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Humor's Effect on Short-Term Memory in Older Adults: An Innovative Wellness Paradigm

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LOMA LINDA UNIVERSITY
School of Allied Health
in conjunction with the
Faculty of Graduate Studies

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Humor's Effect on Short-Term Memory in Older Adults: An Innovative Wellness Paradigm

by

Gurinder Singh Bains

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A Dissertation submitted in partial satisfaction of
the requirements for the degree
Doctor of Philosophy in Rehabilitation Sciences

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Each person whose signature appears below certifies that this dissertation in his/her opinion is adequate, in scope and quality, as a dissertation for the degree Doctor of Philosophy.

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ABBREVIATIONS

PNI	Psychoneuroimmunology
EEG	Electroencephalography
MMSE	Mini Mental State Exam
RAVLT	Rey Auditory Verbal Learning Test
BMI	Body Mass Index
SD	Standard Deviation
T2DM	Type 2 Diabetes Mellitus
HbA1c	Glycated Hemoglobin
AGEs	Advanced Glycation End Products
ANOVA	Analysis of Variance

ABSTRACT OF THE DISSERTATION

Humor's Effect on Short-Memory in Older Adults: An Innovative Wellness Paradigm
by

Gurinder Singh Bains

Doctor of Philosophy, Graduate Program in Allied Health
Loma Linda University, June 2014
Dr. Lee Berk, Chairperson

Context: With ageing, the detrimental effects of stress can impair the ability to learn and sustain memory. Humor and the associated mirthful laughter can reduce stress by decreasing the hormone cortisol. Chronic release of cortisol can damage hippocampal neurons leading to impairment of learning and memory.

Objectives: To examine the effect of watching a humor video on short term memory in older adults.

Design: A randomized, controlled trial.

Setting: Loma Linda University, Loma Linda, CA.

Participants: 30 subjects: 20 normal healthy, older adults, 11 males and 9 females; 10 Type 2 Diabetic older adults, 6 males and 4 females.

Intervention: Two humor groups, healthy elderly (69.9 ± 3.7 years) and diabetics (67.1 ± 3.8 years), self-selected from 1 of 2 humorous videos (20 minutes) - Red Skeleton comedy or a montage of *America's Funniest Home Videos*. The control group (68.7 ± 5.5 years) did not watch a humor video and sat in quiescence.

Outcome Measures: The standardized neuropsychological memory assessment tool, Rey Auditory Verbal Learning Test was used to assess for 1) learning ability, 2) recall ability,

and 3) visual recognition ability. Salivary cortisol measurements, at 5 time points, were obtained.

Results: In the health elderly, diabetic, and control groups: 1) learning ability improved by 38.5%, 33.4%, and 24.0% respectively (p=.025); 2) delayed recall improved by 43.6%, 48.1%, and 20.3% respectively (p=.064); and 3) visual recognition increased by 12.6%, 16.7%, and 8.3% respectively (p=.321). For salivary cortisol levels, there were 1) borderline and significant changes in the healthy elderly group (p=.047, .046, and .062 respectively); 2) significant changes in the diabetic group (p=.047, .025, and .035 respectively); and 3) no significant changes in the control group.

Conclusion: Our research findings offer potential clinical and rehabilitative benefits that can be applied to whole person elderly wellness programs. The cognitive components, learning ability and delayed recall, become more challenging as we age and are essential to older adults for an improved quality of life: mind, body, and spirit. Although older adults have age-related memory deficits, complimentary, enjoyable, and beneficial humor therapies need to be implemented for these individuals.

CHAPTER ONE

INTRODUCTION

The world's population is increasing at a rapid rate. More notably, the elderly population (age 65 and older) is increasing at a even higher rate[1]. There are numerous age-associated challenges older adults will face due to their longevity. Older individuals are affected and have an increased prevalence of short term memory loss[2,3] and are at a greater risk for developing Type 2 Diabetes Mellitus (T2DM)[4]. The effects of age associated diseases and deficits are taxing not only to the individual but also to family members and society. Owing to this upsurge in life expectancy, a new approach of whole-person, mind-body-spirit care would be of great benefit. Instead of merely emphasizing the cause of disease, a salutogenic health paradigm that embodies overall human wellness and promotion is essential, for older adults as well as the young[5]. Native Americans believed in mind-body healing but this belief had been lost in the recent age of pharmacology. Rehabilitation related to short-term memory loss needs to be addressed as a vital element of whole person wellness for older adults, especially in older adults with T2DM.

Age-associated short-term memory deficiencies are expressed in older adults as physical and behavioral compliance issues. For example, noncompliance can be demonstrated as failure to perform at-home, physical-therapy exercises properly, imprecise timings or dosages of medications, or missed healthcare appointments[6]. Poor memory[7], anxiety, depression, and lack of self-motivation are barriers that need to be

overcome to improve compliance [6,8]. In addition, noncompliance leads to a decrease in personal health and the efficacy of treatment[9]. Intervention strategies should be geared toward whole person wellness for older adults and include precursor therapies and modalities for enhancement of memory or reduction of memory deficits; such as humor therapy.

Research has shown that there are numerous methods that can improve short-term memory in older adults. Studies have shown, through measurable brain variations, that cognitive training can enhance short-term memory [10,11]. Furthermore, improving quality of life, incorporating self-improvement approaches, increasing daily activities, and integrating daytime naps are beneficial components of cognitive-stimulation programs[12-14]. Previous studies have also found that applying interventions such as physical activity into a whole person wellness program can decrease cognitive decline in older adults[15,16].

Humor and its associated mirthful laughter affect several systems in the human body. In addition to mirthful laughter having similar physiological effects as exercise, it can also improve cognition, increase natural killer-cell activity, and stimulate circulation[17-19]. Humor therapy has been widely used as a complimentary form of therapy for various medical conditions, including stress reduction during cardiac rehabilitation[20]. Additionally, humor therapy has shown benefits for vascular function[21,22], and can protect against the risk of disease[23,24]. More importantly, humor-associated, mirthful laughter has been associated with quality of life improvement in depressed older adults[25].

Cognitive function can deteriorate in T2DM individuals[26,27]. The precise process of how T2DM is associated with cognitive decline is not clear[28]. Nonetheless, there have been several suggested theories. Poor glucose control, as shown by higher plasma

hemoglobin A1c (HbA1c) and elevated triglyceride levels, contribute to a decline in cognition[26,27]. In addition, poor glycemic control is associated with lower memory test scores[29]. Research has shown that, over a three year span, T2DM individuals can have cognitive decline due to increased insulin resistance and high HbA1c[30]. The formation of advanced glycation end products (AGEs), through elevated glucose concentrations, can have damaging effects on brain neurons and thus, brain synchrony [30,31]. AGEs increase arterial rigidity and enhance oxidative stress [32]. Oxidative stress results in the formation of free radicals which can ultimately lead to cell death. Moreover, oxidative stress has been linked to the development of Alzheimer's disease [33], cardiovascular plaque formation, and blood vessel complications in Diabetics [34,35]. Individuals with T2DM, without dementia, have limitations with daily living activities and have also shown to have tendencies towards cognitive decline [36] . Healthy brain function, especially in diabetics, is a vital element of whole person elderly wellness.

Research has shown that mirthful laughter beneficially affects the body's physiological functions[18]. One approach to such study has been through psychoneuroimmunology (PNI), which focuses on processes linking communication pathways involving the brain to neuroendocrine and immune systems. Mirthful laughter stimulates gamma-wave (31-40 Hz)-band activity in the brain, shown through electroencephalography (EEG), which leads to enrichment of higher cognitive activity [37,38]. Gamma-wave band activity represents the brain at its highest cognitive ability level and mirthful laughter produces this gamma state[37]. Research has also shown that mirthful laughter can decrease the biomarker for cortisol, a neuroendocrine stress hormone[39].

However, a healthy or fulfilling eustress experience can decrease stress[39,40]. Additionally, a decrease in immune function can lead to cognitive decline[41]. Cortisol, released through the adrenal axis of the hypothalamic pituitary, can be neurotoxic to the hippocampus, the site of consolidated memory. With chronic, excess production of cortisol due to continued stressful events, individuals can damage the hippocampus, which in turn can lead to impairment in emotion, learning, and memory[3,42] and in recall of previously stored memories[43]. Cortisol modulates brain function through alterations in brain neuron structure[44]. However, the brain can undergo structural plasticity (new neuron formation) in response to these alterations[45]. This protective feature may prevent permanent damage caused by acute and chronic stress[44]. Functional imaging has shown permanent changes in brain neurons as a result of stressors, such as a test of counting backwards[46]. In addition, research has shown that hippocampal gray matter volume can decrease due to depression[47], poor glucose control in T2DM[48,49] and chronic life stress[50].

Although acute stress and chronic stress affect the same physiological systems of the body, the outcomes are vastly dissimilar. Adaptation and survival is seen with acute stress, while pathophysiology is seen with chronic stress[44]. Allostasis is the change the body undergoes to progress back to homeostasis after daily stress events[51]. Allostatic load is the wear and tear on the body that results from inadequate handling of allostasis[44]. The changes the body goes through as a result of allostatic load can lead to a decrease in physical activity, decrease in sleep, overeating, and deficits in cognition and memory[44].

Several studies have revealed the therapeutic link between mirthful laughter and humor to physiological and behavioral aspects in humans [17,19]. But, there have been no

studies that have shown a link between humor, mirthful laughter, and the enhancement of short term memory in the healthy or diabetic elderly. The purpose of this novel research was to determine the effect of watching a humor video on short term memory in healthy and diabetic elderly cohorts. We hypothesized those healthy and diabetic elderly individuals who watch a humorous video will have an enhancement in their short term memory: *learning ability*, *delayed recall*, and *visual recognition*. Furthermore, we proposed that variations of stress hormone cortisol levels, at predetermined time points, might be seen.

CHAPTER TWO

THE EFFECT OF HUMOR ON SHORT TERM MEMORY IN OLDER ADULTS

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Summary

Background: For older adults, the damaging effects of aging and stress can impair the ability to learn and sustain memory. Humor, with its associated mirthful laughter, can reduce stress as well as cortisol, a stress hormone. Chronic release of cortisol can damage hippocampus neurons, leading to impairment of learning and memory.

Materials/Methods: The primary goal of this study was to determine whether watching a humor video had an impact on short-term memory in an older population. The research team designed a randomized, controlled trial. The study took place at Loma Linda University. The research team recruited 20 normal, healthy, older adults, 11 males and 9 females. The humor group (n=10, mean of 69.3 ± 3.7 years) self-selected one of two humorous videos—a Red Skelton comedy or a montage of America's Funniest Home Videos—and watched it for 20 minutes. A control group (n=10, mean of 68.7 ± 5.5 years) sat calmly for 20 minutes and were not allowed to read, sleep, or talk on a cell phone. The Rey Auditory Verbal Learning Test was used to assess short-term memory—learning ability, delayed recall, and visual recognition. Salivary cortisol levels were measured at predetermined times.

Results: Learning ability improved by 38.5% and 24.0% in the humor and control groups, respectively (p=.014). Delayed recall improved by 43.6% and 20.3% in the humor and control groups, respectively (p=.029). Within the humor group, delayed recall (43.6 %) was significant compared to learning ability (38.5%) (p=.002). At 3 predetermined time points, significant decreases in salivary cortisol were observed in the humor group, p=.047, .046, and .062, respectively.

Conclusion: The study's findings suggest that humor can have clinical benefits and rehabilitative implications and can be implemented in programs that support whole person wellness for older adults. Learning ability and delayed recall are important to these individuals for a better quality of life—mind, body, spirit, socially, and economically. Older adults may have age-associated memory deficiencies. However, medical practitioners now can offer positive, enjoyable, and beneficial humor therapies to improve these deficiencies.

Key Words: Humor, Laughter, Memory, Cortisol, Ageing

Background

Our world is encountering a rapid increase in society's aged population.[1] With this increase in life expectancy, an enhanced need exists for whole-person, mind-body care. Rather than merely emphasizing the cause of disease, an affirmation of a salutogenic paradigm of healthcare that supports overall human wellness and health promotion is needed, for older adults as well as the young.[2] Short-term memory loss affects many older individuals.[3,4] As people become older, numerous concerns about short-term memory loss become much more prevalent,[3] and solutions are vital to individuals, families, and society. These age-associated memory deficits can be precursors to the progression to dementia, indicating a 3-times greater risk.[5] Interventions related to short-term memory loss need to be addressed as an essential component of whole person wellness for older adults.

