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The Relationship Between Dairy Product Consumption and Cognitive Performance in a Group of Community-dwelling Healthy Older Adults

Mariam R. Ismail, *The University of Western Ontario*

Supervisor: Dr. Alan Salmoni, *The University of Western Ontario*
A thesis submitted in partial fulfillment of the requirements for the Master of Arts degree in Kinesiology
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THE RELATIONSHIP BETWEEN DAIRY PRODUCT CONSUMPTION AND COGNITIVE PERFORMANCE IN A GROUP OF COMMUNITY-DWELLING HEALTHY OLDER ADULTS

(Thesis format: Monograph)

by

MARIAM ISMAIL

Graduate Program in KINESIOLOGY

A thesis submitted in partial fulfillment
of the requirements for the degree of
MASTER OF ARTS

The School of Graduate and Postdoctoral Studies
The University of Western Ontario
London, Ontario, Canada

Abstract

Dietary intake is one of the modifiable factors that may affect older adults' cognitive function in their later years. Very little research has considered the potential role of dairy products on cognitive function. The aim of the present study was twofold: first, to investigate whether there is a difference in cognitive performance between older adults who consumed the recommended amount of dairy products (3 servings per day) and individuals who consumed less than the recommended amount (1 or 2 servings per day); second, to examine whether there is an association between the nutrients contained in the dairy products and cognitive performance. To do this, a cross-sectional study was undertaken. The Montreal Cognitive Assessment (MoCA), Rey Complex Figure Test and Recognition Trial (RCFT), Trail-Making Test, Victoria Stroop Test (VST) and the Digit Span Test (forward and backward) were used to assess cognitive function. Dietary intake of food group consumption was also assessed via estimated 5-day food intake records and analyzed for saturated fat, vitamin D, calcium and other macronutrients and micronutrients. In the 32 participants (8 males and 24 females) who completed the study (average age= 70.59± 7.07 years; BMI=27.59±4.45 kg/m^2), no differences were found between the group who consumed <3 servings of dairy products per day and the group who consumed the recommended amount of 3 servings of dairy products per day in their cognitive task performances. However, a number of associations were found between the nutrients (vitamin D, saturated fat, calcium) found in dairy products and cognitive performance. A positive correlation was found between the level of vitamin D and the RCFT, Digit Span test backward version, and the MoCA. Also, a negative correlation was found between the level of saturated fat and performance on the RCFT. However, no association was found between calcium level in dairy products and performance on any of the cognitive tests. In conclusion, dairy products intake is associated with better cognitive performance but underlying mechanisms are still to be determined.

Keywords: dairy, milk, cheese, yogurt, dairy products, cognitive function/dysfunction, cognitive performance, cognitive state, cognitive decline, and cognitive impairment

Dedication

This thesis is entirely dedicated to my mother for her endless support and love as I completed my thesis. I truly appreciate it.

To Dr. Alan Salmoni for his endless support, wisdom, experience and kindness I completed my master program.

To Dr. June Matthews for her abundant support and kindness when I was an undergraduate as well as a graduate student.

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Once again, I would like to thank my parents and siblings' for their support.

Table of Contents

List of Appendices

List of Abbreviations

PEBL	Psychology Experimental Building Language
RCFT	Rey Complex Figure System and Recognition Trial
VST	Victoria Stroop Test
TMT	Trail Making Test
SD	Standard Deviation
DS	Digit Span
DSTF	Digit Span Test Forward
DSTB	Digit Span Test Backward
DST (F+B)	Digit Span Test (Forward +Backward)
DST (F-B)	Digit Span Test (Forward –Backward)
MoCA	The Montreal Cognitive Assessment
CFG	Canada Food Guide
AMDR	Acceptable Macronutrient Distribution Ranges
RDA	Recommended Dietary Allowances
YPAS	Yale Physical Activity Survey
DRI	Dietary Reference Intake
MMES	Mini Mental State Examination
PUFA	Polyunsaturated Fatty Acids
MUFA	Monounsaturated Fatty Acids
SFA	Saturated Fatty Acids

List of Tables

1 Introduction

Older adults are among the fastest growing segments of the world population. It is estimated that by 2015, people aged 65 years and over will outnumber children under the age of five in both developing and developed countries (World Health Organization, National Institute of Aging & National Institute of Health, 2011). This remarkable increase in the number of people aged 65 years or older is projected to grow from an estimated 524 million (8% of the total world's population) to nearly 1.5 billion in 2050 (16% of the world's population) (World Health Organization, National Institute of Aging & National Institute of Health, 2011; Cohen, 2003). In Canada, older adults are also the fastest growing age group. In 2011, the proportion of Canadian older adults aged 65 years and older was 5 million; one in seven was 65 years and over, and this number is expected to increase to 10.4 million older adults, one in four, which will make the older adults age group roughly one -quarter of the population in 2036 (Statistics Canada, 2011).

With the rise in the older adult population, the incidence of cognitive decline is rapidly increasing as well as the risk of losing independence (quality of life), and the associated challenges both economically and socially are all expected to increase concomitantly. Severe cognitive decline and impairment such as dementia can cause devastating effects on both older adults and their families and can exert a significant financial strain on the health care system. So far, there are no therapies available to cure dementia (Massoud & Leger, 2011). However, pharmacological and non-pharmacological approaches can at least relieve symptoms and perhaps delay the progression of the disease. Thus, the successful development of cost-effective and efficient preventative strategies is extremely important since, if available, these strategies could prevent or delay the onset of cognitive decline and impairment and its severe effects on the lives of millions of people. As a result, devising strategies such as identifying modifiable risk factors to prevent cognitive decline and impairment and to maintain an independent elderly population is a priority. Diet is one important non-pharmacological approach to successful aging and in particular the aging cognitive status. Therefore, the impact of consuming dairy products on the

performance of various neuropsychological tests was investigated in a sample of non-demented older adults living in London, Ontario, Canada.

1.1   Statement of Purpose

The aim of the present study was to determine the impact of consuming dairy products on cognitive performance in community-dwelling older adults. The study utilized dietary data and neuropsychological assessment data to address the following research questions.

1.2   Research Questions

This thesis explored three research hypotheses of the effect of dairy products on cognitive function.

The first purpose of the present study was to investigate whether there is a significant difference in the mean cognitive test scores between people who consumed the recommended amount of dairy products (3 servings per day- according to Canada's Food Guide (CFG) for both males and females aged 51+ years old) (Health Canada, 2011) and people who consumed less than the recommended amount of dairy products (1 or 2) servings per day.

Hypothesis (H1): There is a significant difference in the mean cognitive test scores between people who consume the recommended amount of dairy products (3 servings per day) versus people who consume less than the recommended amount.

The second purpose of the study was to explore whether there was an association between scores on cognitive tests and the levels of macronutrients and micronutrients found in participants' dietary intake of dairy products.

Hypothesis (H2): Performance on cognitive tests will be correlated with levels of saturated fat, vitamin D and calcium as derived exclusively from the dairy products consumed by older adults.

The third purpose of the study was to explore whether there was an association between cognitive test scores and levels of macronutrients and micronutrients found in participants' total dietary intake.

Hypothesis (H3): Performance on cognitive tests will be correlated with levels of saturated fat, vitamin D and calcium derived from older adults' total dietary intake.

2   **Literature Review**

2.1   **Cognition**

2.1.1 Aging and Cognitive Domains

As a result of the expected increase in the population of older adults worldwide and in Canada, it has become important to better understand the changes that accompany the aging process. Aging can have diverse effects on cognitive functions. With age, many older adults experience changes in their thinking abilities; for instance, they face difficulties in mathematical calculations, attention-requiring activities, and memory retention. The scientific term for these mental abilities and processes is cognition. Cognition refers to the complex mental processes that enable humans to exert control over their environment and is critical for survival (Dye, LIuch, & Blundell, 2010). It includes domains such as memory (episodic, semantic, and working memory), language, attention, executive function, and processing speed. Most experts agree that preservation of cognitive ability, including language, memory, executive function (the ability to plan and carry out tasks) and attention well into old age is essential to live an independent life (Elias, 1998). However, cognitive decline is expected as a person ages, and has been reported across a variety of cognitive domains. For example, age-related cognitive decline has been frequently reported in areas such as executive functioning, attention, processing speed, episodic memory, and visuo-spatial functioning (Reid & MacLullich, 2006; Park, Lautenschlager, Smith, Hedden & Davidson, 2002; Salthouse, 1996; Verhaeghen & Cerella, 2002). For example, memory-related complaints increase with age, with up to 50% of adults aged 65-74 years, compared to 88% of older adults aged 85 years and over, reporting having difficulties with memory (Reid & MacLullich, 2006). However, for other cognitive functions such as semantic memory (world knowledge) and implicit memory (previously experienced stimuli that do not require conscious or intended retrieval of encountered information such as riding a bicycle), cognitive function remains intact (Ackerman & Rolfus, 1999; Beier & Ackerman, 2001; Kemper & Sumner, 2001).

2.1.2 Cognition between Normal Aging and Dementia

Cognitive decline can be classified into three types. The first is age-related cognitive decline, which is normal cognitive decline that happens as part of a normal aging process and is generally mild and does not interfere with the ability to participate in daily activities (Anderson, Murphy, & Troyer, 2012; American Psychiatric Association, 1994). Second, mild cognitive impairment (MCI) involves a cognitive decline in memory and other cognitive functions beyond that normally expected in a person of the same age and years of education; however, it is not severe enough to cause lifestyle changes (Anderson et al., 2012). Third, dementia, specifically Alzheimer's disease, is severe cognitive decline that can affect one or multiple domains such as memory, executive functioning, and/or language beyond that found in normal aging resulting in impediment of the person's ability to live independently (American Psychiatric Association, 1994). According to the American Psychiatric Association (1994), age-related cognitive decline is an objective decline in cognitive functioning as part of the aging process but within normal limits given the person's age. Whether age-related cognitive decline is a representation of a normal aging process, or expresses a distinctive clinical medical condition, or is eventually a continuum with dementia is still difficult to establish (Brayne & Calloway, 1988). It is vital to consider that people's differences in cognitive decline form a continuum not an entirely discrete group, and that it is far from straightforward to separate non-pathological (age-related cognitive decline) from pathological cognitive decline (mild cognitive impairment and dementia). Of course, it is important to define people into groups such as mild cognitive impairment or dementia for treatment purposes; however, it is vital to recognize that people's differences in cognitive decline can change from non-pathological (age-associated cognitive decline) to pathological (dementia) in old age (Deary et al., 2009).

Severe cognitive decline (dementia) is expected to increase as the population ages. For example, it is estimated that by 2050, 115 million people worldwide will be living with dementia/Alzheimer's disease (World Health Organization, National Institute of Aging & National Institute of Health, 2011). In Canada, the prevalence of dementia will double as well to more than 1 million Canadians over the next 25 years (Naqvi, Liberman,

Rosenberg, Alston, & Straus, 2013). Older people affected by this disease will eventually need constant care and help with basic activities of daily living (ADL), creating a heavy economic and societal burden on the health care system. In 2010, the total worldwide costs of dementia exceeded 600 billion dollars, including informal care provided by family and others, social care provided by community care professionals, and the direct costs of medical care (World Health Organization, National Institute of Aging & National Institute of Health, 2011). In Canada, as in other parts of the world, the health and economic burden of cognitive decline is enormous. In addition to the personal losses experienced by people with severe cognitive decline, due to losing their ability to function independently, the financial costs to society are substantial. It cost Canada an estimated 33 billion dollars per year, including combined direct (medical) and indirect (lost earnings) costs in 2012, and it is projected to increase to $293 billion a year by 2040, putting a significant financial strain on the health care system (Alzheimer Society, 2012).

Unless new and more effective interventions are found to treat or prevent this severe cognitive decline, its prevalence and the costs associated with it are expected to rise dramatically with the aging population. As a result, prevention of cognitive decline in older adults becomes an even higher priority, and making lifestyle changes such as diet modifications to slow or prevent cognitive decline and impairment will become more and more crucial. Thus, identifying non-pharmacological approaches through healthy lifestyle modifications may have the potential to prevent or slow down the deterioration of individuals' cognitive function. There has been a tremendous amount of research on the development of effective dietary interventions that may help inhibit or ameliorate cognitive decline. Diet is one modifiable factor that is associated with the etiology of age-related cognitive decline, and has been a topic of interest over the past two decades. Dietary intervention can be targeted as an appropriate intervention to optimize the cognitive function of an aging person (Crichton, Bryan, Murphy & Buckley, 2010). Whereas diet may not be useful to treat cognitive decline and impairment, it could be useful to enhance cognitive functions by preventing or ameliorating the severity of the cognitive decline and impairment. There are positive associations between cognitive function in older people and a number of nutrients, including antioxidants (vitamins C and E) (Engelhart et al., 2002a; Wengreen et al., 2007), omega 3 polyunsaturated fatty

acids (Gillette-Guyonnet, Secher, & Vellas, 2013), fruits and vegetables (Kang, Ascherio, & Grodstein, 2005; Ortega et al., 1997; Lee et al., 2001), and vitamin B complex (vitamins B6 and B12, and folate) (Morris, Schneider, & Tangney, 2006c). However, little attention has been paid to the potential impact that dairy products might have on cognitive function in comparison to other nutrients. Few studies have examined specifically the influence of dairy products (directly) on cognitive function and others have examined the impact of dietary intake in general with no particular focus on dairy products intake (indirectly) on cognitive function. Of the little conducted dairy research on cognitive function, there are some indications of positive correlation between dairy product intake and cognitive function (Crichton, Murphy, Howe, Buckley & Bryan 2012; Park &Fulgoni, 2013; Crichton, Elias, Dore & Robbins, 2012; Crichton, Murphy & Bryan, 2010, Ozawa et al., 2014). As a result, it would be valuable to assess directly and in more detail the impact of dairy products on cognitive function in community-dwelling older adults.

2.1.3 Morphological Changes at the Brain Level

There are normal changes that occur in the brain as part of the aging process: the most obvious is a steady decrease in brain size (brain atrophy). The brain undergoes normative structural changes in old age, although not uniformly across all brain areas (Raz & Rodrigue, 2006). Some of the brain lobes that show significant changes include the frontal lobe and the temporal lobe, while other lobes such as the parietal and occipital lobes remain relatively preserved across the lifespan (Anderson, Murphy, & Troyer, 2012). The frontal and temporal lobes are most affected when the person ages and they decline at a faster rate, compared to other lobes such as parietal or occipital lobes. The frontal lobe shows notable changes with aging. At a structural level, there is evidence of atrophy in both the gray matter (the neuron cell body) and the white matter (the nerve fibers connecting different brain areas) (Anderson, Murphy, & Troyer, 2012). The decline in gray matter may reflect reductions in the number of cells (due to cell death) or may be a sign of neuronal shrinkage. The white mater changes reflect axonal abnormalities, and may result in slowed neurotransmission. It is plausible that these white matter changes may mediate the larger performance declines in tasks mediated by the frontal lobe that

accompanies healthy aging (Anderson, Murphy, & Troyer, 2012; West, 1996). It has been suggested that the myelinated fibers in this region are more susceptible to breakdown and this could affect the speed for relaying messages from one neuron to another in the brain. Thus, one of the most significant impacts of change in the white matter is a general slowing of the speed of thinking ability of an aged person (Anderson, Murphy, & Troyer, 2012). The temporal lobe, including the hippocampus, is important in retrieval and associating learned information over time, among many other cognitive functions (Anderson, Murphy, & Troyer, 2012). The cause of age-related shrinkage of this lobe includes some loss of neurons (neuronal atrophy or shrinkage) as well as a decrease in the size and number of branches of the existing neurons (Anderson, Murphy, & Troyer, 2012). The hippocampus (an important region in forming vivid and detailed memory) tends to be under-recruited by older adults during both the encoding and retrieval phases of associative memory tasks, which often correlate with the older adults' reduced performance on these tasks (Kensinger, 00; Cabeza, 2002). In addition to the structural changes of the brain with age, there are also chemical changes. Certain neurotransmitters such as acetylcholine and dopamine are important for communication between neurons and these decline with age as part of the aging process (Anderson, Murphy, & Troyer, 2012). For example, the cerebral dopamine receptor density depletes with age, which plays a central role in regulating attention and in modulating response to contextual stimuli (Hedden & Gabrieli, 2004). In summary, several cognitive changes, such as atrophy in the frontal and temporal lobe neurons, alteration in the white matter, and reduction in acetylcholine and dopamine levels have been associated with age.

2.2 Cognitive decline: Non-Dietary Risk and Protective Factors

Several studies have suggested that there are lifestyle-related behaviors that may prevent the development of cognitive decline (Solfrizzi et al., 2011; Dye et al., 2012; Gao et al., 2013). Table 1 summarizes some of the proposed risk and protective factors that may contribute to the deterioration of cognitive function as an individual ages. However, the exact mechanisms by which these factors contribute to the development of cognitive decline and impairment remain partially unclear due to the fact that one cannot perform a randomized controlled trial (RCT) and also the lack of homogenous exposure and

outcome measures (Williams et al., 2010).

Table1: *Non-Modifiable and Modifiable Risk Factors of Cognitive Decline and Impairment*

Non-modifiable risk factors	Modifiable factors	
	Risk factors	Protective factors
Advanced age	*Vascular and metabolic*	*Psychosocial*
Genetic	Cerebrovascular disease	High education
Family history	(CBVD)/Stroke	Social network
ApoE 4	Cardiovascular disease	Cognitive stimulation
Other Novel genes	(CVD)	
(listed at www.alzgene.org)	Diabetes Mellitus (DM)	*Nutrition and lifestyle*
	Hypertension	Mediterranean diet
	Hypercholesterolemia	PUFAs (e.g. in vegetable
	Obesity	and fish oils)
		MUFAs (e.g. in olive oil)
	Psychosocial	Antioxidants (e.g.
	Low education	vitamins C and E)
	Loneliness	Vitamins B12, B6 and
		folate
	Nutrition and lifestyle	Coffee
	Saturated fatty acids	Moderate alcohol/wine
	(SFAs)	consumption
	Homocysteine	Physical activity
	High alcohol consumption	
	Smoking	*Other*
		Anti-hypertensive drugs
	Other	Statins
	Psychological	Hormone replacement
	stress/depression	therapy (HRT)
	Traumatic brain injury	Non-steroidal anti-
	(TBI)	inflammatory drugs
	Respiratory disease	(NSAIDs)
	Inflammation	

Abbreviations: ApoE= Apolipoprotein E; MUFA= monounsaturated fatty acid; PUFA=polyunsaturated fatty acid

Note. From "The Effects of Midlife Diet on Late-Life Cognition: An Epidemiological Approach" by Eskelinen, M., 2014, Publications of the University of Eastern Finland, Dissertations of Health Sciences, p.7.

