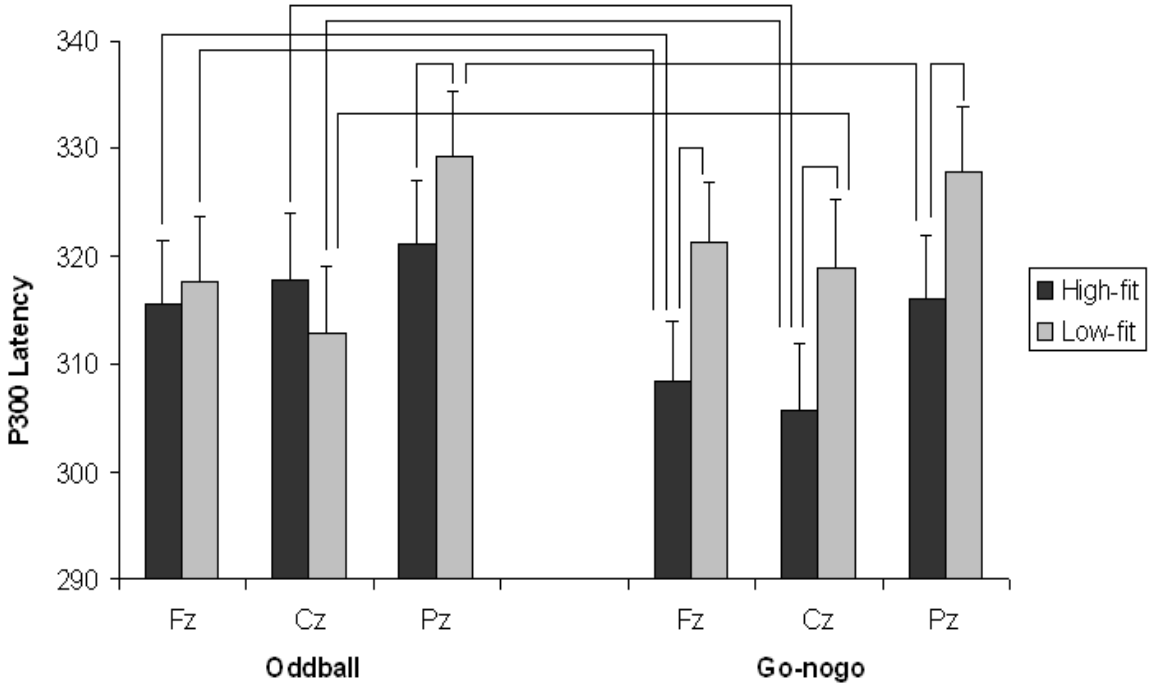

340
330
320
310
300
290
P300 Latency
Fz
Cz
Pz
Fz
Cz
Pz
Oddball
Go-nogo
High-fit
Low-fit

Figure 12.

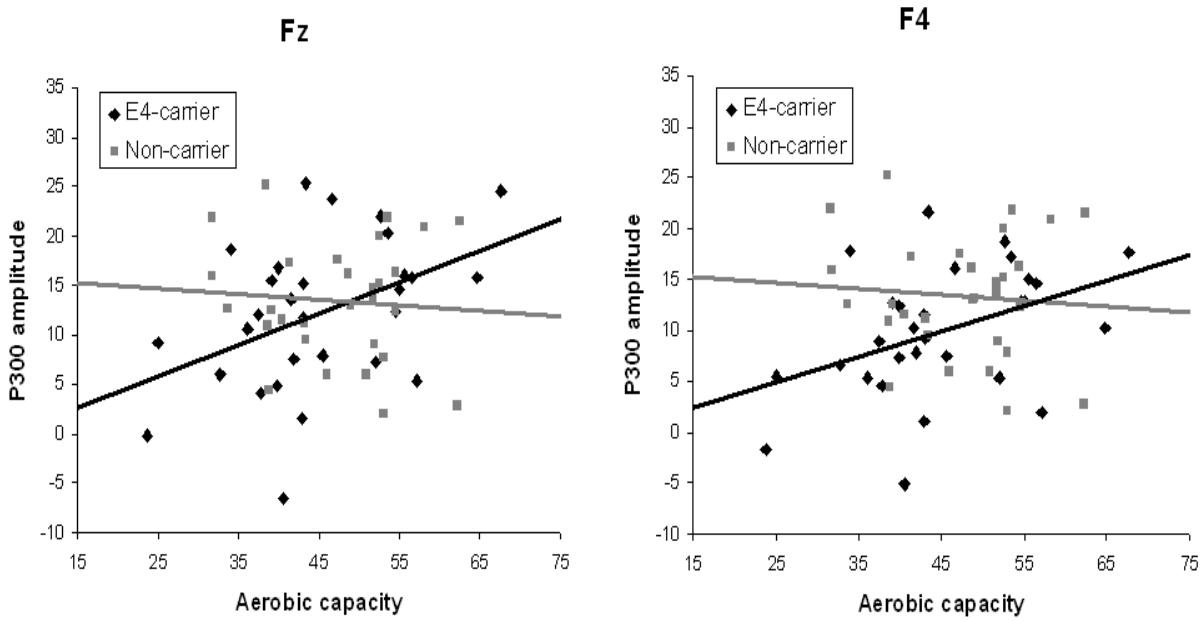


Figure 13.

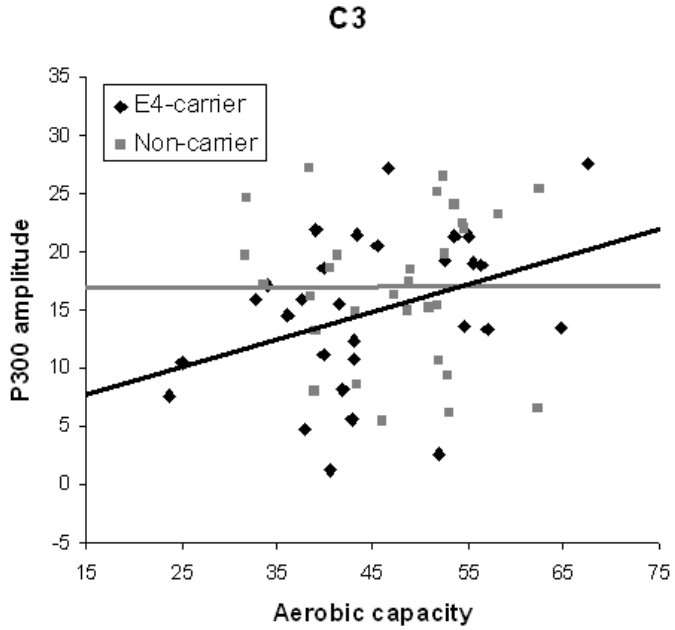
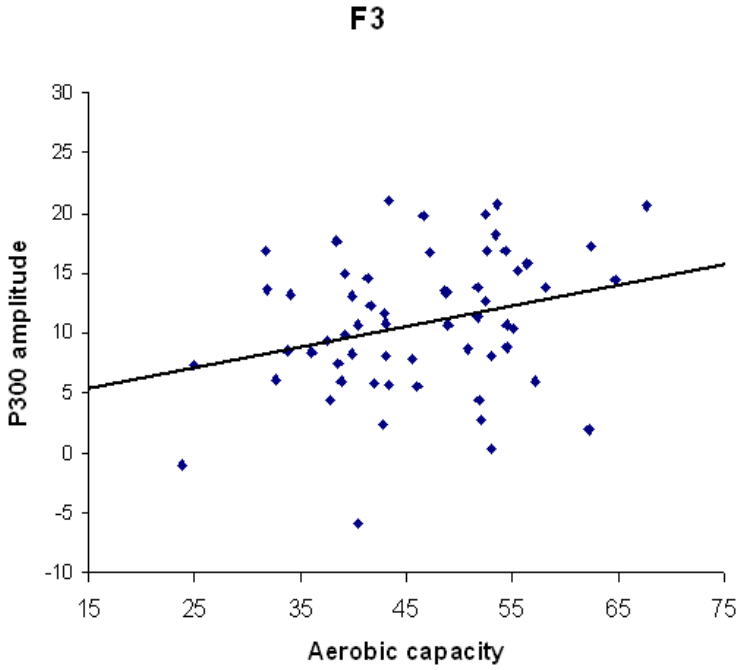


Figure 14.



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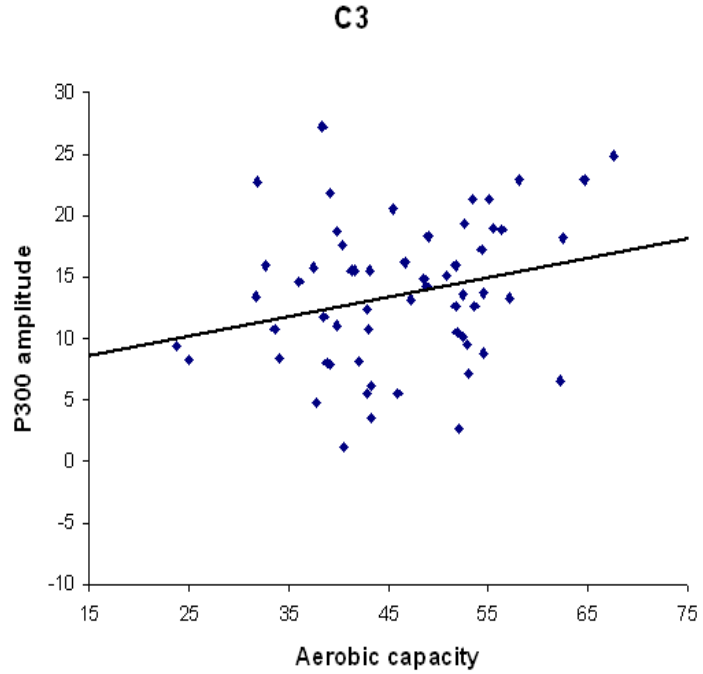


Figure 16.

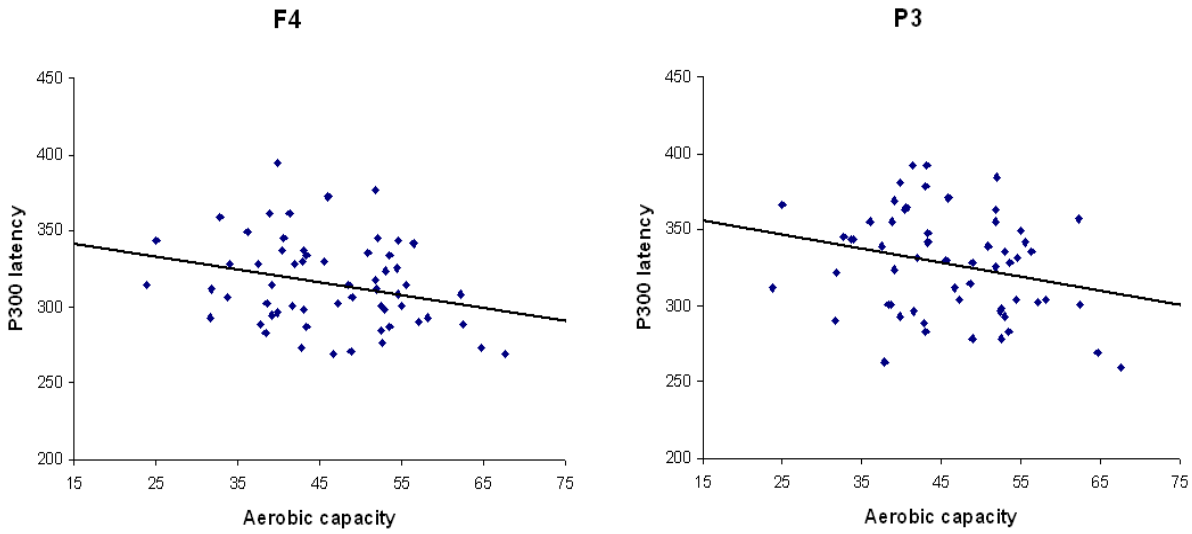
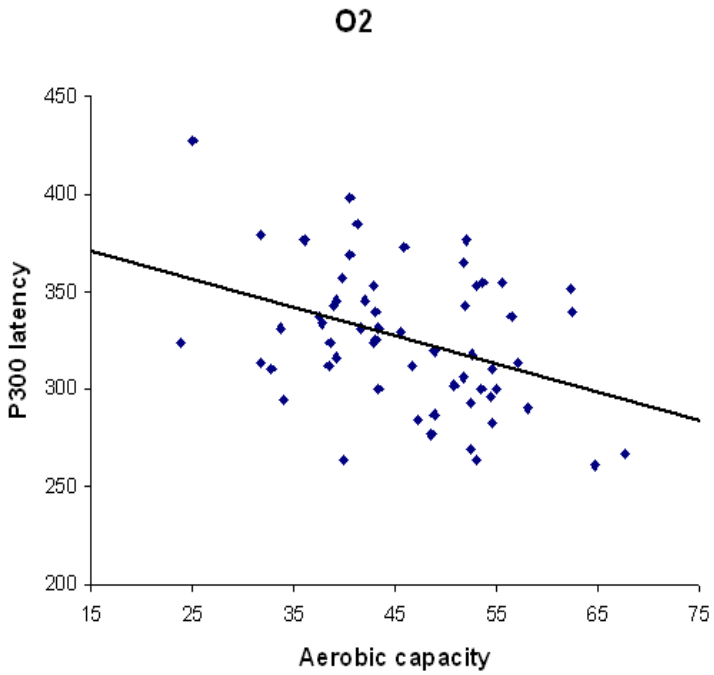


Figure 17.



APPENDICES

Appendix A: Human subjects review application

University of Maryland, College Park

Human Subjects Review Application

Project Title: The interactive effect of cardiovascular fitness and genotype on cerebral cortical responses during executive challenge in young adults

Project Director: Bradley D. Hatfield, Professor, Dept. of Kinesiology
Stephen M. Roth, Assistant professor, Dept. of Kinesiology
Student Investigator: Minjung Woo (Ph.D. student)

1. Abstract:

There are a number of neurobiological benefits of exercise and a positive influence on cognitive function has been observed in children and older adults, particularly for executive processes. Furthermore, the link between cardiovascular fitness and cognitive function is modified by genetic factors. In this regard, the Apolipoprotein (ApoE) e4 allele has been associated with neuro-cognitive degeneration in late adulthood and hypoactivation of brain metabolism has been observed in older individuals who carry the e4 allele, relative to non-carriers. More recently, a similar pattern of hypoactivation has been observed in young adults carrying the e4 allele. Furthermore, neuroimaging studies of children carrying the e4 allele revealed a thinner cerebral cortex (i.e., less cortical thickness), relative to non-carriers. Importantly, the negative effects of this genotype on the central nervous system (CNS) are attenuated by physical activity in elderly men and women. For example, Schuit et al. (2001) reported that the risk of cognitive decline, as measured by change on the Mini mental state examination (MMSE) over a three-year period, was reduced in high-active e4 carriers (> 60 years). However, there have been no studies conducted to date to determine if cardiovascular fitness would compensate for the negative impact of the e4 allele in young healthy adults. Such investigation is important in light of the incomplete development of the frontal lobes, which do not mature until age 25. The neurobiological benefits of exercise would hold particular importance for frontal development particularly in e4 carriers. Therefore, the purpose of the present study is to examine the relationship between cardiovascular fitness and cerebral cortical responses in young e4 carriers and non-carriers (college age male and female) during a frontally-mediated executive challenge.

2. Subject selection:

a. The subject population will consist of college aged (18-25 yrs) male and female. Participants will be recruited from undergraduate classes at UMCP.

b. Participants will be designated based on genotype (APOE e4 present vs. absent). Thirty carriers of a gene (ApoE e4 allele) and thirty non-carriers will be recruited. .

c. Carrying APOE e-4 allele has been identified as a genetic factor associated with early onset of Alzheimer's Disease (AD), cognitive decline and abnormal brain activation in old adult. Even hypoactivation and thinner cortex have been found in young adults and children who carry ApoE e4 allele. It has been shown that regular physical activity attenuates cognitive decline and abnormal brain activity with aging. Thus, it is speculated that cardiovascular fitness reduces negative impact of carrying e4 allele on brain in young adulthood. Accordingly, the purpose of this study is to examine this relationship between cardiovascular fitness and gene with college age young adults.