Short-term memory deficiencies can manifest in older adults as issues with physical and behavioral compliance in individualized, home-rehabilitation treatment programs. For instance, noncompliance might exhibit as a failure to perform at-home, physical-therapy exercises properly, inaccurate timings or dosages of medications, or missed healthcare appointments.[6] Barriers that older adults face that hinder compliance might be poor memory,[7] anxiety, depression, or lack of self-motivation. [6,8] In addition to having a broad impact on society's escalating healthcare costs, noncompliance leads to a decrease in personal health and the efficacy of treatment.[9] Intervention strategies should be geared toward whole person wellness for older adults and include precursor therapies and modalities for enhancement of memory or reduction of memory deficits; i.e., humor therapy.

Several methods have been used to improve short-term memory in older adults. Studies have revealed, with measurable brain variations, that cognitive training can enhance short-term memory.[10,11] Furthermore, cognitive-stimulation programs that incorporate the development and advancement of quality of life, self-improvement approaches, increased daily activities, and the integration of daytime naps have been shown to be beneficial.[12-14] A blend of multivitamin, mineral, and herbal supplements have been shown to improve short-term memory in older women.[15] Implementing natural interventions such as physical activity into a whole person wellness program can reduce cognitive decline in older adults.[16,17]

Humor and its associated mirthful laughter affect several systems in the human body. In addition to mirthful laughter having similar physiological effects as exercise, it can also improve cognition, increase natural killer-cell activity, and stimulate

circulation.[18-20] Humor therapy and the positive emotions that are elicited through mirthful laughter have been used widely as a complimentary therapy for many conditions, including stress reduction during cardiac rehabilitation .[21] Additionally, humor therapy has shown benefits for vascular function,[22,23] and can protect against the risk of disease.[24,25] Furthermore, humor-associated, mirthful laughter has been linked to an improvement in the quality of life of depressed older adults.[26]

Studies have shown that the body's physiological functions are beneficially affected by mirthful laughter.[20] One approach to such studies has been through psychoneuroimmunology (PNI), which focuses on processes linking communication pathways involving the brain to neuroendocrine and immune systems. The approach contributes to our understanding of the link between mirthful laughter and the salutogenic, whole-person lifestyles that support wellness.[27] Mirthful laughter stimulates gamma-wave (31-40 Hz)-band activity in the brain, shown through electroencephalography (EEG), that leads to enrichment of higher cognitive activity.[28,29] It has been shown to decrease the biomarker for cortisol, a neuroendocrine stress hormone.[30]

Moreover, a decrease in one's immune function can lead to a decline in cognition.[31] However, a positive eustress (healthy or fulfilling) experience can reduce stress.[30,32] Cortisol, released through the adrenal axis of the hypothalamic pituitary, can be neurotoxic to the hippocampus, the site of consolidated memory. With chronic, excess production of cortisol due to continued stressful events, individuals can damage the hippocampus, which in turn can lead to impairment in emotion, learning, and memory,[4,33] and in recall of previously stored memories.[34] For healthy older adults being tested on

wordlist memorization, Kircher and colleagues have found that the anterior left hippocampus has the central function in memory encoding and recall.[35]

A number of studies have shown the therapeutic connection of mirthful laughter and humor to physiological and behavioral aspects of humans.[18,20] However, no studies have shown a connection, beneficial or not, between mirthful laughter in humor and the enhancement of short-term memory in older adults. The primary focus of the current research was to determine whether watching a humor video had any impact on short-term memory in a cohort of older adults. The research team proposed that older adults who watch a humorous video might exhibit improvement in their short-term memory—learning ability, delayed recall, and visual recognition—and also show a decrease in levels of the stress hormone cortisol, at predetermined time points.

Materials and Methods

Subjects

Twenty healthy older adults, the initial and final sample size, were recruited and enrolled in the current study (control group, n=10; humor group, n=10). Individuals were recruited through word of mouth on the campus of Loma Linda University and in neighboring communities, including faculty and staff of Loma Linda University, their spouses, and residents of a local senior center. The control group consisted of 6 males and 4 females with a mean age of 68.7 ± 5.5 years. The humor group consisted of 5 males and 5 females with a mean age of 69.3 ± 3.7 years.

Individuals were questioned and excluded if they had any of the following conditions: (1) impaired hearing that would prevent them from understanding a

researcher's verbal instructions, (2) cognitive impairments, (3) neurological disorders, (4) psychiatric disorders, or (5) a history of substance abuse. Furthermore, individuals were excluded if they were taking a corticosteroid. Due to the fact that many older adults do take medications for various conditions, medications were recorded. To screen for cognitive ability, prospective participants were given the Mini Mental State Exam (MMSE). The MMSE is a highly validated exam that tests for cognitive ability. [36] It is subdivided into 5 components: (1) orientation—10 points; (2) registration—3 points; (3) attention and calculation—5 points; (4) recall—3 points; and (5) language and praxis—9 points. The exam provides a total of 30 points and has 11 questions. A score of 24-30 indicates that the person has no cognitive impairment; 18-23 shows mild cognitive impairment; and 0-17 indicates severe cognitive impairment. The duration of the exam is approximately 10 minutes.

For screening of prospective participants, each individual entered a private sitting area and was seated for 10 minutes prior to starting any testing to acclimatize to the room (22°C) and surroundings. The testing area was a quiet and comfortable room to preclude possible distractions, and no interruptions occurred during the process. A bottle of water was provided to each person.

At this point, the informed consent was read by all prospects and verbally explained to each individual. All questions regarding the research study were answered to each person's satisfaction. The participant then signed a statement of informed consent, and the investigator devoted a few minutes to building rapport with the individual to make him or her feel comfortable. Next, a salivary cortisol sample (prebaseline) was obtained.

Finally, the person was given the MMSE by the investigator to check for cognitive ability. All prospects were found to be free of cognitive impairments based on the administration of the MMSE. The Institutional Review Board of Loma Linda University approved all procedures.

Methods

If the individual's cognitive ability was intact as evidenced by the results of the MMSE, he or she was instructed to choose one slip of paper out of 2 from an envelope; one was marked "Control Group" and the other was marked "Humor Group," The investigators were blinded as to the allocation of participants to the humor or control group.

Humor Group: Humor Video

Participants in the humor group were asked to self-select one of two humorous videos: a Red Skelton comedy or a montage of America's Funniest Home Videos. They watched the video on a laptop, wore noise-reduction headphones, and were left alone in the room to watch the video.

Three participants in the humor group selected America's Funniest Home Videos, and 7 selected the Red Skelton comedy. This video was a segment from the DVD titled *Double Feature: The Lucy Show/More Red Skelton* (Vina Distributor 2002, Garden Grove, CA , USA). Each participant watched chapters 1-3 for a total of 20 minutes: Chapter 1—duration of 6 minutes and 33 seconds; Chapter 2—duration of 2 minutes and 52 seconds, and Chapter 3—duration of 11 minutes and 35 seconds. This video was

selected because participants were older adults and could relate to the humor from that era of comedy. The Red Skelton video was part a genre of comedy featuring the American variety show. Chapter 1 consisted of Red Skelton's monologue. Chapters 2 and 3 consisted of Red Skelton with guest stars performing comedy sketches.

America's Funniest Home Videos were a collection of 2 videos found on YouTube. They were entitled *100 Falling People, Part 1*—America's Funniest Home Videos part 538 and *100 Falling People, Part 2*—America's Funniest Home Videos part 540. The duration for part 1 was 11 minutes and 11 seconds and for Part 2 was 8 minutes and 50 seconds, for a total of 19 minutes and 1 second. Both videos consisted of short clips of babies, children, and adults falling in various comical situations.

Control Group: Quiescence

Participants in this group were instructed to sit calmly for 20 minutes. The investigator instructed participants that they would be left alone for that time period. The control group did not watch a video, and participants were not permitted to read, speak, or use cell phones.

Outcome Measures

Rey Auditory Verbal Learning Test (RAVLT)

As shown in Figure 1, the RAVLT was administered twice during the study. The RAVLT was used to determine learning ability, delayed-recall ability, and visual-recognition ability. The test was printed by Western Psychological Services (Torrance, CA, USA) and has been used in clinical practice as well as in research studies. The test is

a widely used neuropsychological assessment and was developed by Andre Rey (1941, 1958). It consists of a 15-item word list that was presented 5 times. A number of studies have shown the high test-retest reliability[37] and validity of this test.[38,39] The instructions are straightforward and can be easily understood by older adults. Examples of words on list A are "drum," "curtain," "school," "color," "house," and "river." The longest word on List A is 7 letters, "curtain." For List B, examples of words are "stove," "desk," "bird," "shoe," and "church." "Mountain" is the longest word on List B, with 8 letters.

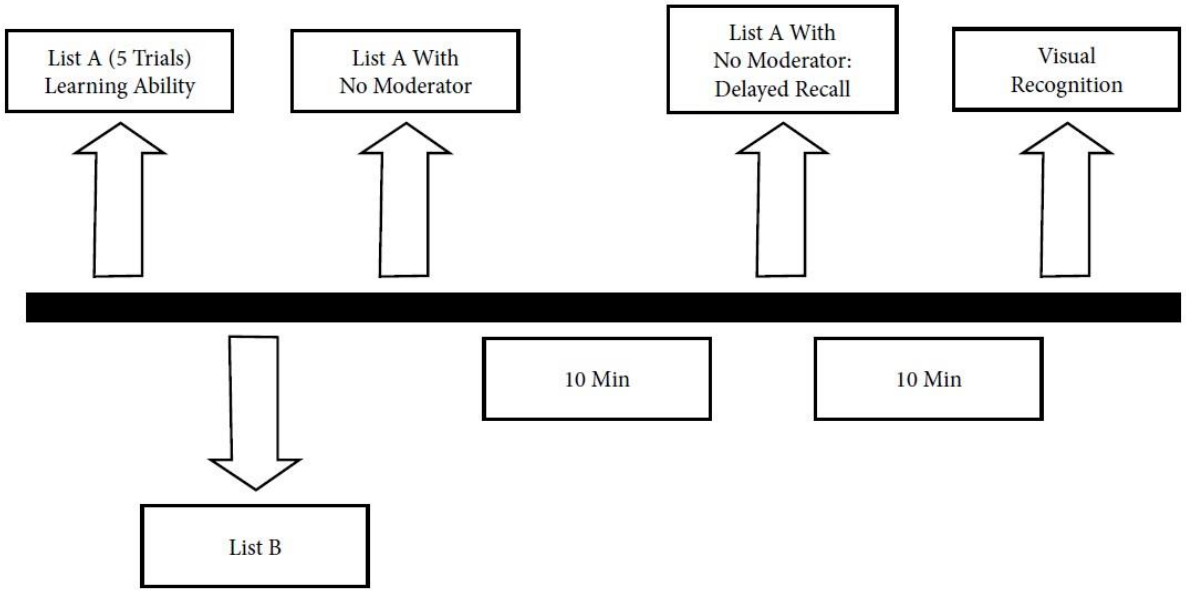

Figure 1. RAVLT protocol

Salivary Cortisol Sampling

As shown in Figure 2, samples were taken at 5 predetermined time points throughout the study. The functioning form of cortisol in blood and saliva is in its unbound form.[40] Salivary flow rates do not have an effect on levels of salivary cortisol.[41] A strong correlation exists between levels of salivary and serum cortisol.[42-44] Day-of-

sampling confounding variables of salivary cortisol were taken into consideration. Twenty-four hours prior to arriving at the lab, participants were informed not to eat or engage in strenuous exercise during the one hour prior to their appointment times.[45,46] They were directed not to drink any coffee or alcohol, to abstain from smoking upon awakening, and not to fall asleep again upon awakening.[47-49] Upon their arrivals, verbal instructions were given to participants explaining the sampling protocol for salivary cortisol. All questions were answered to a participant's satisfaction. When a person is faced with a stressful situation, his or her cortisol levels increase in a manner unrelated to its diurnal rhythm.[50] However, in deference to the diurnal rhythm of cortisol, levels were measured between 10:30 am and 4:00 pm.[46,51] Products for sampling salivary cortisol were purchased from Salimetrics, LLC (State College, PA, USA). Supplies included cryostorage boxes, oral swabs, swab storage tubes, and barcoded sample labels.