2.2.1 Age and cognitive decline: It is well known that age is a risk factor for cognitive decline as it is more prevalent with aging populations (Brayne, 2007). Among individuals aged 60-64 years, the prevalence of dementia is around 1%, but beyond the age of 65, the prevalence of dementia increases exponentially such that up to 50% of 95 year olds are affected (Anderson et al., 2012).

2.2.2 Alcohol and cognitive decline: Moderate alcohol consumption is associated with better cognitive performance and protective effects against cognitive decline and impairment than either abstinence or heavy drinking. This could be due to the protective effect of alcohol on cardiovascular and cerebrovascular health, which are considered major risk factors for cognitive decline (Ganguli, Vander Bilt, Saxton, Shen, & Dodge, 2005).

2.2.3 Smoking and cognitive decline*: smoking, because of its relationship with cardiovascular disease, is a significant risk factor for cognitive decline and impairment. A dose-response effect is evident for the number of cigarettes smoked during one's lifetime and the degree of cognitive decline (Nooyens, van Gelder, & Verschuren, 2008).

2.2.4 Physical activity and cognitive decline*: Cognitive decline is delayed or reduced in people who engage in physical activity. Individuals who walk more may experience less cognitive decline in later life. This is due to the fact that cardiovascular risk factors are known to play a role in the trajectory of cognitive decline in later life, and physical activity lowers these risk factors (Fratiglioni, Paillard-Borg, & Winblad, 2004). Research has shown that physical activity might enhance cognition as cardiovascular fitness increases cerebral blood flow and oxygen delivery to the brain, thereby increasing neuron formation and maintaining brain volume (Etnier, Nowell, Landers, & Sibley, 2006).

2.2.5 Education and social class and cognitive decline: More years spent in formal education and higher social class are associated with less cognitive decline. Mechanisms could include increased "cognitive reserve" through educational and occupational trajectories, safer working and living environments, and access to better health care (Deary et al., 2009; Chodosh, Reuben, Albert, & Seeman, 2002).

2.3 Nutrition: Dietary Assessments and Recommendations

2.3.1 Dietary Assessments

There are several dietary measurement methods that can be used in clinical trials and epidemiological studies to collect dietary intakes from individuals. The most common ones are presented in Table 2. However, it is important to note that despite the various available dietary measurement methods, no single best method exists and diet measurement will always be accompanied by some degree of error (Beaton, 1994). Selecting an appropriate dietary method depends on such considerations as the research design, characteristics of the study participants, and available resources (e.g. skilled personnel, computer facilities, and finances) (Lee & Nieman, 2010). In addition, characteristics such as age, commitment and active participation as well as memory and literacy of the study participants all play a role in choosing the dietary measurement method (Lee & Nieman, 2010). Briefly, the methods can be divided into two categories: record (prospective) and recall (retrospective) (Gibson, 2005; Mahan & Escott-Stump, 2008). Food records or food diaries collect information on current intake over one or more days (usually 1-7 days) (Gibson, 2005). There are two types of food intake records: weighed and estimated. A weighed food intake record is a quantified measure of food and beverage consumption by using scales, while estimated food intake record involves estimating the portion size by using household measures and/or a picture-based portion size booklet (Gibson, 2005; Mahan & Escott-Stump, 2008). All methods must be accompanied by specific data processing software to calculate the food and nutrients intakes, as well as by up-to-date food composition tables.

Table 2: *Dietary Assessment Methods*

Method	Advantages	Disadvantages
Record methods-current intake		
Food record or diary	-Daily recording at the time of food consumption -Can provide recorded information on quantity of food, how food is prepared, and timing of meals/snacks	-Requires literacy skills of participants -Requires ability to measure/judge portion size; underreporting is common -Food intake may be altered -Sex differences exist

		(women more competent) -High costs of coding and analysis
Recall methods-past consumption		
24-hour recall	-Quick -Easy -Inexpensive -Useful in clinical settings -Good reliability between interviewers	-Relies on the short-term memory; in the elderly often unreliable -May not represent usual intake nor reflect nutrient intakes for populations - A tendency to over-and underreport -Selective forgetting (e.g. fat and alcohol) -Requires interviewing skills and comprehensive training of the interviewers
Food frequency questionnaire	-Easily standardized -Good overall picture of food intake; consists usually of 50-150 food items -Suitable for large populations -Inexpensive -Useful in studying the association of a specific food/foods and disease	-Requires literacy skills -Requires knowledge of portion sizes -No meal pattern data, day-to-day variation, longer-term periods of intake, nor quantity-estimation of specific nutrients -Researcher has to know well the current diet before being able to develop an appropriate questionnaire
Dietary history	-Good description of usual intake -Good for longitudinal studies -Observes seasonal variations	-Difficult to standardize -Time-consuming (1-2 h) -High costs of analysis

Note. From "The Effects of Midlife Diet on Late-Life Cognition: An Epidemiological Approach" by Eskelinen, M., 2014, Publications of the University of Eastern Finland, Dissertations of Health Sciences, p.11.

2.3.2 Nutrition recommendations in North America

2.3.2.1 Macronutrient recommendations for older adults

There are set guidelines for macronutrient and micronutrient recommended in North American populations, for example, the Acceptable Macronutrient Distribution Ranges (AMDR). AMDR are values of intake for energy sources that are associated with reduced risk of chronic diseases while providing adequate intakes of essential macronutrients (Institute of Medicine, 2002). AMDR is expressed as percentage of total energy or as percentage of total calories. It has a lower and higher end range. It shows that the incidence of chronic disease will increase if one consumes nutrients above the high boundary of the AMDR, while those who consume less than the lower boundary will increase the risk of consuming inadequate level of macronutrients essential for health (Thompson, Manore, & Sheeshka, 2007). According to AMDR, dietary carbohydrates should be between 45-64% of total energy intake, dietary protein from 10-35% of total energy intake, and dietary fat between 20-35% of total energy intake (Institute of Medicine, 2002). However, the energy needs of older adults are lower than their younger counterparts. This is due to loss of muscle mass and lean tissue, a reduction in thyroid hormones, and a less physically active lifestyle (Thompson, Manore, & Sheeshka, 2007). For example, it is estimated that total daily energy expenditure decreases approximately 10 calories each year for men and 7 calories each year for women aged 19 years and older (Institute of Medicine, 2002). This translates, for instance, to a woman who needs 2000 calories (8370 KJ) at age 20 needing just 1560 calories (6900 KJ) at age 70. Some of this decrease is an inevitable response to the aging process, which can be delayed and minimized by staying physically active and consuming a diet high in nutrients to avoid weight gain. As for micronutrient intake, there are Dietary Reference Intakes (DRIs). According to these guidelines, it is recommended that the intake of saturated fatty acids, trans fatty acids and dietary cholesterol be as low as possible while consuming a nutritionally healthy diet (Health Canada, 2010). However, to reduce the risk for chronic diseases such as heart diseases (and consequently cognitive decline), it is recommended that total fat remain within the 20-35% of the total daily energy intake, with no more than 10% of total energy intake from saturated fat and <300mg per day of cholesterol

(Thompson, Manore, & Sheeshka, 2007; National Institutes of Health, 2002).

2.3.2.1 Micronutrient recommendations for older adults

The vitamins and minerals of particular concern for older adults are calcium, vitamin D, and vitamin B12 (Thompson, Manore, & Sheeshka, 2007). As individuals age, the body undergoes changes that affect the amount of nutrients needed and the way that the body process them. For example, the requirements for calcium and vitamin D, both important for nerve and muscle function, are higher because of the reduced absorption of calcium from the gut, along with reduced production of the active form of vitamin D (which helps with calcium absorption) in older adult's skin as a result of aging (Thompson, Manore, & Sheeshka, 2007). Therefore, it is critical that older adults consume foods that are high in calcium and vitamin D such as low fat cheese, milk, and yogurt. To ensure better health the dietary reference intake recommendations have recently increased from 15mcg/day for those aged 51-70 years old to 20mcg/day for ages 71+ years for vitamin D, and 1000mg/day for males and 1200 mg/day for females aged 51-70 years to 1200mg/day for males and females aged 71+ years old for (Health Canada, 2010).

In addition, vitamin B12 is another nutrient of concern for older adults. Older adults need to consume adequate amounts of vitamin B12 (cobalamin), vitamin B6 (pyridoxine), and folate as inadequate intake of these nutrients are associated with high levels of the amino acid homocysteine (HCy) in the blood. Elevated homocysteine levels are associated with an increased risk for cardiovascular, cerebrovascular, and peripheral vascular diseases, and consequently cognitive decline and impairment (Clarke et al., 2007; La Rue et al., 1997). Accumulation of HCy, in the body has been found to result in oxidative stress, excitation toxicity in neurons, as well as increased DNA strand breakage and mitochondrial membrane damage (Kruman et al., 2000). Vitamin B12 is essential for neurological functioning as it is responsible for enhancing the growth and division of red blood cells (Watanabe, 2007). As a result, deficiency in this vitamin, which tends to affect a large proportion of older adults, is associated with serious health problems such as neurological defects, cognitive decline, and hyperhomocysteinemia, a risk factor for atherosclerosis, which in turn is a risk factor for cardiovascular diseases and cognitive decline later in life (Miller, 2003). Elevated inflammation and vascular disease markers,

such as homocysteine, are implicated in abnormal cognitive decline and dementia. Vitamin B12 deficiency could result from either a low dietary intake of important food sources such as dairy products or problems with its absorption in the intestines (Thompson, Manore, & Sheeshka, 2007). The symptoms of vitamin B12 deficiency include feelings of tiredness, difficulties with attention and memory retention, irritability, and depression. Treatment of this deficiency usually results in improvement in cognitive symptoms, including memory problems. A study involving healthy older adults showed that consumption of 3 cups of milk per day for 12 weeks increased vitamin B12 levels compared to the control group that did not receive milk supplementation (Barr et al., 2000). Also, in a population-based sample of 1183 middle-aged men and women, dietary intake of vitamins B12 and B6 (pyridoxine) was positively associated with self-reported memory function of middle-aged men, and intakes at around the RDI were associated with better memory functioning for women (Bryan & Calvaresi, 2004). Thus, maintaining adequate dietary levels of B12 is essential for healthy brain aging through maintaining proper metabolism of HCy, without which the brain becomes more susceptible to oxidative damage and apoptosis and consequently cognitive decline and impairment.

2.4 Dairy product consumption in North America

Dairy products (milk, cheese, and yogurt) are rich sources of vitamin B12, vitamin D, calcium, and alpha-lactalbumin (whey protein). The latter results in bioactive peptides when partially hydrolyzed and in tryptophan and cysteine when fully hydrolyzed (Chatterton et al., 2006; Camfield, Owen, Scholey, Pipingas, & Stough, 2011) . These various dairy components have been identified as part of the underlying mechanisms that may help with cognitive functioning, either directly or indirectly. Directly, they work via enhanced synthesis of the neurotransmitter "serotonin", which is important for the regulation of mood as well as cognitive function (Camfield, Owen, Scholey, Pipingas, & Stough, 2011). A fully digested α-lactalbumin protein results in tryptophan (precursor of serotonin and cysteine (precursor of glutathione) (Chatterton et al., 2006). By increasing the blood plasma level of tryptophan, there is greater transport of this amino acid across the blood-brain barrier, which results in enhanced serotonin synthesis in the brain

(Lehnert & Wurtman, 1993). Thus, raising brain serotonin level results in enhanced mood and cognitive function. This also results in increased neurogenesis that occurs following stress and the aging process (Jacobs, Van Praag, & Gage, 2000). In addition, serotonin help improve deficiencies and abnormalities in older adults' cognitive function (Jouvet, 1999).

Indirectly, dairy components work via mediating effects on cardiometabolic health. Several studies have revealed that consumption of milk and other dairy products has a beneficial role on cardiovascular health by counteracting cardiometabolic risk factors associated with metabolic syndrome (Pfeuffer & Schrezenmeir 2007; Van Meijl, Vrolix, & Mensink, 2008). Metabolic syndrome is characterized as a cluster of cardiometabolic risk factors including waist obesity, high blood pressure (hypertension), dyslipidemia (hypertriglyceridemia and low high-density lipoprotein (HDL) cholesterol), and high blood sugar/glucose (diabetes) and insulin resistance. For example, whey protein has been found to be insulinotropic (Nilsson, Stenberg, Frid, Holst, & Bjorck, 2004). Milk peptides and calcium have also been found to reduce blood pressure (Nakamura, Yamamoto, Sakai, & Takano, 1995), and they can even reduce plasma (total cholesterol and low-density lipoprotein) cholesterol (Nagaoka et al., 2001). It is recommended that older adults aged 51 years and older consume 3 servings of dairy products daily (Health Canada, 2011). However, milk consumption has fallen over the past 20-25 years in many countries (Elwood, 2005). According to Garriguet (2007), a large proportion of Canadians, in all age groups, are not meeting their recommendations for dairy products. This is supported by the most recent per capita consumption survey indicating an average of 1.3 serving of dairy products per day. Two out of three Canadians are not consuming the minimum number of servings of milk per day due to excessive intake of caffeine, as well as frequent consumption of sweetened beverages and soft drinks (Garriguet, 2007; Weaver, 2010). Moreover, over 70% of those aged 50 years and older are not meeting the recommendation of 3 servings per day of dairy products (Garriguet, 2007).

Dairy products are one dietary component receiving little attention in relation to cognitive function despite numerous studies confirming their beneficial role to cardiovascular health. In addition, research studies have positively associated these cardiometabolic risk

factors including hypertension (Qiu, Winblad, & Fratilioni, 2005), diabetes (Gregg et al., 2005), dyslipidemia (Moroney et al., 1999), and obesity (Whitmer, Sidney, Selby, Johnston, & Yaffe, 2005) with increased risk of cognitive decline and dementia. Thus, the beneficial change in cardiometabolic health imparted by high dairy intake may be the primary mechanism by which dairy products (indirectly) reduce the risk of cognitive decline. The following are the current theories regarding the likely mechanisms of action by which dairy products' individual constituents affect cognitive function directly.

2.4.1 Dietary fat and cognitive health

Fats are important constituents of the human diet due to their high-energy value (9kcal/g), but also because fat-soluble vitamins, such as vitamins A, D, E and K, and carotenoids (vitamin A precursors) play a critical role in the health and functioning of the human body (German &Dillard, 2006). Milk fat as a whole is comprised primarily of triglycerides, in addition to a small amount of phospholipids and sterols, namely cholesterol. Triglyceride is a fat molecule that is made of glycerol and three fatty acids. Absorption of the fatty acid component of the triglyceride molecule requires the hydrolysis of the triglyceride molecule into a glycerol molecule and free fatty acids (Bennion & Schedule, 2004). Fatty acids can be categorized into unsaturated fatty acids (UFA) and saturated fatty acids (SFA). Unsaturated fatty acids can be further categorized into monounsaturated fatty acids (MUFA) (found in olive oil) and polyunsaturated fatty acids (PUFA) (found in vegetable oil) (Bennion & Schedule, 2004). Milk fat contains almost 70% SFA, with myristic (14C) and palmitic acids (16C) combined accounting for almost 50% of long-chain saturated fatty acids (Jensen, 2000). Also, milk fat is a major source of short-medium saturated fatty acids such as butyric (4C), caproic acids (6C), lauric acid (12C) and stearic acids (18C) (Bennion & Schedule, 2004). The different chain length of SFA exerts varied biological effects and has a differential impact on cholesterol level. For example, myristic, palmitic and lauric acids are thought to have a large effect on cholesterol level, whereas butyric and caproic acids have neutral effect on human cholesterol levels (Visioli & Strata, 2014). Although a diet excessively rich in SFA is deleterious because of the increased risk of heart disease, it is important to note that the goal for overall dietary fats is not the elimination of fat, but the maintenance of a

healthy balance of SFA, MUFA and PUFA, and milk fat can contribute toward this balance (German & Dillard, 2006). Dairy products containing milk fat are major food sources of saturated fatty acids (SF), accounting for approximately 21% of total saturated fat intake in the U.S. diet (US Department of Health and Human Services Dietary Guidelines for Americans, 2010). As a result, only low-fat and fat-free dairy products are recommended to enhance healthy plasma lipid profiles to reduce the risk of cardiovascular diseases (Lichtenstein et al., 2006). However, reducing the amount of fat from dairy products only is not effective as other food groups can contribute to total fat in the human diet. High intake of SFA increases blood levels of total cholesterol and LDL cholesterol, which are considered a predictive risk factor for cardiovascular diseases (Thompson, Manore, & Sheeshka, 2007). Certain SFA such as lauric (C12:0), myristic (C14:0), and palmitic acid (C16:0) have been considered hypercholesterolemic and were reported more than 40 years ago to raise total cholesterol and LDL-cholesterol levels (a well-established biomarker of cardiovascular disease risk) (Lichtenstein et al., 1998), whereas, stearic acid (C18:0) has little effect on cholesterol levels (Lichtenstein et al., 1998). The concentration of these SFA in bovine milk varies widely and can range from 11% to 54% by weight in milk fat (German & Dillard, 2006). That is why it is not effective to reduce all types of SFA as some of them can have little or no effects on total cholesterol level. Also, a causal relationship between the intake of saturated fatty acid and cardiovascular disease risk remains controversial (Siri-Tarino, Sun, Hu, & Krauss, 2010). Several prospective cohort studies have shown inconsistent evidence that higher intake of milk and dairy products, regardless of milk fat levels, are associated with an increased risk of cardiovascular disease (Elwood, Pickering, Givens, & Gallacher, 2010; Goldbohm, Chorus, Galindo Garre, Schouten, & van den Brandt, 2011). It is well known that a human diet containing SFA increases the plasma level of cholesterol and therefore can increase the risk of cardiovascular disease (Mensink, Zock, Kester, & Katan, 2003). In addition, the fatty acid composition is not altered when total fat is reduced because other food groups can also provide fat. Also, total fat reduction will contribute to reduced intake of polyunsaturated fatty acids (PUFA), and may result in the augmentation of the cholesterol-raising properties of certain saturated fatty acids, such as myristic fatty acid (in the case that PUFA falls below 5% of total calorie intake) (Hayes, Kohsla, Hajri, & Pronczuk, 1997). Many factors are associated with the variations in the amount and

composition of the fatty acid components in bovine milk: animal origins (related to breed and selection), stage of lactation, mastitis and ruminal fermentation, or feed-related factors such as fiber and energy intake and seasonal and regional effects (Jensen, 2000; Jensen, 2002). Although SFA constitute a large proportion of milk fat, bovine milk contains a mixture of other components, such as lipids, protein, and micronutrients, many of which, when consumed separately or as components of dairy products, have shown favorable or neutral effects on outcome measures of cardiovascular disease risk (Tremblay & Gilbert, 2009; Soedamah-Muthu et al., 2011). For example, there are different constituents in dairy products that have beneficial health effects on metabolic syndrome, a risk factor for cardiovascular disease and cognitive decline later in life. For example, whey protein has been found to be insulinotropic (Nilsson et al., 2004). Other examples are milk peptides and calcium, which have been found to reduce blood pressure (Nakamura et al., 1995) and plasma cholesterol (Nagaoka et al., 2001). The possible mechanism by which calcium can affect plasma cholesterol levels is via fat absorption inhibition in the small intestine (Denke, Fox, & Schulte, 1993), reducing both total cholesterol and LDL cholesterol (bad cholesterol) while increasing HDL cholesterol (good cholesterol) (Van Meijl, Vrolix, & Mensink, 2008). As a result, consumption of dairy products is inversely associated with hypertension, diabetes, and hypercholesterolemia, which are considered components of metabolic syndrome as well as risk factors for cardiovascular diseases and cognitive decline later in life. Thus, reducing the consumption of dairy products due to its fatty acid content may have a negative impact, as other components in dairy products have a positive impact on the wellbeing of an individual. Also, the association between SFA and cardiovascular diseases risk depends mainly on the length of the fat and whether it is naturally present in foods or added from industrially produced sources (hydrogenated). Short-chain fatty acids (SCFA) and medium chain fatty acids (MCFA) comprise approximately 10% of total fatty acids in milk by weight and are generally associated with no effect or a beneficial effect on blood lipids profile (Zaripheh & Miller, 2008). Despite recommendations of lowering saturated fatty acids in human diet, several studies support the role of these dietary fatty acids in enhancing HDL metabolism. Researchers have investigated the impact of reduction of total fat and saturated fatty acids on HDL metabolism in a group of multiracial, young and elderly men and women. Participants

consumed 3 different diets, each for 8 weeks: 34.3% total fat and 15% saturated fat; 28.6% total fat and 9% saturated fat; and 25% total fat and 6% saturated fat. HDL metabolism decreased in a stepwise fashion after the reduction of total fat and saturated fat (Berglund et al., 1999).