3. Procedures:

This study will consist of three days of testing. On day 1, after providing their informed consent (Appendix A), participants will be asked to take part in a non-invasive mouthwash procedure as a method of obtaining a DNA sample and complete a 3-minute brief survey to obtain a preliminary estimate of physical fitness. To complete the mouthwash protocol, participants will be asked to swish 10 ml of Scope mouthwash for 45 sec, after which they spit the mouthwash solution into a 50-ml sterile collection tube. DNA will be analyzed to determine genotype for APOE, which is related to mental functioning and memory. They will not be informed of their genotype. Volunteers who meet the criteria of this project (30 e4 carriers and 30 non-carriers) will be invited to complete Day 2 testing procedures. On day 2, participants will be asked to complete an interview on health history information including family history of AD, the Kaufman Brief Intelligence Test (K-BIT), the Multiple Affect Adjective Check List (MAACL), the Stroop color-word test and the seven day physical activity recall test (PAR) (Appendices B – F, respectively).

To estimate cardiovascular fitness, a submaximal cycle test will be used. Participants will be instructed to pedal, maintaining 50 rpm in 4 different workrate stages. Each stage will last for 3 minutes, and heart rate (HR) at the end of each stage will be recorded to estimate cardiovascular fitness. Workrate for the second stage will be determined based on the subject's HR at the end of the first stage, where the workrate is 0.5 kg. Then in the third and fourth stage, the workrate will be increased 0.5 kg. The test will have the subjects exercising up to a workrate that elicits a HR ~ equal to 85% of their age-predicted maximal HR.

Day 3 testing will include auditory reaction tests (Oddball: attention task and Go-nogo: executive function task) during Electroencephalography (EEG) recording at Cognitive Motor Neuroscience Lab on University of Maryland campus. More specifically, participants will be fitted with an electrode cap housing tin electrodes for the EEG recording. Tin electrodes will be placed above and below the right eye for recording of eye blinks, to the lateral side of each eye for the recording of horizontal eye movement, and on the mastoids to serve as a reference. Eye-channel electrode sites and sites on both ear lobes will be very lightly abraded with a 3M plastic abrasive pad and

then rubbed with alcohol and prepared with an FDA approved non-toxic conducting gel that enables continuous connection between the skin of the scalp and the sensor or electrode surface. The subject's skin will be lightly abraded with the flat end of a wooden q-tip at each electrode site (Fz - mid frontal, F3 – left frontal, F4 – right frontal, Cz – mid central, C3 – left central, C4 – right central, Pz – mid parietal, P3 – left parietal, P4 – right parietal, O1 – left occipital and O2 – right occipital) but the skin will not be broken. Fpz will serve as the ground (International 10-20 system, Jasper, 1958). Using a blunt end needle and syringe, the previously described conducting gel will be applied to each electrode site. Again, the skin will not be broken. All recording will be done in accord with the Society for Psychophysiological Research (SPR) Ad Hoc Committee Report Guidelines for reducing the risk of disease transmission as outlined in Putnam et al. 1992, Vol. 29, (2), 127-141, *Psychophysiology* (see Appendix G) including the fact that all researchers will wear approved latex gloves while working with the subjects. All electrical equipment is UL listed and fully grounded to eliminate any hazard electronic shock. Impedances will be checked and brought below 10 k Ω.

Participants will be instructed to sit quietly in a chair and look straight ahead. In both oddball and go-nogo task, participants will subject to three blocks of 100 tones (20 target and 80 non-target in each block). During the oddball task, participants will be asked to press a button with the dominant hand when they hear the target (rare) tones and to count the number of target tones they hear in each of the trials in order to maintain attention to the auditory stimuli. However, they will be asked to inhibit the button press to the rare tones and will not be asked to count the number of tones during the go-nogo task. The go-nogo task requires response inhibition so it has been used as a measure for executive function. Non-target tones will be 1000Hz and target tones will be 2000 Hz. Tones will be presented in the ear canal via a soft earplug insert. The interstimulus interval will be set at 2.00 seconds. Tones will be 80 dB in the go-nogo task and the oddball task. For each task, accuracy and response latencies (or accurate inhibition of button press) will be recorded for all trials in the experimental blocks. Accuracy will be determined by the percentage correct out of the total number of trials in the block. Failing to respond on a trial will result in an incorrect answer. In addition, responding when instructed to withhold a response also results in an error (i.e., Go-Nogo trials). Continuous EEG will be recorded during the three blocks of 100 trials each during the binaural auditory Go-Nogo task and the oddball task.

EEG will be acquired at a sampling rate of 512 Hz and amplified 20,000 times, while the eye channels will be amplified 5,000 times using Grass model 12A5 Neurodata Acquisition amplifiers with band-pass filter settings of 0.1-100 Hz (96-db/octave). Amplifiers will be calibrated prior to each testing session with a 10-Hz, 50-μV sinusoidal input signal that will be presented to all channels simultaneously. Data sampling will be controlled by Neuroscan software (Neuroscan Labs, Neurosoft, Inc., Sterling, VA) installed on a Gateway 2000 Pentium computer (Gateway, North Sioux City, SD). A subject ground will be placed in the midline frontal pole region (scalp surface) and the amplifiers employed pose no threat of electric shock.

4. Risks and Benefits:

There are no known risks or hazards associated with the mouthwash rinse, though the sensation of the astringent mouthwash can be uncomfortable for some people. The

submaximal cycling test may cause mild discomfort, fatigue or muscle pain after completion the test, even though there are no known risks associated with the test. As a result of wearing the electrode cap to measure brain activity subjects may experience some slight sensation as the scalp is lightly abraded at the electrode sites. To lessen the already minimal risk associated with EEG acquisition we will use standardized laboratory procedures to protect for skin abrasion and risk of infection (Appendix G). This study will confer no direct benefits to the subject but will contribute important health information.

5. Confidentiality:
All information collected in this study is confidential, and participants' name will not be identified at any time. The information collected on the questionnaires and memory tests will be kept in a locked cabinet that can be accessed only by the research team. DNA samples will be stored in a locked refrigerator, which can be accessed only by the research team in the Functional Genomics Laboratory in the Health and Human Performance building at the University of Maryland. All information collected will be kept for a period of 15 years, at which point it will be destroyed.

6. Information and Consent Forms: See Appendix A

7. Conflict of Interest: There are no conflicts of interest in this research

8. HIPAA Compliance: HIPAA regulations do not apply to this project. Participants' health-related information is kept confidential, in a locked cabinet. No individual will be identified, and no individual information will be reported. All study data are expressly separate from any University Health Center functions.

Appendix B: Consent form

Initials _____ Date

Project Title	***The interactive effect of cardiovascular fitness and genotype on cerebral cortical responses during executive challenge in young adults***
Why is this research being done?	*This is a research project being conducted by Bradley D. Hatfield, Ph.D and Minjung Woo(Ph.D student)) in the Department of Kinesiology at the University of Maryland, College Park. We are inviting you to participate in this research project because you belong to one of our target groups of healthy men and women between the age of 18 and 25. Some groups have a high level of regular physical activity while others have a high level of cardiovascular fitness. The purpose of this research project is to explore the relationships between cardiovascular fitness and heredity on brain function in college age group.*
What will I be asked to do?	*The procedure for this study will entail one short session that will take 5 minutes and two sessions that will take 1 and 1/2 hours each on separate days. On day 1, you will be asked to swish 10 ml of Scope mouthwash for 45 sec, after which you will spit the mouthwash solution into a collection tube. Following the mouthwash procedure, you will be asked to complete a 3- minute brief survey to obtain a preliminary estimate of your physical fitness. If you qualify for this study, you will be invited to a second day of testing. On day 2, you will be asked to complete an interview on health history information including family history of Alzheimer's Disease (AD), simple surveys to assess emotion and cognitive ability, and a questionnaire designed to examine physical activity. You will also be asked to complete a color word- naming test. In addition, you will be instructed to cycle for 12 minutes at different work rates, while maintaining a given speed. Heart rate will be recorded every 3 minute to estimate your aerobic capacity, which is an index of cardiovascular fitness. On the third day of testing, you will visit the Cognitive Motor Neuroscience Laboratory to complete auditory reaction tasks which contain six blocks of 100 tones (20 target and 80 non-target in each block). During the three blocks of 1st sound and reaction tasks, you will be asked to press a button when you hear target tones. During the three blocks of 2nd sound and reaction tasks, you will be asked to refrain from pressing the button when you hear the target tones. Tones during tasks will be 80 dB, and a relevant example of such noise levels is an alarm clock. The tone duration will be 0.3 seconds each time they are presented. These tasks will be performed while wearing an electrode cap (like a swim cap with sensors) that measures brain electrical activity. This procedure that measures brain activity is called Electroencephalography (EEG). For EEG recording, you will wear a stretch cap, in which 16 electrodes are embedded and distributed over the entire scalp region, and ear straps, which connect the cap to a chest strap in order to stabilize the position of the cap on your head. The skin beneath the sensors will be lightly rubbed with the wooden end of a cotton swab and gel will be applied to these sensor sites with a tube. Following scalp preparation, the cap will be connected to an EEG device for recording.*

Initials__ Date _____

Project Title	***The interactive effect of cardiovascular fitness and genotype on cerebral cortical responses during executive challenge in young adults***

Day 1	***Day 2***	***Day 3***
5 min	*90 min*	*70 min*
Consent, mouthwash, short physical activity survey	*Health history survey, Emotion survey, Physical activity survey, Intelligence test, Visual reaction task, Cycling test*	*Cap preparation, Baseline brain recording, 1st sound and reaction task, 2nd sound and reaction task*

You will be paid $ 2 for day 1 test, $ 10 for day 2 and $ 30 for day 3 test. You can stop participating at any time.

DNA**: Your DNA will be collected from a mouth wash sample and measured for only the ApolipoproteinE(ApoE) gene, which is related to Alzheimer's disease and cognitive impairment in aging. Your DNA samples will be kept for a period of 15 years before being destroyed since there may be new genes of interest discovered in the near future. If future genes relative to cognitive function/AD are uncovered, then the DNA that was collected from you can be used for an additional experiment. If future investigators want to use DNA material for an un-related research question and set of genes, they can do so only with your written permission. Your **DNA information <u>will not be disclosed to you</u>, but you may be referred to a clinical facility if you wish to seek further information regarding your own risk for cognitive impairment.

You have the right to refuse permission for your DNA to be used in un-related future studies and still participate in the current project. By checking the first option, the investigators have permission to contact you for consent to use additional DNA information, and that by checking the second option you are refusing the right for investigators to use your DNA for future studies:

*___**YES** Investigators may contact me regarding the use of my DNA for future studies.*

*___**NO** Investigators may not contact me to ask permission to use my DNA in future studies.*

Initials _____Date___

Project Title	***The interactive effect of cardiovascular fitness and genotype on cerebral cortical responses during executive challenge in young adults***
What about confidentiality?	*To help protect your confidentiality, this consent document and your health history form will be kept in a locked cabinet, separate from all the other information you provide. All other information collected in this study is numerically coded and identifying information will be removed from all data containing ID number. Thus, your name will not be identified at any time. The information collected on the questionnaires and cognitive tests will be kept in a different locked cabinet that can be accessed only by the research team. Your DNA samples will be stored in a locked refrigerator, which can be accessed only by the research team in the Functional Genomics Laboratory in the Health and Human Performance building at the University of Maryland. All information collected will be kept for a period of 15 years, at which point it will be destroyed. Despite all efforts to keep DNA results confidential, confidentiality cannot be absolutely guaranteed. When we write a report or article about this research project, we will present grouped results only; your identity will be protected to the maximum extent possible.*
What are the risks of this research?	*The sensation of the mouthwash may be uncomfortable for you, though there are no other known risks or hazards associated with the mouthwash rinse. You may feel mild discomfort, fatigue or muscle pain after completion of the 12- min cycle test, even though there are no known risks associated with this test. As a result of wearing the sensor cap to measure brain activity you may experience some slight sensation as the skin beneath the sensors is lightly rubbed.*
What are the benefits of this research?	*The experiment is not designed to help you personally, but the investigators hope to learn more about aging, heredity, physical activity, and brain function. The results from this study will be used to plan future projects based on the most important and relevant findings.*
Do I have to be in this research? May I stop participating at any time?	*Your participation in this research is completely voluntary. You may choose not to take part at all. If you decide to participate in this research, you may stop participating at any time. If you decide not to participate in this study or if you stop participating at any time, you will not be penalized or lose any benefits to which you otherwise qualify.*
Is any medical treatment available if I am injured?	*The University of Maryland does not provide any medical, hospitalization or other insurance for participants in this research study, nor will the University of Maryland provide any medical treatment or compensation for any injury sustained as a result of participation in this research study, except as required by law.*

Initials _____ Date _____

Project Title	***The interactive effect of cardiovascular fitness and genotype on cerebral cortical responses during executive challenge in young adults***	
What if I have questions?	*This research is being conducted by Dr. Bradley Hatfield, in Department of Kinesiology at the University of Maryland, College Park. If you have any questions about the research study itself, please contact: Dr. Bradley Hatfield (Primary investigator)at : 2134C HHP Bldg. Dept. of Kinesiology, College Park, MD 20742; office (301)-405-2485; email bhatfiel@umd.edu or Minjung Woo (Student investigator) at: 2237 HHP Bldg. Dept. of Kinesiology, College Park, MD 20742; office (301)-405-2466; email mjwoo@umd.edu* *If you have questions about your rights as a research subject or wish to report a research-related injury, please contact:* ***Institutional Review Board Office, University of Maryland, College Park, Maryland, 20742; (e-mail) irb@deans.umd.edu; (telephone) 301-405-0678*** *This research has been reviewed according to the University of Maryland, College Park IRB procedures for research involving human subjects.*	
Statement of Age of Subject and Consent	*Your signature indicates that:* *you are at least 18 years of age;,* *the research has been explained to you;* *your questions have been fully answered; and* *you freely and voluntarily choose to participate in this research project.*	
Signature and Date	**NAME OF SUBJECT**	
	SIGNATURE OF SUBJECT	
	DATE	

Appendix C: Medical History

Name_____ Telephone_____
Address_____
_____
Date of Birth_____ Age_____ Gender M____F____
Race, ethnicity: _____Caucasian _____Black _____Hispanic
_____Asian _____Other
Color Blind Yes____ No____

Medical History Are you currently taking or have you taken any of the following medications within the past two months?

Aspirin, Bufferin, Anacin	Tranquilizers
Blood pressure pills	Weight reducing pills
Cortisone	Blood thinning pills
Cough medicine	Dilantin
Digitalis	Allergy shots
Hormones	Water pills
Insulin or diabetic pills	Antibiotics
Iron or blood medications	Barbituates
Laxatives	Phenobarbital
Sleeping pills	Thyroid medicine

Other medications not listed

_____
_____
_

Do you currently or have you ever had any of the following medical disorders?

Heart attack	Yes___	No___
Chest pain	Yes___	No___
Hardening of the arteries	Yes___	No___
Irregular heart beat	Yes___	No___
Kidney disease	Yes___	No___
Diabetes	Yes___	No___
Cancer	Yes___	No___
Gout	Yes___	No___
Asthma	Yes___	No___
Epilepsy or seizure disorder	Yes___	No___
Migraine headaches	Yes___	No___ if yes, frequency, intensity____
Psychiatric disorder	Yes___	No___ if yes, what diagnosis_____

Physical Activity

_____Has your physical activity level remained consistent during the previous 5 years?
a. very consistent b. somewhat consistent c. inconsistent

Have you had any injuries or medical conditions that caused you to be physically inactive for more than 3 months during the past 5 years?

_____Yes _____No If yes explain_____

_____

We are interested in how stable your physical activity level has been over the course of your lifetime. Please use the scale below to rate your level of physical activity for the next five questions.

1 Very physically active, regular aerobic exercise and sports
2 Fairly physically active, sports and active leisure
3 Moderately physically active, hobbies, active leisure activities
4 Fairly physically inactive, very few sports, light physical leisure activities
5 Very physically inactive, no sports, non-physical leisure activities

____How would you characterize your physical activity level between the ages of 11 and 15?
____Between the ages of 16 and 20?
____Between the ages of 21 and current?