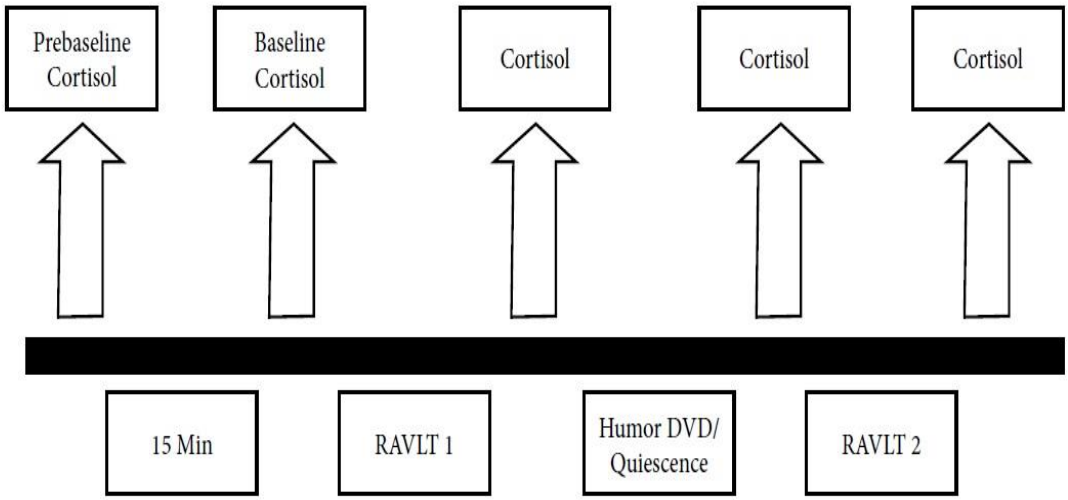

Figure 2. Salivary cortisol sampling protocol

Procedures

After the participant's assignment to either the humor or the control group when the investigator and participant were ready, the second sample of salivary cortisol was obtained and the RAVLT 1 was administered to check learning ability, delayed recall, and visual recognition. Two investigators administered the test. Investigator 1 read the words aloud, while Investigator 2 kept a tally of the words repeated by the participant, on a premade word-list form. Investigator 1 and the participant were seated face-to-face at a table, to facilitate better hearing and understanding of the words spoken. Investigator 2 sat to the side and at a distance from the participant and Investigator 1 to prevent the participant from seeing or hearing tallying of the correctly repeated words. For reliability, the same Investigator 1 was used to speak and the same investigator 2 was used to record throughout the study. After the RAVLT 1 was administered, the third sample of salivary cortisol was obtained.

Next the humor group watched the chosen video and the control group sat calmly for 20 minutes. The investigator then obtained the fourth sample of salivary cortisol and administered the RAVLT 2, using the same words as for RAVLT 1. Lastly, after the RAVLT 2 was finished, the final sample of salivary cortisol was obtained.

Measurements of Salivary Cortisol

The oral swab was placed directly under participants' tongues by the investigator, and they were instructed to keep their mouths closed for 90 seconds, timed with a stopwatch. Subsequently, the oral swab was removed by the investigator and placed directly into an oral-swab tube, capped, and then immediately stored in a -20˚C

refrigerator-freezer. After the participant completed the study and all salivary cortisol samples had been obtained, the storage tubes were immediately placed in a -80°C freezer. Each of the storage tubes was pre labeled by Salimetrics with a barcode representing the participant's number and the predetermined time point that the sample was taken. The same investigator obtained all salivary cortisol samples.

Participants were allowed to drink water throughout the study. However, when the time for a salivary-cortisol sampling was approaching, participants were informed 10 minutes prior to it not to take any sips of water until after the sample was made. When all experiments were completed, the samples were shipped overnight to Salimetrics for analysis. Two cortisol readings (μg/dL) were analyzed per sample, and an average was reported.

RAVLT-Learning Ability

As shown in Figure 2, participants were instructed that Investigator 1 would read aloud the list of 15 words (List A) and that they should listen carefully and not interrupt the sequence. The speed was approximately one spoken word per second. When all 15 words had been read, participants were asked to repeat as many words as they could recall. The order in which participants repeated words was not of importance. To facilitate the jogging participants' memories, they were encouraged to repeatedly speak the words aloud once they could not recall words to any further extent. They were allowed to repeat words previously spoken. Investigator 2 used a premade list of words from List A and checked the list as words were spoken by the participant. After the participant could no longer recall any words, the same words were tested again, and in

the same procedure, for a total of 5 trials. The investigators moved onto the next trial after participants stated that they could not recall any additional words or until at most a minute of silence had occurred. No time break took place between the 5 trials.

After the fifth trial, investigator 1 immediately read aloud a set of 15 new words from a different list (List B) and instructed participants to repeat as many of the List-B words that they could recall. Investigator 2 again tallied the correct words on a premade list. As per the RAVLT protocol, List B was given to intentionally confuse the participant. Following the reading of list B, investigator 1 asked the participant to repeat the words from the original list (List A). At this juncture, the imperative requirement was that investigator 1 should not speak the words prior to having the participant recall and state the words. The participant was then given a 10-minute break. During the break, the participants were not allowed to sleep, read, or talk on their cell phones or to the investigators. When the 10-minute break concluded, delayed recall was tested.

RAVLT-Delayed Recall

Investigator 1 asked the participant to recall and repeat the words from list A once more. Again, prior to the participant stating the words, Investigator 1 did not read the words from List A. The participant was then given a 10-minute break. During the break, the participant again was not allowed to sleep, talk on the cell phone, talk to investigators, or read. When the 10-minute break was completed, visual recognition was tested.

RAVLT-Visual Recognition

Investigator 2 handed the participant a pen and a one-page form consisting of a

list of words. Participants were instructed to locate the words that were spoken to them and learned and recalled by them from List A and to check off only those words. Participants were told to check off a maximum of 15 words. The form contained 50 words and consisted of words from List A, List B, and various other words. Participants were given a maximum of 3 minutes to complete this task.

Statistical Analysis

Statistical analysis was performed using the statistical package SPSS for Windows version 22 (SPSS Inc., Chicago, IL, USA). Data was summarized using frequencies and means and standard deviations. The demographic characteristics of the two groups were compared using the Mann-Whitney U test and Fisher's Exact test. The Mann-Whitney U test was also used to compare the RAVLT scores between the 2 groups, and the Kruskal-Wallis Test was used to compare the change in scores within the groups. The Wilcoxon Signed Ranked test was conducted to assess changes in cortisol levels as a result of the intervention. The significance level was set at $P \leq 0.05$.

Results

The demographics of all 20 participants are displayed in Table 1. No significant differences existed between the control and the humor groups in gender, age, height, weight, BMI, and MMSE score. Figure 3 shows percentage changes in learning ability, delayed recall, and visual recognition due to the 20-minute humor intervention for the humor group and the rest period the control group, as measured by the RAVLT.

Table 1. Mean (SD) of demographic characteristics by study group (N=20)

	Control Group (n = 10)	Humor Group (n = 10)	*P* Value
Gender, %			
Male	60	50	.50[a]
Female	40	50	
Age, y	68.7 (5.5)	69.3 (3.7)	.78[b]
Height, cm	170.5 (8.8)	168.8 (9.6)	.67[b]
Weight, kg	76.9 (17.7)	83.9 (19.8)	.41[b]
BMI, kg/m^2	26.2 (4.1)	29.5 (6.8)	.22[b]
MMSE	29.4 (0.7)	28.7 (0.9)	.08[b]

Abbreviations: SD = standard deviation; MMSE = Mini-Mental State Exam.

[a]Determined using Fisher's exact test (χ^2).
[b]Determined using independent *t* tests.

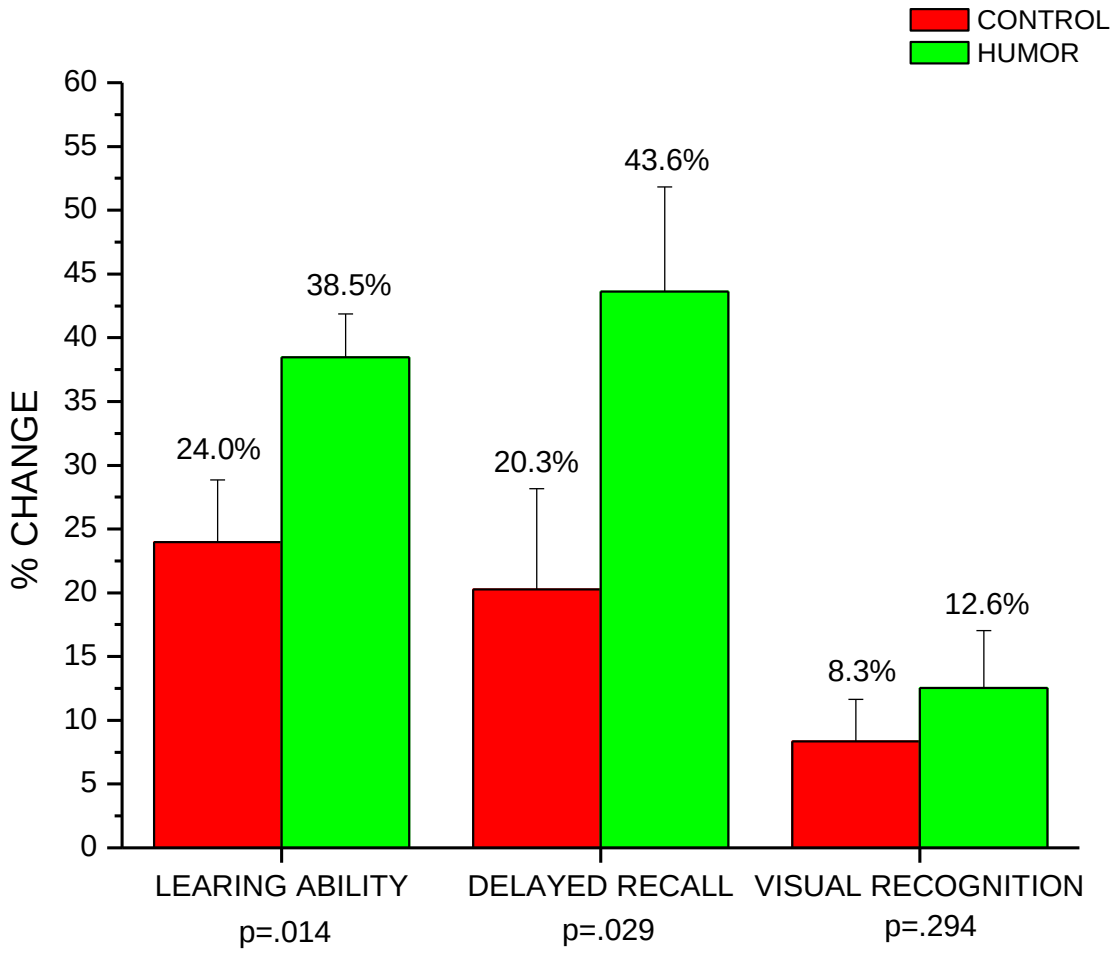

Figure 3. Mean (SD) % Change of RAVLT Scores

Results indicated that learning ability, delayed recall, and visual recognition were enhanced in both groups. When examining the changes in scores on the RAVLT between pre- and postintervention testing, however, a significant difference existed between the humor and control groups in scores for delayed recall (p=.029), as shown in Figure 3. The percentage increase was more than double for the humor group, a 43.6% increase as compared to a 20.3% increase for the control group. This category shows the greatest

improvement for the humor group, indicating that the video was most influential in improving delayed recall.

Furthermore, significant differences also were found when comparing changes in learning ability between the two groups. As shown in Figure 3, although the control group increased their learning ability by 24.0%, the humor group had a larger increase, 38.5% (p=.014). Although both groups experienced an increase in the percentage change in visual recognition, 8.3% in the control and 12.6% in the humor group, the difference between these increases was not significant (P=.294).

Percentage changes within each group for the 3 parts of the RAVLT are shown in Figure 4. For the control group, the greatest percentage change was seen for learning ability vs delayed recall and visual recognition, p=.099. The opposite was observed in the humor group. The humor group showed a greater increase in delayed recall vs. learning ability and visual recognition. p=.002.