Literature has shown mixed associations between SFA in dairy products and cognitive function. Some studies have shown negative associations, and others have shown no associations. In a population-based prospective study on 1449 individuals aged 65-80 years, older adults using moderate amount of fats from spreads and milk products had a decreased risk of developing dementia compared to individuals who use little or no fats at all (Laitinen et al., 2006). This indicates that moderate intake of fat in the diet may be beneficial. However, the type of fat in the diet should be taken into consideration too. In the same study, moderate intake of saturated fatty acids were associated with increased risk of dementia, while moderate intake of PUFA and MUFA were actually protective (Laitinen, et al, 2006). These associations were seen only among the Apo E epsilon 4 group (gene responsible for dementia) even after adjusting for this factor (Laitinen et al., 2006). In addition, the study showed that higher intake of SFA from milk products and spreads were associated with poorer global cognitive function and prospective memory, while higher intake of polyunsaturated fatty acids were associated with better semantic memory (Laitinen et al., 2006). In a French epidemiological cohort study to determine the potential long-term impact of dietary habits on age-related cognitive decline among 4809 female older adults, cognitive decline was associated with lower intake of animal fats, and higher intake of dairy fats and vitamin A (Vercambre, Bortron-Ruault, Ritchie, Clavel-Chapelon, & Berr, 2009). In this study, participants consumed 70% of animal fat as butter and the intake was generally low with 95% of the women consuming less than 6 grams per day (Vercambre et al., 2009). As a comparison, 90% of the population included in the Chicago Health and Aging Project consumed more than 16 grams of animal fat/day (Morris, Evans, Bienias, Tangney, & Wilson, 2004). The latter study concluded that diets high in SFA (16g of saturated fat) are associated with cognitive decline in older adults, whereas there was no association between the intake of animal fat and cognitive function in the French study. This could be due to the fact that the level of SFA (6g) in this study was below the level of which could be detrimental to cognitive

function. Also, in 2012, a randomized, two-way crossover dietary intervention trial examined the effect of high intake of reduced-fat dairy products on cognitive function. Fifty-nine overweight or obese individuals aged 18-75 years old, whose usual dairy product intake was low (2 servings/day) were randomized to either high dairy (4 serves/day) or low-dairy (1 serves/day) treatment groups, and crossed over to the alternate diet after 6 months. Participants were tested at the end of each 6- month diet period on multiple measures of cognitive functions, including memory, information processing speed, executive functioning, attention, and abstract reasoning. In the 38 participants who completed the trial, spatial working memory performance was marginally better following 6 months of consuming a high dairy products diet compared with a low dairy product diet (Crichton, Murphy, Howe, Buckley & Bryan, 2012). Another study involved a retrospective cross-sectional analysis using data from food frequency questionnaires on 432 men and 971 women, aged 39-65 years old. It investigated the association between consumption of dairy products and cognitive function, assessed by Cognitive Failures and Memory Functioning questionnaires. Results indicated that consumption of low-fat yogurt was associated with better memory recall in men only. Also, consumption of whole fat dairy products, including ice cream and cream (high in saturated fat) were associated with increased depression and poorer memory functioning (Crichton, Murphy & Bryan, 2010)

Other studies have examined saturated fat from the total diet as opposed from dairy products only, and this literature has also shown mixed results. Some studies have shown a negative association and others have shown no association at all. For example, a study from Spain assessed the relationship between dietary intake and global cognitive functions using the Mini Mental State Examination and the Pfeiffer's Mental State Questionnaire (PMSQ) in 260 community-dwellers who were mentally healthy individuals, aged 65-90 years old. Subjects with adequate scores in the MMSE (>28 points) had lower intake of SFA, MUFA, and cholesterol (Ortega et al., 1997). However, whereas this study included other macro and micronutrients assessments, it did not include TFA intake. The Chicago Health and Aging Project followed 3718 subjects aged 65 years old for over six years and evaluated their dietary intake with a 139-item FFQ. Cognitive change was measured at 3 and 6 year follow-ups with the East Boston Memory

Test (EBT) of immediate and delayed recall, the MMSE and the Symbol Digit Modalities test for perceptual-motor speed (SDMT). Results show that a diet high in saturated and trans fats may be associated with cognitive decline in older adults (Morris et al., 2006). This is supported by another study by The Cardiovascular Risk Factors, Aging and Dementia (CAIDE) longitudinal study in Finland examined the association between cognitive health and diet. A population of 1449 people aged 65 to 80 years was followed for a total average of 21 years. An FFQ test was used for dietary assessment. Evaluation of cognitive status through the assessment of cognitive and executive global functions, memory, and psychomotor skills were conducted. Results show that abundant saturated fat at midlife was associated with poorer global cognitive function and prospective memory after adjusting for confounders. In contrast, high intake of PUFA was associated with better semantic memory. Furthermore, frequent fish consumption was associated with better global cognitive function and semantic memory. Higher PUFA-SFA ratio was also associated with better psychomotor speed and executive function (Eskelinen et al., 2008). Also, a cross-sectional population-based study among 1613 adults aged 45-70 years old examined dietary intake and cognitive function. Dietary intake was assessed via a self-administered food frequency questionnaire while cognitive function was assessed via neuropsychological tests including the visual (verbal) learning test, the Concept Shifting task, an abbreviated Stroop Color-Word test, the Letter Digit Substitution test, and a Category Fluency test. Results show that fatty fish and marine omega -3 polyunsaturated fatty acids (PUFA) consumption was associated with a reduced risk of cognitive dysfunction while the intake of cholesterol and saturated fat were associated with increased risk of impaired cognitive function in this population group (Kalmijn et al., 2004). However, a three-year long prospective study examined the effects of dietary fatty acids on cognitive function in 482 women, failed to show any association between saturated fatty acids and cognitive function. Dietary intake was assessed with food frequency questionnaire while cognitive function was evaluated with various tests including Consortium to Establish a Registry for Alzheimer's disease (CERAD), word list learning constructions, and word fluency tests. Results showed no association between cognitive function and intake of saturated fat, trans fat, or dietary cholesterol intake. MUFA intake was associated with lower cognitive decline, particularly in the visual and memory domains (Naqvi et al., 2011).

On the other hand, other studies have shown a positive association between dairy products consumption and cognitive function and longevity, without taking into account the fat level of the dairy products. For example, a cross-sectional study by Crichton et al. (2012) examined the association between dairy and cognition, based on data from 972 adults aged 23-98 years from the community-based Maine-Syracuse Longitudinal study. The results demonstrated a significant association between the self-reported frequency of consumption of dairy products and cognitive function, assessed by a battery of eight cognitive tests. Highest scores for all cognitive tests were observed for high dairy product consumers: executive function, scanning and tracking, global and visual-spatial memory and organization, MMES, and verbal memory were better when compared with those who never or rarely consumed dairy products. However, the study did not account for the fat content of dairy products (Crichton, Elias, Dore & Robbins, 2012). This is supported by a 17-year longitudinal prospective population-based cohort study that examined the effects of milk and dairy intake on the development of dementia, especially Alzheimer's disease in 1081 cognitively healthy Japanese older adults aged 60 years and older. Milk and dairy intake were estimated using a 70-item semi-quantitative food frequency questionnaire while cognitive function was assessed using the Hasegawa Dementia Scale or MMSE. Results show that higher intake of milk and dairy products were associated with lower incidence of all-cause dementia, especially Alzheimer's disease and vascular dementia (Ozawa et al., 2014). Other studies have shown a positive correlation between longevity and dairy product consumption in older adults. A cross-sectional study investigated the relationship between dietary patterns and longevity in 104 centenarian (>100 years old) Japanese subjects. Dietary intake was assessed via a 3-day food intake record recorded by caregivers while cognitive function was assessed via a classified semi-quantitively scale into 5-categories ranging from (0-normal, 0.5- subnormal, 1-mild demented, 2- moderately demented, and 3- severely demented). Results showed that individuals with a dietary preference for dairy products tended to have greater longevity compared to the other dietary patterns in centenarian individuals (Shimizu et al., 2002). Also, in a 5-year cohort study among 162 self-sufficient, healthy older adults aged 65 years and older residing in an elderly home in Rome, Italy, the relationship between specific food groups and nutrients and overall 5-year survival were assessed. Dietary intake was measured via a validated, semi-quantitative food frequency questionnaire.

Results showed that frequent consumption of citrus fruit, milk and yogurt, low consumption of meat, and high intake of vitamin C, riboflavin and linoleic acid is associated with longevity (Fortes et al., 2000).

2.4.2 Dietary calcium and cognitive health

Approximately 53% of daily nutrient intake of calcium is contributed by the consumption of dairy products in a typical Canadian diet (Agriculture and Agri-Food Canada, 2009). Calcium is an important mineral that is required in amounts greater than 100 mg per day in the human diet (Thompson, Manore, & Sheeshka, 2007). Dairy products are the most common source of calcium in the Canadian diet, with one cup of plain yogurt and one cup of 2% milk contributing 450 mg and 300mg of calcium, respectively (Thompson, Manore, & Sheeshka, 2007). If dairy products are excluded from the diet, then it is difficult to meet the recommended daily intake (RDI)-adequate intake of calcium, which is 1200mg/day for males and females aged 50 years and older (Thompson, Manore, & Sheeshka, 2007). Older adults have a higher need for calcium; however, their ability to absorb calcium diminishes with age due to their reduced intestinal capacity (only 25%) to absorb calcium (Thompson, Manore, & Sheeshka, 2007). This change in calcium absorption with age has been taken into account by increasing the recommendation to 1200mg/day, compared to the 1000 mg/day for adults aged 19-50 years old (Thompson, Manore, & Sheeshka, 2007). Calcium is critical for normal transmission of nerve impulses. It enters nerve cells and stimulates the release of molecules called neurotransmitters, which conduct nerve impulses from one neuron to another. Therefore, calcium deficiency inhibits the ability to transmit messages between neurons (Thompson, Manore, & Sheeshka, 2007). This is why a person can experience convulsions when blood calcium levels fall dangerously low (Thompson, Manore, & Sheeshka, 2007). However, in the case that a person's calcium needs are high, the body will generally increase its absorption rate from the small intestine. However, as adults get older, their small intestine capacity to absorb calcium diminishes as part of the aging process along with their higher need of calcium. These individuals, in turn, will turn to calcium supplements to fulfill their higher need for calcium than can be obtained in a balanced

diet. This can lead consequently to various health conditions, such as mental confusion (Thompson, Manore, & Sheeshka, 2007).

Hypercalcemia, or high serum levels of calcium, usually due to supplement use, is a disease condition in which serum (blood) calcium levels reach abnormally high concentrations (Thompson, Manore, & Sheeshka, 2007). This abnormal serum calcium level is associated with a cognitive decline in older adults (Schram et al., 2007). In 2007, one prospective cohort study of two independent, population-based studies, the Rotterdam study and the Leiden 85-plus study, have shown that high serum calcium levels are associated with a faster decline in cognitive function over the age of 75 years old (Schram et al., 2007). In the Rotterdam study, high serum calcium levels were associated with worse global cognitive function on the MMES and a faster rate of cognitive decline during 11 years of follow-up in older adults aged 75 years and over. In the Leiden 85-plus study, high serum calcium levels were also associated with worse test scores on the MMES and this held true for the specific cognitive domains that were tested: attention, psychomotor speed, and memory (Schram et al., 2007). In these two sub-studies, the authors did not mention the reason for high serum calcium levels, which could be either from calcium supplements use or disease conditions. The possible mechanism led to the observed association between serum calcium level and cognitive decline through calcium dysregulation. Calcium dysregulation or disrupted calcium homeostasis, plays a role in the pathophysiology of cognitive decline or dementia (Tolppanen, Williams & Lawlor, 2010). Calcium ions can diffuse easily through the blood brain barrier. Serum calcium levels are directly associated with the calcium levels in the cerebrospinal fluid in the brain or extracellular calcium levels in the brain (Tolppanen, Williams, & Lawlor, 2011). High calcium levels in the brain activate calcium sensor receptors, enhancing calcium influx into neurons, and leading to calcium overload and subsequently neuronal death (Breitwieser & Gama, 2007). Numerous studies have demonstrated that elevation of calcium levels in the neurons ultimately lead to neuronal cell death, and as a result, it is well known that maintaining serum calcium levels within a narrow limit within individuals is important in preventing cognitive decline and dementia (Schram et al., 2007). In summary, these research studies have suggested that high serum calcium levels due to supplement use is associated with greater

cognitive decline in older adults (Schram et al., 2007; Tilvis et al., 2004). While consuming too many calcium supplements can lead to toxicity and even death (Thompson, Manore & Sheeshka, 2007), consuming more than the recommended intake of calcium from dietary sources does not lead to toxicity as excessive dietary calcium can not be absorbed from the small intestine and will end up excreted in feces (Thompson, Manore & Sheeshka, 2007).

Numerous studies have shown a positive association between dietary intake of calcium and cognitive function. For example, in a cross-sectional and a short-term prospective (9 months) cohort study on active and mentally healthy community dwellers (n=187) over 65 years of age in Portugal, researchers investigated the association between the adequacy of dietary intake of both calcium and omega-3 PUFA and cognitive function. Dietary intake was assessed using a 3-day food intake record, while cognitive function was assessed using the MMSE. Results showed that diets high in calcium are related to improvement in cognitive function, while high intake of omega-3 PUFA might prove to be beneficial (Velho, Marques-Vidal, Baptista, & Camilo, 2008). This is also supported by another cross-sectional study on cognitive healthy (n=449) community- dwellers aged 60 years and older in Korea, which examined the association between dietary intake and cognitive function. Dietary intake was assessed using a 24-hour recall, while cognitive function was assessed using the MMSE. Results showed that a diet with low intake of calcium and dairy products was associated with lower cognitive test scores (Lee et al., 2001). Lastly, a 17-year long prospective population-based cohort study, examined the effects of milk and dairy intake on the development of dementia, especially Alzheimer's disease, on 1081 cognitively healthy Japanese older adults aged 60 years and older. Milk and dairy intake was estimated using a 70-item semi-quantitative food frequency questionnaire while cognitive function was assessed using the Hasegawa Dementia Scale or the Mini-Mental Status Examination (MMSE). Results showed that higher self-reported dietary intake of potassium, calcium and magnesium were associated with lower incidence of all-cause dementia, especially Alzheimer's disease and vascular dementia (Ozawa et al., 2012). Also, a cross-sectional study was conducted on 240 elderly individuals (120 institutionalized and 120 free living) selected randomly from institutions and from different community sites in Alexandria, Egypt. The purpose of the study was

to assess the nutritional status, measured using 24-hour recall for 3 consecutive days of those living in institutions and those living independently in the community. Total daily energy intake, vitamin A and calcium (the nutrients least adequately supplied in the elderly diets) were found below the recommended dietary allowance for all subjects, with lower intake among free-living, compared with institutionalized individuals (Shabayek & Saleh, 2000). According to the research study results, this lower intake is due to the fact that free-living subjects had lower intake of calcium- rich food, such as milk and cheese, compared to institutionalized individuals, who were served milk and its products on daily basis at both meals and snacks (Shabayek & Saleh, 2000). Also, the study showed a low intake of vitamin A (important for enhanced cognitive function) among free-living compared to institutionalized individuals. This may be due to the low intake of fresh vegetables-rich in carotene, and low- fat dairy products such as milk and yogurt, which were mainly consumed by elderly subjects (Shabayek & Saleh, 2000). This study indicated that community-dwellers in Egypt had lower dietary intake of calcium because they consumed less dairy products (rich sources of calcium) compared to institutionalized individuals. This indicates that dairy products are rich source of calcium and that community-dwelling older adults usually consume less than the recommended, which is in line with previous research studies. Thus far, research has shown that higher dietary intake of calcium is associated with improved cognitive function.