Family History

Do you have any biological parents, siblings, or grandparents who have been clinically diagnosed with Alzheimer's disease? ____Yes No____ If yes, please list them below.
Parents_____

Siblings_____

Grandparents_____

Appendix D.1. : 7-Day Physical Activity Recall

Follow-up 3

Data: _____ Participant ID#: _____

Worksheet

NOTE: If participant was sick last past week, DO NOT complete the worksheet.

Sleep Time: ____.____ *From* *bedtime to* *wake-up time*			
Physical Activity			
	Moderate	Hard	Very Hard
Morning Time	____.____	____.____	____.____
Afternoon Time	____.____	____.____	____.____
Evening Time	____.____	____.____	____.____

Rounding Time: 10 – 22 min = .25 1:08 – 1:22 hr/min = 1.25 23 – 37 min = .50
38 – 52 min = .75 53 – 1:07 hr/min = 1.0

Appendix D.2. : Description of physical activity categories for the 7-day physical activity recall

Please think about **moderate** intensity activities like the following:

Sports/Recreational Activities:	Brisk walking, Volley ball, Badminton, Cheerleading, Dancing, and Marching in the band
At home:	Ranking the lawn, Weeding in the garden, Housework (i.e., moping, cleaning windows, and sweeping) and Mowing the lawn with a walking mower.

Please think about **hard** intensity activities like the following:

Sports/Recreational Activities:	Aerobics, Doubles tennis, Speed Walking, Swimming (casually), and Skateboarding.
At home:	Scrubbing the floors, Digging in the garden, Stair climbing (moderate pace), and Wrestling with siblings.

Please think about **very hard** intensity activities like the following:

Sports/Recreational Activities:	Running (fast pace), Singles tennis, Backpacking (hilly country or rough trails), Rope jumping, Soccer, Basketball, and Roller-blading or roller-skating or ice-skating.
At home:	Lifting and carrying heavy loads (more than 50 lbs.)

Appendix E: 7-Day Physical Activity Recall

STROOP
COLOR AND WORD TEST

Name: _____

Age: _____ Sex: _____ Date: _____

FOR PROFESSIONAL USE ONLY

	RAW SCORE	AGE CORRECTED RAW SCORES*	T-SCORES*
WORD SCORE (W)			
COLOR SCORE (C)			
COLOR-WORD SCORE (CW)			
PREDICTED COLOR-WORD SCORE* (CW´) $\frac{W \times C}{W + C} = \frac{\_\_ \times \_\_}{\_\_ + \_\_} = \_\_\_\_ = CW'$			
CW – CW´ = INTERFERENCE * ___ – ___ = _____			

* INDICATES THAT AGE CORRECTED RAW SCORES SHOULD BE USED IF APPROPRIATE

DO NOT OPEN THE BOOKLET UNTIL YOU ARE INSTRUCTED TO DO SO.

RED	BLUE	GREEN	RED	BLUE
GREEN	GREEN	RED	BLUE	GREEN
BLUE	RED	BLUE	GREEN	RED
GREEN	BLUE	RED	RED	BLUE
RED	RED	GREEN	BLUE	GREEN
BLUE	GREEN	BLUE	GREEN	RED
RED	BLUE	GREEN	BLUE	GREEN
BLUE	GREEN	RED	GREEN	RED
GREEN	RED	BLUE	RED	BLUE
BLUE	GREEN	GREEN	BLUE	GREEN
GREEN	RED	BLUE	RED	RED
RED	BLUE	RED	GREEN	BLUE
GREEN	RED	BLUE	RED	GREEN
BLUE	BLUE	RED	GREEN	RED
RED	GREEN	GREEN	BLUE	BLUE
BLUE	BLUE	RED	GREEN	RED
RED	GREEN	BLUE	RED	GREEN
GREEN	RED	GREEN	BLUE	BLUE
RED	BLUE	RED	GREEN	RED
GREEN	RED	GREEN	BLUE	GREEN

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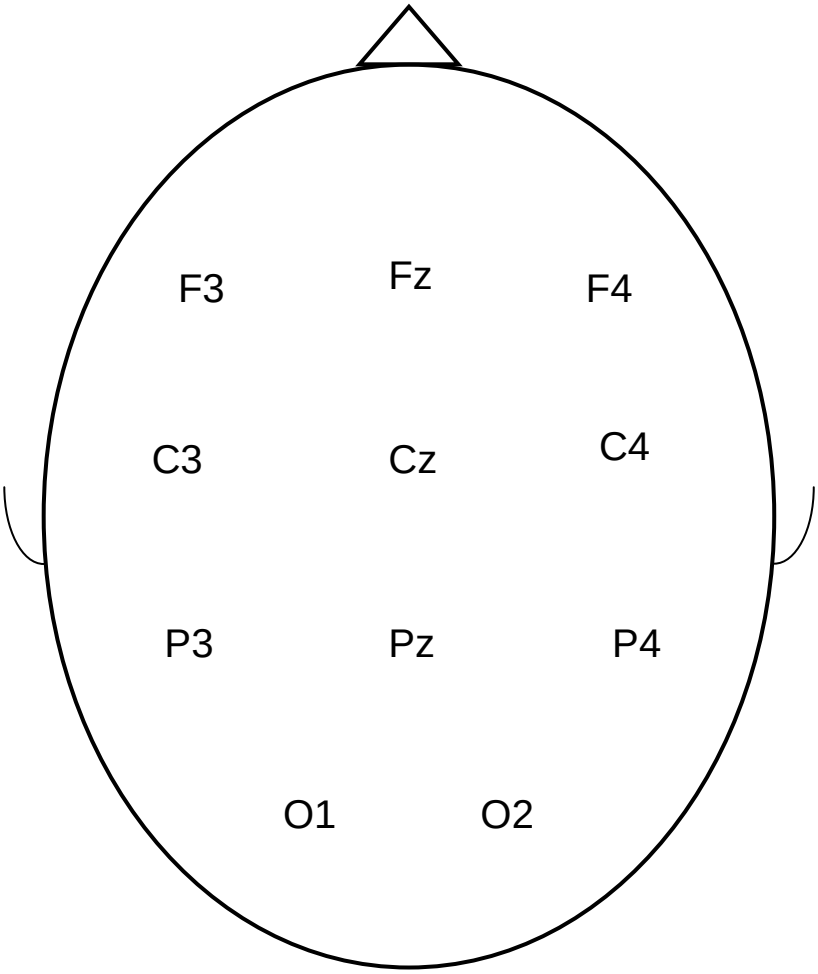
RED	BLUE	GREEN	RED	BLUE
GREEN	GREEN	RED	BLUE	GREEN
BLUE	RED	BLUE	GREEN	RED
GREEN	BLUE	RED	RED	BLUE
RED	RED	GREEN	BLUE	GREEN
BLUE	GREEN	BLUE	GREEN	RED
RED	BLUE	GREEN	BLUE	GREEN
BLUE	GREEN	RED	GREEN	RED
GREEN	RED	BLUE	RED	BLUE
BLUE	GREEN	GREEN	BLUE	GREEN
GREEN	RED	BLUE	RED	RED
RED	BLUE	RED	GREEN	BLUE
GREEN	RED	BLUE	RED	GREEN
BLUE	BLUE	RED	GREEN	RED
RED	GREEN	GREEN	BLUE	BLUE
BLUE	BLUE	RED	GREEN	RED
RED	GREEN	BLUE	RED	GREEN
GREEN	RED	GREEN	BLUE	BLUE
RED	BLUE	RED	GREEN	RED
GREEN	RED	GREEN	BLUE	GREEN

Appendix F: Topographical map of EEG recording sites



Appendix G: Flyer for subject recruitment

Are you a healthy 18-23 years old MALE interested in participating in a research study?

We are seeking individuals to participate in a study examining the relationship between cardiovascular fitness and heredity on brain activity during the simple task.

What will you be asked to do?

Screening

45 second Mouthwash for DNA sampling

You will receive **$ 2** for 45 second mouthwash

If you qualify to the study, you will be invited for further testing

Test

Simple surveys

10 min easy cycling test

Brain activity recording

You will receive **$ 40** for 2 hour 15 min Test

Where? At the Cognitive Motor Neuroscience Laboratory, Department of Kinesiology, University of Maryland, College Park

INTERESTED IN PARTICIPATING?

Mirjung Woo
mjwoo@umd.edu
301-405-2466

Appendix H: Directions for Collecting DNA sample

PLEASE
DO NOT **EAT**, **DRINK**, **CHEW GUM**, or **RINSE** YOUR MOUTH FOR **ONE HOUR**
BEFORE COLLECTING THE SALIVA SAMPLE!

Directions for Collecting DNA Sample:

1) Open bottle of Scope. Open the collection container and fill with mouthwash to black fill line (10mL).

2) SWISH the mouthwash from the container around in your mouth vigorously for **45 seconds. Watch clock when doing this!**

   DO NOT GARGLE or CLEAR THROAT! SWISH ONLY.

3) Holding the container close to your mouth, spit the mouthwash back in the container. Replace the top on the container and screw it on tightly.

4) Mail sample within 24 hours of collection! Keep sample at room temperature.

Appendix I: Payment Voucher

UNIVERSITY OF MARYLAND
DEPARTMENT OF KINESIOLOGY

2351 HHP Building
College Park, Maryland 20742-2611
TEL 301.405.2450 FAX 301.405.5578
www.hhp.umd.edu/knes

PHYSICAL ACTIVITY, GENE AND BRAIN STUDY
PAYMENT VOUCHER

Instructions: Take this completed voucher to Ms. Regina Clary, KNES Business Office, 2334 HHP, x5-2506

_____ _____ _____
(Print Name) **(Date)** **(Amount)**

_____
Permanent Address: Street, City, State, Zip

_____
Signature **Social Security Number**

Signature of Authorized Research Staff _____ **DATE:** _____

Appendix J.1 ~ J.6 : Summary of hierarchical regression analyses

Appendix J.1. Summary of hierarchical regression analyses for P300 amplitude in Go-nogo task

Measure	Model	Standardized Beta Coefficients			R^2	Change in R^2	df	F change	P
		Gene	VO_2max	Gene × VO_2max					
Fz	1	-.095			.009	.009	1/57	.523	.473
	2	-.064	.237		.064	.055	1/56	3.31	.074
	3	-1.355	-.081	1.313	**.128**	**.064**	**1/55**	**4.023**	**.05**
F3	1	-.077			.006	.006	1/57	.342	.561
	2	-.041	.278		**.082**	**.076**	**1/56**	**4.615**	**.036**
	3	-1.234	-.017	1.214	.136	.054	1/55	3.469	.068
F4	1	-.16			.026	.026	1/57	1.499	.226
	2	-.132	.21		.069	.043	1/56	2.607	.112
	3	-1.452	-.115	1.342	**.136**	**.067**	**1/55**	**4.237**	**.044**
Cz	1	-.198			.039	.039	1/57	2.329	.132
	2	-.171	.208		.082	.042	1/56	2.585	.114
	3	-1.26	-.061	1.108	.127	.045	1/55	2.859	.097
C3	1	-.168			.028	.028	1/57	1.654	.204
	2	-.14	.209		.071	.043	1/56	2.579	.114
	3	-.976	.003	.849	.098	.027	1/55	1.626	.208
C4	1	-.205			.042	.042	1/57	2.507	.119
	2	-.173	.245		.101	.059	1/56	3.66	.061
	3	-1.228	-.016	1.073	.143	.043	1/55	2.732	.104
Pz	1	-.089			.008	.008	1/57	.45	.502
	2	-.083	.044		.010	.002	1/56	.106	.746
	3	-.695	-.107	.622	.024	.014	1/55	.807	.373
P3	1	-.091			.008	.008	1/57	.478	.492
	2	-.082	.072		.013	.005	1/56	.293	.591
	3	-.443	-.017	.368	.018	.005	1/55	.281	.598
P4	1	-.188			.035	.035	1/57	2.083	.154

	2	-.182	.044		.037	.002	1/56	.112	.74
	3	-.593	-.057	.418	.044	.006	1/55	.371	.545
O1	1	.032			.001	.001	1/57	.058	.811
	2	.035	.022		.002	.000	1/56	.028	.869
	3	.969	.253	-.951	.035	.033	1/55	1.904	.173
O2	1	-.056			.003	.003	1/57	.177	.676
	2	-.067	-.087		.011	.007	1/56	.422	.519
	3	.775	.121	-.857	.038	.027	1/55	1.551	.218

Appendix J.2.. Summary of separate regression analysis for P300 amplitude at Fz, F3 and F4 sites in e4 carrier and non-carrier in Go-nogo task.

	Standardized Beta Coefficients	**R^2**	**df**	***F***	***P***
Non-carrier					
Fz	-.082	.007	1/28	.188	.668
F4	-.117	.014	1/28	.386	.539
E4-carrier					
Fz	.436	.19	1/27	6.352	**.018**
F4	.418	.175	1/27	5.725	**.024**

Appendix J.3. Summary of hierarchical regression analyses for P300 amplitude in Oddball task

Measure	Model	Standardized Beta Coefficients				R^2	Change in R^2	df	F change	P
		I.Q.	Gene	VO_2max	Gene × VO_2max					
Fz	1	.300				.090	.090	1/57	5.64	.021
	2	.283	.114			.103	.013	1/56	..791	.378
	3	.301	.139	.214		.147	.045	1/55	2.888	.095
	4	.290	-.852	-.032	1.010	.185	.038	1/54	2.491	.12
F3	1	.217				.047	.047	1/57	2.827	.098
	2	.194	.156			.071	.024	1/56	1.434	.236
	3	.215	.185	.247		.131	.060	1/55	3.774	.057
	4	.206	-.641	.042	.841	.157	.026	1/54	1.672	.202
F4	1	.173				.030	.030	1/57	1.767	.189
	2	.169	.027			.031	.001	1/56	.040	.842
	3	.183	.046	.159		.055	.025	1/55	1.430	.237
	4	.17	-1.081	-.121	1.147	.104	.049	1/54	2.923	.093
Cz	1	.299				.090	.090	1/57	5.612	.021
	2	.309	-.064			.094	.004	1/56	.248	.620
	3	.328	-.038	.221		.141	.048	1/55	3.045	.087
	4	.316	-1.136	-.051	1.118	.187	.046	1/54	3.063	.086
C3	1	.307				.094	.094	1/57	5.942	.018
	2	.308	-.002			.094	.000	1/56	.000	.988
	3	.332	.033	.294		.179	.084	1/55	5.637	.021
	4	.317	-1.372	-.055	1.431	.254	.076	1/54	5.469	.023
C4	1	.167				.028	.028	1/57	1.640	.206
	2	.172	-.032			.029	.001	1/56	.056	.814
	3	.189	-.007	.204		.070	.041	1/55	2.402	.127
	4	.175	-1.281	-.112	1.298	.132	.062	1/54	3.861	.055
Pz	1	.171				.029	.029	1/57	1.722	.195
	2	.182	-.072			.034	.005	1/56	.292	.591
	3	.191	-.059	.111		.046	.012	1/55	.690	.410

	4	.189	-.254	.062	.199	.048	.001	1/54	.083	.775
P3	1	.206				.042	.042	1/57	2.520	.118
	2	.219	-.088			.050	.008	1/56	.445	.507
	3	.233	-.068	.169		.078	.028	1/55	1.652	.204
	4	.230	-.380	.091	.317	.081	.004	1/54	.218	.642
P4	1	.115				.013	.013	1/57	.766	.385
	2	.133	-.122			.028	.014	1/56	.832	.386
	3	.140	-.113	.074		.033	.005	1/55	.304	.584
	4	.137	-.327	.021	.218	.035	.002	1/54	.098	.755
O1	1	.053				.003	.003	1/57	.162	.689
	2	.059	-.038			.004	.001	1/56	.078	.780
	3	.062	-.033	.038		.006	.001	1/55	.079	.779
	4	.069	.547	.182	-.591	.019	.013	1/54	.708	.404
O2	1	-.003				.000	.000	1/57	.001	.980
	2	.013	-.105			.011	.011	1/56	.614	.437
	3	.005	-.116	-.086		.018	.007	1/55	.407	.526
	4	.009	.267	.008	-.389	.024	.006	1/54	.309	.581

Appendix J.4. Summary of separate regression analysis for p300 amplitude at C3 site in e4 carrier and non carrier in Oddball task.