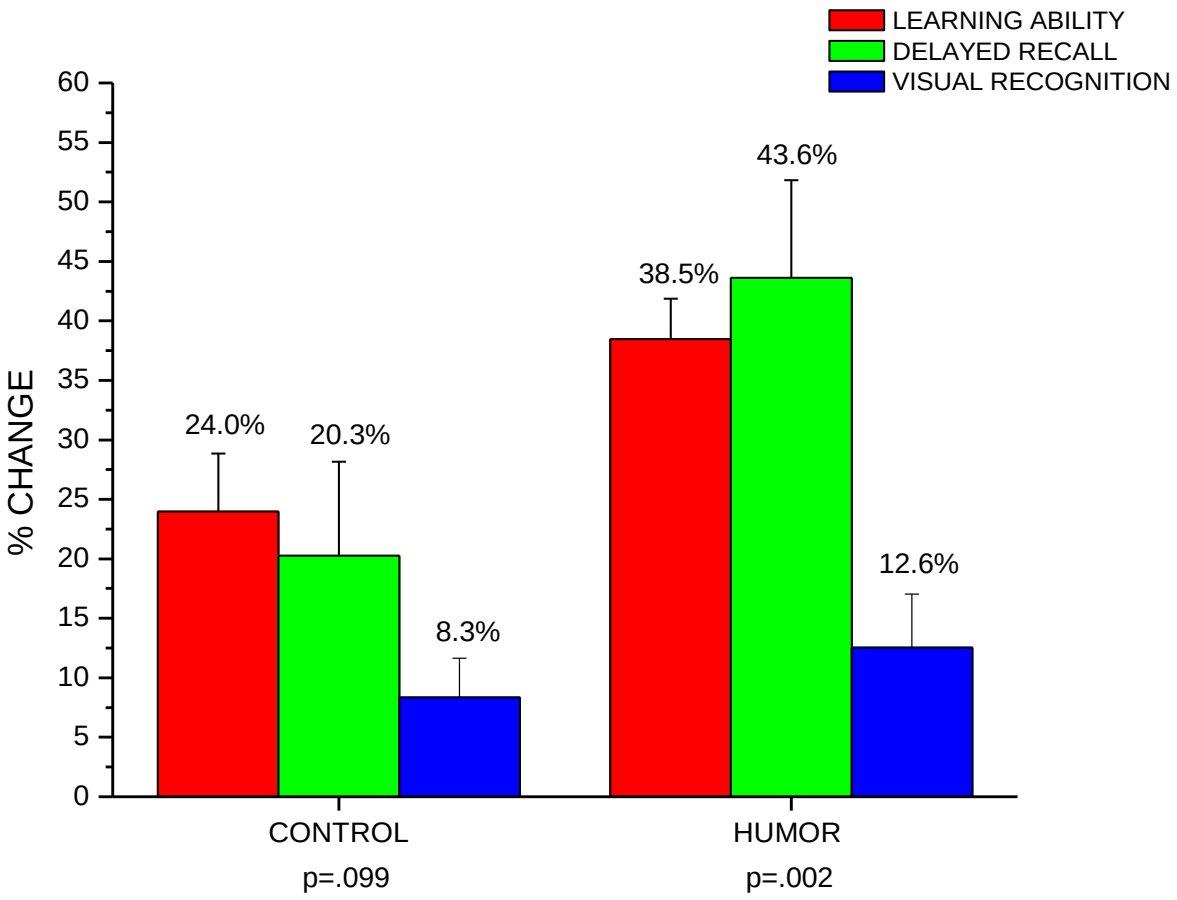

Figure 4. Mean (SD) % Change of RAVLT Scores by Study Group

For the control and humor groups, Table 2 shows the levels of salivary cortisol at the 5 predetermined time points throughout the study. It illustrates the mean (SD) for the cortisol levels (µg/dL) in the control and humor groups over time. One control participant's cortisol level appeared to be an outlier. After using the Grubbs' Test, it was determined to be 2.8 standard deviations away from the mean, and thus, was deleted. Watching a humor video significantly decreased salivary cortisol levels in the humor group. The most relevant drop in that group's levels occurred from measurements at baseline /pre-RAVLT 1 to those immediately after watching the humor video/pre-

RAVLT 2 p=.046. For the control group, during the same time frame, no significant change occurred in levels of salivary cortisol, p=.130.

In addition, a significant change occurred in the levels of salivary cortisol for the humor group from baseline/pre-RAVLT 1 to just immediately before watching the humor video (post-RAVLT 1/pre-humor or quiescence) ,p=.047). Again, no significant changes in the level of salivary cortisol occurred during this same period for the control group, p=.477. Borderline significant changes occurred in the humor group from baseline/pre=RAVLT 1 to immediately after administering the RAVLT 2 (post-RAVLT 2 or end of study), p=.062. In the control group, a significant change also occurred during the same time frame, p=.047. Prebaseline measurements compared to baseline measurements were not significant for the control or the humor group, p=.257 and p=.242, respectively.

Table 2. Mean (SD) cortisol levels (µg/dL) by study group

	CONTROL GROUP	HUMOR GROUP
(A) PRE BASELINE	.20 (.08)	.25 (.20)
(B) BASELINE / PRE RAVLT 1	.20 (.08)	.27 (.20)
(C) POST RAVLT 1 / PRE HUMOR & QUIESCENCE	.22 (.12)	.23 (.14)
(D) POST HUMOR & QUIESCENCE / PRE RAVLT 2	.17 (.10)	.21(.13)
(E) POST RAVLT 2	.14 (.06)	.17 (.07)
	P value*	P value*
A-B	.257	.242
B-C	.477	.047
B-D	.130	.046
B-E	.047	.062

Abbreviations: SD=Standard Deviation,* Wilcoxon Signed Rank Test, RAVLT=Rey Auditory Verbal Learning Test

Discussion

The current study examined whether short-term memory in older adults could be enhanced by watching a humor video and also whether levels of the stress hormone cortisol could be modulated in this process. The results showed that the humor group had a substantial increase in learning ability and delayed recall when compared to the control group. In addition, the current study has shown that pre-baseline levels of cortisol measured 10 minutes before baseline measurements and before the start of RAVLT 1, were not significantly different for the humor and control groups. This result suggests that no anticipatory effect existed at the onset of the memory test. After cortisol

measurements were taken at pre-baseline, participants were randomly placed into one of the two groups.

The research team might ask whether individuals' anticipation that they would be watching and enjoying a humor video could diminish the possible stress response associated with taking a memory test, and thus, affect test scores. Immediately following RAVLT 1, the humor group's cortisol levels significantly decreased. That decrease could be attributed to the humor group's anticipation that they would be in a mirthful state. However, in the control group, cortisol levels actually increased following RAVLT 1. A stressful state can be either a eustress or a distress state. Results for the humor group seem to indicate they experienced a eustress state while taking the memory test, as shown by their decreased cortisol levels. On the other hand, the control group appears to have experienced a distress state, as shown by their increased cortisol levels.

Even though both groups were aware that a second memory test (RAVLT 2) was pending, as predicted cortisol levels did decrease during the 20-minute time frame when the humor group was watching the humor video and the control group was sitting quietly and calmly. A previous study had shown, not surprisingly, that humor and the associated mirthful laughter decreases cortisol levels just as readily as a calm meditative state.[30] More astonishing was the effect that humor and laughter had on modulating cortisol levels as compared to baseline for the humor group (Table 2). After watching the humor video, cortisol levels were significantly lower than participants' baseline levels. Although the control group's cortisol levels also decreased while sitting calmly for 20 minutes, the change was not significant.

The research team suggests that the hippocampus, which helps consolidate short-term memory, possibly sustained less overall, cortisol-induced suppression in the humor group, potentially explaining improved cognitive outcomes for that group.[4,33] Throughout the study, the humor group showed a consistent decrease in cortisol levels. Moreover, at the end of the study, at completion of RAVLT 2, the humor group showed a borderline statistically significant decrease in cortisol, from baseline measurements (Table 2).

The research teams further suggests the humor group was continuously in a eustress state throughout the study, potentially accounting for the fact that the group's overall scores for memory tests were significantly higher after the humor and laughter session. However, although the cortisol levels had decreased in both groups during the 20-minute period, the increases in scores on the memory test were significantly greater in the humor group (Figure 2). The impact of watching the humor video had the greatest influence on delayed recall in the humor group, with the percentage increase more than doubling and being significantly greater than that of the control group. Also, learning ability showed a significantly greater improvement in the humor group than in the control group.

Furthermore, throughout the study, variations occurred within the control group's cortisol levels. At this time, the research team lacks sufficient data that might suggest if this variability did or did not impact hippocampal influence over cognitive outcomes. However, although the control group showed a significant decrease in cortisol levels from baseline measurements, the results show that the decreases did not significantly affect the group's scores on the memory tests and that the control group's scores for learning ability and delayed recall were affected.

Within the humor group, delayed recall showed a more significant increase than observed for learning ability; note that delayed recall was tested after learning ability. Not surprisingly, delayed recall should increase after a session of learning. Due to the research team's view that the humor group was in a continuous eustress state, the enhancement to learning ability may have translated into better delayed recall. However, within the control group, no significant difference existed between the increases in learning ability and delayed recall, perhaps because of the continuum of distress. This finding strongly supports the claim that humor may play a role in enhancing short-term memory, and specifically, learning ability and delayed recall in older adults.

This current study has some potential limitations. First, the sample size was small. Although the sample size had over a 90% power based on results of a pilot study, nonetheless a larger sample size would have allowed for better representation of an older-adult population as a whole. Second, due either to the small sample size or the assay testing for salivary cortisol, more variability may have been evident in the cortisol levels. It remains to be seen whether a larger sample size will reduce this variability. Third, the participants included in this study were limited to healthy older adults. Future studies should include cohorts of older adults with diabetes and obesity. Diabetic and obese individuals should be included because poor glucose tolerance and obesity have been associated with greater risks for cognitive impairment.[52,53]

Conclusions

The study's findings suggest that humor has clinical benefits and rehabilitative implications and can be implemented into salutogenic, whole person wellness programs

for older adults. Difficulties with learning ability and delayed recall become more burdensome as individuals age. Learning ability and delayed recall are important to older adults for a better quality of life mind, body, spirit, socially, and economically. Older adults have age-associated memory deficiencies. However, healthcare practitioners now can offer positive, enjoyable, and beneficial humor therapies to improve these deficiencies.

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CHAPTER THREE

HUMOR'S EFFECT ON SHORT-TERM MEMORY IN HEALTHY AND DIABETIC OLDER ADULTS: AN INNOVATIVE APPROACH FOR WHOLE-PERSON WELLNESS

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Summary

Background: With ageing, the damaging effects of stress can impair the ability to learn and sustain memory. Humor and the associated mirthful laughter can reduce stress by decreasing the stress hormone cortisol. Excess cortisol can damage hippocampal neurons leading to impairment of learning and memory.

Materials/Methods: The purpose was to examine the effect of watching a humor video on short term memory in 3 age matched elderly groups: elderly (69.9 ± 3.7 years), diabetic (67.1 ± 3.8 years), and control (68.7 ± 5.5 years) (no video). The standardized neuropsychological memory assessment tool, Rey Auditory Verbal Learning Test (RAVLT) was used to assess for 1) learning ability, 2) recall ability, and 3) visual recognition ability. Salivary cortisol measurements, at 3 predetermined time points, were obtained. RAVLT was given to 30 elderly individuals before and after watching a humor video of their choice for 20 minutes vs. no humor video.

Results: Results showed that 1) learning ability improved by 38.45% , 33.38%, and 23.96% in the elderly, diabetic, and control groups respectively (p=.025); 2) delayed recall improved by 43.61%, 48.10%, and 20.25% in the elderly, diabetic, and control groups respectively (p=.064); and 3)visual recognition increased by 12.55%, 16.72%, and 8.33% in the elderly, diabetic, and control groups respectively (p=.321). Results, for the changes in salivary cortisol levels, at the predetermined time points, indicated there were 1) borderline significant changes in the elderly group (p=.047, .046, and .062 respectively), 2) significant changes in the diabetic group (p=.047, .025, and .035 respectively), and 3) no changes in the control group (p=.323, .323, and .187 respectively).

Conclusion: Due to decreased cortisol levels, elderly and diabetic elderly individuals that watch a humor video that induces mirthful laughter vs. not watching a humor video have greater enhancement in: 1) capability to learn, 2) have greater recall, and 3) improve visual recognition in short term memory function.

Key Words: Humor, Laughter, Memory, Cortisol, Ageing, Diabetes

Background

Although the world's population rate and life expectancy is increasing, the health related well-being of the population is lagging behind[1]. As a result of the upsurge in life expectancy, there is an obligation towards a heightened awareness for whole person, mind-body-spiritual care. A salutogenic paradigm of health care (prevention, education, and promotion) is necessary to promote overall human wellness [2,3]. Accordingly, commitment for health promotion of mind-body-spiritual wellness should target the elderly. Older adults face abundant challenges in regards to age related loss of short term memory [4,5] that affect quality of life. These challenges are encountered by the individuals, families, and society. This is complicated by the fact that elderly encounter age associated diseases such as Diabetes Mellitus that affect short term memory [6]. The growth of the overall elderly population is a major influencing factor in the Type 2 Diabetes Mellitus (T2DM) pandemic [7]. Short term memory loss therapies need to be a crucial element of whole person elderly wellness as well as being applied in the clinical setting.