2.4.3 Dietary vitamin D and cognitive health

Vitamin D (calciferol) is a fat-soluble seco-steroid synthesized in skin (as hormone) or ingested with food (as vitamin) (Kalueff & Tuohimaa, 2007). Vitamin D is biologically inactive; however, it undergoes bioactivation by double hydroxylation in the liver and kidney, leading to the formation of 1,25-dihydroxyvitamin D, the main biological active form of vitamin D (Kalueff & Tuohimaa, 2007). Vitamin D crosses the blood- brain barrier and binds to vitamin D receptors in the brain. Vitamin D receptors and the enzyme responsible for formation of the active vitamin in the human brain, 1 alpha hydroxylase, are widespread in both brain neurons and glial cells within brain regions critical for cognition (Kalueff & Tuohimaa, 2007). This indicates that, as vitamin D is a fat-soluble vitamin, it can cross the blood- brain barrier. It is also the likely mechanism by which

vitamin D might have an effect on cognitive function. Beyond the age of 50 years, the need for vitamin D increases. It is estimated that we experience a four-fold decrease in our capacity to synthesize vitamin D from the sun by the time we reach 65 years of age and older (Thompson, Manore, & Sheeshka, 2007). Thus the adequate intake (AI) of vitamin D for men and women aged 50-70 years old is 10 µg/day, and the AI increases to 15 microgram per day for adults aged 70 years and older. Most foods naturally contain very little vitamin D. Thus, our primary source of this vitamin in the diet comes from fortified milk. Due to high vitamin D insufficiency in the Canadian population, the government has proposed changes to both the levels of vitamin D added to foods, and the type of foods fortified. For example, currently, cow's milk and goat's milk have approximately 53 IU (1.3microgram) of vitamin D per 100 ml, but yogurt, buttermilk, cottage cheese, and most cheeses are not fortified with vitamin D (Thompson, Manore, & Sheeshka, 2007). Several studies have shown a positive association between lower vitamin D level and lower cognitive function in older adults. In a population-based, cross-sectional study on 3369 European men aged 40-79 years, researchers have linked low serum 25-hydroxyvitamin D levels with slower processing speed on the Digit Symbol Substitution Test (DSST) cognitive test (Lee et al., 2008). Similar results were shown in another cross-sectional study on 3325 older adults aged 65 years and older which linked vitamin D deficiency with increased odds of cognitive decline and poor sustained attention, but not memory or visuospatial ability (Liewellyn, Lang, Langa, & Melzer, 2011). Similarly, low levels of biomarker 25(OH) of vitamin D in homebound older adults in Boston were associated with poor executive functioning and processing speed but not memory (Buell & Dawson-Hughes, 2009). The study indicated that a low level of vitamin D is associated with lower cognitive decline. However, it is important to consider that the study did not account for whether this lower level was related to diet or supplements use or just not getting enough amount of sun exposure. Another study considered supplement use. A population-based cross-sectional study on 5596 female community-dwelling older adults in France, with a mean age of 80 years and free of vitamin D supplements, has shown that women with inadequate weekly dietary intake of vitamin D (5 microgram/day or 35 microgram/week) were associated with a lower score on Pfeiffer Short Portable Mental State Questionnaire (SPMSQ), a test for global cognitive function (Annweiler, Schott, Rolland, Blain, Hermann, & Beaucher, 2010).

On one hand, several mechanisms have been proposed to explain how vitamin D deficiency may increase the odds of cognitive decline. One of the mechanisms is that vitamin D is involved in the metabolism of several neurotransmitters in the central nervous system (CNS). Such neurotransmitters include, but are not limited to acetylcholine, dopamine, serotonin and gamma-aminobutryic acids (Kalueff & Tuohimaa, 2007). Also, vitamin D may exert its neuroprotective role through its antioxidative properties, which in turn can stabilize cognitive functioning and prevent the onset of neurodegenerative diseases such as dementia (Kalueff &Tuohimaa, 2007). On the other hand, the exact mechanism by which dietary intake of vitamin D influences cognitive functioning is still unclear. It has been proposed that older women with poor cognition and loss of autonomy usually eat nutrient-poor foods, with subsequent inadequate dietary intake of vitamin D (Annweiler et al., 2010). Alternatively, a low vitamin D intake might be surrogate for other nutrient abnormalities, which could result in cognitive decline (Annweiler et al., 2010). According to the same study, disability is often seen among older women with cognitive impairment and inadequate dietary intake of vitamin D, compared to those with normal cognitive functioning and those taking the recommended dietary intake of vitamin D (Annweiler, et al., 2010). A cross-sectional study was conducted on 69 free-living urban healthy Italian elderly people aged 70-89 years. Dietary analysis over 3 days before study entry and observed nutrient intake were compared with the Italian Recommended Dietary Allowance (RDA). Cognitive function was assessed using the MMSE. Results showed a significant negative correlation between dietary intake of vitamin D and poor performance on cognitive tests. This inverse relationship is due to low mean intake (below RDA) for calcium, vitamin D and folate. This inadequacy between the observed intake of those nutrients in this age group and the RDA was the result of low consumption of milk and by-products, fish, fruits and vegetables containing vitamin D (Rondanelli, Trotti, Opizzi, & Solerte, 2007). A 13-year population-based study in France looked at cognitively healthy adults aged 35-60 years and examined the association between midlife dietary intake of vitamin D and subsequent performance. Dietary intake was assessed using computerized 24-hour dietary records on randomly assigned days for a total of 6 records per year. Cognitive function was assessed 13 years later with a comprehensive neuropsychological test battery that assessed: episodic memory using the RI-48 cued recall test (consisting of a list of 48 words divided

into 12 categories); semantic memory using the semantic fluency task (consisting of naming as many animals as possible); short-term memory using the forward and backward digit span test; and executive function using the Trail Making test. Results show that neither of these domains were associated with midlife dietary intake of vitamin D. However, the study did show a significant and positive association between midlife dietary intake of vitamin D and the forward digit span task measuring short-term memory. This indicates that dietary intake of vitamin D might have different effects on various cognitive domains (Andreeva et al., 2014).

2.5 Cheese consumption

In a state-wide survey of 1056 older adults aged 69 years in Albama, intake of cheese was found to be inversely correlated with cognitive decline (Rahman, Baker, Allman & Zamrinin, 2007). Study participants who ate cheese at least once a week were more than 40% less likely to have cognitive decline than those who ate cheese less than once a week or never (Rahman, Baker, Allman & Zamrinin, 2007). The potential mechanism by which cheese consumption may affect cognitive function is through vitamin A (retinol), an antioxidant, or tyramine. Tyramine is an amino acid found in many cheeses. It is a precursor of major neurotransmitters (such as dopamine and norepinephrine) that play a role in cognitive functioning through anti-amnestic actions (Izquierdo & Dias, 1983). Another possible mechanism that may explain the effect of cheese on cognitive functioning involves the process of cheese aging. During cheese aging, bioactive peptides are formed which could have a beneficial effect on memory. These compounds need to be small to cross the blood brain barrier in the brain (Ortego et al., 1997; Wu, Ying, & Comez-Pinilla, 2004)). A single serving of aged cheese provides a high amount of certain proteins, lipids, vitamins and minerals, which continuously replenish the central nervous system (CNS). For instance, cheese provides specific amino acids such as cysteine that are important for glutathione production during periods of inflammatory stress. This amino acid allows the CNS to regulate inflammation and maintain homeostasis in the brain. Also, cheese provides lipids to the CNS, which allows neuronal cells signaling to function optimally as lipids play important roles in the synthesis and fluidity of the nerve cell membrane, synaptic plasticity and neuronal regulations (German & Dillard, 2006).

This could be the secondary mechanism of action of cheese on cognitive function (Park & Fulgoni, 2013). A study using a cross-sectional approach drew on data from the National Health and Nutrition Examination Surveys (NHANES 1988-1994 and 1999-2002), and set out to determine the potential relationship between intake of dairy products (total dairy products, milk and cheese) and cognitive function in adults aged 60 years and older (Park &Fulgoni, 2013). In the first dataset, a story recall was used as the only test of cognitive functions (attention and delayed verbal memory). However, in the later dataset (NHANES 1999-2002), response speed, sustained attention, visual spatial skills and associative learning and memory were assessed using the digit-symbol substitution test (DSST). In both datasets, an in-person 24-hour dietary recall was obtained from the participants. The analyses showed that consumers of dairy products were associated with a higher score on the story recall test and the digit-symbol substitution test when compared with non-consumers. Even among groups, cheese consumption (high in saturated fat) showed significantly higher cognitive scores in comparison to milk consumption (Park & Fulgoni, 2013). This indicates that cheese consumption might have an effect on cognitive function.

2.6 Yogurt consumption

Yogurt, a fermented milk product, is a nutrient-dense food. It is abundant in calcium, zinc, B-vitamin, and probiotics (El-Abbadi, N., Dao, M., & Meydani, 2014). A 100-gram serving of plain low-fat yogurt contains 0.56 mg vitamin B12, 183 mg of calcium among many other dietary minerals and vitamins (USDA, 2013). Yogurt is also a good source of protein and it may be supplemented with vitamin D and additional probiotics, which are associated with a range of beneficial health outcomes at the level of musculoskeletal, immune, cardiometabolic, and cognitive systems. Research has shown that consuming yogurt (an average of 2.3 servings yogurt/week) is associated with a greater adherence to healthy dietary habits, such as eating fruits, vegetables, nuts, fish, and whole grains (Wang, Livingston, Fox, Meigs, & Jacques, 2013). These results further show that yogurt consumers have a significantly reduced prevalence of nutrient deficiencies for riboflavin, vitamin B12, calcium, magnesium, and zinc (Wang, Livingston, Fox, Meigs, & Jacques, 2013). There is limited research on the influence of yogurt on cognitive function and

long-term mental health. This is why it is an important area for future research to explore, as yogurt is a rich source of vitamin B12 due to evidence of its anti-inflammatory effects that may be protective against cognitive impairment. A longitudinal study by Vercambre et al (2009) was conducted on French women born between 1925 and 1930 to examine the relationship between dietary habits and age-related mental health. The study showed that cognitive health and instrumental activities of daily living declined significantly with lower concentrations of riboflavin, vitamin B6 and vitamin B12. This result was supported by another cross-sectional study conducted in Australia by Crichton et al. (2010), on 432 men and 971 women, aged 39-65 years old. Results indicated that consumption of low-fat yogurt was associated with better memory recall in men only (Crichton, Murphy, & Bryan, 2010).

In summary, the above research shows inconclusive results from studies regarding the impact of dairy products on cognitive function. This is due to several methodological issues that need to be taken into consideration. First, only a small number of studies have investigated the potential role of dairy products alone on cognitive function; most studies included other food groups. Second, some studies had a wide age range (e.g. 20-90 years of age), were not limited to older adults, and had a highly disproportionate number of participants between males and females. Third, dairy intake and total dietary intake were self-reported. Also, the researchers did not account for the fat content of dairy products and measurement of dietary intake; using a questionnaire and a single 24- hour recall may not provide a true representation of the impact of long-term consumption of dairy products. In addition, most of the studies did not use a comprehensive neuropsychological battery to test cognitive functions. Moreover, some studies did not adjust for cardiovascular health, lifestyle, and other dietary factors that may affect cognitive functions. Some studies did not account for a wide range of dairy products, such as dairy products that are involved in the preparation of mixed dishes. This inconclusive result could also be due to the study design (cross-sectional vs. longitudinal), or because the fat content was not only examined from dairy products food group only, but also from other food groups. Also, some studies have examined different dairy products (milk, cheese, yogurt). These studies have also been conducted in different geographical regions. For instance, most of the studies that have been conducted to

investigate the relationship between dairy products fat and cognition are in countries such as Sweden, Finland, Japan, United States, Australia and Korea. These countries may have different percentages of fat incorporated in their individual processes of manufacturing dairy products, and other food groups in comparison to Canada.

3   Methods

3.1   Participants

A total of 35 older adults (9 males, 26 females) participated in this study (mean age 70.23±6.86, range 61-86). Study participants' were recruited on a voluntary basis from the London community. Individuals were recruited using a recruitment flyer, referral and face-to-face meetings. Recruitment flyers were posted at Western Retired Academic Associations (RAA), public libraries, church groups and the Western senior alumni lecture series. Permission was obtained to place the flyers along with sign-up sheets so that interested participants could leave their contact information if they wished to participate. Also, a phone number and email address was put on the flyers so that interested participants could contact the researcher if they chose not to leave their name and phone number/email address on the sign-up sheets. Potential participants were excluded if they were less than 60 years old, unable to communicate in English (read/write), lived in long- term care or assisted living facilities, or required assistance with activities of daily living. Also, they were excluded if they had visual impairment such as color blindness and/or auditory impairment uncorrected by aids. The study protocol was explained fully to participants (Appendix A) and all participants provided written, informed consent (Appendix B). The study employed a cross- sectional design to investigate the potential relationship between community dwelling older adults' consumption of dairy products and cognitive performance on five cognitive tasks. Most testing was conducted in a quiet room at a public library. This study was approved by the Health Sciences Research Ethics Board (Appendix C).

3.2   Measures

3.2.1   Demographic Measures

Demographic measures, including age, height, weight, gender, marital status, education, employment history and other factors were measured through a self-administered demographic questionnaire (Appendix D). These demographic factors have been used in past studies on cognitive function in older adults to describe the sample. In this

questionnaire, lifestyle factors, history of diseases and use of medication and supplements were also included.

3.2.2 Cognitive Function Measures

A battery of neuropsychological tests was administered to all participants by the researcher. The tests were selected based on previous literature. To assess the fundamental cognitive domains, five (2 computerized and 3 paper-based) neuropsychological assessments were conducted. These five tasks included the Montreal Cognitive Assessment (MoCA), the Rey Complex Figure Test and Recognition Trial (RCFT) (Appendix G), the Trial-Making Test (TMT), the Digit Span Test; both forward and backward (DST) and the Victoria Stroop Test (VST). Table 3 depicts the order, duration and delay for cognitive assessment and table 4 shows the cognitive tests and the domains they each examined whilst subsequent sections describe each of the tests administered.

Table 3: *Order, duration, and delay for cognitive assessments*

Task	Trial	Time to next part of task	Duration (approximately)
Rey Complex Figure Test	Copy	3-minutes	2-10 minutes*
The Stroop test			3 minutes
Rey Complex Figure Test	3 minute delayed recall	27 minutes	2-10 minutes*
The Digit Span Test			4 minutes
The Trail Making Test			3 minutes
The Montreal Cognitive Assessment			10 minutes
Rey Complex Figure Test	30 minutes delayed recall and recognition		5-14 minutes*
			Total ~60 minutes

Note. Time of task variable based on participant's completion speed

Table 4: *Neuropsychological tests used to measure cognitive domains*

Neuropsychological test	Cognitive domain	Outcome measures	Descriptions
Rey Complex Figure Test and Recognition Trial (RCFT)	Visual Memory	RCFT-3 minute delay (Immediate recall) (raw score)	Number of elements correctly recalled
		RCFT-30 minute delay (Delay recall) (raw score)	Number of elements correctly recalled
		RCFT- recognition trial (raw score)	Number of elements correctly recognized
Victoria Stroop Test (VST)	Executive function/selective attention	VST- Efficiency Score (C/D) (seconds).	Speed of Stroop Colors/Speed of Stroop Dots
Digit Span Test (DST)	Attention/working memory	DST- Forward (raw score)	Total Correct Forward Trials (raw score)
		DST- Backward (raw score)	Total Correct Backward Trials (raw score)
		DST -(Forward +Backward) (raw score)	Total correct trials forward and backward) (F+B)
		DST- (Forward – Backward) (raw score)	Total correct trials forward and backward (F-B)
Trail-Making Test (TMT)	Visual attention/task switching/processing speed/cognitive flexibility		Speed of Trial
		TMT- (minutes)	
Rey Complex Figure Test and Recognition Trial (RCFT)	Visuospatial functioning	RCFT-copy (raw score)	Number of elements drawn correctly
The Montreal Cognitive Assessment (MoCA)	Global cognitive function	MoCA (raw score)	

3.2.2.1 The Montreal Cognitive Assessment (MoCA)

The Montreal Cognitive Assessment (MoCA, 2014) is used as a screening measure to detect the general cognitive status and Mild Cognitive Impairment (MCI) clinical condition in the elderly population (Nasreddine et al., 2005). The items included in the MoCA measure were visuospatial/executive functioning, naming, memory, language, abstraction, delayed recall and orientation. A score was given for each correct answer, with a maximum possible score of 30 points. MoCA was included in the cognitive battery, as it has been used previously to assess the relationship between cognition and dietary interventions (De Jager, Oulhaj, Jacoby, Refsum, & Smith 2012; Sydenham, Dangour, & Lim, 2012; Nasreddine et al., 2005; Markwick, Zamboni, & De Jager, 2012).

3.2.2.2 Rey Complex Figure Test and Recognition Trial (RCFT)

The Rey Complex Figure Test and Recognition Trial (RCFT); (Meyers & Meyers, 1995) is used as a measure of visuospatial functioning and visual memory. RCFT was included in the cognitive battery, as it has been employed previously to assess the relationship between cognition and dietary interventions (Ho et al., 2007; Gleason et al., 2009). Participants were presented with a complex figure and instructed to accurately copy it (RCFT copy). Three minutes following the copy, participants were asked to re-draw the figure from memory to produce the (RCFT 3- minute- delay) score. Thirty minutes following the initial copy, participants were asked to again re-draw the figure from memory to produce the (RCFT 30- minute- delay) score. After the 30-minute delayed recall, participants were presented with 24 different figure designs and asked to recognize which of these 24 designs were parts of the original stimulus figure (recognition trial) score. All three drawings were scored via standardized procedures to produce the test scores. Two cognitive outcome measures were used from this test: First, the RCFT (composite score), an outcome measure of visual memory including immediate recall, delayed recall, and recognition trial was computed. This score was calculated by converting the raw score for each of these measures to overall sample-based Z-scores then Z-scores were averaged for the relevant measures to compute a single composite score for that domain. Second, the RCFT (copy score), an outcome measure used for visuospatial functioning was computed. Please refer to Appendix G for a copy of the

stimulus figure.

3.2.2.3 The Victoria Stroop test

The Stroop test is a timed measure of selective attention and cognitive flexibility (Stroop, 1935; Regards, 1981). Various versions of the Stroop test are available; in this study the Victorian version was used (Spreen & Strauss, 1998). The Stroop test was included in the cognitive battery, as it is has been employed previously to assess the relationship between cognition and dietary interventions (Fournier et al., 2007; Pipingas et al., 2008). There were three parts to the task; the first part (part D (dots)) required participants to name the color of dots. They were given a card with six lines of four dots of four colors (red, green, yellow and blue). Each color was used six times, and the four colors were arranged in a pseudo-random order within the array with each color appearing once in each row. Participants had to read across the rows as quickly as possible reading out the color of each dot. The time to complete the task and number of errors made was recorded. For the second task, (part W (neutral/non-color words) a new card with six rows each with four words (When, And, Hard and Over) written in four different colors was used. Common words were printed in color and in lower case. Again, time to complete task and number of errors was recorded. Participants had to read across the rows as quickly as possible reading out the colors. The third task, the colors task, was the most difficult of the three Victorian Stroop tasks, and assesses the examinees' ability to suppress usual responses for a less favorable, yet correct, response. In this task, a card was presented with words printed in any of the four colors; however, the words were the four aforementioned colors (e.g., the word green printed in the color red). Participants were asked to name the colors of ink of the printed words, not the actual word. This task required the participant to inhibit the automatic response of reading the word, for the more difficult task of naming the color. Time to complete the task and number of errors were recorded. Before the actual test began, the individual had the opportunity to learn what they were required to do on each part of the test. The first and second part of the test was used to measure cognitive speed, whereas the third part of the test was used to measure response inhibition (Verhaeghen & Cerella, 2002). Paper-based administration of the test required that, if an error was made, the participant had to correct the error before continuing to the next item and this was reflected in the completion time of the test. The approximate time

to administer this test was five minutes. The Victoria Stroop Test (VST) efficiency score was calculated by dividing C (colored words part) by D (dots part).