	Standardized Beta Coefficients	**R^2**	**df**	***F***	***P***
Non-carrier	-.099	.01	1/28	.277	.603
E4-carrier	.51	.26	1/27	9.478	.005

Appendix J.5 Summary of hierarchical regression analyses for P300 latency in Go-nogo task

Measure	Model	Standardized Beta Coefficients			R^2	Change in R^2	df	F change	P
		Gene	VO_2max	Gene × VO_2max					
Fz	1	-.148			.022	.022	1/57	1.278	.263
	2	-.175	-.205		.063	.041	1/56	2.474	.121
	3	1.029	.092	-1.225	.119	.055	1/55	3.462	.068
F3	1	-.094			-.009	.009	1/57	.503	.481
	2	0.127	-.257		.041	.065	1/56	3.932	.052
	3	.943	.007	-1.089	.070	.044	1/55	2.733	.104
F4	1	-.102			.010	.010	1/57	.602	.441
	2	-.141	-.295		**.096**	**.086**	**1/56**	**5.303**	**.025**
	3	.716	-.084	-.872	.124	.028	1/55	1.766	.189
Cz	1	-.054			.003	.003	1/57	.166	.685
	2	-.083	-.223		.052	.049	1/56	2.889	.095
	3	.521	-.074	-.614	.066	.014	1/55	.822	.369
C3	1	-.107			.011	.011	1/57	.661	.42
	2	-.14	-.252		.074	.062	1/56	3.769	.057
	3	.563	-.078	-.715	.093	.019	1/55	1.147	.289
C4	1	-.095			-.008	.009	1/57	.523	.472
	2	-.126	-.231		.028	.053	1/56	3.138	.082
	3	.657	-.038	-.796	.035	.023	1/55	1.408	.24
Pz	1	-.078			.006	.006	1/57	.345	.559
	2	-.107	-.223		.055	.049	1/56	2.883	.095
	3	.166	-.155	-.277	.058	.003	1/55	.166	.685
P3	1	-.092			.008	.008	1/57	.483	.49
	2	-.128	-.274		**.082**	**.074**	**1/56**	**4.5**	**.038**
	3	.265	-.177	-.4	.088	.006	1/55	.356	.553
P4	1	-.057			.003	.003	1/57	.184	.67
	2	-.086	-.224		.053	.049	1/56	2.921	.093
	3	.356	-.115	-.089	.060	.007	1/55	.437	.511
O1	1	-.11			.012	.012	1/57	.695	.408

	2	-.143	-.254		.075	.063	1/56	3.833	.055
	3	.296	-.146	-.446	.083	.007	1/55	.442	.509
O2	1	-.023			.001	.001	1/57	.029	.864
	2	-.056	-.251		.063	.062	1/56	3.711	.059
	3	.235	-.18	-.296	.066	.003	1/55	.19	.664

Appendix J.6 Summary of hierarchical regression analyses for P300 latency in Oddball task

Measure	Model	Standardized Beta Coefficients			R^2	Change in R^2	df	F change	P
		Gene	VO_2max	Gene × VO_2max					
Fz	1	-.056			.003	.003	1/57	.178	.675
	2	-.072	-.123		.018	.015	1/56	.85	.361
	3	.659	.057	-.743	.038	.02	1/55	1.169	.284
F3	1	-.078			.006	.006	1/57	.346	.559
	2	-.094	-.126		.022	.016	1/56	.89	.35
	3	.585	.042	-.69	.039	.018	1/55	1.009	.32
F4	1	-.08			.006	.006	1/57	.365	.548
	2	-.096	-.123		.021	.015	1/56	.856	.359
	3	.796	.097	-.907	.052	.03	1/55	1.764	.19
Cz	1	-.09			.008	.008	1/57	.465	.498
	2	-.097	-.052		.011	.003	1/56	.152	.698
	3	.628	.126	-.737	.031	.020	1/55	1.141	.29
C3	1	-.085			.007	.007	1/57	.418	.521
	2	-.100	-.112		.02	.012	1/56	.7	.406
	3	.729	.093	-.843	.046	.026	1/55	1.515	.224
C4	1	-.022			.000	.000	1/57	.028	.868
	2	-.039	-.128		.017	.016	1/56	.923	.341
	3	1.01	.13	-1.067	.059	.042	1/55	2.458	.123
Pz	1	-.078			.006	.006	1/57	.347	.558
	2	-.106	-.217		.052	.046	1/56	2.743	.103
	3	.439	-.093	-.554	.064	.011	1/55	.667	.418
P3	1	-.085			.007	.007	1/57	.419	.52
	2	-.114	-.213		.052	.045	1/56	2.646	.109
	3	.815	.016	-.944	.085	.033	1/55	1.983	.165
P4	1	-.076			.006	.006	1/57	.335	.565
	2	-.110	-.257		.071	.065	1/56	3.918	.053
	3	.823	-.027	-.949	.104	.033	1/55	2.044	.158
O1	1	-.032			.001	.001	1/57	.06	.807

	2	-.066	-.257		.068	.065	1/56	3.892	.053
	3	.398	-.142	-.472	.074	.008	1/55	.49	.487
O2	1	.062			.004	.004	1/57	.218	.642
	2	.011	-.382		**.147**	**.143**	**1/56**	**9.411**	**.003**
	3	.26	-.321	-.253	.150	.002	1/55	.153	.697

Appendix K.1. Behavior data of non-carrier

Sub ID	Fitness	Age	VO_2max	Energy	K-BIT	Stroop	NG_AC	NG_RT	OD_AC	OD_RT
1	1	20	48.95	44.34	102	-7.30	96.33	0.37	98.33	0.38
2	1	19	51.77	42.27	98	14.05	98.33	0.41	98.33	0.44
3	1	20	51.78	44.25	102	11.81	96.67	0.43	98.67	0.65
4	1	21	54.43	40.45	81	5.15	96.67	0.35	99.33	0.32
5	1	19	53.02	43.05	114	0.97	95.33	0.37	99.00	0.35
6	1	22	58.18	41.21	112	-12.09	95.67	0.33	96.67	0.33
7	1	20	52.50	41.20	116	-14.03	98.00	0.48	100.00	0.46
8	1	21	54.58	60.30	100	1.84	95.33	0.38	99.00	0.40
9	1	22	52.47	43.25	103	10.27	97.33	0.34	99.67	0.31
10	1	21	51.85	42.38	96	1.60	96.33	0.44	99.33	0.34
11	1	21	62.47	37.79	115	18.47	97.00	0.30	98.67	0.36
12	1	20	53.60	45.07	114	17.05	94.67	0.38	99.00	0.44
13	1	22	52.99	47.89	103	-6.98	94.33	0.36	99.00	0.34
14	1	20	62.25	38.57	104	12.43	97.00	0.37	99.67	0.39
15	1	22	50.86	41.96	103	11.33	96.33	0.31	99.33	0.38
16	2	19	33.66	48.00	115	12.82	99.00	0.33	99.67	0.40
17	2	19	40.48	43.71	103	-5.47	96.00	0.46	99.33	0.41
18	2	20	43.13	35.05	109	-0.57	95.67	0.37	100.00	0.39
19	2	21	39.13	44.36	92	-9.09	97.67	0.37	99.67	0.30
20	2	20	43.35	36.34	102	20.29	96.67	0.39	99.67	0.41
21	2	19	38.56	44.45	116	10.03	95.00	0.37	99.00	0.33
22	2	21	31.66	47.75	109	5.47	96.33	0.36	99.33	0.35
23	2	21	41.32		82	2.78	98.33	0.39	99.33	0.43
24	2	21	47.24	34.79	109	9.84	97.33	0.31	99.67	0.32
25	2	26	48.87	34.07	99	10.27	98.33	0.37	100.00	0.39
26	2	20	31.78	37.36	113	24.73	97.33	0.34	98.00	0.43
27	2	21	38.41	42.03	116	8.61	98.00	0.28	99.67	0.38
28	2	21	38.85	37.16	115	27.20	96.33	0.38	99.00	0.51
29	2	22	48.60	46.80	114	10.47	95.67	0.27	99.33	0.47
30	2	20	45.96	34.09	104	11.59	95.33	0.39	97.67	0.72

Appendix K.2. P300 latency during oddball task in non-carrier

Sub ID	Fz	F3	F4	Cz	C3	C4	Pz	P3	P4	O1	O2
1	290.62	298.44	288.67	300.39	306.25	300.39	312.11	316.02	312.11	321.87	319.92
2	300.39	302.34	302.34	304.3	308.2	310.16	308.2	310.16	312.11	306.25	306.25
3	368.75	380.47	394.14	374.61	384.37	382.42	376.56	378.52	376.56	370.7	364.84
4	317.97	319.92	319.92	302.34	304.3	306.25	298.44	296.48	298.44	290.62	296.48
5	345.31	370.7	362.89	331.64	374.61	360.94	364.84	366.8	364.84	353.12	353.12
6	312.11	316.02	314.06	306.25	310.16	312.11	312.11	310.16	314.06	296.48	290.62
7	290.62	288.67	292.58	276.95	280.86	290.62	284.77	284.77	292.58	312.11	292.58
8	327.73	321.87	327.73	341.41	343.36	312.11	331.64	337.5	335.55	351.17	282.81
9	276.95	275	282.81	263.28	275	273.05	269.14	267.19	273.05	271.09	269.14
10	259.37	265.23	280.86	325.78	329.69	319.92	333.59	333.59	331.64	335.55	343.36
11	329.69	339.45	331.64	329.69	333.59	333.59	339.45	341.41	341.41	314.06	339.45
12	349.22	351.17	345.31	355.08	358.98	353.12	355.08	357.03	355.08	357.03	355.08
13	288.67	290.62	288.67	282.81	292.58	292.58	275	370.7	355.08	263.28	263.28
14	339.45	335.55	339.45	355.08	358.98	351.17	353.12	355.08	353.12	349.22	351.17
15	376.56	374.61	368.75	364.84	366.8	360.94	335.55	343.36	321.87	276.95	302.34
16	304.3	306.25	304.3	304.3	317.97	312.11	323.83	327.73	323.83	302.34	331.64
17	329.69	331.64	321.87	351.17	355.08	349.22	374.61	366.8	378.52	398.05	398.05
18	349.22	347.27	349.22	339.45	335.55	341.41	349.22	347.27	341.41	345.31	325.78
19	314.06	321.87	304.3	278.91	308.2	282.81	302.34	310.16	314.06	308.2	316.02
20	372.66	380.47	378.52	376.56	382.42	372.66	386.33	386.33	343.36	370.7	331.64
21	310.16	312.11	308.2	306.25	302.34	306.25	314.06	314.06	312.11	325.78	323.83
22	292.58	296.48	292.58	286.72	286.72	288.67	290.62	288.67	290.62	300.39	314.06
23	374.61	384.37	372.66	382.42	386.33	378.52	386.33	384.37	390.23	392.19	384.37
24	292.58	290.62	294.53	286.72	286.72	292.58	280.86	284.77	288.67	284.77	284.77
25	298.44	294.53	294.53	288.67	290.62	286.72	288.67	290.62	288.67	284.77	286.72
26	331.64	337.5	337.5	327.73	339.45	327.73	351.17	345.31	362.89	319.92	378.52
27	265.23	269.14	271.09	267.19	284.77	282.81	306.25	312.11	312.11	321.87	312.11
28	333.59	337.5	343.36	339.45	351.17	341.41	347.27	353.12	347.27	347.27	343.36
29	296.48	302.34	298.44	300.39	310.16	323.83	316.02	325.78	321.87	282.81	276.95
30	310.16	314.06	314.06	300.39	329.69	300.39	358.98	351.17	360.94	364.84	372.66

Appendix K.3. P300 latency during Go-nogo task in non-carrier

Sub ID	Fz	F3	F4	Cz	C3	C4	Pz	P3	P4	O1	O2
1	304.3	310.16	306.25	310.16	314.06	316.02	327.73	327.73	329.69	339.45	345.31
2	314.06	316.02	317.97	317.97	323.83	325.78	323.83	325.78	317.97	304.3	306.25
3	366.8	362.89	376.56	357.03	360.94	362.89	349.22	355.08	347.27	349.22	339.45
4	323.83	321.87	325.78	312.11	312.11	317.97	302.34	304.3	312.11	292.58	296.48
5	298.44	296.48	323.83	312.11	327.73	339.45	329.69	335.55	349.22	339.45	337.5
6	292.58	292.58	292.58	290.62	290.62	296.48	300.39	304.3	308.2	335.55	302.34
7	300.39	306.25	300.39	290.62	294.53	296.48	290.62	298.44	300.39	310.16	308.2
8	343.36	339.45	343.36	327.73	339.45	333.59	329.69	331.64	325.8	345.32	337.59
9	280.86	286.72	284.77	267.19	294.53	271.09	288.67	296.48	288.67	337.5	351.17
10	310.16	273.05	312.11	265.23	271.09	325.78	362.89	362.89	370.7	366.8	372.66
11	286.72	290.62	288.67	275	282.81	280.86	286.72	300.39	294.53	329.69	290.62
12	333.59	335.55	333.59	321.87	327.73	325.78	321.87	327.73	327.73	329.69	331.64
13	296.48	304.3	298.44	288.67	302.34	294.53	294.53	292.58	290.62	288.67	290.62
14	398.05	398.05	308.2	362.89	380.47	345.31	353.12	357.03	349.22	343.36	343.36
15	331.64	329.69	335.55	323.83	331.64	327.73	314.06	339.45	296.48	296.48	300.39
16	306.25	306.25	306.25	308.2	317.97	310.16	337.5	343.36	341.41	366.8	353.12
17	341.41	343.36	337.5	351.17	353.12	349.22	358.98	362.89	358.98	378.52	370.7
18	333.59	339.45	337.5	357.03	329.69	351.17	364.84	392.19	355.08	353.12	341.41
19	319.92	333.59	314.06	314.06	366.8	308.2	364.84	368.75	353.12	378.52	360.94
20	333.59	331.64	333.59	335.55	331.64	337.5	343.36	341.41	343.36	372.66	358.98
21	308.2	310.16	302.34	298.44	292.58	300.39	300.39	300.39	306.25	316.02	319.92
22	294.53	296.48	292.58	286.72	288.67	288.67	290.62	290.62	294.53	298.44	312.11
23	360.94	362.89	360.94	376.56	374.61	382.42	382.42	392.19	372.66	372.66	353.12
24	304.3	302.34	302.34	300.39	300.39	304.3	304.3	304.3	306.25	310.16	308.2
25	273.05	276.95	271.09	269.14	276.95	269.14	273.05	278.91	276.95	296.48	290.62
26	312.11	317.97	312.11	302.34	323.83	310.16	294.53	321.87	296.48	308.2	284.77
27	271.09	278.91	282.81	263.28	282.81	278.91	294.53	300.39	296.48	306.25	308.2
28	347.27	362.89	360.94	347.27	349.22	349.22	355.08	355.08	351.17	353.12	353.12
29	310.16	306.25	314.06	314.06	310.16	317.97	316.02	314.06	316.02	310.16	335.55
30	376.56	378.52	372.66	374.61	368.75	372.66	374.61	370.7	374.61	370.7	376.56