In older adults, issues with short term memory become notably apparent through the lack of health care compliance. These behavioral and physical compliance concerns

are observed in home rehabilitation treatment programs and are also noted in day to day events. For example, non-compliance can manifest as reduced compliance for therapeutic exercise, inaccurate timings and dosages of medications, and neglecting health care appointments [8]. Non-compliance often leads to escalation of T2DM outcomes, and thus, a diminished quality of life among the elderly. To successfully manage Diabetes and avoid such a diminished quality of life, individuals need to comply with physicians' orders [6]. Yet, poor memory retention remains a barrier that impedes compliance [9]. Subsequently, this directly impacts society's ever-increasing health care cost and efficacy of treatment [10].

Memory impairments, in the elderly, can lead to mild cognitive decline [11]. Furthermore, these deficits, which impose a 3 fold greater risk, can be precursors to the progression towards dementia [12]. Also, in T2DM individuals, the risk for dementia is approximately doubled [13]. Numerous approaches have been developed and employed to improve short term memory and cognition in the elderly. Studies have shown that cognitive training can enhance short term memory [14,15]. Programs such as cognitive stimulation that incorporate increased physical activity, proper nutrition, advancement of quality of life, increased social interaction, and the integration of day time naps have been shown to have benefit for this population [16-18].

Humor and the resultant mirthful laughter affect the human body in numerous beneficial ways. In addition to promoting cognition, humor and mirthful laughter can protect against disease [19], decrease stress [20], improve the quality of life in depressed older adults [21], and boost the immune system [22]. Humor therapy, through mirthful laughter, elicits positive emotions that have been used widely as a component of complimentary

therapy [23]. It has also been shown to improve vascular function and has promoted similar physiological changes as those observed during physical activity [24,25].

Psychoneuroimmunology (PNI) can explain the beneficial effects that mirthful laughter induces in the human body. PNI is a discipline that explains the network of communication pathways between the brain, neuroendocrine, and the immune systems. During electroencephalography, mirthful laughter stimulates the gamma wave band of the brain [26]. This leads to the development of higher cognitive activity and has been shown to decrease the neuroendocrine stress hormone cortisol [20]. In addition, PNI supplements our knowledge base of the relationship between the salutogenic whole-person wellness lifestyle and mirthful laughter [27].

Cortisol is released via the hypothalamic pituitary adrenal axis. Although cortisol can be neurotoxic to the hippocampus, the site of consolidated memory, a positive eustress experience can reduce stress and thus decrease the levels of cortisol [20,28]. However, during a state of chronic excess cortisol secretion, due to continued stress events, the hippocampus can become damaged. This in turn can lead to impairment in emotion, learning, memory [5,29] and recall of prior stored memory [30]. During testing on wordlist memorization in the elderly, the anterior left hippocampus has the vital role in memory encoding and recall [31].

Studies have revealed that cognitive function can decline in individuals with T2DM [32,33]. The exact mechanism of T2DM associated cognitive dysfunction has not yet been clarified [34]. However, there have been numerous suggested concepts. Poor glucose control, as evidenced by higher hemoglobin A1c (HbA1c) and elevated triglyceride levels in the plasma, contribute to decline in cognition [32,33]. Greenwood et al. showed that poor

glycemic control is related to lower memory test scores [13]. Umegaki et al. showed that over a three year span, T2DM individuals may have cognitive dysfunction due to increased insulin and HbA1c [35]. Elevated glucose concentrations can have harmful effects on brain neurons through the formation of advanced glycation end products (AGEs) [35,36]. AGEs increase arterial rigidity and enhance oxidative stress [37]. Oxidative stress leads to the formation of free radicals which can eventually lead to cell death. In addition, oxidative stress has been implicated in the genesis of Alzheimer's disease [38], cardiovascular plaque formation, and blood vessel complications in Diabetics [39,40]. T2DM individuals without dementia have shown tendencies towards cognitive decline and have limitations with daily living activities [41]. Healthy brain function, especially in diabetics, is one essential component of the salutogenic whole person elderly wellness programs.

Several studies have shown the therapeutic relationship between humor and the resultant mirthful laughter, with regard to behavioral and physiological connections [22,24]. However, there have been no studies that have shown a link - beneficial or not - between mirthful laughter, humor, and the improvement of short term memory amongst the healthy or diabetic elderly. The aim of this novel research was to examine the effect of watching a humor video on short term memory in healthy and diabetic elderly cohorts. We hypothesized those healthy and diabetic elderly individuals who watch a humorous video will demonstrate enhancement in their short term memory: *learning ability*, *delayed recall*, and *visual recognition*. Additionally, we proposed that variations of stress hormone cortisol levels, at predetermined time points, might be observed.

Materials and Methods

Subjects

Thirty elderly subjects were enrolled in this study [10 controls (no humor); 10 humor group (humor video); 10 diabetic group (humor video)]. Subjects were recruited through word of mouth from the Loma Linda University campus, the Loma Linda University Diabetes Treatment Center, and neighboring communities, including Loma Linda University faculty, staff, their spouses, and residents of a local senior retirement center. Thirty subjects composed the initial and final sample size. The control group consisted of 6 males and 4 females with mean age of 68.7 ± 5.5 years. The humor group consisted of 5 males and 5 females with mean age of 69.3 ± 3.7 years. The diabetic group consisted of 6 males and 4 females with mean age of 67.1 ± 3.8 years. For the diabetic group, the mean diabetes duration was 7.7 ± 6.5 years and the mean HbA1c was 6.8 ± 1.0. Subjects were excluded if they had impaired hearing that would prevent them from understanding the researcher's verbal instructions. In addition, subjects were questioned and excluded if they had any of the following conditions: cognitive impairments, neurological disorders, psychiatric disorders, or a history of substance abuse. Furthermore, subjects were excluded if they were taking a corticosteroid. Due to the fact that many elderly do take medications for various conditions, medications were recorded. To screen for cognitive ability, the Mini Mental State Exam (MMSE) was given. The MMSE is a highly validated exam that tests for cognitive ability [42]. All subjects were found to be free of cognitive impairments based on the administration of the MMSE. The informed consent was read by all subjects and verbally explained to each subject. All questions regarding the research study were answered to the subject's

satisfaction. Subjects signed a statement of informed consent prior to their participation. The Institutional Review Board of Loma Linda University approved all procedures.

The investigators were blind to the humor/control group allocation. After signing the informed consent, the non-diabetic subjects were instructed to choose one slip of paper out of two from an envelope to randomly determine which group he/she would be assigned. One slip had the words "Control Group" and the other slip had the words "Humor Group".

Methods

Humor Video

Determination of which video each subject watched was established by asking the subject to self-select between two humorous videos: Red Skelton comedy or a montage of America's Funniest Home Videos. Three elderly humor group subjects selected America's Funniest Home Video and 7 elderly humor group subjects selected Red Skelton comedy. Seven elderly diabetic subjects selected America's Funniest Home Video and 3 elderly diabetic group subjects selected Red Skelton comedy. The elderly control group did not watch a video and was not permitted to read, speak, or use their cell phones for the 20 minutes between tests.

The Red Skelton video was a segment from the DVD titled *Double Feature: The Lucy Show/More Red Skelton* (Vina Distributor 2002). The subject watched Chapters 1-3 for a total of 20 minutes: Chapter 1 duration was 6 minutes and 33 seconds; Chapter 2 duration was 2 minutes and 52 seconds, and Chapter 3 duration was 11 minutes and 35 seconds. This video was selected because subjects were elderly and could relate to the

humor from that chronological comedy era. The Red Skelton video was part of the American variety show genre of comedy. Chapter 1 consisted of Red Skelton's monologue. Chapters 2 and 3 consisted of Red Skelton with guest stars performing comedy sketches.

America's Funniest Home Videos were a collection of two videos on YouTube. They were entitled *100 falling people Part 1-America's Funniest Home Videos part 538* and *100 falling people Part 2-America's Funniest Home Videos part 540*. The duration of the two videos were 11 minutes and 11 seconds for part 1 and 8 minutes and 50 seconds for Part 2 for a total of 19 minutes and 1 second. Both videos consisted of short clips of babies, children, and adults falling in various comical situations.

Outcome Measures

Rey Auditory Verbal Learning Test (RAVLT)

The RAVLT was used to determine *learning ability*, *delayed recall* ability, and *visual recognition* ability. The test was printed by Western Psychological Services (Torrance, CA). It has been used in clinical practice as well as in research studies. The test is a widely used neuropsychological assessment test. It was developed by Andre Rey (1941, 1958). A number of studies had shown high test-retest reliability [43] and validity [44,45]. It consists of a 15 item word list that is presented five times. The instructions are straightforward and can be easily understood by the elderly. Examples of words on the list A are: "drum", "curtain", "school", "color", "house" and "river". The longest word on List A is 7 letters ("curtain"). For List B, examples of words are: "stove", "desk", "bird", "shoe", and "church". "Mountain" is the longest word on List B with 8 letters.

The RAVLT is administered twice; once immediately before the humor/quiescence period (RAVLT1) and once immediately after the humor/quiescence period (RAVLT 2).

Mini Mental State Exam (MMSE)

The MMSE is a highly validated exam that tests for cognitive ability [42]. The MMSE is divided into five components: orientation 10 points, registration 3 points, attention and calculation 5 points, recall 3 points, and language and praxis 9 points. The exam is scored out of a total of 30 points with 11 questions. A score of 24 - 30 indicates having no cognitive impairment; 18 – 23 mild cognitive impairment; and 0-17 severe cognitive impairment. The duration of the exam is approximately 10 minutes.

Salivary Cortisol Sampling

The functioning form of cortisol in blood and saliva is in its unbound form [46]. Salivary flow rates do not have an effect on salivary cortisol levels [47]. There is a strong correlation between salivary and serum cortisol levels [48-50]. To control for confounding, the day of sampling of salivary cortisol was taken into consideration. Twenty four hours prior to arriving at the lab, the subject was informed not to eat or engage in strenuous exercise one hour prior to their appointment time [51,52]. They were directed to not drink any coffee or alcohol and abstain from smoking upon awakening and to not fall asleep again upon awakening [53-55]. Upon arriving, verbal instructions were given to the subject explaining the salivary cortisol sampling protocol. All questions were answered to the subject's satisfaction. When faced with a stressful situation, cortisol levels increase separately from diurnal rhythm [56]. However, in deference to the diurnal rhythm of

cortisol, levels were taken between 10:30 am and 4:00 pm [51,57]. Salivary cortisol sampling products were purchased from Salimetrics, LLC (State College, PA). Supplies included cryostorage boxes, oral swabs, swab storage tubes, and bar coded sample labels. As shown in Figure 5, samples were taken at 5 pre-determined time points throughout the study. The oral swab was placed directly under the subject's tongue by the investigator and the subject was instructed to keep their mouth closed for 90 seconds (timed with a stopwatch). Subsequently, the oral swab was removed by the investigator and placed directly into an oral swab tube, capped, and then immediately stored in a -20˚C refrigerator freezer. Once the subject completed the study and all salivary cortisol samples were obtained, the storage tubes were immediately placed in a -80° C freezer. Each of the storage tubes was pre labeled by Salimetrics with a bar code including the subject number and predetermined time point that sample was taken. The same investigator obtained all salivary cortisol samples. The subject was allowed to drink water throughout the research study. However, when a salivary cortisol sampling was approaching, the subject was informed 10 minutes prior to not take any sips of water until after the sample was made. When all experiments were completed, samples were shipped over night to Salimetrics for analysis. Two cortisol readings (μg/dL) were analyzed per sample and an average was reported.