3.2.2.4 PEBL- Digit Span (DS) test

Digit span is a measure of attention and concentration, and is incorporated in the Wechsler Memory Scale (WMS) and the Wechsler Adult Intelligence Scale (WAIS-III); (Wechsler, 1997). The digit span test was included in the cognitive battery, as it has been employed previously to assess the relationship between cognition and dietary interventions (Eussen et al., 2006; Dangour et al., 2010; Fournier et al., 2007). The digit span task comprises two separate trials: digits forward and digits backward. Before the actual test administration, the participant was given a practice trial. Then, the actual test was administered using a laptop computer running the Psychological Experimental Building Language software program (Mueller & Piper, 2014). First, in the digits forwards task, a random number sequence consisting of three numbers was presented visually on the computer to the participant. After the numbers disappeared, the participant was required to type in the digits in the space provided on the computer by using the keyboard. This was followed by another sequence of the same length. Following correct performance on the pervious shorter sequences, sequences with one extra number were generated by the computer and the process continued until the participant made an error on both of the two sequences of the same length. In the digits backwards task, the process was the same as digits forwards except that the participant had to type in the numbers backwards. Both tests continued to a maximum of 10 numbers unless the participant failed earlier. Digit Span Forward scores reflected the highest number of correctly repeated consecutive digits (DST- F). Digit Span Backward scores reflected the highest number of correctly repeated consecutive digits in the backward order (DST-B). The sum of correct digits forward and digits backward made up the total digit span score (F+B), whereas the difference of correct digits forward and digits backward made up the total digit span score (F-B).

3.2.2.5 PEBL-Trail-Making test

The Trail Making Test (TMT) has been incorporated into prominent neuropsychological batteries such as the Army Individual Test and Halstead-Reitan Neurological Battery as an assessment of visual search, psychomotor speed and cognitive flexibility (Reitan & Wolfson, 1985; Crowe, 1998; Tombaugh, 2004). Visual search is a measure of indvidual ability to scan an environment for a specific target amongst distractions (Eckstein, 2011), while cognitive flexibility is defined as the individual's ability to shift and adapt cognitive processing and speed in response to different circumstances (Eslinger & Gratton, 1993). TMT was included in the cognitive battery, as it has been employed previously to assess the relationship between cognition and dietary interventions (Gleason et al., 2009; Crews et al., 2005; Karr et al., 2012). TMT is a timed measure of processing speed, executive function, visual attention, cognitive flexibility and task switching. Before the test began, the participants were given a paper-based practice session to ensure understanding and familiarity with the test. Then, the actual test was administered using the Psychological Experimental Building Language (PEBL version 0.14). There were three parts to the task; the first part (TMT- Numbers) of the test required the participant to click on each circle of the 26 circles numbered from 1-26 in numeric order as quickly and accurately as possible. The circles were arranged in a random order on the screen. The time to complete each of these trials was recorded and if errors were made, they were corrected and this was reflected in the completion time of the trial. This part of the test is considered an assessment of visuomotor speed. For the second task (TMT- Letters), the participants were presented with 26 circles with letters A-M arranged in a random order on the screen and required the participant to click on each circle in alphabetical order as quickly and accurately as possible. Also, if errors were made, they were reflected in the completion time of the trial. This part of the test is also used to assess the visuomotor speed. The third task, the numbers and letters task, is the most difficult of the three parts of the Trial-Making test, and assesses the ability to switch between numbers and letters as quickly and accurately as possible. In this task, participants were presented with 26 circles arranged in random order on the screen. These 26 circles alternated between numbers (1-13) and letters (A-L), and participants had to click on them in alternating order (1-A-2-B-3-C and so on). This part of the test was used to evaluate cognitive flexibility in addition to visual scanning (Crowe, 1998). The time to complete each of these trials was recorded and error correction was reflected in the

completion time of the task. Participants were instructed to use the computer mouse and click circles in sequential numeric (1-2-3-4---), alphabetic (A-B-C-D-----) or alphanumeric (1-A-2-B-3-C----) order. When the correct sequential circle was clicked, a computer-generated line (trail) connected the circles together. The process continued until all 26 circles were joined together. Timing did not begin until the first circle was clicked, which is labeled "1" on every trial. There were six trials in total; with each part having two trials, alternating between numbers, letters and numbers and letters. The average of these six trials made up the total time in seconds required to complete the Trail-Making test. The approximate time to administer this test was 5-10 minutes.

3.2.3 Physical activity measures

3.2.3.1 The Yale Physical Activity Survey (YPAS)

The Yale Physical Activity Survey (Dipietro et al., 1993) was used to assess the participants' physical activity level. YPAS was included in the battery because it is sensitive enough to detect the lower ranges of physical activities engaged in by older adults (Dipietro et al., 1993). The YPAS contains a wide range of physical activities including those involved in household chores, recreation, and structural exercises. It is a measure of physical activity level in adults over 60 years of age. Participants were required to report the frequency and duration of physical activity in five categories (vigorous, leisurely walking, moving standing and sitting) during a typical 7-day period and over the previous month. An index score for each of these categories was computed as a product of frequency, duration, and a weighting factor for each of the physical activities in a category. A summary score, known as the Yale index, was computed as the sum of the index scores across the above- mentioned five categories.

3.2.4 Dietary measures

3.2.4.1 Food Intake Record (FIR)/Diary

Dietary intake, with a particular focus on dairy product intake (whether consumed directly or incorporated into mixed dishes) was collected using a five-day food intake record/diary including a 2-weekend days and 3- weekdays prior to the cognitive testing to capture habitual or "usual" dietary intake. This food intake record/diary was used to collect information about the time of meal intake, the amount and type of all food and beverages, and how it was prepared (i.e., raw, fried, boiled), as well as all main ingredients included in mixed dishes. In addition, any medications and/or supplements eaten or drank by the participants were recorded on the diary for each day. The food diary required the participants to write down everything they consumed from the time they woke up until the time they went to sleep. Participants were given a folder, which included pamphlets that contained sets of photographs of types and portion sizes of foods to help them determine the quantities ingested and a set of instructions to help them record their dietary intake. Also, the diary (five pages of food intake record; one page for each day) was included in this folder for participants to record their dietary intake. This diary was structured with headings relating to the time of consumption, actual food eaten, and preparation of food, brand name and amount eaten. A "comment" section was included at the end of each day for participants to write down any relevant additional information. A sample practice of how to fill in the diary (1-day food intake record) was practiced with the participants to ensure that they could record information as accurately as possible. This practice session was conducted during the first visit/interview between the participant and the researcher. In the second visit, the researcher inspected the food intake record for completeness and that sufficient details had been recorded. Following input of this diary into the computer software program, the independent variables were the saturated fat (g), calcium (mg), and vitamin D (mcg) over the five days. This was done to obtain a more reliable estimate of the "usual food consumption" .The diary was inputted by the researcher into a computerized nutritional data analysis software program (ESHA), which uses the USDA food composition table databank as the reference source for nutrient composition. All dairy products ingested were coded and converted into energy and nutrients. The tables of recommended intakes of energy and nutrients for the Canadian population were used to calculate the Recommended Dietary Allowances

(RDA) for vitamin D, calcium and other nutrients and the Acceptable Macronutrient Distribution Range (AMDR) for saturated fat and other nutrients for this population group. Comparisons between observed intakes and RDA and AMDR were used to assess the adequacy of the diet as well.

3.3 Procedures

Participants who were willing and eligible to participate in the study were scheduled for an initial study visit of one hour. This visit was determined upon mutual agreement of a suitable time and place to meet. This first-visit began with a brief overview of the study to ensure understanding and to review the consent form. Next, participants were trained on how to fill in the food diary. The participants were also given a phone number to call if they had any questions or concerns when filling out the diary. Instructions on how to fill in the Yale Physical Activity Survey were given to participants. The participants were then given the demographic questionnaire, food intake records/food diary and the physical activity questionnaire (YPAS) to be completed at home and briefly debriefed about the cognitive testing which would take place at the next visit. Also in this visit, a time and place for the second visit was arranged one week later. The participants returned the following week for a second and final one-hour visit. In this visit, the participants were given a copy of their signed informed consent and advised to keep it for their records. The participants submitted their completed demographic questionnaires, food intake records/ diaries and the Yale Physical Activity Assessment. To eliminate any potential distractions, cognitive testing was done in a quiet room at the public library. Also, cognitive testing was done individually to eliminate any potential influential observational behavior of one participant over the other. To standardize testing, participants were required to sit up straight in front of the computer screen at a distance of 60-65 cm. The researcher monitored participant compliance routinely. Prior to conducting the PEBL battery, participants were provided standardized instructions and reminded that both speed and accuracy of each task were important. There were five separate cognitive tasks: two computerized using the PEBL battery tests (TMT and DST) and three paper-based formats (MoCA, RCFT and VST). The PEBL (Michigan Tech University, Houghton, MI), a widely available Windows computer software program,

provided the platform for the cognitive battery. Previous research conducted by Piper and colleagues (2014) concluded that PEBL demonstrated similar accuracy as other previously validated cognitive tests when evaluating executive function. These five cognitive tasks were administered individually and in the same order for each participant. For the computer format, the tasks were administered using a personal computer running the Microsoft Windows operating system. The tasks were displayed on a 20-inch monitor and the "Fullscreen" option box was checked on the PEBL Launcher. The instructions were displayed on the screen and read by the experimenter to each participant and each participant was asked to complete each task as fast and as accurately as possible. For the paper format, the tasks were administered on paper and instructions were read to participants before the actual test. Following the completion of each task participants were asked if they would like a short break and were also given the opportunity to ask questions. Practice sessions for the cognitive tasks (VST, TMT and DST) were administered to each participant before the actual test. Please see table 3 for details of the testing. On completing the protocol, the participants were thanked for their participation and asked if they would like to receive a final research report of the study findings. Participants were reminded that their data would remain confidential.

3.4   Data Analysis

3.4.1 Nutritional

Dietary intakes were analyzed using the Elizabeth Stewart Hands and Associates nutritional analysis software ESHA (version 2014). Total dietary intake and dairy product intake only were obtained from the participant food diary and analyzed using this software. The USDA food composition databank was selected as the reference source for nutrient composition. Following the input of participant total dietary intake as well as dairy product intake only, a mean daily nutrient intake of vitamin D (mcg), calcium (mg), saturated fat (g) and other nutrients were acquired. Three servings of dairy products per day was assumed as the cut-off criterion diet score for dairy products as recommended by Canada's Food Guide (CFG) for adults aged 51 years and over. The researcher, in accordance with Canada's Food Guide, calculated the number of servings of dairy product intake from participants' food diary. Dairy products were determined as the

consumption of any of the following food items: cheese, yogurt, dairy deserts, cream, ice-cream, and milk. Thus, a serving of dairy product intake was calculated as follows: 1 serving of milk (reconstituted or powdered) =250 ml (1 cup)

1 serving of milk (evaporated)= 125ml (.5 cup)

1 serving of fortified soy beverage= 250 ml (1cup)

1 serving of yogurt= 175 g (.75 cup)

1 serving of Kefir= 175 g (.75 cup)

1 serving of cheese= 50 g (1.5 oz)

1 serving of yogurt drinks=200 ml

Nutrient content and scores from the cognitive battery of tests were used to complete statistical analyses used to answer the research hypotheses.

3.4.2 Statistical

Data were analyzed using the Statistical Package for the Social Sciences (SPSS) version 17.0 for Windows. Independent (unpaired) t tests were used to investigate group differences in the number of servings of dairy products (<3) group and (>=3) group and neuropsychological tests. The independent variable was the number of servings of dairy products (<3) group (>=3) group and the dependent variables were MoCA, RCFT composite and copy scores, VST Efficiency score, TMT score and DST scores. Exploratory analyses were conducted using the Pearson's product- moment correlation coefficient to investigate if cognitive performance on the neuropsychological tests was associated with the nutrients intake from dairy products only and/or from the total diet. Independent variables were saturated fat (g), vitamin D (mcg) and calcium (mg) and the dependent variables were MoCA, RCFT composite and copy scores, VST Efficiency score, TMT score and DST score. Linear Regression was also conducted to determine whether performance on cognitive tests could be predicted by the individuals' nutrient intake. Independent variables were saturated fat (g), vitamin D (mcg) and calcium (mg)

and the dependent variables were MoCA, RCFT composite and copy scores, VST Efficiency score, TMT score and DST score. Each regression analyses included age and total energy intake as confounding variables. Significance was set at $p \leq 0.05$. Caution should be taken when interpreting the results reported below. Since a large number of analyses were performed, there is an elevated experiment wide probability of type I errors. However, because of the exploratory nature of this study, the traditional $p \leq 0.05$ significance level was retained. Preliminary analyses were performed to ensure no violation of the assumptions of normality. The interpretation of the association's strength between the nutrient and cognitive test was determined according to Cohen's (1988) guidelines. These guidelines are as follows: r=. 10 to .29 or r=-.10 to -.29 is small; r=. 30 to .49 or r=-.30 to -.49 is medium; r=. 50 to1.0 or r=-.50 to -1.0 is large.

4   Results

4.1   Sample Characteristics

Of the original 35 study participants, three were unable to complete the entire study protocol. Two participants had unforeseen family obligations and one participant had a fear of being diagnosed with dementia. The three participants were excluded from data

analysis resulting in a final sample of 32 participants. The sample consisted of 8-males and 24-females with an average age of 70.59 years (SD=7.07, range 61-86). All participants were community dwelling older adults residing in London, Ontario. The demographic and medical characteristics of the sample are presented in table 5.

Table 5: *Demographic and Health Characteristic of Older Adults*

Characteristics	N (%)
Age Group	32
60-65	4 (12.5%)
65-74	19 (59.3%)
75+	9 (28.13%)
Gender	
Male	8 (25%)
Female	24 (75%)
Marital Status	
Single	3 (9.4%)
Widowed	3 (9.4%)
Separated	0 (0%)
Divorced	3 (9.4%)
Common-law	3 (9.4%)
Married	20 (62.5%)
Education	
Elementary	0 (0%)
High School	4 (12.5%)
Vocational/Technical School	2 (6.25%)
College	4 (12.5%)
University: Bachelor Degree	10 (31.25%)
University: Post-Graduate Degree	10 (31.25%)
Professional Degree	2 (6.25%)
Most Frequent Self-Reported Health Problems	
High Blood Pressure (Hypertension)	14 (43.75%)
High Blood Sugar (Diabetes)	3 (9.4%)
High Blood Cholesterol (Dyslipidemia)	8 (25%)
Obesity (High Waist Circumference)	2 (6.25%)
Depression	4 (12.5%)
Anxiety	2 (6.25%)
Stress	3 (9.4%)
Taking Prescription Medications	
Yes	20 (62.5%)
No	10 (31.25%)
Taking Dietary Supplements	
Yes	27 (84.4%)
No	5 (15.63%)

Consume Soft Drinks	
Yes	12 (37.5%)
No	20 (62.5%)
Consume Alcohol	
Yes	26 (81.25%)
No	6 (18.75%)
Smoking	
Yes	1 (3.13%)
No	31 (96.88%)
Physical Activity	
Part I (Energy Expenditure Summary Index)	4023.1Kcal/week
Part II (Yale Physical Activity Index)	60.16

4.2 Cognitive Performance on Neuropsychological Battery

All participants had normal global cognitive function as evidenced by having a score of 26 out of 30 on the Montreal Cognitive Assessment (MoCA), which corresponds to the criterion cut-off score used to classify individuals' cognitive function as either normal or impaired cognitive function. The sample had normal scores on digit span forward and digit span backward as healthcare professionals usually use 6+-1 as a normal range for Digit Span Forward and 5+-1 for Digit Span (Hodges, 2005). The sample also had within the average scores for copy (33.42-29.63), immediate recall (17.65-9.00), delayed recall (17.29-8.60) and recognition total correct (20.23-19.33) for adults aged 60-89 years old for the RCFT (Meyers & Meyers, 1995). The sample had above the range score of (95.9-204) seconds reported by (Tombaugh, 2004). This is due to the fact that the test was measured using the computer timer. However, when the timer was manually started, the average test score of 202 seconds was obtained, which is within the range scores associated with this age group. As for the Victoria Stroop test, the sample performed better than the average efficiency scores of (2.55-2.95) for adults aged 60-80 years and over reported by (Bullock, Brulot & Strauss, 1996, Spreen & Strauss, 1998). For the Dots part of the test (12.56-19.31) seconds, Words part of the test (16.16-23.91) seconds and Colors part of the test (31.32-56.98) seconds for adults aged 60-80 years and over reported by (Regards, 1981). Please refer for table 6 for test results.

Table 6: *Neuropsychological Outcomes for Older Adults*

Cognitive domain	Neuropsychological test	Mean (+-SD)
Visual memory	RCFT-3 minute delay	13.27 (6.87)

	(Immediate recall) (raw score)	12.86 (6.86)
	RCFT-30 minute delay (Delay recall) (raw score)	19.38 (1.99)
	RCFT- recognition trial (raw score)	-0.069 (0.85)
	RCFT- Composite score (Immediate recall+ Delayed recall+ Recognition)	
Executive function/selective attention	Stroop Test Efficiency Score (C/D) (seconds)	1.56 (0.37)
	Stroop Dots (seconds)	.1925 (.043)
	Stroop Words (seconds)	.208 (.058)
	Stroop Colors (seconds)	.293 (.700)
Attention/working memory/processing speed/visual attention/cognitive flexibility	Digit Span Test Forward (raw score)	6.25 (1.50)
	Digit Span Test Backward (raw score)	4.84 (1.27)
	Digit Span Test (Forward+ Backward) (raw score)	11.09 (2.44)
	Digit Span Test (Forward- Backward) (raw score)	1.41 (1.34)
	Trail Making Test (TMT) (seconds)	250.59 (87.09)
Visuospatial functioning	RCFT-copy (raw score)	32.11 (4.82)
Global cognitive functioning	MoCA (raw score)	26.1 (2.32)

Note. In cognitive tests used, the higher result means better performance, except for Victoria Stroop Test and the Trail Making Test where the best performance is the least amount of time used.