Appendix K.4. P300 amplitude during oddball in non-carrier

Sub ID	Fz	F3	F4	Cz	C3	C4	Pz	P3	P4	O1	O2
1	12.40	8.31	11.79	22.23	18.30	18.60	25.63	21.89	19.97	12.57	10.36
2	13.68	11.99	11.99	24.15	15.91	19.26	28.54	24.64	24.26	22.13	20.41
3	7.47	3.47	11.37	17.32	12.62	16.53	20.13	17.22	19.42	15.13	17.84
4	11.17	11.50	10.41	18.15	17.28	18.88	25.13	23.40	22.48	20.59	20.36
5	2.46	2.62	1.69	9.61	7.07	6.39	14.25	13.02	13.57	10.81	10.70
6	19.92	15.40	16.67	27.27	22.89	21.07	26.20	22.77	19.83	14.83	14.69
7	1.23	1.83	0.31	10.09	10.16	5.62	15.54	14.25	10.08	7.19	6.18
8	-3.10	0.11	3.36	8.66	8.76	8.54	17.70	16.54	15.25	8.45	9.53
9	11.12	10.36	14.18	16.28	13.57	14.09	14.33	14.66	13.14	9.34	2.77
10	2.83	5.85	5.86	10.10	10.47	12.72	18.44	15.26	19.21	10.73	13.78
11	15.37	11.85	10.31	23.78	18.09	17.30	23.30	21.07	19.10	8.10	7.12
12	6.36	5.41	5.72	14.56	12.66	13.52	17.76	14.85	15.12	11.04	11.60
13	7.81	8.08	4.31	12.49	9.42	10.36	12.43	9.06	13.46	6.32	9.15
14	2.79	1.90	2.24	13.86	6.59	11.01	17.64	16.06	14.32	12.88	10.00
15	5.97	8.59	8.32	19.36	15.18	13.61	19.64	17.65	15.83	5.34	4.45
16	6.69	2.29	9.48	18.03	10.79	16.37	18.51	15.45	16.31	4.79	5.77
17	10.07	10.82	9.32	19.53	17.65	16.35	22.34	19.30	18.51	10.73	11.07
18	13.69	9.32	16.24	20.55	15.47	20.21	12.33	17.44	17.72	9.36	12.82
19	6.56	5.81	11.40	9.97	7.84	16.87	12.36	10.56	15.85	6.88	9.36
20	0.50	-1.06	-0.68	6.40	3.56	4.77	9.93	6.10	8.45	7.06	8.74
21	3.12	1.29	7.39	15.49	11.67	16.75	19.63	16.68	20.78	15.34	17.59
22	10.48	8.73	9.51	15.39	13.39	15.13	20.58	17.47	17.85	17.82	15.66
23	9.98	8.45	10.91	20.73	15.55	16.31	19.97	15.40	14.88	5.43	5.33
24	11.31	7.27	10.90	14.61	13.18	12.02	16.77	13.70	13.63	9.58	9.81
25	12.63	9.79	12.24	19.76	14.21	20.42	19.62	16.25	20.07	12.10	13.67
26	11.16	9.63	8.98	28.60	22.78	16.73	23.57	21.33	16.16	5.95	9.74
27	25.25	17.69	18.33	34.22	27.20	24.74	31.67	27.12	23.78	12.87	12.68
28	4.45	5.99	1.77	6.26	8.03	4.16	13.92	14.43	13.39	11.37	11.19
29	16.17	13.48	11.78	15.31	14.89	14.82	17.03	15.61	13.59	5.64	5.09
30	6.03	5.50	5.18	7.28	5.46	7.94	6.59	7.63	7.36	2.94	3.72

Appendix K.5. P300 latency during Go-nogo task in non-carrier

Sub ID	Fz	F3	F4	Cz	C3	C4	Pz	P3	P4	O1	O2
1	13.15	10.62	10.37	23.60	18.43	17.81	28.07	22.93	21.83	15.72	14.47
2	14.71	13.81	14.81	27.14	25.12	24.46	25.09	23.85	27.97	19.80	21.54
3	13.75	11.40	13.07	19.76	15.38	17.43	18.89	16.74	17.21	10.07	11.16
4	16.25	16.91	13.42	23.62	22.41	20.37	22.79	22.33	19.78	16.57	17.27
5	2.08	0.30	2.09	10.11	6.15	9.38	11.85	10.49	12.49	9.09	11.22
6	20.89	13.82	13.89	33.65	23.21	21.35	24.62	20.67	20.77	10.13	11.43
7	15.24	12.63	14.40	24.87	19.88	20.47	27.38	20.86	19.09	8.94	9.44
8	12.30	8.80	13.06	22.02	22.00	21.90	21.16	20.22	19.83	13.32	10.37
9	19.92	19.90	18.60	33.27	26.43	25.08	21.59	19.86	15.19	10.60	9.32
10	8.94	4.34	5.99	13.75	10.63	10.25	12.63	15.12	12.96	10.89	13.31
11	21.49	17.29	17.25	31.55	25.36	23.60	22.21	21.39	21.69	8.82	5.29
12	21.87	20.77	17.77	27.77	24.06	22.81	23.55	20.56	21.11	14.61	14.26
13	7.81	8.08	4.31	12.49	9.42	10.36	12.43	9.06	13.46	6.32	9.15
14	2.79	1.90	2.24	13.86	6.59	11.01	17.64	16.06	14.32	12.88	10.00
15	5.97	8.59	8.32	19.36	15.18	13.61	19.64	17.65	15.83	5.34	4.45
16	12.58	8.53	13.35	26.59	17.11	20.75	27.35	21.24	22.85	8.14	9.21
17	11.54	10.69	9.17	22.91	18.54	18.19	20.33	17.83	17.48	8.28	7.74
18	11.16	8.17	16.64	22.35	14.81	19.65	19.34	15.81	19.09	9.57	11.36
19	12.57	9.89	13.60	17.30	13.15	18.10	15.23	14.23	15.31	7.01	7.46
20	9.44	5.76	8.22	14.93	8.53	11.99	13.78	6.59	12.52	3.65	6.93
21	10.90	7.51	11.72	21.58	16.18	19.69	21.46	17.58	20.44	13.06	14.90
22	21.91	16.81	17.05	25.55	19.69	20.56	21.39	19.98	18.78	13.59	10.40
23	17.21	14.62	13.41	23.93	19.67	20.24	19.69	19.09	17.90	5.42	8.80
24	17.54	16.70	15.54	21.40	16.23	17.77	17.92	16.19	16.93	10.85	11.86
25	13.03	13.30	12.28	19.80	17.34	17.12	17.76	16.58	17.94	11.82	11.45
26	15.95	13.65	12.42	34.17	24.60	19.26	25.27	20.93	19.62	3.81	7.37
27	25.25	17.69	18.33	34.22	27.20	24.74	31.67	27.12	23.78	12.87	12.68
28	4.45	5.99	1.77	6.26	8.03	4.16	13.92	14.43	13.39	11.37	11.19
29	16.17	13.48	11.78	15.31	14.89	14.82	17.03	15.61	13.59	5.64	5.09
30	6.03	5.50	5.18	7.28	5.46	7.94	6.59	7.63	7.36	2.94	3.72

Appendix K.6. P300 latency during Go-nogo task in non-carrier

Sub ID	Fz	F3	F4	Cz	C3	C4	Pz	P3	P4	O1	O2
1	13.15	10.62	10.37	23.60	18.43	17.81	28.07	22.93	21.83	15.72	14.47
2	14.71	13.81	14.81	27.14	25.12	24.46	25.09	23.85	27.97	19.80	21.54
3	13.75	11.40	13.07	19.76	15.38	17.43	18.89	16.74	17.21	10.07	11.16
4	16.25	16.91	13.42	23.62	22.41	20.37	22.79	22.33	19.78	16.57	17.27
5	2.08	0.30	2.09	10.11	6.15	9.38	11.85	10.49	12.49	9.09	11.22
6	20.89	13.82	13.89	33.65	23.21	21.35	24.62	20.67	20.77	10.13	11.43
7	15.24	12.63	14.40	24.87	19.88	20.47	27.38	20.86	19.09	8.94	9.44
8	12.30	8.80	13.06	22.02	22.00	21.90	21.16	20.22	19.83	13.32	10.37
9	19.92	19.90	18.60	33.27	26.43	25.08	21.59	19.86	15.19	10.60	9.32
10	8.94	4.34	5.99	13.75	10.63	10.25	12.63	15.12	12.96	10.89	13.31
11	21.49	17.29	17.25	31.55	25.36	23.60	22.21	21.39	21.69	8.82	5.29
12	21.87	20.77	17.77	27.77	24.06	22.81	23.55	20.56	21.11	14.61	14.26
13	7.81	8.08	4.31	12.49	9.42	10.36	12.43	9.06	13.46	6.32	9.15
14	2.79	1.90	2.24	13.86	6.59	11.01	17.64	16.06	14.32	12.88	10.00
15	5.97	8.59	8.32	19.36	15.18	13.61	19.64	17.65	15.83	5.34	4.45
16	12.58	8.53	13.35	26.59	17.11	20.75	27.35	21.24	22.85	8.14	9.21
17	11.54	10.69	9.17	22.91	18.54	18.19	20.33	17.83	17.48	8.28	7.74
18	11.16	8.17	16.64	22.35	14.81	19.65	19.34	15.81	19.09	9.57	11.36
19	12.57	9.89	13.60	17.30	13.15	18.10	15.23	14.23	15.31	7.01	7.46
20	9.44	5.76	8.22	14.93	8.53	11.99	13.78	6.59	12.52	3.65	6.93
21	10.90	7.51	11.72	21.58	16.18	19.69	21.46	17.58	20.44	13.06	14.90
22	21.91	16.81	17.05	25.55	19.69	20.56	21.39	19.98	18.78	13.59	10.40
23	17.21	14.62	13.41	23.93	19.67	20.24	19.69	19.09	17.90	5.42	8.80
24	17.54	16.70	15.54	21.40	16.23	17.77	17.92	16.19	16.93	10.85	11.86
25	13.03	13.30	12.28	19.80	17.34	17.12	17.76	16.58	17.94	11.82	11.45
26	15.95	13.65	12.42	34.17	24.60	19.26	25.27	20.93	19.62	3.81	7.37
27	25.25	17.69	18.33	34.22	27.20	24.74	31.67	27.12	23.78	12.87	12.68
28	4.45	5.99	1.77	6.26	8.03	4.16	13.92	14.43	13.39	11.37	11.19
29	16.17	13.48	11.78	15.31	14.89	14.82	17.03	15.61	13.59	5.64	5.09
30	6.03	5.50	5.18	7.28	5.46	7.94	6.59	7.63	7.36	2.94	3.72

Appendix K.7. Behavior data of e4-carrier

Sub ID	Fitness	Age	VO2max	Energy	K-BIT	Stroop	NG_AC	NG_RT	OD_AC	OD_RT
1	1	22	64.75	42.43	114	4.76	94.33	0.29	98.33	0.29
2	1	22	43.34	46.29	108	8.89	96.67	0.34	99.33	0.40
3	1	20	46.62	62.09	102	4.94	96.33	0.32	99.67	0.31
4	1	19	42.97	39.09	129	9.77	96.00	0.35	99.67	0.39
5	1	21	52.64	37.52	115	-1.56	95.00	0.33	98.67	0.34
6	1	21	52.00	44.23	104	-3.31	97.00	0.50	99.33	0.53
7	1	20	55.06	40.13	117	9.77	93.67	0.27	98.00	0.30
8	1	21	67.64	34.34	105	17.21	96.33	0.34	99.67	0.36
9	1	22	43.06	35.57	99	-5.23	96.67	0.43	99.67	0.44
10	1	20	45.57	36.29	119	5.51	96.00	0.30	99.00	0.34
11	1	23	55.58	33.86	104	-1.86	94.67	0.26	99.67	0.35
12	1	20	53.51	33.43	118	3.47	96.67	0.30	100.00	0.45
13	1	19	56.43	42.63	98	0.49	95.33	0.42	99.33	0.51
14	1	23	57.10	35.25	98	-4.43	97.00	0.31	99.67	0.34
15	1	20	54.53	39.68	104	-0.72	93.33	0.31	99.33	0.37
16	2	22	41.95	47.39	103	5.24	96.00	0.29	99.33	0.36
17	2	19	39.12	36.27	114	19.84	95.67	0.28	99.67	0.37
18	2	21	32.74	39.13	114	-29.94	96.33	0.44	99.67	0.42
19	2	21	39.82	35.34	102	-6.89	92.67	0.42	99.33	0.51
20	2	22	42.87	67.38	96	-3.52	97.67	0.43	100.00	0.62
21	2	21	37.53	37.53	116	8.53	95.33	0.28	99.33	0.41
22	2	21	37.85	43.70	98	5.57	97.00	0.30	99.67	0.35
23	2	18	36.11	33.21	112	8.63	97.33	0.43	99.00	0.40
24	2	20	39.86	35.32	109	-5.76	98.67	0.33	100.00	0.38
25	2	21	40.54	37.27	93	4.02	96.00	0.40	98.67	0.48
26	2	20	41.57	38.05	124	7.95	96.67	0.27	100.00	0.35
27	2	20	23.76	35.30	109	15.02	94.33	0.39	98.67	0.32
28	2	19	25.00	35.23	109	-9.40	97.00	0.52	99.00	0.40
29	2	19	34.00	42.86	102	12.29	95.00	0.31	98.67	0.40

Appendix K.8. P300 latency during oddball task in e4 carrier

Sub ID	Fz	F3	F4	Cz	C3	C4	Pz	P3	P4	O1	O2
1	271.09	265.23	269.14	249.61	259.37	257.42	257.42	261.33	257.42	259.37	261.33
2	278.91	284.77	286.72	284.77	280.86	284.77	308.20	302.34	302.34	298.44	300.39
3	261.33	261.33	259.37	319.92	316.02	275.00	294.53	306.25	317.97	304.30	312.11
4	316.02	327.73	312.11	325.78	349.22	341.41	337.50	345.31	343.36	341.41	353.12
5	357.03	360.94	358.98	345.31	347.27	353.12	321.87	310.16	308.20	329.69	317.97
6	347.27	347.27	347.27	351.17	357.03	355.08	364.84	382.42	362.89	376.56	376.56
7	294.53	306.25	292.58	286.72	302.34	290.62	306.25	312.11	304.30	312.11	300.39
8	275.00	275.00	276.95	271.09	271.09	275.00	271.09	269.14	273.05	267.19	267.19
9	321.87	316.02	317.97	325.78	333.59	337.50	343.36	345.31	343.36	337.50	339.45
10	345.31	335.55	347.27	335.55	331.64	341.41	331.64	327.73	329.69	327.73	329.69
11	360.94	364.84	355.08	357.03	364.84	355.08	355.08	358.98	355.08	353.12	355.08
12	306.25	310.16	308.20	298.44	308.20	304.30	306.25	310.16	308.20	302.34	300.39
13	319.92	321.87	323.83	321.87	331.64	335.55	333.59	337.50	335.55	335.55	337.50
14	308.20	308.20	308.20	317.97	316.02	316.02	323.83	316.02	317.97	317.97	314.06
15	329.69	337.50	325.78	331.64	337.50	327.73	327.73	331.64	323.83	302.34	310.16
16	333.59	335.55	337.50	327.73	323.83	335.55	341.41	333.59	345.31	269.14	345.31
17	302.34	308.20	312.11	271.09	302.34	323.83	331.64	333.59	335.55	337.50	345.31
18	329.69	321.87	337.50	304.30	306.25	310.16	302.34	308.20	306.25	306.25	310.16
19	312.11	316.02	327.73	341.41	353.12	341.41	349.22	351.17	353.12	362.89	357.03
20	232.03	230.08	233.98	259.37	257.42	282.81	276.95	271.09	300.39	255.47	323.83
21	335.55	339.45	331.64	337.50	341.41	333.59	337.50	341.41	335.55	339.45	337.50
22	280.86	288.67	282.81	280.86	288.67	282.81	304.30	304.30	323.83	325.78	333.59
23	339.45	337.50	341.41	321.87	325.78	323.83	321.87	325.78	319.92	374.61	376.56
24	300.39	304.30	302.34	288.67	296.48	292.58	312.11	329.69	323.83	267.19	263.28
25	333.59	337.50	333.59	345.31	355.08	368.75	360.94	362.89	370.70	368.75	368.75
26	308.20	310.16	302.34	304.30	321.87	308.20	321.87	337.50	325.78	325.78	331.64
27	319.92	321.87	323.83	316.02	316.02	319.92	317.97	317.97	319.92	323.83	323.83
28	370.70	374.61	368.75	380.47	392.19	390.23	403.91	409.77	407.81	409.77	427.34
29	337.50	339.45	333.59	259.37	321.87	321.87	290.62	325.78	302.34	294.53	294.53