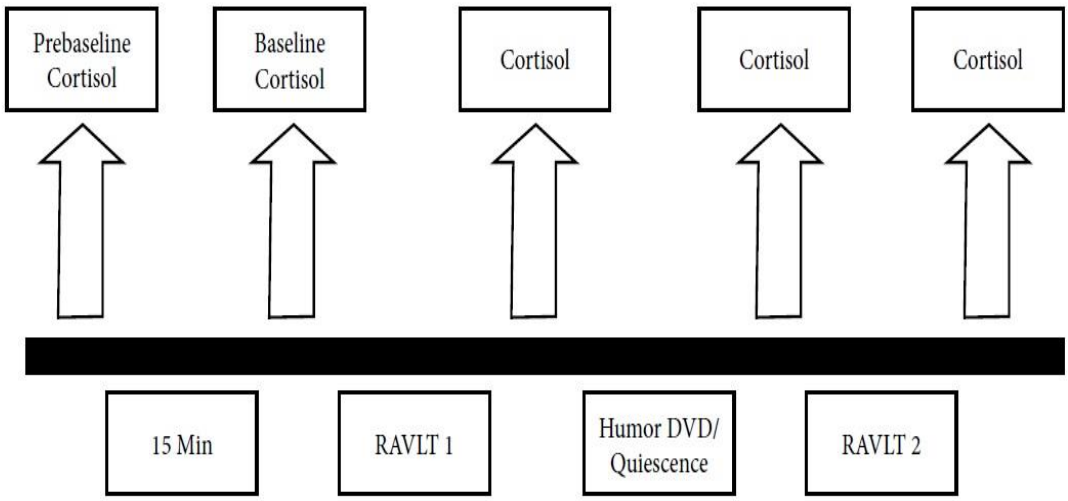

Figure 5. Salivary cortisol sampling protocol

Procedures

Each subject entered a private sitting area and was seated for 10 minutes prior to starting the RAVLT to acclimatize to the room (22°C) and surroundings. The testing area was a quiet and comfortable room to preclude possible distractions. There were no interruptions during the session. A bottle of water was provided to each potential subject. After informed consent was obtained, a few minutes were devoted to build rapport with the subject in order to make him/her feel comfortable. Next, the first of five salivary cortisol samples was obtained. The subject was then given the MMSE by the investigator to check for cognitive ability. If the subject's cognitive ability was intact, the non-diabetic subject randomly chose, by slip of paper, to determine whether he/she would be in the humor video group or the control group (no video). The diabetic group also watched a humor video. If the subject was selected into the humor video group, he/she and the diabetic group were given an option of two humor videos to choose from (Red

Skelton Comedy/American Funniest Home Videos). Once the investigator and subject were ready, the second salivary cortisol sample was obtained and the RAVLT 1 was administered to check for *learning ability*, *delayed recall*, and *visual recognition*. Two investigators administered the test. Investigator 1 read aloud the words, while Investigator 2 kept a tally of the words repeated by the subject, on a premade word list form. Investigator 1 and the subject were seated face to face at a table, to facilitate better hearing and understanding of the words spoken. Investigator 2 sat to the side and at a distance from the subject and Investigator 1 to prevent the subject seeing or hearing tallying of the correctly repeated words. For reliability purposes, throughout the study, the same Investigator 1 was used to speak, and the same investigator 2 was used to record. After the RAVLT 1 was administered and before the start of the 20 minutes of either watching a humor video or sitting calmly, the third salivary cortisol sample was obtained. After the 20 minutes concluded, the fourth salivary cortisol sample was obtained and then the RAVLT 2 was administered. Lastly, after the RAVLT 2 was finished, the final salivary cortisol sample was obtained.

RAVLT-Learning Ability

As shown in Figure 6, Investigator 1 would read aloud a list of 15 words (List A), subjects were to listen carefully, and not interrupt the sequence. The speed of the words spoken was approximately one word per second. When all fifteen words had been read, the subject was to repeat as many words as he/she could recall. The order of repeated words by the subject was not of importance. To facilitate jogging their memory, subjects were encouraged to repeatedly speak the words aloud once he/she could not recall to any

further extent. They were allowed to repeat words previously spoken. Investigator 2 used a pre made list of words from List A and List B and checked the list as words were spoken by the subject. Once the subject could no longer recall any words, the same words were tested again, and in the same procedure, for a total of five trials. We moved onto the next trial once the subject stated he/she could not recall any additional words or until there was at most a minute of silence. There was no time break between the five trials.

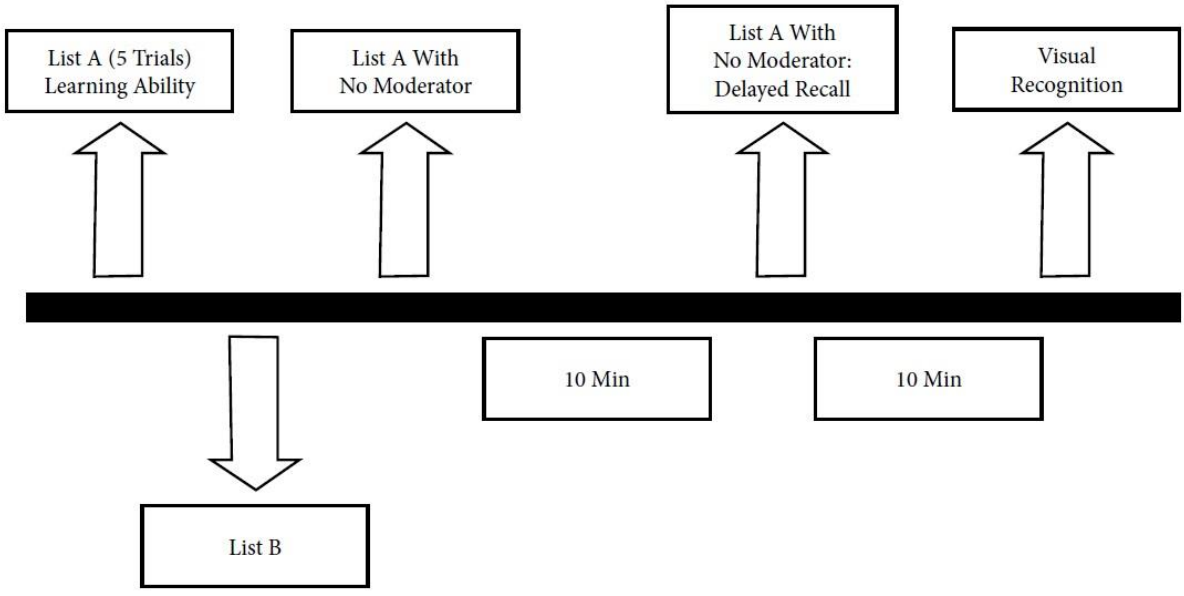

Figure 6. RAVLT protocol

After the fifth trial, investigator 1 immediately read aloud a set of 15 new words from a different list (List B) and instructed the subject to repeat as many of List B words as they can recall. Investigator 2 again tallied the correct words. As per RAVLT protocol, List B was given to intentionally confuse the subject.

Following the reading of list B, investigator 1 asked the subject to repeat the words from the original list (List A). At this juncture, the imperative point was investigator 1 did not speak the words prior to having the subject recall and state the words. The subject was then given a 10 minute break. During the break, the subject was not allowed to sleep, read, or talk on their cell phone, or to the investigators. After the ten minutes break concluded, *delayed recall* was tested.

RAVLT-Delayed Recall

Investigator 1, once more, asked the subject to recall and repeat the words from list A. Once again, prior to the subject stating the words, Investigator 1 did not read List A words. The subject was then given a 10 minutes break. During the break, the subject again was not allowed to sleep, talk on the cell phone, talk to investigators, or read. After the ten minutes break was completed, *visual recognition* was tested.

RAVLT-Visual Recognition

Investigator 2 handed the subject a pen and a one page form consisting of a list of words. The subject was instructed to locate the words that were spoken to him/her and learned and recalled by him/her from List A and to check off merely these words. Subjects were told to check off a maximum of 15 words. There were 50 words on the form and consisted of words from List A, List B, and various other words. Subjects were given a maximum of 3 minutes to complete this task.

Humor Group/Diabetic Group: Watching humor video

After being administered the RAVLT 1, the subject watched the comedy video that he/she chose for 20 minutes on a laptop. The subject was left alone in the room to watch the comedy video. The subject wore noise reduction headphones. Once the 20 minute comedy viewing session was completed, the RAVLT 2 was given following the same procedure as above utilizing the same words from RAVLT 1.

Control Group: Quiescence

After being administered the RAVLT 1, the subject was instructed to sit calmly for 20 minutes. We instructed the subject that he/she would be left alone for that time period. Subjects were not allowed to read, sleep, talk on the cell phone, etc. Once the 20 minutes of sitting calmly were finished, the RAVLT 2 was given following the same procedure as above, utilizing the same words from RAVLT 1.

Statistical Methods

Statistical analysis was performed using the statistical package SPSS for Windows version 22. Data was summarized using frequencies and means and standard deviations. The demographic characteristics of the three elderly groups were compared using Kruskal-Wallis Analysis of Variance (ANOVA) and Pearson Chi-square. Kruskal-Wallis ANOVA was also used to compare the RAVLT scores among the three groups. Wilcoxon Signed Ranked was conducted to assess changes in cortisol levels as a result of the intervention. The significance level was set at $p \leq 0.05$.

Results

The demographics of all 30 subjects are displayed in Table 3. There were no significant differences among the control group, humor group, and diabetic group in terms of gender, age, height, weight, and Body Mass Index (BMI). Percent changes in *learning ability*, *delayed recall*, and *visual recognition* due to the 20 minute humor interventions for the humor group, diabetic group, and the control group (no humor) are shown in Figure 7. Results indicated that *learning ability*, *delayed recall*, and *visual recognition* were enhanced in all groups.

Table 3**.** Mean (SD) of demographic characteristics by study group (N=30)

	CONTROL (n=10)	HUMOR (n=10)	DIABETIC (n=10)	p value**
Gender %				
male	60	50	60	.873*
female	40	50	40	
AGE(yrs)**	68.7 (5.5)	69.3 (3.7)	68.3 (3.2)	.490
HEIGHT(cm)**	170.5 (8.8)	168.8 (9.6)	174.3 (9.9)	.958
WEIGHT(kg)**	76.9 (17.7)	83.9 (19.8)	87.7 (16.2)	.326
BMI(kg/m^2)**	26.2 (4.1)	29.5 (6.8)	28.8 (4.0)	.287

Abbreviations: SD=Standard Deviation, * Pearson Chi-Square, ** Kruskal-Wallis ANOVA

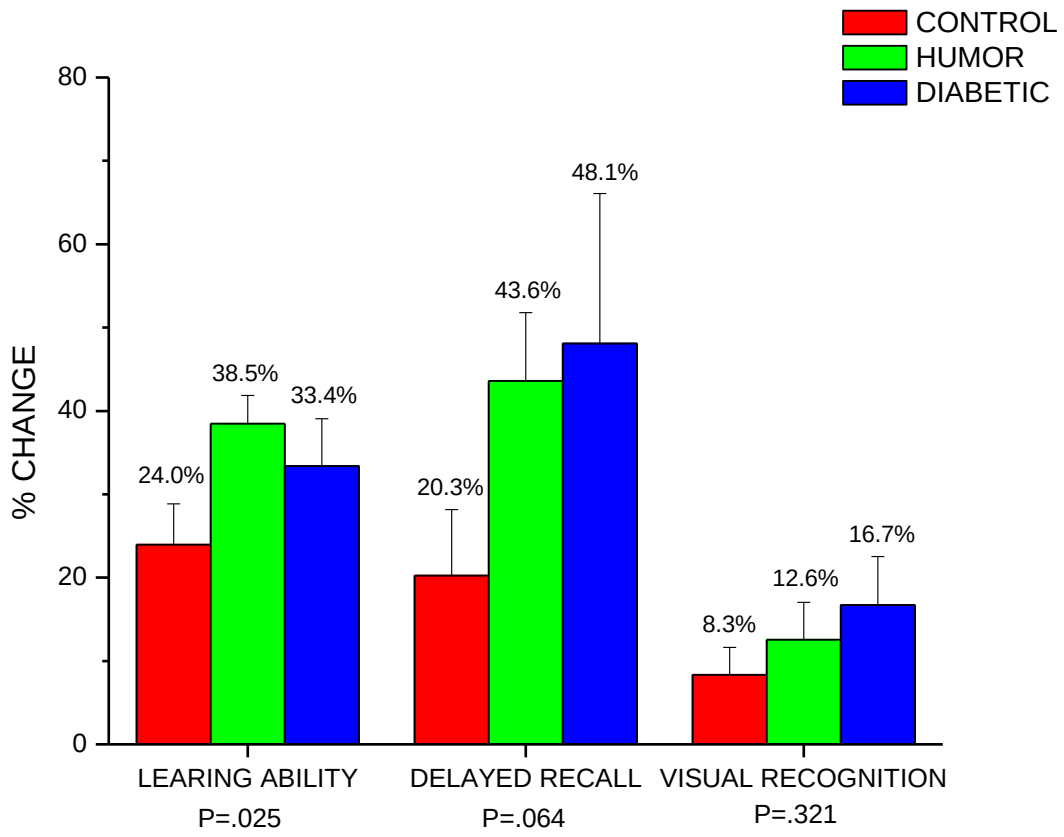

Figure 7. Mean (SD) % Change of RAVLT Scores

When examining the differences between pre and post score percent changes of the RAVLT, there were significant changes in the *learning ability* portion of the test among the three groups (p=.025), as shown in Figure 7. Watching a humor video of choice, when compared to the control group (no video), demonstrated the greatest improvement for the *learning ability* portion of the test. The percent increase in the elderly humor group was more than 1.5 times that of the control group as presented in Figure 7. The elderly humor group showed a 38.5% increase in their *learning* ability after watching a humor video compared to a 33.4 % increase in the elderly diabetic group

and a 24.0% increase in the control group. The impact of the humor video seems to have been most influential in improving *learning ability*.