4.3   Dietary Patterns

Only 10 participants (31.25%) reported consumption of the recommended amount of dairy products per day (3 servings) with one of these ten participants reporting having

four servings of dairy products per day. The remaining 22 participants (68.75%) reported having either one or two servings per day. Also, 84.4% of the participants were regular users of dietary supplements. Most common supplements taken were vitamin D, multivitamin, calcium and vitamin B12. Almost half of the participants (53.13%) had vitamin D supplements, 43.75% reported having multivitamin, 18.75% reported having calcium and 9.3% reported having vitamin B12. The most common reasons of taking these supplements were just an extra to the dietary intake or due to deficiency. On average, participants had 2079.74 kcal per day, which is considered high, as the energy needs of older adults should be lower than those of younger adults (Thompson, Manore & Sheeshka, 2007). Also, participants' intake of saturated fat was 13 % of total calories intake, which is well over the recommendations of (<7% of total calories from saturated fat). Most participants (56.25%) had their cholesterol level within the recommendations of <300mg per day. However, participants' level of vitamin D and calcium were well below the DRI at 100% and 84.4%, respectively. However, it is important to note that the current study did not take into account the supplements taken. Please refer to table 7 for an average intake of key nutrients from participants' total dietary intake.

Table 7: *Average Dietary Intake of Key Nutrients from Total Diet Over Five Days*

Nutrients	Mean (SD)
Energy (kcal)	2079.74 (616.80)
Saturated fat (g)	29.23 (11.09)
Cholesterol (mg)	275.71 (121.69)
Vitamin D (mcg)	2.32 (1.79)
Calcium (mg)	813.02 (293.74)

As for physical activity, all participants were considered physically active. Part I of the YPAS assesses the amount of physical activity/exercise during a typical week in five categories: work, yard work, care taking, exercise and recreation. Participants spent an average of 3.73 (2.23) hours per day doing exercise/physical activity during a typical week. Part II of the YPAS assesses the individual's physical activity level over the pervious month. It included specific activities such as (vigorous, leisurely walking, moving, standing and sitting). Participants scored on average 60.16 (Yale Physical Activity Index)

4.3.1 Hypothesis (H1): Number of Servings of Dairy Products and Cognitive Performance

Independent t tests were used to investigate group (< 3 servings versus >=3 servings of dairy products) differences in cognitive test scores. Results showed that there were no significant differences in any of the performed cognitive tests. Therefore, visual memory as measured by the RCFT-composite score; visuospatial functioning as measured by the RCFT-copy score; executive functioning and selective attention as measured by the VST; working memory as measured by the DST; visual attention, processing speed and cognitive flexibility as measured by the TMT and the global cognitive functioning as measured by the MoCA were none significantly different for participants who consumed less than 3 servings of dairy products per day compared with participants who consumed 3 or more servings of dairy products per day. Thus, hypothesis (H1) was not supported. Please refer to table 8 for neuropsychological tests scores between both groups.

Table 8: *Comparison of mean scores on neuropsychological tests between (< 3) and (>=3) servings of dairy products*

Cognitive test outcome measures	(<3 servings of dairy products) group N=22	(>=3 servings of dairy products) group N=10	T-test (p-value)
RCFT-Copy Score	32.614 (3.65)	31.00 (6.85)	.874 (.389)
RCFT Composite Score	.00 (1.02)	-.143(.77)	.394 (.696)
Trial-Making Test (TMT)	4.04 (1.23)	4.54 (1.95)	-.876 (.388)
Digit Span Test			
Forward (F)	6.00 (1.38)	6.80 (1.69)	-1.419 (.166)
Backward (B)	4.59 (1.01)	5.40 (1.65)	-1.719 (.096)
Forward +Backward (F+B)	10.59 (1.89)	12.20 (3.19)	.036 (.164)
Forward –Backward (F-B)	1.41 (1.50)	1.40 (.966)	.017 (.986)
MoCA	26.14 (1.98)	26.00 (3.06)	.152 (.880)
Victoria Stroop Test (Efficiency Score)	1.59 (.43)	1.50 (.22)	.791 (.435)

Note. RCFT=Rey Complex Figure Test and Recognition Trail, TMT=Trail Making Test, MoCA=The Montreal Cognitive Assessment, VST=Victoria Stroop Test.

4.3.2 Hypothesis (H2): Relationship between dairy product nutrient intake and cognitive performance

Bivariate, Pearson-Product Moment correlations were computed between cognitive measures and saturated fat, vitamin D and calcium levels found exclusively in the dairy products consumed. Please refer to Appendix E for all of the dairy product correlational results. Specifically, performance on the Rey Complex Figure Test and Recognition Trial (as measured by the composite score) was negatively correlated with saturated fat [$r=-0.361, n=32, p=0.042$]. A high level of this nutrient was associated with fewer items recalled and recognized by participants from the RCFT stimulus figure. There was a positive correlation between the level of vitamin D intake and performance on three cognitive tests [$r=0.367, n=30, p=0.046$] for RCFT; [$r=0.390, n=30, p=0.033$] for DSTB and [$r=0.362, n=30, p=0.049$] for MoCA. However, no correlation was found between the level of calcium intake in dairy products and any of the performed cognitive tests.

To control for the potential confounding effects of age and total calorie intake, linear regression analyses were also employed to predict which of the following nutrients

(saturated fat, vitamin D and calcium) best-predicted performance on the cognitive tests. Analyses revealed that none of these nutrients were significant predictors on any of the performed cognitive tests. This means that neither of these nutrients derived from dairy products only have an impact on any of the cognitive domains measured by these cognitive tests.

4.3.3 Hypothesis (H3): Relationship between total dietary nutrient intake and cognitive performance

Bivariate Pearson–Product Moment correlations were computed between nutirents from participants' total dietary intake and their performance on cognitive tests. Please refer to Appendix F for all of the total dietary nutrients correlation results. There was a positive correlation between vitamin D level and RCFT composite score [*r=0.358, n=32, p=0.044*] and MoCA [*r=0.372, n=32, p=0.036*], with a higher level of vitamin D associated with better scores on RCFT and MoCA. Another important finding was that a higher level of calcium was associated negatively with the Victoria Stroop test (VST). This indicated that a higher level of calcium was associated negatively with performance on the VST *[r=-0.457, n=32, p=0.009]*. Higher calcium levels enhanced executive function and selective attention as better performance on this test is associated with less time taken to complete the task. No other meaningful associations were found between any of the other nutrients in total diet and any of the cognitive tests outcome measures.

Linear regression analyses were also employed to determine whether saturated fat, vitamin D or calcium levels predicted performance on the cognitive tests, taking into account both age and total calories from the total diet as confounding variables. Analyses revealed that both calcium and vitamin D levels were significant predictors for the digit span test (DST); both forward and backward as evidenced by having adjusted R^2=19.9%, F=2.545, p= .053. Also, analyses revealed that saturated fat was a significant predictor for MoCA performance as evidenced by having adjusted R^2=32.5%, F=3.978, p=.008. In addition, calcium level was found to be a significant predictor for performance on Victoria Stroop test performance as evidenced by having adjusted R^2=22.5%, F=2.801, p=.037.

The justification for inclusion of the variables entered into the linear regression analyses for both dairy and total dietary intake was empirically based on attainment of significant correlations between the dependent measures of interest and each of the predictor variables. Thus, TMT, RCFT; both copy and composite scores, VST, DST; both forward and backward and MoCA were regressed for calcium, vitamin D and saturated fat, taking into account age, total calories from total dietary intake as cofounding variables. Also the chosen nutrients were based on the fact that vitamin D, calcium and saturated are considered the most abundant nutrients found in dairy products. Justification for choosing age as a confounding variable was because age typically has an effect on cognitive test performance (Meyers & Meyers, 1995; Tombaugh, 2004; Regards, 1981; Gregoire & Van der Linden, 1997). It is also important to control for total calories because according to Willett, most nutrients are associated with total energy intake either because they contribute directly to energy intake or because individuals who consume more total energy also eat, on average, more of all specific nutrients and, as a result, it is important to control for the confounding variable total calorie intake (Willett, Howe & Kushi, 1997).

4.4 Adequacy of Individuals Total Dietary Intake

Dietary data showed that 100% of the participants met the Acceptable Macronutrient Distribution Range (AMDR) recommendation for protein, 60% of participants met the AMDR for fat and 54% of the participants met AMDR for carbohydrates. In addition, the dietary data indicated that 100% of the participants were over the recommended intake of saturated fat (<7%), whereas 100% were within the recommended amount of dietary intake for poly fat (<10%). For cholesterol, 56.25% of the participants reported having their cholesterol level within the recommended amount, which is <300mg. As for the micronutrient level of vitamin D and calcium, participants in general were well below the recommended amount of dietary intake according to the dietary reference intake guidelines for vitamins and minerals with 100% and 84.4% for vitamin D and calcium, respectively. Table 9 presents the adequacy of dietary intake of macronutrients (carbohydrates, fat and protein), Polyunsaturated fatty acids and major vitamins and

minerals. In general, the micronutrients of vitamin and minerals levels were below the
Dietary Reference Allowances (RDA).

Table 9: *Adequacy of dietary intake*

Nutrients	Recommendations	Within	Below	Over
Carbohydrates	45-65%	53.1%	46.9%	
Protein	10-35%	100%		
Fat	20-35%	59.4%		40.6%
Saturated Fat	<7%			100%
Monounsaturated Fat	10-15%	3.1%	96.9%	
Polyunsaturated Fat	<10%	100%		
Cholesterol	<300mg		53.1%	46.9%
Poly Fatty Acids				
Omega-3 Fatty Acids	M= 1.6g/day F=1.1g/day	3.1%	62.5%	34.4%
Omega-6 Fatty Acids	M=14g/day F=11g/day		90.6%	9.4%
Ratio 6/3 2:1			6.25%	93.8%
Vitamins				
Vitamin B12	2.4 mcg/day		53.1%	46.9%
Vitamin D	51-70 years 15mcg/day >70 years 20mcg/day		100%	
Vitamin A	M=900mcg/day F=700mcg/day		71.9%	28.1%
Folate	400 mcg/day		87.5%	12.5%
Vitamin B2 (Riboflavin)	M=1.3 mg/day F=1.1mg/day	3.1%	37.5%	59.4%
Minerals				
Calcium	M=51-70 years 1000mg/day F=51-70 years 1200mg/day >70 years (M and W) 1200mg/day		84.4%	15.6%
Sodium	51-70 years (M and F)= 1300mg/day >70 years= 1200mg/day		3.1%	96.9%
Iron	8mg/day		15.6%	84.4%

Magnesium	M=420mg/day	96.9%	3.1%
	F=320mg/day		
Phosphorus	700mg/day	56.3%	43.8%
Potassium	4700mg/day	96.9%	3.1%
Zinc	M=11mg/day	93.8%	6.25%
	F=8mg/day		

5 Discussion

The purpose of this study was twofold: first, to examine whether there is a difference in cognitive performance between individuals who consume the recommended amount of dairy products (3 servings per day) and individuals who consume less than the recommended amount (<3 servings per day); second, to investigate whether there were associations between levels of dietary nutrients and performance on a battery of cognitive tests. The final sample consisted of older adults (8 males, and 24 females) with a mean age of 70.59 ±7.07 years. Most had at least a high school diploma. The mean participants' body mass index (BMI) was 27.59kg/m^2, which, according to the World Health Organization (2000), is considered a normal healthy body mass index for an older adult aged 60 years and over (WHO, 2000; Health Canada, 2003). Most of the participants had some form of metabolic syndrome, which is defined as a clustering of cardiometabolic risk factors such as abdominal obesity (high waist circumference for males >=102 cm (40inch) and for females >=88cm (35inch)) (National Institute of Health, 1998), dyslipidemia (high blood cholesterol), hypertension (high blood pressure) and diabetes (high blood glucose). Some participants had either one or a combination of these medical conditions. About half (14) of the participants reported having hypertension, 3 reported having diabetes, 8 reported having dyslipidemia and 2 reported having high waist circumference. These cardiometabolic risk factors may contribute to a 1.7 fold increase in the risk of developing cardiovascular diseases (CVD), which are considered risk factors for cognitive decline (Crichton, Bryan, Buckley, &Murphy, 2010). According to Whitmer et al. (2005), the risk of dementia increases from 1.27 for having one risk factor to 2.37 for having all four cardiovascular risk factors (Whitmer et al., 2005). Many (22) of the participants (68.7%) were found to consume less than the recommended amount of dairy products (3 servings per day) according to Canada's Food Guide (CFG) for adults aged 51 + years and older. This is consistent with a 2004 Canadian national nutritional survey finding that the average intake of those aged 19 years or older was 1.5 serving of dairy products per day (Garriguet, 2007). This low consumption pattern of dairy products is consistent with intakes in other parts of the world. For example, according to the most recent Australian national nutritional survey, the average Australian adult consumes between 1 and 1.5 servings of dairy products per day (Australian Bureau of Statistics,

1998) while in the United Kingdom, milk consumption has fallen by 33% over the past 25 years (Elwood, 2005). The potential implications of the present research study are now discussed.

5.1 Effect of Number of Servings of Dairy Products on Cognitive Performance

With respect to the first hypothesis, there were no significant differences on mean cognitive test scores between the group that consumed less than 3 servings of dairy products per day versus the group that consumed 3 or more servings per day. This finding of the present research study is incongruent with the study conducted by Crichton who reported that participants who consumed at least one dairy product serving per day had significantly higher scores on multiple domains of cognitive function compared with those who never or rarely consumed dairy products (Crichton, Elias, Dore & Robbins, 2012). In another study, Crichton reported that participants who consumed 4 servings of dairy products per day had better performance on spatial working memory following six months compared to individuals who consumed 1 serving per day (Crichton, Murphy, Howe, Buckley & Bryan, 2012). According to Crichton and colleagues, this means that consuming at least one serving of dairy products per day can have an impact on cognitive performance when compared to non-consumers or that impact on cognitive performance was observed when comparing higher amounts (4 servings) to lower amounts (1serving) of dairy products per day. The difference in cut-off values may be one reason for the difference between the present study and those of Crichton. Another reason may be the fact that, in the present study, there were only 10 participants who consumed the recommended amount of dairy products per day compared to 22 participants who did not. Also, the insignificant result in the present study could be due to the fact we did not control for any demographic, lifestyle, other dietary consumption, supplement use and cardiovascular factors. Such factors should potentially be considered for future research examining the impact that dairy products might have on cognitive function.

5.2 Relationship between Dairy Product/Total Dietary Intake Nutrient Levels and Cognitive Performance

With respect to the second and third hypotheses, several associations were found between the levels of nutrients found in dairy products/ total dietary intake and older adults' performance on the cognitive tests.

5.2.1 Vitamin D and Cognitive Function

The level of vitamin D in dairy products and in the total diet was associated with higher scores on the Montreal Cognitive Assessment test (MoCA). This is congruent with Rondanelli et al. (2007) who reported that lower dietary intake of vitamin D from total diet was associated with lower scores on the MoCA (Rondanelli et al., 2007). In addition, the present study is the first to find an association between vitamin D level in dairy products only and performance on MoCA. This suggests that further research is needed to support the present study finding. Also, the present study showed that the level of vitamin D was associated with performance on the digit span backward test. This is congruent with Andreeva (2014) who reported that midlife vitamin D intake was positively associated with scores on the forward digit span test (Andreeva et al. 2014). Although the present study results found an association with the backward version as opposed to the forward version, this shows a promising result of the possible beneficial impact that dairy products' level of vitamin D might have on working memory; specifically, the storage and executive processing components of the working memory. However, after controlling for total calories and age, that both vitamin D and calcium from total diet (in combination) were significant predictors on both Forward and Backward digit span test. The present study found a positive association between the level of vitamin D contained in dairy and the number of correctly recalled elements immediately (for the immediate recall), after 30 minutes (for the delay recall) and also correctly recognized elements (for the recognition trial) on the Rey Complex Figure Test and Recognition Trial (RCFT). The study also found an association between total dietary intake of vitamin D and RCFT performance. This is considered the first study to examine the level of vitamin D contained in dairy products only and/or from total diet and performance on RCFT performance, which suggests that vitamin D might have an impact on visual memory as

well. In summary, the present study indicates that the level of vitamin D contained in the diet of older adults might have a positive effect on global cognitive function as measured by MoCA, working memory as measured by digit span backward test and visual memory as measured by RCFT.

Currently, little is known about the exact mechanisms by which vitamin D can affect cognitive performance. However, vitamin D is a fat-soluble vitamin and as a result, can cross the blood brain barrier and can bind to vitamin D receptors located in the human cortex and hippocampus, key areas for cognition (Kalueff & Tuohimaa, 2007). These receptors, along with the enzyme 1 alpha hydroxylase are responsible for transforming the inactive form of vitamin D to the active form in the brain region, which are widespread in both neuron and glial cells within the brain (Kalueff & Tuohimaa, 2007). The neuro-protective effect of vitamin D is associated with a reduction in calcium ions in the brain as a high level of calcium is associated with neurotoxicity and consequently neuronal death (Shinpo et al., 2000; Kalueff, Eremn, & Tuohimaa, 2004). This could occur via two mechanisms: first, vitamin D could stimulate the expression of calcium binding proteins or inhibit the expression of calcium channels in the hippocampus (Losel & Wehling, 2003). Both mechanisms protect neurons against toxic damage by reducing calcium levels in the neuron cell (Losel & Wehling, 2003). The second mechanism by which vitamin D might impart its effects on the nervous system is through its role in autoimmune processes. Vitamin D acts as an inducer of anti-inflammatory neurotrophin proteins such as nerve growth factors and decreases the pro-inflammatory proteins such as tumor necrosis factor (Garcion et al., 2002; Garcion et al., 2003). For example, the induction of the neurotrophin such as nerve growth factor decreases the degenerative processes in hippocampus in rats during aging and the progression of Alzheimer disease (Brown, Dusso & Slatopolsky, 1999; Langub, Herman, Maluche & Koszewski, 2001). This suggests the importance of the anti-neurodegenerative activity of vitamin D and that it might be the mechanism by which vitamin D has an effect on cognitive function (Kalueff & Tuohimaa, 2007).