Appendix K.9. P300 latency during Go-nogo task in e4 carrier

Sub ID	Fz	F3	F4	Cz	C3	C4	Pz	P3	P4	O1	O2
1	271.09	275.00	273.05	265.23	255.47	265.23	269.14	269.14	271.09	267.19	273.05
2	284.77	290.62	286.72	284.77	284.77	286.72	282.81	347.27	282.81	355.08	360.94
3	269.14	273.05	269.14	276.95	265.23	269.14	314.06	312.11	308.20	316.02	312.11
4	306.25	357.03	329.69	384.37	374.61	364.84	368.75	378.52	368.75	386.33	374.61
5	269.14	267.19	276.95	263.28	271.09	273.05	271.09	278.91	278.91	261.33	261.33
6	341.41	339.45	345.31	355.08	349.22	358.98	378.52	384.37	372.66	382.42	380.47
7	296.48	302.34	300.39	294.53	312.11	298.44	343.36	349.22	316.02	357.03	339.45
8	269.14	269.14	269.14	263.28	263.28	267.19	261.33	259.37	267.19	259.37	267.19
9	290.62	288.67	298.44	280.86	282.81	288.67	292.58	282.81	296.48	298.44	298.44
10	329.69	329.69	329.69	323.83	323.83	327.73	331.64	329.69	331.64	329.69	331.64
11	319.92	319.92	314.06	321.87	327.73	317.97	339.45	341.41	343.36	349.22	355.08
12	286.72	288.67	286.72	273.05	280.86	284.77	282.81	282.81	290.62	263.28	284.77
13	341.41	339.45	341.41	345.31	341.41	349.22	349.22	335.55	358.98	341.41	357.03
14	290.62	292.58	290.62	288.67	300.39	292.58	296.48	302.34	302.34	310.16	317.97
15	306.25	316.02	308.20	329.69	339.45	331.64	327.73	331.64	329.69	333.59	329.69
16	325.78	345.31	327.73	325.78	347.27	323.83	331.64	331.64	329.69	319.92	319.92
17	294.53	300.39	294.53	286.72	294.53	294.53	294.53	323.83	306.25	357.03	341.41
18	351.17	349.22	358.98	355.08	353.12	357.03	353.12	345.31	357.03	337.50	347.27
19	398.05	394.14	394.14	386.33	376.56	384.37	378.52	380.47	386.33	360.94	364.84
20	269.14	271.09	273.05	275.00	276.95	284.77	288.67	288.67	300.39	278.91	298.44
21	331.64	331.64	327.73	331.64	335.55	335.55	337.50	339.45	339.45	341.41	343.36
22	288.67	288.67	288.67	276.95	288.67	280.86	269.14	263.28	276.95	253.52	257.42
23	339.45	343.36	349.22	339.45	343.36	347.27	360.94	355.08	364.84	362.89	380.47
24	298.44	300.39	296.48	286.72	286.72	292.58	319.92	292.58	317.97	351.17	325.78
25	339.45	347.27	345.31	345.31	349.22	355.08	362.89	364.84	364.84	360.94	370.70
26	296.48	294.53	300.39	282.81	286.72	292.58	290.62	296.48	296.48	314.06	319.92
27	316.02	327.73	314.06	308.20	312.11	310.16	306.25	312.11	308.20	310.16	308.20
28	343.36	351.17	343.36	337.50	351.17	341.41	339.45	366.80	335.55	335.55	333.59
29	327.73	327.73	327.73	314.06	323.83	321.87	317.97	366.80	323.83	358.98	355.08

Appendix K.10. P300 amplitude during oddball in e4 carrier

Sub ID	Fz	F3	F4	Cz	C3	C4	Pz	P3	P4	O1	O2
1	6.87	6.67	8.97	19.50	22.83	19.24	22.90	22.74	21.78	15.26	13.29
2	11.60	6.79	6.49	9.77	6.24	11.94	12.22	12.33	10.41	9.70	7.00
3	12.16	9.71	8.20	19.01	16.23	14.75	18.57	13.31	17.83	11.98	10.87
4	15.23	11.57	11.44	16.94	12.31	15.19	13.04	10.99	13.02	7.44	6.42
5	21.93	16.90	18.66	25.15	19.34	17.70	21.49	18.34	16.28	13.28	10.12
6	7.22	2.70	5.33	7.45	2.60	5.20	2.78	3.74	3.58	-4.14	2.72
7	14.58	10.44	12.92	24.55	21.33	22.69	23.24	19.65	19.83	11.82	13.27
8	22.13	18.14	16.74	30.04	24.88	22.79	27.22	23.71	21.09	14.54	12.19
9	11.74	10.82	9.26	13.58	10.75	12.44	19.38	14.67	17.37	10.54	12.96
10	7.88	7.81	7.57	20.29	20.51	18.19	22.53	23.80	18.08	14.67	11.85
11	16.03	15.27	15.03	21.62	18.96	19.50	21.54	18.79	17.96	9.68	9.39
12	20.24	18.19	17.19	20.86	21.37	18.20	20.10	19.46	17.85	11.50	10.67
13	15.89	15.76	14.54	21.65	18.88	18.96	21.15	18.17	16.16	10.95	7.62
14	5.38	5.92	2.00	12.15	13.32	9.94	13.23	13.28	9.37	5.69	4.97
15	12.42	10.73	12.84	16.27	13.64	17.11	17.05	12.36	14.24	6.18	6.97
16	7.65	5.80	7.81	11.93	8.18	11.61	10.85	7.49	10.09	2.18	2.65
17	15.44	14.93	12.76	24.12	21.85	17.77	29.13	26.37	23.83	18.41	17.11
18	5.97	6.16	6.58	18.33	15.86	14.87	23.91	21.07	18.78	13.08	14.33
19	4.95	8.28	7.29	11.16	11.07	11.16	14.88	15.56	14.03	10.32	9.01
20	1.57	2.41	1.05	4.90	5.59	4.40	5.03	6.48	4.39	1.51	1.56
21	12.01	9.36	8.83	24.53	15.85	17.62	25.81	19.64	21.77	10.82	11.97
22	4.04	4.31	4.65	8.07	4.73	8.67	10.35	10.22	10.49	8.46	7.99
23	10.64	8.39	5.36	17.43	14.56	13.16	23.59	20.74	18.90	11.01	8.96
24	16.85	13.10	12.39	24.76	18.64	18.17	23.42	20.59	17.64	9.31	7.79
25	-6.46	-5.87	-5.09	-1.23	1.21	1.31	12.29	11.91	11.23	12.61	11.90
26	13.56	12.25	10.35	18.56	15.48	14.67	17.68	15.79	14.92	10.61	9.04
27	3.90	3.53	2.86	10.29	9.40	10.56	17.35	15.87	15.95	14.81	15.45
28	10.70	8.65	14.10	10.32	8.32	9.71	11.04	5.31	8.92	8.01	7.92
29	10.38	9.33	11.68	14.36	8.43	17.50	25.57	21.07	23.74	14.94	18.39

Appendix K.11. P300 amplitude during Go-nogo task in e4 carrier

Sub ID	Fz	F3	F4	Cz	C3	C4	Pz	P3	P4	O1	O2
1	15.90	14.40	10.24	31.14	13.39	25.44	23.18	18.71	21.81	11.49	8.00
2	25.38	20.99	21.63	27.76	21.42	14.32	21.64	22.23	18.69	20.79	19.32
3	23.77	19.77	16.04	32.41	27.18	25.68	30.34	22.91	22.78	10.46	10.57
4	15.23	11.57	11.44	16.94	12.31	15.19	13.04	10.99	13.02	7.44	6.42
5	21.93	16.90	18.66	25.15	19.34	17.70	21.49	18.34	16.28	13.28	10.12
6	7.22	2.70	5.33	7.45	2.60	5.20	2.78	3.74	3.58	-4.14	2.72
7	14.58	10.44	12.92	24.55	21.33	22.69	23.24	19.65	19.83	11.82	13.27
8	24.59	20.59	17.64	33.45	27.49	22.59	26.31	24.00	21.08	13.28	12.79
9	11.74	10.82	9.26	13.58	10.75	12.44	19.38	14.67	17.37	10.54	12.96
10	7.88	7.81	7.57	20.29	20.51	18.19	22.53	23.80	18.08	14.67	11.85
11	16.03	15.27	15.03	21.62	18.96	19.50	21.54	18.79	17.96	9.68	9.39
12	20.24	18.19	17.19	20.86	21.37	18.20	20.10	19.46	17.85	11.50	10.67
13	15.89	15.76	14.54	21.65	18.88	18.96	21.15	18.17	16.16	10.95	7.62
14	5.38	5.92	2.00	12.15	13.32	9.94	13.23	13.28	9.37	5.69	4.97
15	12.42	10.73	12.84	16.27	13.64	17.11	17.05	12.36	14.24	6.18	6.97
16	7.65	5.80	7.81	11.93	8.18	11.61	10.85	7.49	10.09	2.18	2.65
17	15.44	14.93	12.76	24.12	21.85	17.77	29.13	26.37	23.83	18.41	17.11
18	5.97	6.16	6.58	18.33	15.86	14.87	23.91	21.07	18.78	13.08	14.33
19	4.95	8.28	7.29	11.16	11.07	11.16	14.88	15.56	14.03	10.32	9.01
20	1.57	2.41	1.05	4.90	5.59	4.40	5.03	6.48	4.39	1.51	1.56
21	12.01	9.36	8.83	24.53	15.85	17.62	25.81	19.64	21.77	10.82	11.97
22	4.04	4.31	4.65	8.07	4.73	8.67	10.35	10.22	10.49	8.46	7.99
23	10.64	8.39	5.36	17.43	14.56	13.16	23.59	20.74	18.90	11.01	8.96
24	16.85	13.10	12.39	24.76	18.64	18.17	23.42	20.59	17.64	9.31	7.79
25	-6.46	-5.87	-5.09	-1.23	1.21	1.31	12.29	11.91	11.23	12.61	11.90
26	13.56	12.25	10.35	18.56	15.48	14.67	17.68	15.79	14.92	10.61	9.04
27	-0.25	-1.04	-1.75	8.84	7.56	7.10	14.01	13.43	12.89	12.34	11.98
28	9.14	7.38	5.51	12.78	10.43	10.87	13.05	10.46	12.12	8.71	10.53
29	18.74	13.16	17.85	25.67	17.07	23.27	25.61	22.65	22.24	16.23	17.20

REFERENCES: Chapter 1 & 2

Allan D. M., Praag, H.C., Ray, J., Weaver, Z., Winrow, C. J., Carter, T. A., Braquet, R., Harrington, E., Ried, T., Brown, K. D., Gage, F. H., & Barlow, C. (2001). Ataxia telangiectasia mutated is essential during adult neurogenesis, *Genes and development*, 15, 554-566.

Ally. B. A., Jones, G. E., Cole, J. A., & Budson, A. E. (2006). The P300 component in patients with Alzheimer's disease and their biological children, *Biological Psychology*, 72, 180-187.

Anwar, N., Lovestone,S., and Cheetham,M.et.al. (1993). Apolipoprotein Eepsilon4 allele and Alzheimer's disease. *Lancet* 342, 1308-1310.

Bashore, T. (1989). Age, physical fitness, and mental processing speed. *Annual Review of Gerontology and Geriatrics,* 9, 120-144.

Batty, M. & Taylor, M.J. (2002). Visual categorization during childhood: an ERP study. *Psychophysiology,* 39, 482–490.

Beck, E. C., Swanson, C., Dustman, R. E. (1980). Long latency components of the visually evoked potential in man: Effects of aging. *Experimental Aging Research*, 6**,** 523–545

Black, J. E., Isaacs K. R., B. J. Anderson, A. A. Alcantara, & W. T. Greenough. (1990). Learning causes synaptogenesis, whereas motor activity causes angiogenesis, in cerebellar cortex of adult rats. *Proceeding of the National Academy of Sciences of the United States of America*, 87*,* 5568-5572.

Blakemore, S., & Choudhury, S. (2006). Development of the adolescent brain: implications for executive function and social cognition. *Journal of child psychology and psychiatry*, 47, 296-312.

Bookheimer, S. Y., Strojwas, M. H., Cohen, M. S., Saunders, A. M., Pericak-Vance, M. A., Mazziotta, J. C., Small, G. W. (2000). Patterns of Brain Activation in People at Risk for Alzheimer's Disease. *The New England journal of medicine.* 343, 450-456

Borgaonkar,S., Schmidt,L., and Martin,S., et.al. (1993). Alzheimer's disease with apolipoproteinE. *Lancet* 342, 625.

Bourgeois, J. P., Goldman-Rakic, P. S., & Rakic, P. (1994). Synaptogenesis in the prefrontal cortex of rhesus monkeys, *Cerebral Cortex*, 4, 78-96.

Brown, W. S., Marsh, J. T., & LaRue, A. (1983). Exponential electrophysiological aging: P3 latency. *Electroencephalography and clinical neurophysiology*, 55**,** 277–285

Buck, S. M., Hillman, C. H., & Castelli, D. M. (2007). The relation of aerobic fitness to strop task performance in preadolescent children. *Journal of Cognition and Development*, 40, 166-172.

Buckner, R. (2004). Memory and executive function in aging and AD: Multiple factors that cause decline and the reserve factors that compensate. *Neuron*, 44, 195-208.

Casey, B. J., Trainor, R. J., Orendi, J. L., Schubert, A. b., Nystrom, L. E., Cohen, J. D., Noll, D. C., Giedd, J., Castellanos, X., Haxby, J., Forman, S. D., Dahl, R. E., & Rapoport, J. L. (1997). A pediatric functional MRI study of prefrontal activation during performance of a Go-No-Go task. *Journal of cognitive Neuroscience*, 9, 835-847.

Casey, B. J., Tottenham, N., Liston, c., & Durston, S. (2005). Imaging the developing brain: what have we learned about cognitive development: *Trends in Cognitive Sciences*, 9, 104-110.

Chugani, H. T., Phelps, M. E., & Mazziotta, J. C. (1987). Positron emission tomography study of human brain functional development. *Annals of Neurology*, 22, 487-297.

Changeuz, J. P., & Danchin, A. (1976) Selective stabilization of developing synapses as a mechanism for the specification of neuronal networks. *Nature*, 264, 705-712.

Chodzko-Zajko, W.J., & Moore, K.A. (1994). Physical fitness and cognitive functioning in aging. *Exercise and Sport Science Review*, 22, 195-220.

Churchill, J., Galvez, R., Colcombe, S., Swain, R., Kramer, A., & Greenough, W., (2002). Exercise, experience and the aging brain. *Neurobiology of Aging,* 23, 941-955.

Colcombe, S. J., Erickson, K. I., Raz, N., Webb, A. G., Cohen, N. J., McAuley, E. et al. (2003). Aerobic fitness reduces brain tissue loss in aging humans. *The journals of gerontology. Series A, Biological sciences and medical sciences*, 58, 176-180.

Colcombe, S. & Kramer, A. F. (2003). Fitness effects on the cognitive function of older adults: a meta-analytic study. *Psychological sciences: a journal of the American Psychological Society*, 14, 125-130.

Colcombe, S. J., Kramer, A. F., Erickson, K. I., Scalf, P., McAuley, E., Cohen, N. J. et al. (2004). Cardiovascular fitness, cortical plasticity, and aging. *Proceedings of the National Academy of Sciences of the United States of America*, 101, 3316-3321.

Corder, E.H., Saunders, A.M., Strittmatter, W.J., Schmechel, D.E., Gaskell, P.C., Small, G.W., Roses, A.D., Haines, J.L., & Pericak-Vance, M.A. (1993). Gene dose of apolipoprotein E type 4 allele and the risk of alzheimer's disease in late onset families. *Science*, 261, 921-923.

Cotman, C. W. & Berchtold, N. C. (2002). Exercise: a behavioral intervention to enhance brain health and plasticity. *Trends in neurosciences*, 25*,* 295-301.

Cotman,C.W. and Engesser-Cesar,C. (2002). Exercise enhances and protects brain function. *Exercise Sport Science Review*, 30, 75-79.

Curry, J.G., and Polich, J. (1992). P300, global probability, and stimulus sequence effects in children. *Developmental Neuropsychology*, 8, 185–202

Davis, C. L., Tomporowski, P. D., Boyle, C. A., Walker, J. L., Miller, P. H., Naglieri, J. A., et al. (2007). Effects of aerobic exercise on overwiehgt children's cognitive functioining: A randomized controlled trial. *Research Quarterly for Exercise and Sport*, 78, 510-519.