Furthermore, there were also borderline significant differences comparing changes in *delayed recall* among the three groups (p=.064). As shown in Figure 7, although the control group increased their *delayed recall* by 20.3%, the elderly humor group and elderly diabetic group had greater increases in *delayed recall* by 43.6% and 48.1% respectively. Although there was an increase in the percent change in visual recognition in all three groups, 8.3% in the control and 12.6% in the elderly humor group, and 16.7% in the elderly diabetic group, yet this increase was not significantly different among groups (p=.321).

Percent changes within each group individually for the 3 parts of the RAVLT (A: *learning ability*, B: *delayed recall*, and C: *visual recognition*) are shown in Figure 8. For the control group, the greatest percent change was seen during Part A of the RAVLT: *learning ability* (24.0%) vs. Part B of the RAVLT: *delayed recall* (20.3%); (p=.099). The opposite was observed in the humor group and diabetic group. The humor group had a 43.6% increase in Part B of the RAVLT: *delayed recall* vs. a 38.5% increase in Part A of the RAVLT: *learning ability*; (p=.002). The diabetic group had a 48.1% increase in Part B of the RAVLT: *delayed recall* vs. a 33.4% increase in Part A of the RAVLT: *learning ability*; (p=.090).

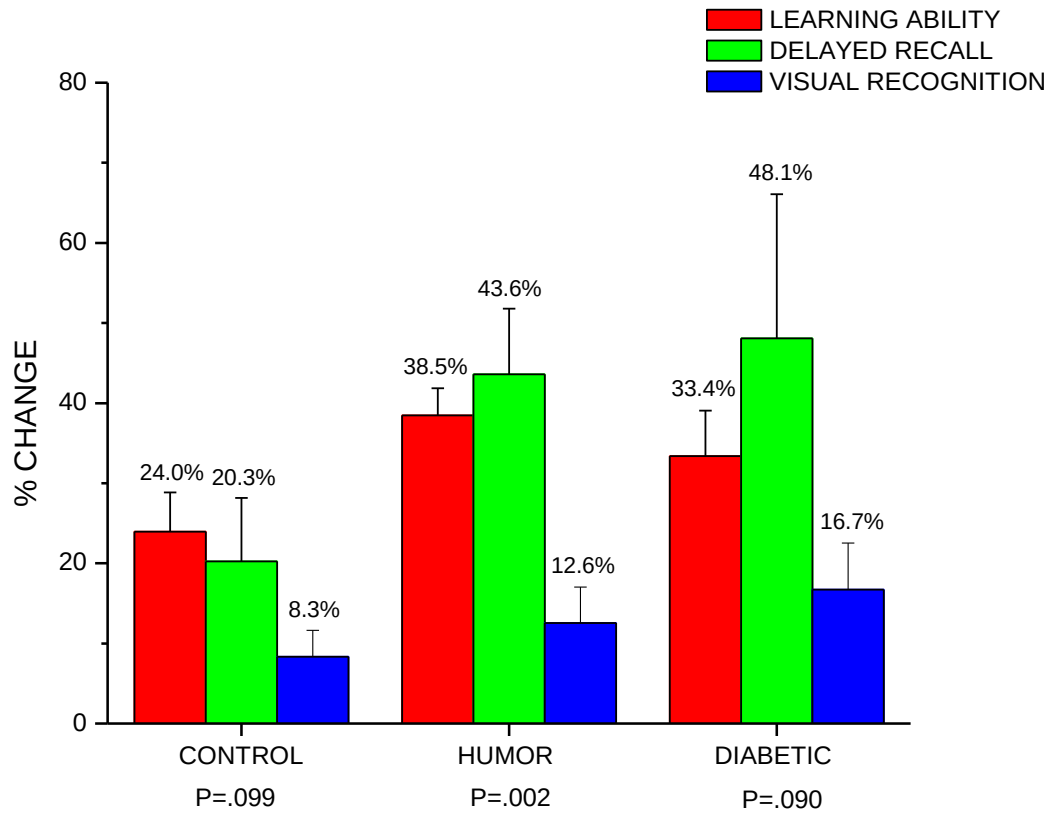

Figure 8. Mean (SD) % Change of RAVLT Scores by Study Group

The Mean (SD) of salivary cortisol level (µg/dL) at the five predetermined time points for all three groups is shown in Table 4. One control subject's cortisol level appeared to be an outlier and after utilizing the Grubbs' Test it was determined to be 2.8 standard deviations away from the mean and thus deleted. Watching a humor video had a significant effect on salivary cortisol levels in the elderly humor and diabetic groups. In both the humor and diabetic group, salivary cortisol levels dropped after watching a humor video. The most relevant drop in the humor and diabetic group's salivary cortisol levels occurred from baseline measurements/pre RAVLT 1 to immediately after watching the humor video (p=.046 and p=.025 respectively). For the control group, during the

same time frame, there was no significant change in salivary cortisol levels (p=.130). In addition, there was a significant change in the salivary cortisol levels for the humor and diabetic group from baseline/pre RAVLT 1 to just immediately before watching the humor video (p=.047 and p=.047 respectively). Also, there was no significant change in the salivary cortisol level during the same time frame for the control group (p=.477). Although, there were significant changes in the control group from baseline/pre RAVLT 1 to immediately after administering the RAVLT 2 (end of study) (p=.047), the greatest change was observed in the diabetic group (p=.035). In addition, there was a significant change in the humor group during the same time frame (p=.047). Pre baseline measurements compared to baseline measurements were not significant in the control, humor, and diabetic groups, (p=.257, p=.242, and p=.242 respectively).

Table 4. Mean (SD) cortisol level (µg/dL) by study group (N=29)

	CONTROL (n=9)	HUMOR (n=10)	DIABETIC (n=10)
(A) Prebaseline	.20 (.08)	.25 (.20)	.18 (.09)
(B) Baseline / pre- RAVLT 1	.20 (.08)	.27 (.20)	.18 (.13)
(C) Post RAVLT 1 / prehumor or quiescence	.22 (.12)	.23 (.14)	.13 (.05)
(D) Post humor or quiescence / pre- RAVLT 2	.17 (.10)	.21(.13)	.12 (.03)
(E) Post- RAVLT 2	.14 (.06)	.17 (.07)	.11 (.03)
	p value*	p value*	p value*
A-B	.257	.242	.242
B-C	.477	.047	.047
B-D	.130	.046	.025
B-E	.047	.062	.034

Abbreviations: SD=Standard Deviation,* Wilcoxon Signed Rank Test, RAVLT=Rey Auditory Verbal Learning Test

Discussion

As a result of the elderly population increasing at a fast rate, there is a growing need to address the challenges of short term memory deficiencies. The damaging effects of aging and stress can impair the ability to learn and sustain memory. Salutogenic whole person elderly wellness programs must include a component on improving short term memory. Previous studies have shown that various interventions such as cognitive stimulation programs, exercise, and multivitamin/herbal supplementation can improve short term memory [16,17,58]. In this study, we examined whether short term memory in the healthy and diabetic elderly can be improved by watching a humor video and

additionally, whether cortisol can be modulated during the course of the study. To our knowledge, this research is the first to examine the effects of humor and its resultant mirthful laughter on the enhancement of short term memory in the healthy and diabetic elderly.

The results showed that the elderly, humor and diabetic groups had a substantial increase in *learning ability* and *delayed recall* when compared to the control group. In addition, our findings showed that pre baseline cortisol levels taken 10 minutes before baseline measurements, taken immediately before the start of RAVLT 1, were not significantly different for the three groups. This suggests that there was no indication of an anticipatory effect at the onset of the memory test for the three groups.

One must ask whether anticipating watching and enjoying humor might lessen the potential stress response associated during a memory test, and therefore, affect test scores. Directly following RAVLT 1, the diabetic and humor groups' cortisol levels significantly decreased. The decrease in cortisol levels may be attributed to the anticipatory effect of the awareness that they will be in a mirthful state. In contrast, in the control group, there was an increase in cortisol levels following RAVLT 1. A stressful state can be either a eustress or distress state. As shown by their decreased cortisol levels, the results for the diabetic and humor groups appear to suggest they underwent a state of eustress while participating in the memory test. On the other hand, as indicated by their increased cortisol levels, the control group seems to have experienced a state of distress.

Despite having the knowledge that a second memory test (RAVLT 2) was expected, cortisol levels, as expected, decreased during the 20 minute time frame when

the diabetic and humor group were watching the humor video, and the control group was seated in quiescence. Furthermore, humor and the associated mirthful laughter decreases cortisol levels just as readily as a quiescent state [20]. More surprising, for the diabetic and humor group, was the effect humor/laughter had on modulating cortisol levels compared to their baseline (Table 4). Cortisol levels for the diabetic and humor group, after watching the humor video, were significantly lower than their baseline levels. Albeit the control groups' cortisol levels decreased, while seated in a quiescence state, it was not significant.

We propose the hippocampus, the site of short term memory consolidation, feasibly endured less cortisol-induced overall suppression in the diabetic and humor groups, possibly explaining the improved cognitive outcomes [5,29]. There was a consistent decrease in cortisol levels, throughout each time point, in the diabetic and humor groups. By the conclusion of the study (completion of RAVLT 2), the diabetic group showed a statistically significant decrease in cortisol from baseline. In addition, the humor group showed a statistically significant - although borderline - decrease in cortisol from baseline measurements (Table 4).

Moreover, we propose the diabetic and humor groups were in a continuous state of eustress. This possibly may justify the greater overall memory test scores in both groups after the humor/laughter session. Significantly greater memory test scores were observed in the diabetic and humor groups, although cortisol levels had decreased in the all three groups during the 20 minute period (Table 4). The impact of watching the humor video had the greatest influence on *delayed recall* in the diabetic and humor groups where the percent increase more than doubled for each group as compared to that of the

control group. Also, the diabetic and humor groups showed a significantly greater improvement in *learning ability* than in the control group.

Additionally, the control group showed, throughout the research study, variations in cortisol levels. Currently, we lack adequate data that can suggest this variability did or did not impact hippocampal effect over cognition. However, we observed that although the control group exhibited a significant decrease in cortisol levels from baseline measurements, this decrease was not associated with increases in memory test scores. Consequently, the control group's *learning ability* and *delayed recall* scores would be expected to be affected.

Within the humor group, *delayed recall* showed a greater significant increase than observed for *learning ability* (note: *delayed recall* was tested after *learning ability*). Following a learning session, delayed *recall* should increase. Due to the continuous eustress state of the humor group, the enhancement to *learning ability* would be expected to translate into better *delayed recall* ability. In contrast, within the diabetic group, there was no significant difference between the increases of *learning ability* and *delayed recall*. The expectation of the enhancement of delayed recall due to the improvement in learning ability was not observed in the diabetic group. Oxidative stress has been linked to cognitive impairment [59]. In the brain, oxidative stress results in a decrease in neurotransmitters,(i.e. acetylcholine), which in turn leads to poor communication between brain neurons. Additionally, acetylcholine plays a major role in the development of memory and learning [60]. We further suggest that, because the RAVLT acted as an acute short term stressor, the diabetic group was not able to overcome the damaging effects of previous advanced glycation end products. This again further

illustrates the importance of having humor therapy as a component in a salutogenic whole person elderly wellness program. In addition, within the control group, possibly due to the continuum of distress, there was no significant difference between the increase of *learning ability* and *delayed recall*. This conclusion supports the assertion that humor may play a role in improving short term memory, specifically, learning *ability* and *delayed recall* in the elderly.