5.2.2 Calcium and Cognitive Function

No bivariate correlations were found between the level of calcium in dairy products and performance on any of the cognitive tests. This is surprising as calcium is considered the most abundant nutrient in dairy products with 53% of calcium in Canadian's diets contributed by the consumption of dairy products (Agriculture and Agri-Food Canada, 2009). However, after controlling for age and total caloric intake the regression analyses showed an association between the total dietary intake of calcium and performance on the Victoria Stroop Test (VST), with a higher calcium intake was associated with better performance. This indicates that calcium levels might have an effect on executive functioning and selective attention as measured by VST efficiency score. In addition and after controlling for total calories and age, that both vitamin D and calcium from total diet (in combination) were significant predictors on both Forward and Backward digit span test. These finding is supported by numerous studies used to investigate whether there was an association between dietary calcium and performance on cognitive tests. Velho et al. (2008) found that a diet high in calcium was associated with improved cognitive function as measured by the Mini-Mental Status Examination (MMSE). Another study conducted by Lee et al. (2001) showed that a low intake of calcium and dairy products was associated with lower scores on global cognitive function as measured by the MMSE. Ozawa et al. (2012) revealed that a higher self-reported dietary intake of potassium, calcium and magnesium was associated with a lower incidence of all-cause dementia, especially Alzheimer's disease and vascular dementia. This is supported by another prospective, longitudinal study in Japan on 1774 non-demented subjects. Dietary intake was assessed via self-administered questionnaire, while cognitive function was assessed via Cognitive Ability Screening Instrument (CASI) and cognitive questionnaires. Results showed that increased blood pressure, a risk factor for cardiovascular and cognitive decline later in life, as well as lower milk intake in midlife, was associated with Vascular dementia (VaD) detected 25 to 30 years later (Yamada, Kasagi, Sasaki, Masunari, Mimori, & Suzuki, 2003). Consuming more than the recommended dietary intake of calcium does not lead to neuronal death due to toxicity (because this excess dietary consumption will not be absorbed from the intestine), but is rather excreted in the feces (Thompson, Manore, & Sheeshka, 2007). This indicates that

low cognitive performance could only be due to low dietary intake of calcium as opposed to an excess amount. This low consumption is due to many factors: first, as adults get older, their capacity to absorb calcium from the intestine decreases. Second, unbalanced eating habits due to health issues, along with a higher need for calcium than can be obtained from balanced diet, will promote individuals to consume calcium supplements to fulfill their needs.

Little is known about the exact mechanisms by which dietary calcium has an impact on cognitive function. However, it has been suggested that the primary mechanisms by which dietary calcium imparts its effects on cognitive function is indirectly via mediating effects on cardiometabolic health. Hypertension has been recognized as a significant risk factor for cardiovascular diseases, stroke and vascular dementia (VaD) (Sharp et al., 2011). It has been reported that dietary calcium has favorable effects against cardiovascular diseases through the antihypertensive properties of this mineral (Houston & Harper, 2008;Nakamura et al, 1995; McCarron & Reusser, 2002), improvement of dyslipidemia (Karanja et al., 1987) and lower rates of obesity (Astrup, Chaput, Gilbert, & Lorenzen, 2010). According to Resnick, a reduction in dietary calcium may result in calcium depletion from all membrane storage sites, resulting in less stability of the vascular smooth muscle cell membrane (Resnick, 1991; Resnick, 1992). However, when there is an optimal concentration of dietary calcium, it stabilizes vascular cell membrane, inhibits its own entry into cells, and reduces vasoconstriction, reducing blood pressure (BP) (Undurti, 2001). Calcium has been found to have a particular benefit on serum lipid profiles, with increased levels of calcium, increasing HDL-cholesterol while decreasing both total and LDL-cholesterol (Van Meijl, Vrolix & Mensink, 2008). A possible mechanism by which calcium may affect lipoprotein metabolism is through the inhibition of fat absorption in the small intestine (Denke, Fox & Schulte, 1993). In addition, dietary calcium has been associated with lower rates of obesity, which results in weight reduction through an increase in satiety due to consumption of dairy proteins (Astrup et al., 2010) and to the impact of calcium on fat excretion (Astrup et al., 2010). According to Astrup et al. (2010), increasing dairy calcium consumption by 1200mg/day resulted in increased fecal fat excretion by 5.2g/day as calcium forms insoluble fatty acids soaps and other

hydrophobic aggregations of bile acids, phosphorous, and fatty acids in the small intestine, resulting in greater fat excretion (Astrup et al., 2010).

5.2.3 Saturated Fat and Cognitive Function

The level of saturated fatty acid in dairy products was associated negatively with RCFT performance as measured by the composite score. A higher amount of saturated fat contained in dairy products was associated with fewer items remembered immediately (immediate recall), after 30 minutes (delayed recall) and recognized (recognition trial). This indicates that saturated fatty acids might have an impact on the visual memory domain. This finding is supported by a wealth of research (Laitinen et al. 2006; Morris et al., 2004; Eskelinen et al., 2008; Kalmijn et al., 2004; Vercambre et al., 2009; Ortega, et al., 1997; Crichton, et al., 2010; Morris et al., 2006). These research studies suggest that high intake of saturated fatty acids from total dietary intake was associated with poorer global function and worse scores on multiple cognitive domains (Eskelinen et al., 2008; Kalmijn et al., 2004; Vercambre et al., 2009; Ortega et al., 1997; Crichton, et al., 2010; Almeida, Norman, Hankey, Jamrozik, & Flicker, 2006; Gu, Nieves, Stern, Luchsinger, & Scarmeas, 2010). There is also a plethora of research studies that have examined the effect of saturated fatty acids from dairy products on cognitive function. Studies have shown that moderate to high levels of saturated fatty acids from dairy products and spreads were associated with poorer global cognitive function and prospective memory (Laitinen et al., 2006), and cognitive decline (Morris et al., 2004; Solfrizzi et al., 2008). This is supported by the present study and after controlling for age and total calories from total diet, saturated fat was found to be a significant predictor on MoCA; with higher level of saturated fat associated with poorer global cognitive function. The potential mechanisms by which saturated fatty acids affect cognitive functioning in the elderly cannot be derived from the present study. However, there are findings from other studies that suggest increasing level of SFS lead to increases in low-density lipoprotein, which is considered a risk factor for dyslipidemia, a component of metabolic syndrome and a risk factor of cardiovascular diseases (NIH, 2002;Huth & Park, 2012), and, therefore for cognitive decline, VaD and AD (Solfrizzi et al., 2005). A diet rich in SFA is deleterious and positively associated with cardiometabolic risk factors such as diabetes,

hypertension, dyslipidemia, and obesity (components of metabolic syndrome), which are also risk factors for cognitive decline, dementia and lower scores on tests of cognitive functions through white matter lesions, which are critical for cognition (Breteler, 1994). Further, in experimental studies on rats, a Western diet with 40% saturated fatty acids and 1% cholesterol may increase hippocampal accumulation of amyloid beta-peptide (AB), a pathogenic marker of Alzheimer's disease (Oksman et al., 2006). In addition, a diet high in SFA may lead to cognitive impairment through alteration of the fatty acids "phosphatidylcholine" profile in the brain (Greenwood & Winocur, 1996) and cerebral oxidative stress or neural inflammation via increased level of reactive oxygen species (ROS) (Zhang, Dong, Ren, Driscoll, & Culver, 2005). Also, SFA has decreased the levels of hippocampal brain-derived neurotrophic factor (BDNF), which is relevant for neuronal plasticity, and the maintenance of cognitive function (Molteni, Barnard, Ying, Roberts, & Gomez-Pinilla, 2002). According to that study (on rats), SFAs decreased the level of neurotrophins such as brain–derived neurotrophic factor (BDNF), which is important for neurotransmitter release, learning and memory in the hippocampus and thus low performance on spatial learning memory tasks. This indicates that an individual should practice caution when consuming saturated fat as it had a negative impact on cognitive function.

5.3 Limitations

There are several limitations associated with the present study that should be acknowledged. First, the independent sample t- tests did not reach statistical significance between the less than 3 serving and greater than 3 servings of dairy products per day on any mean cognitive test scores. The inability to detect a significant difference could be attributed to the relatively small sample size in general and specifically the number of people who consumed the recommended amount of dairy products. Second, the sample size may not be representative of the general older adults population and this would affect external validity (generalizability). The current sample was predominately highly educated with at least high school diploma and was therefore, homogenous. The majority were white females who did not smoke and only consumed small to moderate amounts of alcohol. Therefore, conclusions drawn from the current study cannot be generalized to

other groups. These associations need to be replicated in a larger population sample. Also, participants who are interested or concerned about their health may be more likely to participate in health research, and subsequent reporting bias may therefore exist. Third, residual confounding variables by unknown risk factors, such as gender, education, number of supplements, number of medications, body mass index (BMI), depression status and genetics may play an important role in altering the outcomes. It was not possible to control for these because of the limited sample size. As a result, further research into the area should, where possible, control for as many of these variables as possible and will need rather a large sample to do so. Fourth, the sample included more women (24 out of 32 participants), which likely stems from the fact that women's life expectancy is higher than men's and the risk of dementia increases sharply with age (World Health Organization, National Institute of Aging & National Institute of Health, 2011). Thus, recruiting an equal numbers of males and females could result in significant results on cognitive test between both genders. Fifth, the current study is a cross-sectional analysis and cannot address cause-and-effect relationships, namely cognitive performance affecting dietary intake or dietary intake affecting cognitive performance. Thus, longitudinal study and specifically intervention studies would be beneficial to provide a clearer profile of causality and underlying mechanisms of the role of specific nutrients on cognitive function.

Sixth, the dietary measurement method is based on self-reported data, which could be problematic in older adults due to their cognitive abilities, or underestimation or overestimation of true associations. Furthermore, measurement at only one point in time may not reflect long-term consumption patterns. Thus, administering a variety of dietary assessment methods as well as at different time points would act as a valid method. Seventh, the neuropsychological test battery was not as comprehensive (measuring every cognitive domain) as it could have been due to time and expertise limitations. Thus, including more neuropsychological tests would be beneficial to measure every aspect of cognitive function. Finally, no blood sample was collected and having only one measure (dietary intake measure) might not be of accurate representative of the nutrient status of an individual. Thus, biochemical assessments based on blood or tissue concentrations will reflect the nutrient life as some nutrients such as fatty acids have long life compared

to others. As a result, a combination of dietary, biochemical, clinical, and anthropometric methods can be regarded as the "gold standard" in estimating individual nutritional status. In addition, study participants had a wide age range and, as a result, the observed association between nutrient levels and task performance were somewhat overshadowed by the wide age range. Thus, selecting a narrower age range would help to omit the effects of particular age on task performance will help better identify the precise role of nutrients during the aging process. While this would make the process of recruitment significantly more time consuming and may reduce the sample size even more, it would help construct a set of age ranges by which cognitive function may be more susceptible to level of nutrients.

5.4 Strengths

Firstly, diet and cognitive function were assessed using previously validated dietary and neuropsychological instruments that have been widely used in both cross-sectional and epidemiological studies. These cognitive tests have been used in other studies investigating dairy products (response to the dairy products) and, as a result, they are sensitive and reliable to the measured cognitive domain. Second, most of the study participants were recruited via face-to-face and it was the most effective means of getting a higher number of participants. The present study is in line with Resnick et al. (2003) who reported that face-to-face contact was the most effective recruitment method for older women because it facilitated easier communication. Third, the neuropsychological test battery included both paper and computer-based cognitive tests. This ensures there was a variety to avoid any boredom or inconvenience caused by using the computers only and in case older adults were not familiar with computer usage. Finally, the five-day food intake records were able to capture participants' food intake. This is evidenced by the fact that the participants did not appear to change their eating patterns by reducing the number of foods and snacks consumed or decreasing the complexity of their diet by substituting food that is simpler to record. Keeping food intake records can result in significant changes in eating patterns, which could affect the ability of the researcher to capture an accurate picture of usual dietary intake. To minimize changes in eating patterns, it is suggested that food intake records should avoid lengthy periods of consecutive days (e.g.

7-day food intake records) (Block, 1989; Feskanich et al., 1993; Rebro, Patterson, Kristal, & Cheney, 1998).

5.5 Conclusion

The results of the present study revealed that nutrient levels from dietary intake were uniquely associated with specific cognitive functions in older men and women. Although the explained variance in visual memory and processing speed and flexibility was modest, it is important to remember that participants in this study were already functioning at a high level. It is suggested that future research about the effect of dairy products on cognitive performance should be investigated with a wider range of participants. Prevention strategies are needed to impede an increased prevalence of cognitive dysfunction. Intervening to prevent cognitive decline and impairment can help prevent adverse health outcomes such as dementia. Intervention through simple steps such as dietary intake could have important implications for health care and the economic system as dementia costs the North American care system billions of dollars in direct and indirect costs. These results contribute to increased evidence indicating that nutrient intake could influence cognitive function in the older adult population.

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Appendices

Appendix A: Letter of Information

Research Title: Relationship between Dairy product consumption and cognitive performance in a group of community-dwelling healthy older adults living in London, Ontario, Canada

Members of the Research Team:
Dr. Alan Salmoni (principle investigator), Health Sciences, School of Kinesiology, Western University, 519 661 3541 x 83541
Mariam Ismail (Master student), Health Sciences, School of Kinesiology, Western University, 519 870 2102

Background:
I am a master student at Western University in the school of kinesiology conducting a study on the effect of dairy product consumption on cognitive performance in community-dwelling healthy older adults living in London, Ontario, Canada. I would like to invite you to participate in this research study to investigate the potential relationship of consuming dairy products on cognitive function. The purpose of this letter is to provide you with the information needed to make an informed decision about participating in this research study. Please read the following information carefully and do not hesitate to ask the research team any questions to clarify any points or phrases. If after reading the below information and you wish to participate in this research, please sign the consent form provided at the end of the package.

Why should you take part in this study? Why are we doing this study?
You are being invited to partake in this research study because you are an older adult living in the community. The purpose of this research study is to investigate the potential effect of consuming dairy products on cognitive performance in community-dwelling healthy older adults living in London, Ontario, Canada.

Why is this study being done?
As the proportion of adults aged 60 years and older is expected to increase rapidly in the near future, it has become important to better understand the cognitive changes that accompany normal aging. With age, many older adults experience changes in their thinking abilities, for instance, they face difficulties in mathematical calculations, attention-requiring activities, and memory retention. In response to the increasing aging population, and the great societal impact of age-related cognitive decline combined with the absence of curative treatment, there has been a tremendous amount of research on the development of effective dietary interventions that may help ameliorate age-related cognitive decline and diet is one of the modifiable factors. The association between diet and cognitive function has been a topic of interest over the past two decades. Dietary habit is a modifiable factor that can be targeted as an appropriate intervention to optimize

the cognitive function of an aging person. There are positive associations between cognitive function and a number of nutrients, including antioxidants (vitamin C, and E), omega 3 and omega 6 polyunsaturated fatty acids, fruits and vegetables, and vitamin B complex (vitamin B6 and B12, and folate). However little research has been paid to the potential impact dairy products have on cognitive function in comparison to other nutrients. As a result, it is time to investigate the potential relationship of dairy products have on cognitive performance.

Who is eligible for this study?
If you are 65 years of age or older, have the ability to understand and read English, community-dweller and live independently at home, then you are eligible to participate in this study. If your age is less than 65 years of age, inability to understand or read English, live in an assisted living or long term care facility, have significant visual or hearing impairment uncorrected by aids, then you will be excluded.

What is being asked of you as a participant?

If you agree to take part in this research study, the procedures and visits you can expect will include the following: you will be asked to take part in two visits that will take approximately 2 hours in total of your time; 1-hour for each visit at a previously arranged and agreed upon location and time between the researcher student and the participating participant. These two visits will take part place over a 1-week period.

If you are eligible and willing to participate in the study, you will be scheduled for an **initial (first study visit1) of 1-hour**. In this visit: The researcher student will review the informed consent form with you and answer any questions you have about the study. Next, a short cognition test called the Montreal Cognitive Assessment (MoCA) will be administered by the researcher student. Also, an educational session by the researcher student on how to fill in the food intake record/diary via completing a sample of 1-day food intake record will be practiced to ensure that you can accurately and sufficiently recorded your dietary intake. Also, instructions on how to fill in the Yale Physical Activity Survey (Yale) will be given to you. Then, you will be given the following forms: demographic questionnaire, food intake records (FIRs), and Yale Physical Activity Survey (Yale) to be completed at home. Also, in this visit, time and place for the second visit will be scheduled between the researcher student and the participating participant upon a mutual agreement to conduct the cognitive testing.

You will be asked to fill in the following paper forms at home:
- *Demographic Questionnaire***:** You will be asked to provide basic information about your age, sex, education level, employment status, etc. This will take approximately 3 minutes to complete
- *5-Day Food Intake Record:* This form asks you to record all beverages, foods, and supplements consumed and all medication taken on 4 weekdays and 1 weekend day, and it takes about 12 minutes to complete per day.
- *The Yale Physical Activity Questionnaire (Yale):* You will be asked to complete a short questionnaire to determine your current physical activity level, which will take about 10 minutes.

Visit2: (second and final study visit of 1-hour), you will return the following week for 1- hour visit. In this visit, the participants will be given a copy of informed consent for their own records. Participants will also submit their completed demographic questionnaire, food intake record/diary and Yale and will also be checked by the researcher student for completion and clarity. Next cognitive testing will be administered by the researcher student, which will take approximately 30 minutes to complete both the actual tests and the practice session associated with each test. These tests will run in the following sequence-Victoria Stroop test, Trial Making test (part A and part B) and the digit span test (forward and backward) by using a personal computer. After each test, the participants will be asked whether they would like to take break or not and the opportunity to ask any questions. On completion of the protocol, you will be thanked for your time and if you would like to receive a research report summary of the study findings.

In this visit, you will be asked to sit in front of a laptop computer screen to do the cognitive tasks in the following sequence: Victoria stroop test, Trial Making Test (part A +Part B) and the Digit span test (forward and backward)
Below is a brief description about what you will encounter when you do each task:

- *Victoria Stroop test***:** before the test begins, you will do a brief practice session to familiarize yourself with the test. In this test, you will press the key buttons {1,2, 3 and 4} that corresponds to each of the following color {red, blue, yellow and green}. Next, the actual test will begin which will ask to press the correct key button that correspond to the color of the stimuli that you will see on the screen. For example, if the color of the stimuli is red, then, you have to press 1 that corresponds to the color of the stimuli. This test will take approximately 10 minutes for both the practice session and the actual test to complete.
- *Trial Making Test (part A+ Part B)*: before the test begins, you will do a brief practice session to familiarize yourself with the test. In this test, you will have to connect 26 blue circles "dots" arranged in a random order in the correct sequence. The test usually begins by clicking circle 1. For part A of this test, you have to connect numbers from 1-26 in the correct order. For example, 1-2-3-4 -----26. For part B, you will have to click on the circles in an alphanumeric order. For example,1-A-2-B-3-C and so on. If you click on the correct circle, a line will be drawn and the same process goes on until you connect the 26 dots. However, if you click on the wrong circle, no line will be drawn until you click on the correct one. This test will take approximately 7 minutes to complete both parts
- *Digit Span test (forward and backward):* before the actual test begins, the researcher student will perform a practice session on a piece of paper with you. In the forward version of this test, you will hear and visually see 3 numbers on the screen and you have to type in these numbers in the order you saw them. For example, if you see 123 on the screen, you have to type in 123 by using the keyboard. For backward version of this test, you will be asked however, to type in these numbers in the reverse order 321. If you answer correct, you will move on to the next random numbers of 4 digits. This test will take approximately 8 minutes to complete both versions.