Deeny, S. P. (2005). *Exercise behavior and maintenance of cerebral cortical activity during cognitive challenge in middle-aged men and women genetically at risk for dementia.* Unpublished doctoral dissertation, University of Maryland, College park.

Deeny, S. P., Poeppel, D., Zimmerman, J. B., Roth, S. M., Brandauer, J., Witkowski, S., Hearn J. W., Ludlow, A. T., Contreras-Vidal, J. L., Brandt, J., & Hatfield, B. D. (2008). Exercise, APOE, and working memory: MEG and behavioral evidence for benefit of exercise in epsilon4 carriers, *Biological Psychology*, 78, 179-187.

Duncan-Johnson, C.C. (1981) P300 latency: a new metric of information processing. *Psychophysiology*, 18, 207–215.

Dustman, R. E., Emmerson, R. Y., Ruhling, R. O., Shearer, D. E., Steinhaus, L. A., Johnson, S. C. et al. (1990). Age and fitness effects on EEG, ERPs, visual sensitivity, and cognition. *Neurobiology of.aging,* 11*,* 193-200.

Filbey, F. M., Slack, K. J., Sunderland, T. P., & Cohen, R. M. (2006). Functional magnetic resonance imaging and magnetoencephalography differences associated with ApoE e4 in young healthy adults. *NeuroReport*, 17, 1585-1590

Fjell, A. M. & Walhovd, K. B. (2001). P300 and Neuropsychological Tests as Measures of Aging: Scalp Topography and Cognitive Changes. *Brain Topography,* 14, 25-40.

Fuster, J. M. (2001). The prefrontal cortex – An update: Time is of the essence. *Neuron* 30, 319-333.

Giedd, J. N., Blumenthal, J., Jeffries, N. O., castellanos, F. Z., Liu, H., Sijdenbos, A., Paus, T., Evans, A. C., & Rapaport, J. L (1999). Brain development during childhood and adolescence: A longitudinal MRI study. *Nature Neuroscience* 2, 861-863.

Goodin, D. S., Squires, K. C., Henderson, B. H., & Starr, A. (1978). Age-related variations in evoked potentials to auditory stimuli in normal human subjects. *Electroencephalography and clinical neurophysiology*, 44, 447–458

Green, J., & Levey, A. I. (1999). Event-related potential changes in groups at increased risk for Alzheimer disease. *Archives of neurology*, 56, 1421-1422.

Hansen, A. L., Johnsen, B. H., Sollers III, J. J., Stenvik, K., & Thayer, J. F. (2004). Heart rate variability and its relation to prefrontal cognitive function: the effects of training and detraining. *European Journal of Applied Physiology*, 93, 263-272.

Hillman, C. H., Belopolsky, A. V., Snook, E. M., Kramer, A. F., & McAuley, E. (2004). Physical activity and executive control: implications for increased cognitive health during older adulthood. *Research Quarterly for Exercise and Sport*, 75, 176-185.

Hillman, C. H., Kramer, A. G., Belopolsky, A.V., & Smith, D. P. (2006). A cross-sectioanl examination of age and physical activity on performance and event-related brain potentials in a task switching paradigm. *International Journal of Psychophysiology*, 59, 30-39.

Hillman, C.H., Castelli, D., & Buck, S.M. (2005). Aerobic fitness and neuro-cognitive function in healthy preadolescent children. *Medicine & Science in Sports & Exercise*, 37, 1967-1974

Hillman, C. H., Weiss, E. P., Hagberg, J. M., & Hatfield, B. D. (2002). The relationship of age and cardiovascular fitness to cognitive and motor processes. *Psychophysiology*, 39, 303-312.

Hooper, C. J., Luciana, M., Conklin, H. M., & Yager, R. S. (2004). Adolescents' performance on the development of decision making and ventromedial prefrontal cortex. *Developmental Psychology*, 40, 1148-1158.

Houlden, H., Crook, R., & Duff, K. E. A. (1993). Confirmation that the apolipoprotein E4 allele is associated with late-onset, familial Alzheimer's-disease. *Neurodegeneration*, 2, 283-286.

Howard, L., Polich, J. (1985) P300 latency and memory span development. *Developmental Psychology*, 21, 283-289.

Huttenlocher, P. R. (1990). Morphometric study of human cerebral cortex development. *Neuropsychologia*, 28, 517-527.

Jernigan, T. L. & Tallal. P. (1990). Late childhood changes in brain morphology observable with MRI. *Developmental Medicine and Child Neurology,* 32, 379-385.

Johnstone, S. J., Barry, R. J., Anderson, J. W., & Coyle, S. F. (1996). Age-related changes in child and adolescent event-related potential component morphology, amplitude and latency to standard and target stimuli in an auditory oddball task. *International Journal of Psychophysiology*, 24, 223-238.

Kempermann, G, Kuhn, H. G., & Gage, F. H. (1997). More hippocampal neurons in adult mice living in an enriched environment. *Nature*, 386, 493-495.

Kramer, A.F., Hahn, S., & Gopher, D. (1999) Task coordination and aging: Explorations of executive control processes in the task switching paradigm. *Acta Psychologica,* 101,339-378

Kutas, M., McCarthy, G., & Donchin, E. (1977). Augmenting mental chronometry: The P300 as a measure of stimulus evaluation time. *Science*, 197, 792-795.

Luciana, M., Conklin, H. M., Cooper, C. J., & Yarger, R. S. (2005). The development of nonverbal working memory and executive control processes in adolescents. *Child Development*, 76, 697-712.

Luna, B., Garver, K. E., Urban, T. A., Lazar, N. A., & Sweeney, J. A. (2004). Maturation of cognitive processes from late childhood to adulthood. *Child Development*, 75, 1357-1372.

Luria, A. R. (1996). *Higher cortical functions in man.* Oxford, UK: Basic Books Inc.

Mackinlay, R., Charman, T., & Karmiloff-Smith, A. (2003). *Remembering to remember: A developmental study of prospective memory in a multitasking paradigm.* Tampa, Florida: Poster presented at the Society for Research in Child Development, Biennial Meeting, 24-27 April.

McCarthy, G. & Donchin, E. (1981). A metric for thought: A comparison of P300 latency and reaction time. *Science*, 211, 77-79.

McDowell, K., Kerick, S.E., Santa Maria, D.L., & Hatfield, B.D. (2003). Aging, physical activity, and cognitive processing: an examination of P300. *Neurobiology of Aging* 24**,** 597-606.

Moceri, V. M., Kukull, W. A., Emanuel, I., van Belle, G., Larson, E. B. (2000) Early-life risk factors and the development of Alzheimer's disease, *Neurology*, 54, 415.

Mrzljak, L., Uylings, H. B. M., Van Eden, C. G. & Judas, M. (1990) Neuronal development in human prefrontal cortex in prenatal and postnatal stagees. *In the prefrontal cortex: its Structure, Function and Pathology* pp.185-222. Amsterdam: Elsevier.

Naglieri, J. A., & Das, J. P. (1997). *Cognitive assessment system*. Itasca, IL: Riverside.

Noguchi, S., Murakami,K., & Yamada,N. (1993). Apolipoprotein E genotype and Alzheimer's disease. *Lancet*, 342, 737.

Paus, T. (1989). The development of sustained attention in children might be related to the maturation of frontal cortical functions. *Acta Neurobiologiae Experimentalis*, 49, 51-55.

Pfefferbaum, A., Mathalon, D. H., Sullivan, E. V., Rawles, J. M., Sipursky, R. B. & Lim, K. O. (1994). A quantitative magnetic resonance imaging study of changes in brain morphology from infancy to late adulthood. *Archives of Neurology*, 51, 874-887.

Podewils, L. J., Guallar, E., Kuller, L. H., Fried, L. P., Lopez, O. L., Carlson, M. C., & Lyketsos, C. G. (2005). Physical activity, apoe genotype, and dementia risk: Findings from the cardiovascular health cognition study. *American Journal of Epidemiology*, 161, 639-651.

Poirier, J., Davignon, J., Boutheillier, D., Kogan, S., Bertrand, P., Gauthier, S. (1993). Apolipoprotein E polymorphism and Alzheimer's disease. *The Lancet*, 342, 697-699.

Polich, J. (1992). On the correlation between P300 amplitude and latency. *Bulletin of the Psychonomic Society*, 30, 5-8.

Polich, J. (1996). Meta-analysis of P3 normative aging studies. *Psychophysiology*, 33, 334-353.

Polich, J. (1997). EEG and ERP assessment of normal aging. *Electroencephalography and Clinical Neurophsiology*, 104, 244-256.

Polich, J. & Lardon, M. T. (1997). P300 and long-term physical exericse. *Electroencephalography and Clinical Neurophsiology*, 103, 493-498.

Procyk, E & Goldman-Rakic, P. S. (2006). Modulation of dorsolateral prefrontal delay activity during self-organized behavior. *The journal of neuroscience,* 26(44), 11313-23.

Purves, D. (1988). *Body and mind; A trophic Theory of Neural Connections.* Harvard University Press, Cambridge.

Rebeck,G., Reiter,J., Strickland,D., & Hyman,B. (1993). Apolipoprotein E in sporadic Alzheimer's disease: Allelic variation and receptor interactions. *Neuron*, 11, 575-580.

Reinvang, I. (1999). Cognitive event-related potentials in neuropsychological assessment. *Neuropsychology review,* 9, 231–248.

Reiman, E.M., Caselli, R.J., Chen, K., Alexander, G.E., Brandy, D., & Frost, J. (2001). Declining brain activity in cognitively normal apolipoprotein E e4 heterozygotes: A foundation for using positron emission tomography to efficiently test treatments to prevent Alzheimer's disease. *Proceedings of the National Academy of Sciences*, 98, 3334-339.

Reiman,E.M., Caselli,R.J., Yun,L.S., Chen,K., Bandy,D., Minoshima,S., Thibodeau,S.N., & Osborne,D. (1996). Preclinical evidence of Alzheimer's disease in persons homozygous for the E4 allele for apolipoprotein E. T*he New England Journal of Medicine*, 334, 752-758.

Reiman, E. M., Chen, K., Alexander, G. E., Caselli, R. J., Bandy, D., Osborne, D. et al. (2004). Functional brain abnormalities in young adults at genetic risk for late-onset Alzheimer's dementia. *Proceeding of the National Academy of Sciences of the United States of America*, 101, 284-289.

Reiman, E. M., Uecker, A., Caselli, R. J., Lewis, S., Bandy, D., de Leon, M. J. et al. (1998). Hippocampal volumes in cognitively normal persons at genetic risk for Alzheimer's disease. *Annals of neurology*, 44, 288-291.

Reiss, A. L., Abrams, M. T., Singer, H. S., Ross, J. L., & Denckla, M. B. (1996). Brain development, gender and IQ in children: A volumetric imaing study. *Brain*, 119, 1763-1774.

Riley, K.P., Snowdon, D. A., Desrosiers, M. F., & Markesbery, W. R. (2005). Early life linguistic ability, late life cognitive function, and neuropathology: findings from the Nun study. *Neurobiology of aging*, 26, 341-347.

Robertson, E.M., Tormos, J.M., Maeda, F., & Pascual-Leone, A. (2001). *The Role of the Dorsolateral Prefrontal Cortex during Sequence Learning is Specific for Spatial Information*, *Cerebral Cortex*, 11(7), 628-635.

Rogers, R. L., Meyer, J. S., & Mortel, K. F. (1990). After reaching retirement age physical activity sustains cerebral perfusion and cognition. *Journal of the American Geriatrics Soceity*, 38, 123-128.

Rubia, K., Smith, A. b., Brammer, M. J., & Taylor, E. (2003). Right inferior prefrontal cortex mediates response inhibition while mesial prefrontal cortex is responsible for error detection. *NeuroImage*, 20, 351-358.

Scarmeas, N., Habeck, C. G., Hilton, J., Andersonn, K. E., Flynn, J., Park. A., & Stern. Y. (2005). APOE related laterations in cerebral activation even at college age. *Journal of neurology, neurosurgery, and psychiatry*, 76, 1440-1444.

Scheibel, A.B. (1979). The hippocampus: Organizational patterns in health and senescence. *Mechanisms of Ageing and Development*, 9, 89-102.

Scisco, J. L., Leynes, P. A., Kang, J. (2008). Cardiovascular fitness and executive control during task-swithing: An ERP study. *International Journal of Psychophysiology*, 69, 62-60.

Scheibel, A. B. (1990). Dendritic correlates of higher cognitive function. *In neurobiology of Higher Cognitive Function* (edited by Schiebel, A. B. & Wechsler, A.) pp 239-270. New York: The Guilford Press.

Scholey, A. B., Moss, M. C., Neave, N., Wesnes, K. (1999) Cognitive performance, Hyperoxia, and heart rate following oxygen administration in healthy young adults. *Physiology and Behavior*, 67(5),783-789.

Schuit, A. J., Feskens, E. J., Launer, L. J., & Kromhout, D. (2001). Physical activity and cognitive decline, the role of the apolipoprotein e4 allele. *Medicine & Science in Sports & Exercise*, 33, 772-777.

Semlitsh, H. V., Anderer, P., Schuster, P., & Presslich, O. (1986). A solution for reliable and valid reduction of ocular artifacts, applied to the P300. *Psychophysiology*, 223, 695-703.

Shallice, T. (1982). Specific impairments of planning, Philosophical Transactions of the Royal Soceity of London, *Series B, Biological Sciences*, 298, 199-209.

Shaw, P., Lerch, J. P., Pruessner, J. C., Tylor, K. N., Rose, A. B., Greenstein, D., Clasen, L., Evans, A., Rapoport, J. L., & Giedd, J. N. (2007). Cortical morphology in children and adolescents with different apolipoprotein E gene polymorphisms: an observational study. *Lancet neurology*, 6, 494-500.

Shay, K. & Roth, D. (1992). Association between aerobic fitness and visuospatial performance in healthy older adults. *Psychology and Aging*, 1, 15-24.

Sibley, B. A. & Etnier, J. L. (2003). The relationship between physical activity and cognition in children: A meta-analysis. *Pediatric Exercise Science*, 15, 243-256.

Sowell E. R., Peterson, B. S., Thompson, P. M., Welcom, S. E., Henkenius, A. L., & Toga, A. W. (2003). Mapping cortical change across the human life span, *Nature neuroscience*, 6(3), 309-315.

Sowell, E. R., Thompson, P. M., Holmes, C. J., Jernigan, T. L., & Toga, A. W. (1999). In vivo evidence for post-adolescent brain maturation in frontal and striatal regions. *Nature Neuroscience,* 2, 859-861.

Sowell, E. R., Thompson, P. M., Tessner, K. D., & Toga, A. W. (2001). Mapping continued brain growth and gray matter density reduction in dorsal frontal cortex; Inverse relationships during postadolescent brain maturation. *Journal of Neuroscience,* 21, 8819-8829.

Spirduso, W. W. (1980). Physical fitness, aging, and psychomotor speed: A review. *Journal of Gerontology*, 35, 850-865.

Spirduso, W. W. (1983). Exercise and the aging brain. *Research quarterly for exercise and sport*, 54, 208-18.

Stones, M. & Kozma, A. (1989). Age, exercise and coding performance. *Psychology and Aging*, 4, 190-194.

Strittmatter., W., Weisgraber, K., Goedert, M., et al., (1993). Binding of human Apolipoprotein E to synthetic amyloid beta peptide: Isoform-specific effects and implications for late-onset Alzheimer's disease. *Proceedings of the National Academy of Sciences*, 90, 8098-8102.

Swain, R. A., Harris, A. B., Wiener, E. C., Dutka, M. V., Morris, H. D., Theien, B. E. et al. (2003). Prolonged exercise induces angiogenesis and increases cerebral blood volume in primary motor cortex of the rat. *Neuroscience*, 117, 1037-1046.

Tomporowski, P. D., Davis, C. L., Miller, P. H., & Naglieri, J. A. (2007). Exercise and children's intelligence, cognition and academic achievement. *Educational Psychology Review*, 20, 111-131.

West,R.L. (1996). An application of prefrontal cortex function theory to cognitive aging. *Psychological Bulletin*, 120, 272-292.

Wickens, C., Kramer, A., Vanasse, L., & Donchin, E. (1983). The performance of concurrent tasks: a psychophysiological analysis of the reciprocity of information processing resources. *Science,* 221, 1080–1082

Woo, T. U., Pucak, M. L., Kye, C. H., Matus, C. V., & Lewis, D, A. (1997). Peripubertal refinement of the intrinsic and associational circuitry in monkey prefrontal cortex. *Neuroscience*, 80, 1149-1158.

Yakovlev, P.I. & Lecours, A. R. (1967). The myelogenetic cycles of regional maturation of the brain. *In regional Development of the Brain in Early Life* (edited by Minkowski, A.), 3-70. Oxford:Blackwell.

Yu, Y.W., Lin, C.H., Chen, S.P., Hong, C.J., & Tsai, S.J. (2000). Intelligence and event-related potentials for young female human volunteer apolipoprotein E epsilon4 and non-epsilon4 carriers. *Neuroscience Letters***,** 294**,** 179–181

Zecevic, N., & Rakic, P. (2001). Development of layer I neurons in the primate cerebral cortex. *Journal of Neuroscience*, 21, 5607-5619.

REFERENCES: Chapter 3

Allan D. M., Praag, H.C., Ray, J., Weaver, Z., Winrow, C. J., Carter, T. A., Braquet, R., Harrington, E., Ried, T., Brown, K. D., Gage, F. H., & Barlow, C. (2001). Ataxia telangiectasia mutated is essential during adult neurogenesis, *Genes and development*, 15, 554-566.

Ally. B. A., Jones, G. E., Cole, J. A., & Budson, A. E. (2006). The P300 component in patients with Alzheimer's disease and their biological children, *Biological Psychology*, 72, 180-187.

American College of Sports Medicine, Guidelines for graded exercise testing and exercise prescription. 2nd ed. Philadelphia, PA:Lea & Febiger, 1980

Balady, G. J., Berra, K. A., Golding, L. A., Gordon, N. F., Mahler, D. A., Myers, J. N., & Sheldahl, L. M. (2000). Physical fitness testing and interpretation. In Franklin, B. A., Whaley, M. H., & Howley, E. T. (Eds.), *ACSM's guidelines for exercise testing and prescription; Sixth edition*. (pp. 71-75). American College of Sports Medicine. Baltimore, Maryland

Bashore, T. (1989). Age, physical fitness, and mental processing speed. *Annual Review of Gerontology and Geriatrics,* 9, 120-144.

Beekely, M. D., Brechue, W. F., deHoyos, D. V., Garzarella, L., Werber-Zion, G., & Pollock, M. L. (2004). Cross-Validation of the YMCA submaximal cycle erogmeter test to predict VO_2 max. *Research Quarterly for Exercise and Sport*, 75, 337-342.

Bixby, W. R., Spalding, T. W., Haufler, A. J., Deeny, S. P., Mahlow, P. T., Simmerman, J. B., & Hatfield, B. D. (2007). The unique relation of physical activity to executive function in older men and women. *Medicine & Science in Sports & Exercise*, 39, 1408-1416.

Black, J. E., Isaacs K. R., B. J. Anderson, A. A. Alcantara, & W. T. Greenough. (1990). Learning causes synaptogenesis, whereas motor activity causes angiogenesis, in cerebellar cortex of adult rats. *Proceeding of the National Academy of Sciences of the United States of America*, 87, 5568-5572.

Blair, S. N., Haskell, W. L., Ho, P., Paffenbarger, R. S., Vranizan, K. M., Farquhar, J. W., & Wood, P. D. (1985). Assessment of habitual physical activity by a seven-day recall in a community survey and controlled experiments. *American Journal of Epidemiology*, 122, 794-804.

Blakemore, S., & Choudhury, S. (2006). Development of the adolescent brain: implications for executive function and social cognition. Journal of child psychology and psychiatry, 47, 296-312.

Bookheimer, S. Y., Strojwas, M. H., Cohen, M. S., Saunders, A. M., Pericak-Vance, M. A., Mazziotta, J. C., Small, G. W. (2000). Patterns of Brain Activation in People at Risk for Alzheimer's Disease. *The New England journal of medicine.* 343, 450-456

Buck, S. M., Hillman, C. H., & Castelli, D. M. (2007). The relation of aerobic fitness to strop task performance in preadolescent children. *Journal of Cognition and Development*, 40, 166-172.

Chugani, H. T., Phelps, M. E., & Mazziotta, J. C. (1987). Positron emission tomography study of human brain functional development. *Annals of Neurology*, 22, 487-297.

Colcombe, S. J., Erickson, K. I., Raz, N., Webb, A. G., Cohen, N. J., McAuley, E. et al. (2003). Aerobic fitness reduces brain tissue loss in aging humans. *The journals of gerontology. Series A, Biological sciences and medical sciences*, 58, 176-180.

Colcombe, S. & Kramer, A. F. (2003). Fitness effects on the cognitive function of older adults: a meta-analytic study. *Psychological sciences: a journal of the American Psychological Society*, 14, 125-130.

Colcombe, S. J., Kramer, A. F., Erickson, K. I., Scalf, P., McAuley, E., Cohen, N. J. et al. (2004). Cardiovascular fitness, cortical plasticity, and aging. *Proceedings of the National Academy of Sciences of the United States of America*, 101, 3316-3321.

Corder, E.H., Saunders, A.M., Strittmatter, W.J., Schmechel, D.E., Gaskell, P.C., Small, G.W., Roses, A.D., Haines, J.L., & Pericak-Vance, M.A. (1993). Gene dose of apolipoprotein E type 4 allele and the risk of alzheimer's disease in late onset families. *Science*, 261, 921-923.

Cotman, C. W. & Berchtold, N. C. (2002). Exercise: a behavioral intervention to enhance brain health and plasticity. *Trends in neurosciences*, 25, 295-301.

Cotman,C.W. and Engesser-Cesar,C. (2002). Exercise enhances and protects brain function. *Exercise Sport Science Review*, 30, 75-79.

Deeny, S. P., Poeppel, D., Zimmerman, J. B., Roth, S. M., Brandauer, J., Witkowski, S., Hearn J. W., Ludlow, A. T., Contreras-Vidal, J. L., Brandt, J., & Hatfield, B. D. (2008). Exercise, APOE, and working memory: MEG and behavioral evidence for benefit of exercise in epsilon4 carriers, *Biological Psychology*, 78, 179-187.

Dishman, R. K. & Steinhardt, M. (1988). Reliability and concurrent validity for a 7-day recall of physical activity in college students. *Medicine and Science in Sports and Exercise*, 20, 14-25.

Dustman, R. E., Emmerson, R. Y., Ruhling, R. O., Shearer, D. E., Steinhaus, L. A., Johnson, S. C. et al. (1990). Age and fitness effects on EEG, ERPs, visual sensitivity, and cognition. *Neurobiology of.aging*, 11, 193-200.

Filbey, F. M., Slack, K. J., Sunderland, T. P., & Cohen, R. M. (2006). Functional magnetic resonance imaging and magnetoencephalography differences associated with ApoE e4 in young healthy adults. *NeuroReport*, 17, 1585-1590.

Fuster, J. M. (2001). The prefrontal cortex – An update: Time is of the essence. *Neuron* 30, 319-333.

Giedd, J. N., Blumenthal, J., Jeffries, N. O., castellanos, F. Z., Liu, H., Sijdenbos, A., Paus, T., Evans, A. C., & Rapaport, J. L (1999). Brain development during childhood and adolescence: A longitudinal MRI study. *Nature Neuroscience* 2, 861-863.

Golden, C. J. (1978*). Stroop Color and Word Test: Manual for clinical and experimental uses*. Chicago, IL:Stoelting Co.

Green, J., & Levey, A. I. (1999). Event-related potential changes in groups at increased risk for Alzheimer disease. *Archives of neurology*, 56, 1421-1422.

Hansen, A. L., Johnsen, B. H., Sollers III, J. J., Stenvik, K., & Thayer, J. F. (2004). Heart rate variability and its relation to prefrontal cognitive function: the effects of training and detraining. *European Journal of Applied Physiology*, 93, 263-272.

Hillman, C. H., Belopolsky, A. V., Snook, E. M., Kramer, A. F., & McAuley, E. (2004). Physical activity and executive control: implications for increased cognitive health during older adulthood. *Research Quarterly for Exercise and Sport*, 75*,* 176-185.

Hillman, C.H., Castelli, D., & Buck, S.M. (2005). Aerobic fitness and neuro-cognitive function in healthy preadolescent children. *Medicine & Science in Sports & Exercise*, 37, 1967-1974

Hillman, C. H., Erickson, K. I., & Kramer, A. F. (2008). Be smart, exercise your heart: exercise effects on brain and cognition. Nature reviews. Neuroscience. 9, 58-65.

Hillman, C. H., Kramer, A. G., Belopolsky, A.V., & Smith, D. P. (2006). A cross-sectioanl examination of age and physical activity on performance and event-related brain potentials in a task switching paradigm. *International Journal of Psychophysiology*, 59, 30-39.

Hillman, C. H., Weiss, E. P., Hagberg, J. M., & Hatfield, B. D. (2002). The relationship of age and cardiovascular fitness to cognitive and motor processes. *Psychophysiology*, 39*,* 303-312.

Hudspeth, W. J., & Pribram, K. H. (1992) Physiological indices of cerebral maturation. *International Journal of Psychophysiology*, 12, 19-29.

Huttenlocher, P. R. (1990). Morphometric study of human cerebral cortex development. *Neuropsychologia*, 28, 517-527.

Jasper, H. H. (1958). The ten twenty electrode system of the international federation. *Electroencephalography and Clinical neurophysiology*, 10, 370-375

Kaufman, A. S., & N.L. Kaufman. (1990) *Kaufman Brief intelligence Test Manual*. Circle Pines, MN: American Guidance services.

Kramer, A.F., Hahn, S., & Gopher, D. (1999) Task coordination and aging: Explorations of executive control processes in the task switching paradigm. *Acta Psychologica,* 101,339-378

McDowell, K., Kerick, S.E., Santa Maria, D.L., & Hatfield, B.D. (2003). Aging, physical activity, and cognitive processing: an examination of P300. *Neurobiology of Aging* 24**,** 597-606.

Moceri, V. M., Kukull, W. A., Emanuel, I., van Belle, G., Larson, E. B. (2000) Early-life risk factors and the development of Alzheimer's disease, *Neurology*, 54, 415.

Neville, H.J., & Bavelier, D. (2000). Specificity and plasticity in neurocognitive development in humans. In Gazzaniga, M.S. (Ed). *The New Cognitive Neurosciences. (2nd ed.)* (pp.83-99), Cambridge, MA: The MIT Press.

Jacobs, D. R. Jr., Ainsworth, B. E., Hartman, T. J., & Leon, A. S. (1993). A simultaneous evaluation of 10 commonly used physical activity questionnaires. *Medicine & Science in Sports and Exercise*, 25, 81-91.

Jaušovec, H., & Jaušovec, K. (2000). Correlations between ERP parameters and intelligence: reconsideration. *Biological Psychology*, 50, 137-154.

Pfefferbaum, A., Mathalon, D. H., Sullivan, E. V., Rawles, J. M., Sipursky, R. B. & Lim, K. O. (1994). A quantitative magnetic resonance imaging study of changes in brain morphology from infancy to late adulthood. *Archives of Neurology*, 51, 874-887.

Polich, J. (1997). EEG and ERP assessment of normal aging. *Electroencephalography and Clinical Neurophsiology*, 104, 244-256.

Polich, J. & Lardon, M. T. (1997). P300 and long-term physical exericse. *Electroencephalography and Clinical Neurophsiology*, 103, 493-498.

Putnam, L. e., Johnson, R. J., & Roth, W. T. (1992). Guidelines for reducing the risk of disease transmission in the psychophysiology laboratory. *Psychophysiology*, 29, 127-141.

Reiman, E. M., Chen, K., Alexander, G. E., Caselli, R. J., Bandy, D., Osborne, D. et al. (2004). Functional brain abnormalities in young adults at genetic risk for late-onset Alzheimer's dementia. *Proceeding of the National Academy of Sciences of the United States of America*, 101, 284-289.

Reiman, E. M., Uecker, A., Caselli, R. J., Lewis, S., Bandy, D., de Leon, M. J. et al. (1998). Hippocampal volumes in cognitively normal persons at genetic risk for Alzheimer's disease. *Annals of neurology*, 44, 288-291.

Riley, K.P., Snowdon, D. A., Desrosiers, M. F., & Markesbery, W. R. (2005). Early life linguistic ability, late life cognitive function, and neuropathology: findings from the Nun study. *Neurobiology of aging*, 26, 341-347.

Rogers, R. L., Meyer, J. S., & Mortel, K. F. (1990). After reaching retirement age physical activity sustains cerebral perfusion and cognition. *Journal of the American Geriatrics Soceity*, 38, 123-128.

Sallis, J. F., Buono, M. J., Roby, J. J., Micale, F. G., & Nelson, J. A. (1993). Seven-day recall and other physical activity self-reports in children and adolescents. *Medicine & Science in Sports & Exercise*, 25, 99-108.

Scarmeas, N., Habeck, C. G., Hilton, J., Andersonn, K. E., Flynn, J., Park. A., & Stern. Y. (2005). APOE related laterations in cerebral activation even at college age. *Journal of neurology, neurosurgery, and psychiatry*, 76, 1440-1444.

Schuit, A. J., Feskens, E. J., Launer, L. J., & Kromhout, D. (2001). Physical activity and cognitive decline, the role of the apolipoprotein e4 allele. *Medicine & Science in Sports & Exercise*, 33, 772-777.

Scisco, J. L., Leynes, P. A., Kang, J. (2008). Cardiovascular fitness and executive control during task-swithing: An ERP study. *International Journal of Psychophysiology*, 69, 62-60.

Semlitsh, H. V., Anderer, P., Schuster, P., & Presslich, O. (1986). A solution for reliable and valid reduction of ocular artifacts, applied to the P300. *Psychophysiology*, 223, 695-703.

Shaw, P., Lerch, J. P., Pruessner, J. C., Tylor, K. N., Rose, A. B., Greenstein, D., Clasen, L., Evans, A., Rapoport, J. L., & Giedd, J. N. (2007). Cortical morphology in children and adolescents with different apolipoprotein E gene polymorphisms: an observational study. *Lancet neurology*, 6, 494-500.

Sibley, B. A. & Etnier, J. L. (2003). The relationship between physical activity and cognition in children: A meta-analysis. *Pediatric Exercise Science*, 15, 243-256.

Sowell, E. R., Thompson, P. M., Holmes, C. J., Jernigan, T. L., & Toga, A. W. (1999). In vivo evidence for post-adolescent brain maturation in frontal and striatal regions. *Nature Neuroscience,* 2, 859-861.

Sowell E. R., Peterson, B. S., Thompson, P. M., Welcom, S. E., Henkenius, A. L., & Toga, A. W. (2003). Mapping cortical change across the human life span, *Nature neuroscience*, 6(3), 309-315.

Spirduso, W. W. (1980). Physical fitness, aging, and psychomotor speed: A review. *Journal of Gerontology*, 35, 850-865.

Spirduso, W. W. (1983). Exercise and the aging brain. *Research quarterly for exercise and sport*, 54, 208-18.

Strittmatter., W., Weisgraber, K., Goedert, M., et al., (1993). Binding of human Apolipoprotein E to synthetic amyloid beta peptide: Isoform-specific effects and implications for late-onset Alzheimer's disease. *Proceedings of the National Academy of Sciences*, 90, 8098-8102.

Swain, R. A., Harris, A. B., Wiener, E. C., Dutka, M. V., Morris, H. D., Theien, B. E. et al. (2003). Prolonged exercise induces angiogenesis and increases cerebral blood volume in primary motor cortex of the rat. *Neuroscience*, 117, 1037-1046.

Tomporowski, P. D., Davis, C. L., Miller, P. H., & Naglieri, J. A. (2007). Exercise and children's intelligence, cognition and academic achievement. *Educational Psychology Review*, 20, 111-131.

Yu, Y.W., Lin, C.H., Chen, S.P., Hong, C.J., & Tsai, S.J. (2000). Intelligence and event-related potentials for young female human volunteer apolipoprotein E epsilon4 and non-epsilon4 carriers. *Neuroscience Letters***,** 294**,** 179–181