Will being in this study help you in any way?
We do not anticipate direct benefits to you from taking part in this research study. We hope that the information learned from this study can be used in the future to benefit other older adults residing in community. By doing this research, society will uncover the importance of consuming dairy products in relation to cognitive function. Also, it will help health professionals, especially dietitians to encourage and identify strategies to improve the consumption of dairy products in this population group in order to promote healthy brain functions.

Participation:
Participation in this study is completely voluntary. The participant may refuse to participate and withdraw from the study at anytime with no consequences in the future. Also, there will be few questions that are of personal nature and may not be comfortable for the participant to answer. You are free to refuse to answer these questions. If you should choose to withdraw from the study, the data collected from your visits will be discarded. You do not waive any legal rights by singing the consent form.

Benefits and risks:
Participants will not be paid for taking part in this research study. There will be no direct cost to the participant for being involved in this research study. As researchers, we do not anticipate any personal risk from participant participation in this study, but if you feel uncomfortable or upset at any time, please notify the interviewer. Your participation in the study is highly appreciated and it will help the research team gain and share knowledge in both academia and professional fields as well as benefit other older adults.

Privacy and Confidentiality:
Maintaining participant research records is our most important priority. However, we cannot guarantee an absolute confidentiality because your personal information may be disclosed if it is required by law. As research investigators, we have taken the following measures to keep your information and identity secure. Your name will be coded with a participant identification number throughout the study; your records will be locked in a cabinet in a secure office at the university for 5 years, no information that discloses your identity will be released without your specific consent to the disclosure when results reported in meetings, presentations, or publications. We will keep all study material for at least 5 years after the study results have been published and will be destroyed at the end of that time. Participants have the right to withdraw from the research study at any time. Also, if you chose to leave the research study, you may choose to have your collected information excluded from the study. However, if the participant completed the study and the results are published in a scientific journal, the research investigators cannot allow your data to be withdrawn at a later date. If the research study results have been published, none of your personal information will be identified at any point. This measure has been taken to ensure that no one has the ability to tell whether you have participated in this research study or not.

Contact Information:
If you have any questions about your rights, as a participant, you may contact: the office of Research Ethics at Western University, 519 661 3036.

If you have any further questions or concerns about this research, please feel free to contact us at the principle investigator: Dr. Alan Slamoni at 519 661 2111 ext. 83541 OR Mariam Ismail at 519 870 2102

Thank you in advance for considering participation in this study! This letter is yours to keep for future reference.

Appendix B: Letter of Informed Consent
Relationship between dairy product consumption and cognitive performance in a group of community-dwelling healthy older adults living in London, Ontario, Canada

I have read the letter of information; have had the ability to understand the nature of the research study. The research study has been explained to me and I agree to participate in this study. All questions have been answered to my satisfaction.

I consent to participate in this research study as outlined in the letter of information
Initial----------------------------
Name of participant (please print)-----------------------------------
Signature------------------------------
Date--

Pearson obtaining the consent form (please print)---------------------------------
Initial----------------------
Signature-------------------
Date----------------------

Appendix C: Human Ethics Approval

Western Research

Western University Health Science Research Ethics Board
HSREB Delegated Initial Approval Notice

Principal Investigator: Dr. Alan Salmoni
Department & Institution: Health Sciences\Kinesiology, Western University

HSREB File Number: 105710
Study Title: The relationship between dairy product consumption and cognitive performance in a group of community-dwelling healthy older adults
Sponsor:

HSREB Initial Approval Date: November 20, 2014
HSREB Expiry Date: August 31, 2015

Documents Approved and/or Received for Information:

Document Name	Comments	Version Date
Other	Daily Food Record	
Recruitment Items	Participant Recruitment Flyer	2014/09/15
Instruments	Demographics Questionnaire (ADDED SEPT 17, 2014)	
Instruments	MoCA	2014/10/17
Instruments	Yale Physical Activity	2014/10/17
Recruitment Items	Script for Verbal Consent (ADDED SEPT 17, 2014)	
Other	References	
Letter of Information & Consent		2014/10/17
Western University Protocol		2014/09/15

The Western University Health Science Research Ethics Board (HSREB) has reviewed and approved the above named study, as of the HSREB Initial Approval Date noted above.

HSREB approval for this study remains valid until the HSREB Expiry Date noted above, conditional to timely submission and acceptance of HSREB Continuing Ethics Review. If an Updated Approval Notice is required prior to the HSREB Expiry Date, the Principal Investigator is responsible for completing and submitting an HSREB Updated Approval Form in a timely fashion.

The Western University HSREB operates in compliance with the Tri-Council Policy Statement Ethical Conduct for Research Involving Humans (TCPS2), the International Conference on Harmonization of Technical Requirements for Registration of Pharmaceuticals for Human Use Guideline for Good Clinical Practice Practices (ICH E6 R1), the Ontario Personal Health Information Protection Act (PHIPA, 2004), Part 4 of the Natural Health Product Regulations, Health Canada Medical Device Regulations and Part C, Division 5, of the Food and Drug Regulations of Health Canada.

Members of the HSREB who are named as Investigators in research studies do not participate in discussions related to, nor vote on such studies when they are presented to the REB.

The HSREB is registered with the U.S. Department of Health & Human Services under the IRB registration number IRB 00000940.

Ethics Officer, on behalf of Dr. Joseph Gilbert, HSREB Chair

Ethics Officer to Contact for Further Information

Erika Basile ebasile@uwo.ca	Grace Kelly grace.kelly@uwo.ca	Mina Mekhail mmekhail@uwo.ca	Vikki Tran vikki.tran@uwo.ca

Western HealthSciences

Kinesiology

Appendix D: Demographic Questionnaire

Demographics Questionnaire   DO NOT put your name on this sheet

Subject Code----------------------   Date------------------------------

Demographics Questionnaire

Please answer the following questions about yourself. This personal information will be held in strict confidence. Only averages from a large group will be reported at the end of data collection period.

1. What is your age? ------------- Years old
2. Weight------------lbs or Kg   Height-------------cm or m
   BMI (weight (kg)/Height (m^2) (calculated by the researcher)
3. Gender: please circle
   o   Males
   o   Females
4. What is your current marital status? Please circle
   o   Single
   o   Married
   o   Common-law
   o   Separated
   o   Divorced
   o   Widowed
   o   Would prefer not to say---------
5. What is the highest level of education you have completed? Please specify Less than elementary school
   o   Elementary
   o   High school
   o   Vocational/technical school
   o   College
   o   University: Bachelor's degree
   o   University: Post-graduate degree
   o   Professional degree (MD, JD, etc.)
   o   Other, please specify------------------
6. What is your current employment status? Please circle
   o   Employed Full-Time
   o   Employed Part-Time
   o   Retired
   o   Would prefer not to say
7. What is your current household income in Canadian dollars?
   o   Under $25,000
   o $25,000-$49,000
   o $50,000-$74,000
   o $75,000-$99,000

- o $100,000-$124,000
- o $125,000-$149,000
- o $150,000 or more
- o Would prefer not to say

8. How many adults are supported by this household income (including yourself)?
    - o 1
    - o 2
    - o 3
    - o 4
    - o 5
    - o 6 or more

9. Do you consume alcohol?
    - o Never, former/current (please circle)
    - o Yes, amount------------

10. Are you smoker- tobacco smoke
    - o Current, if yes how many cigars per day--------------
    - o Past smoker
    - o Non-smoker

11. Do you engage in physical activity
    - o Yes
    - o No

12. Are you taking any supplements (e.g. vitamin D, multivitamin, iron, fish oil, etc.)? Yes, No (please circle). If so what kind------------- and why are you taking it (deficiency, just an extra, other)--------------- Please specify

13. Have you ever been told by a doctor that you have (please circle all that apply)
    - o Alzheimer disease,
    - o Dementia
    - o Memory problems
    - o Hypertension (High blood pressure)
    - o Dyslipidemia (High blood cholesterol)
    - o Diabetes (High blood glucose)
    - o Obesity (High waist circumference)
    - o Depression
    - o Anxiety
    - o Stress
    - o Physical disability such as vision or hearing problem
    - o Other, please specify---------------------

14. Please list all of the medications that you are taking to treat the above-mentioned medical conditions (For example, for hypertension; Thiazide, for diabetes, Metformine, etc. or any other medical conditions --
    ----

15. Are you consuming soft drinks?
    - o Yes, how often------------
    - o No

16. How would you rate your dietary pattern of consuming other food groups (fruits and vegetables, grain and meat)?
    - o Good

- o Bad
- o Moderate

17. Do you engage in extracurricular activities (e.g. volunteering, book clubs, cognitive activities, others, please specify-----------------------------------)
    - o Yes
    - o No

Appendix E: Pearson Product Moment Correlations (Dairy Products Intake)

	RCFT-Composite Score	DSTB	DST (F+B)	MoCA	Stroop
Fat					-.360 (.043)
Saturated fat	-.361 (.042)				
Mono fat					-.408 (.031)
Poly fat					-.494 (.008)**
Cholesterol	-.358 (.044)				
Vitamin D	.367 (.046)	.390 (.033)			
Magnesium			.386 (.042)	.362 (.049)	
Phosphorous				.470 (.012)	
Potassium		.370 (.041)		.376 (.042)	
Zinc				.402 (.034)	-.431 (.022)
Vitamin A		.390 (.027)		.400 (.023)	
Vitamin B2		.378 (.039)		.409 (.025)	

Note. Data are presented as Pearson-Product Moment correlation coefficient r (p-value)
** Correlation is significant at the 0.01 level (2-tailed)
* Correlation is significant at the 0.05 level (2-tailed)

Appendix F: Pearson Product Moment Correlations (Total Dietary Intake)

	RCFT-Composite Score	MoCA	Stroop
Poly fat			-.412 (.019)
Vitamin D	.358 (.044)	.372 (.036)	
Calcium			-.457 (.009)**
Magnesium		.507 (.003)**	
Phosphorus			-.370 (.037)
Potassium		.430 (.014)	

Note. Data are presented as Pearson-Product Moment correlation coefficient r (p-value)
** Correlation is significant at the 0.01 level (2-tailed)
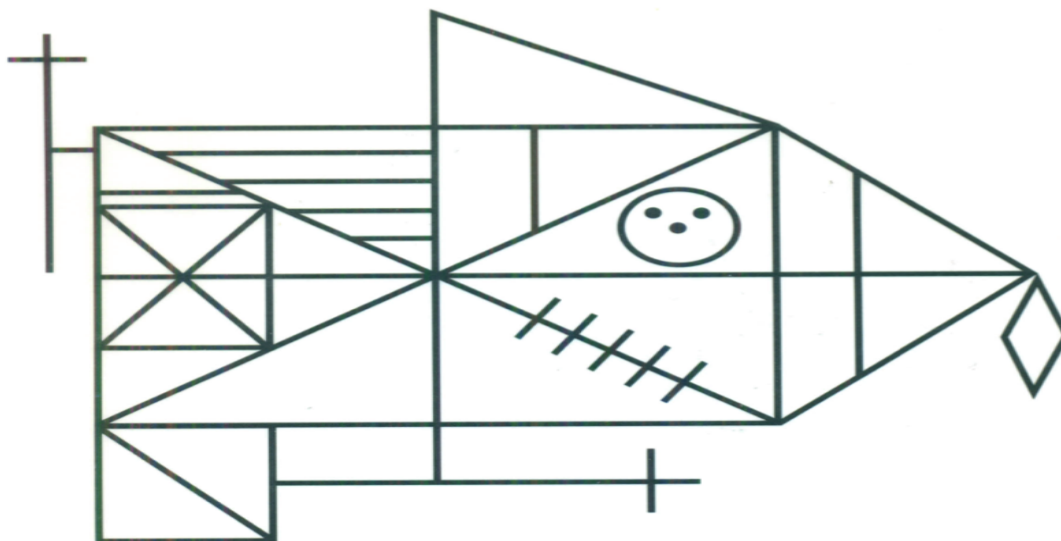* Correlation is significant at the 0.05 level (2-tailed)

Appendix G: Rey Complex Figure Test and Recognition Trial (RCFT)

Meyers, J., & Meyers, K. (1995). *Rey Complex Figure Test and Recognition Trial Professional Manual:* Psychological Assessment Resource, Inc.

Appendix H: Yale Physical Activity Survey (YPAS)

Part 1			
We are interested to learn about the types of activities which are part of your regular routine. I am going to show you lists of common types of physical activities. Please tell me how much time (in minutes or hours) you spent during the past week.			

Activity	Time		Intensity code
	Hours	**Minutes**	
Work			
Shopping (eg, grocery, clothes)	_____	_____	3.5
Stair climbing while carrying a load.	_____	_____	8.5
Laundry:	_____	_____	
Unloading/ loading machine, hanging, folding only	_____	_____	3.0
Washing clothes by hand	_____	_____	4.0
Light housework: tidying, dusting, sweeping, collecting rubbish in the home, polishing, ironing.	_____	_____	3.0
Heavy housework: vacuuming, mopping, scrubbing floors and walls, moving furniture, boxes or rubbish bins.	_____	_____	4.5
Food preparation: chopping, stirring, moving about to get food items and pans.	_____	_____	2.5
Food service: setting table, carrying food, serving food			2.5
Dish washing: clearing the table, washing / drying dishes, putting dishes away.	_____	_____	2.5
Light home repair. Small appliance repair, light home maintenance / repair.	_____	_____	3.0
Heavy home repair: painting, carpentry, washing/polishing car.	_____	_____	5.5
Other: _____	_____	_____	_____

Yard work	Hours	Minutes	
Gardening, pruning, planting, weeding, digging, hoeing	_____	_____	4.5
Lawn mowing (walking only)	_____	_____	4.5
Clearing walks/driveways: sweeping, shoveling, raking	_____	_____	5.0
Other: _____	_____	_____	_____

Care taking	Hours	Minutes	
Older or disabled person (lifting, pushing wheelchair)	_____	_____	5.5
Child care (lifting, carrying, pushing pram)	_____	_____	4.0

Exercise	Hours	Minutes	
Brisk walking	_____	_____	6.0
Pool exercises, stretching, yoga	_____	_____	3.0
Vigorous calisthenics, aerobics	_____	_____	6.0
Cycling	_____	_____	6.0
Swimming (laps only)	_____	_____	6.0
Other	_____	_____	_____

Recreation	Hours	Minutes	
Leisurely / slow walking	_____	_____	3.5
Needlework: knitting, sewing, needlepoint, etc	_____	_____	1.5
Dancing: line, ballroom, tap, square etc	_____	_____	5.5
Bowling	_____	_____	3.0
Golf	_____	_____	5.0
Racquet sports: tennis, squash	_____	_____	7.0
Billiards	_____	_____	2.5
Other	_____	_____	_____

Part two

I would now like to ask you about certain types of activities that you have done during the past month. I will ask you about how much vigorous activity, leisurely walking, sitting, standing and some other things you usually do.

1. About how many times during the month did you participate in **vigorous** activities, that lasted at least **10 minutes** and caused large increases in breathing, heart rate, or leg fatigue, or caused you to perspire?

Score: Not at all (go to Q3)	0
1-3 times per month	1
1-2 times per week	2
3-4 times per week	3
5 + times per week	4
Refused	7
Don't know	8
Frequency score	_____

2. About how long do you do this vigorous activity/ies each time?

Not applicable	0
10-30 minutes	1
31 – 60 minutes	2
60 + minutes	3
Refused	7
Don't know	8
Duration Score	_____
Weight	5

Vigorous activity index score:
Frequency score _____ X Duration score _____ X weight _____ = _____
(Responses 7 or 8 are scored as missing)

3. Think about the walks you have taken in the past month. About how many times per month did you walk **for at least 10 minutes** or more **without stopping** which was not strenuous enough to cause large increases in breathing, heart rate, or leg fatigue or cause you to perspire?

Score: Not at all (go to Q5)	0
1-3 times per month	1
1-2 times per week	2
3-4 times per week	3
5 + times per week	4
Refused	7
Don't know	8

Frequency score _____

4. When you did this walking, for how many minutes did you do it?

Not applicable	0
10-30 minutes	1
31 – 60 minutes	2
60 + minutes	3
Refused	7
Don't know	8

Duration Score _____

Weight: 4

Leisurely walking index score:

Frequency score _____ X Duration score _____ X weight _____ = _____

(Responses 7 or 8 are scored as missing)

5. About how many hours per day do you spend moving around on your feet while doing things? Please report only the item that you are **actually moving**.

Not at all	0
Less than 1 hour per day	1
1 to 3 hours per day	2
3 to 5 hours per day	3
5 to 7 hours per day	4
7+ hours per day	5
Refused	7
Don't know	8

Moving score _____

Weight 3

Moving Index Score

Moving score _____ X Weight _____ = _____

(Responses 7 or 8 are scored as missing)

6. Think about how much time you spend standing or moving around on your feet on an average day during the past month. About how many how per day do you **stand**?

Not at all	0
Less than 1 hour per day	1
1 to 3 hours per day	2
3 to 5 hours per day	3
5 to 7 hours per day	4
7+ hours per day	5
Refused	7
Don't know	8

Standing score _____
Weight   3

Standing Index Score
Standing score _____ X Weight _____ = _____
(Responses 7 or 8 are scored as missing)

7. About how many hours did you spend sitting on an average day during the past month?

Not at all	0
Less than 3 hours	1
3 hours to less than 6 hours	2
6 hours to less than 8 hours	3
8 + hours	4
Refused	7
Don't Know	8

Sitting Score _____
Weight   1

Sitting Index Score
Sitting score _____ X Weight _____ = _____
(Responses 7 or 8 are scored as missing)

8. About how many flights of stairs do you climb up **each** day? (let 10 steps = 1 flight

_____

9. Please compare the amount of physical activity that you do during other seasons of the year with the amount of activity you just reported for a typical week in the past month. For example, in the summer, do you do more or less activity than what you reported in the past month?

please circle the appropriate score for each season.

	Lot more	Little more	Same	Little less	Lot less	Don't know
Spring	1.30	1.15	1.00	0.85	0.70	.
Summer	1.30	1.15	1.00	0.85	0.70	.
Autumn	1.30	1.15	1.00	0.85	0.70	.
Winter	1.30	1.15	1.00	0.85	0.70	.

Seasonal adjustment score: = sum of all seasons / 4 = _____

DiPietro, L., Caspersen, C.J., Ostfeld, A.M. ,& Nadel, E.R. (1993). A survey for assessing physical activity among older adults. *Medicine and Science in Sports and Exercise, 25*(5), 628-642

Curriculum Vitae

Name: Mariam Ismail

Post-secondary Education and Degrees:
Brescia University College, Western University
London, Ontario, Canada
2008-2012 BSc. (Honours Specialization in Nutrition and Dietetics)

Western University
London, Ontario, Canada
2013-2015 M.A. (Psychological Basis of Kinesiology)

Related Work Experience
Teaching Assistant
Western University
2013-2015