There are potential limitations in this study. The sample size was small; however, based on pilot study results, the current sample size had a 90% power. However, a larger sample size would better represent the older adult population. The variability in cortisol levels may be due to salivary cortisol assay testing. The subjects that participated were limited to the elderly (healthy and diabetic). Future studies should include elderly cohorts with other medical conditions, such as obesity and poorly controlled diabetics. Obese individuals should be involved because obesity has been associated with greater risks for cognitive deficiencies [61].

Conclusions

Our research findings offer suggestive clinical and rehabilitative benefits that can be applied to the salutogenic whole person elderly wellness programs. The cognitive components, *learning ability* and *delayed recall*, become more challenging as we age. *Learning ability* and *delayed recall* are essential to older adults for an improved quality of life: health wise, socially, and economically. Although older adults have age-related memory deficits, complimentary, enjoyable, and beneficial humor therapies should be considered for these individuals.

Acknowledgements

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CHAPTER FOUR

DISCUSSION

The 65 year old and over population is projected to double by the year 2040[1]. As an outcome of the older adult population increasing at this exponential rate, there is an essential need to address the burdens of short term memory deficiencies. The detrimental effects of aging and stress can impair the ability to learn and sustain memory[3,42]. Whole person elderly wellness programs should be incorporated to include enhancements in quality of life in the elderly. An essential component of this salutogenic paradigm must include modalities to improve short term memory. Several interventions such as cognitive stimulation programs, increased daily activities, increased physical activity, and multivitamin/herbal supplementation can improve short term memory[14,16,52]. In this study, I examined whether short term memory in the healthy and diabetic elderly can be improved by watching a humor video and additionally, whether cortisol can be modulated during the course of the study. To my knowledge, this research is the first to examine the effects of humor and its resultant mirthful laughter on the enhancement of short term memory in the healthy and diabetic elderly.

The study results indicated that the two groups that watched humor (healthy elderly and diabetics) had considerable increases in *learning ability* and *delayed recall* when compared to the control group. Furthermore, the findings indicated that pre-baseline cortisol levels obtained 10 minutes before the immediate start of RAVLT 1 (baseline) were not significantly different for the three groups. This strongly indicates

that there was no suggestion of an anticipatory effect at the onset of the memory test for the three groups.

I hypothesized that memory test scores can be affected through the individuals' anticipation that they will be watching and enjoying a humor video and thus, lessen the potential stress response related with undertaking a memory test. Immediately following RAVLT 1, the diabetic and healthy elderly groups' cortisol levels significantly decreased. Although an anticipatory effect was not observed from pre baseline to baseline cortisol measurements, an anticipatory effect was suggestive directly following the administration of RAVLT 1. The decrease in cortisol levels observed may be attributed to the anticipatory effect of the awareness that subjects will be in a mirthful state. However, in the control group, cortisol levels actually increased following RAVLT 1. Stress can be physiologically divided into good stress (eustress) and bad stress (distress). Therefore, a stressful state can be either a eustress or distress state. As indicated by the subjects' decrease in cortisol levels while participating in the memory test, the results for the diabetic and elderly healthy groups seem to suggest they underwent a state of eustress. In contrast, as shown by their increased cortisol levels, the control group appeared to have undergone a distress state.

As expected, cortisol levels decreased during the 20 minutes during which the diabetic and elderly healthy groups were watching the humor video, and the control group was seated in quiescence, while knowing that a second memory test (RAVLT 2) was pending. Additionally, humor and the associated mirthful laughter decrease cortisol levels as quickly as a calm meditative state (quiescence)[39]. However, humor and laugher had a remarkable effect on modulating cortisol levels when compared to their baseline,

for the diabetic and elderly healthy group. After watching the humor video, cortisol levels were significantly lower than baseline levels in both humor groups. Unforeseen, although the control groups' cortisol levels did decrease during their quiescence state, the change was not significant.

It is possible that the hippocampus, which aids in consolidating short-term memory, potentially sustained less overall, cortisol-induced suppression in the healthy elderly and diabetic groups, potentially explaining improved cognitive outcomes for both groups[3,42]. There was a constant decrease in cortisol levels, throughout the study, in the diabetic and elderly healthy groups. By the conclusion of the study (completion of RAVLT 2), the diabetic group showed a statistically significant decrease in cortisol from baseline. In addition, the humor group showed a statistically significant - although borderline - decrease in cortisol from baseline measurements.

Furthermore, it is suggested that, from the data, the diabetic and elderly healthy groups were in a continuous eustress state. This conceivably may justify the greater overall memory test scores observed in both humor groups after the humor/laughter session. Although cortisol levels had decreased in the all three groups during the 20 minute period, significantly greater memory test scores were observed in the diabetic and healthy elderly groups. The impact of watching the humor video had the greatest influence on *delayed recall* in the diabetic and humor groups where the percent increase more than doubled for each group as compared to that of the control group. Also, the diabetic and healthy elderly groups showed a significantly greater improvement in *learning ability* when compared to the control group.

Additionally, the control group showed, throughout the research study, variations in cortisol levels. Currently, there is a lack of adequate data that can suggest this variability did or did not impact hippocampal effect over cognition. However, although the control group exhibited a significant decrease in cortisol levels from baseline measurements, this decrease was not associated with an increase in memory test scores. Consequently, the control group's *learning ability* and *delayed recall* scores would be expected to be affected.

Within the elderly healthy group, *delayed recall* showed a greater significant increase than observed for *learning ability* (note: *delayed recall* was tested after *learning ability*). Following a learning session, delayed *recall* should increase. Due to the continuous eustress state of the healthy elderly group, the enhancement to *learning ability* would be expected to translate into better *delayed recall* ability. In the healthy elderly, the ability to handle the allostatic load of stressors and the plasticity of the hippocampal neurons over time may contribute to the better *delayed recall* ability. In contrast, within the diabetic group, there was no significant difference between the increases of *learning ability* and *delayed recall*. The expectation of the enhancement of delayed recall due to the improvement in learning ability was not observed in the diabetic group. Oxidative stress has been linked to cognitive impairment[53]. In the brain, oxidative stress results in a decrease in neurotransmitters, (i.e. acetylcholine), which in turn leads to poor communication between brain neurons. Additionally, acetylcholine plays a major role in the development of memory and learning[54]. Because the RAVLT acted as an acute short term stressor, the diabetic group was not able to overcome the damaging effects of previous advanced glycation end products. As reported in prior research[48,49], individuals

with T2DM have a decrease in hippocampal volume that can affect cognition and memory. T2DM individuals are in a continuous allostatic overload phenomenon. This overload may explain the insignificant changes in delayed recall and learning ability within the diabetic group. This again further illustrates the importance of having humor therapy as a component in a salutogenic whole person elderly wellness program. In addition, within the control group, possibly due to the continuum of distress, there were no significant increases in *learning ability* and *delayed recall*. This conclusion supports the assertion that humor may play a role in improving short term memory, specifically, learning *ability* and *delayed recall* in the elderly.

In the present research study, there are several limitations. Firstly, the sample size was small. Thirty subjects comprised the initial and final sample size. The control group consisted of 6 males and 4 females with mean age of 68.7 ± 5.5 years; the healthy elderly group consisted of 5 males and 5 females with mean age of 69.3 ± 3.7 years; and, the diabetic group consisted of 6 males and 4 females with mean age of 67.1 ± 3.8 years. However, based on pilot study results, the current sample size provided greater than 90% power. Nonetheless, a larger sample size would be a better representation of the older adult population. Secondly, owing to either to the small sample size or the salivary cortisol assay testing, variability may have been evident in the cortisol levels. It remains to be seen whether a larger sample size will reduce this variability. The subjects we recruited for the study were limited to healthy elderly and diabetic elderly with glucose control. For the diabetic group, the mean diabetes duration was 7.7 ± 6.5 years and the mean HbA1c was 6.8 ± 1.0. Additional cohorts with diverse health issues should be included in future studies. Such cohorts should include obese individuals and diabetics

with poor glucose control. Obese individuals have increased risk for cognitive deficiencies[55]. Thirdly, we did not consider education levels. A highly educated individual may handle stress (i.e., memory test) in a more productive manner and the resultant allostatic overload to homeostasis more easily.

The degree to which an individual, healthy or non-healthy, can handle their allostatic load is dependent on quality of life[56]. Individuals are able to modulate their cortisol levels during stress based on prior experiences, upbringing, and habituation[57]. Social interaction, with family and friends, and having positive self-esteem had been shown to have a beneficial effect on allostasis and thus, a lower allostatic load. Being able to return to homeostasis through a decreased level of allostatic load can lead to an improved quality of life.

Our research study's results advocate humor and the resultant mirthful laughter can potentially have clinical benefits and rehabilitative implications. Whole person wellness programs for older adults should incorporate humor modalities into their salutogenic therapies. For older adults (healthy and diabetic), the most important risk factor for disease is quality of life. Mind, body and spirit need to be in synchrony in order for an improved quality of life. This enhancement will translate into increases in social and economic well-being for older adults. As adults become older, there will be inevitable age-associated memory deficiencies. In addition, numerous challenges, such as difficulties with the cognitive components, learning ability and delayed recall, will become more burdensome. However, healthcare practitioners now can implement positive, enjoyable, and beneficial humor therapies to improve these deficiencies for these individuals, and thus, improve quality of life.

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APPENDIX A

INFORMED CONSENT FORM

LOMA LINDA UNIVERSITY

School of Allied Health Professions

Informed Consent to Participate in Research

"Does therapeutic humor enhance the learning, recall, and recognition of short term memory in elderly and age matched elderly Type 2 diabetics?"

PURPOSE AND PROCEDURES

You are invited to participate in a research study to examine how your short term memory is affected by watching a humorous video. You have been asked to participate because you are an adult in the age range of 55-99 years old with or without Type 2 Diabetes and do not have any known neurological or cognitive disorders. Except for Type 2 Diabetes, you do not have any disorders or disease that affects your memory or learning ability and your hearing is not impaired. You will be given the Mini Mental State Exam (MMSE) which tests for cognitive impairment. You will be asked to participate for one day. If you are an elderly non diabetic, you will be randomly assigned to one of two groups (one that watches a humor DVD or one that does not). We will test your learning ability, recall ability, and recognition ability. This will be accomplished using the Rey Auditory Verbal Learning Test (RAVLT). You will hear 15 words and then be asked to recall the words. This will happen five times. After the fifth time, a new set of words will be given and then you will be asked to repeat the words from that list. And finally, you will be asked to see a list of words and check off the words that you recognize from the original list. After five minutes you will watch two episodes of a humorous video of your choice (we will provide an age matched selection to choose from such as "I Love Lucy", "Victor Borge", "Seinfeld", "America's Funniest Home Videos", "Red Skelton", "Russell Peters", or "The Three Stooges"). Once that is completed, you will take the RAVLT again. We will also be collecting your salivary cortisol samples at 5 time points (15 minutes prior to the RAVLT, immediately before and after the first RAVLT, immediately after the Humor DVD, and immediately after the second RAVLT). The salivary cortisol collection will be done with an oral swab placed under your tongue for 1-2 minutes at each collection time. There will only be one day of data collection per subject (approximately 30 subjects in total). Your day will take about 2 hours for participating in this study. The experiments will take place in room A640 Nichol Hall, Loma Linda University.

RISKS

There are some risks involved in this study, no greater than day to day life. You might feel uncomfortable if you do discover your memory is not as well as you had thought. You might also feel uncomfortable with the salivary oral swab or temporary dryness inside your mouth. There also might be a breach of confidentiality. The Institutional Review Board of Loma Linda University has determined the risks for this study are minimal.

_____Initial
_____Date

A Seventh-day Adventist Institution
DEPARTMENT OF PHYSICAL THERAPY | Nichol Hall, Loma Linda, California 92350
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"Does therapeutic humor enhance the learning, recall, and recognition of short term memory in elderly and age matched elderly Type 2 diabetics?"

BENEFITS

There will probably be no personal benefit to you from participation; however, the information we learn from this research study may benefit the elderly in the future in giving society and the scientific community a way to improve short term memory in the elderly.

PARTICIPANTS RIGHTS

Participation is voluntary. You may leave the study at any time. If at any time during a procedure you experience tiredness or discomfort beyond what you are willing to endure, just tell the person conducting the procedure you want to stop. This decision will NOT affect your standing with those conducting the study or loss of benefits that you are entitled to.

CONFIDENTIALITY

All records will be confidential. Confidentiality will be protected by not disclosing your participation without your written permission. We cannot guarantee absolute confidentiality. Your personal information may be disclosed if required by law. You will not be identified by name in any publications describing the result of this study. Any publication resulting from this study will refer to you by ID number. All data collected will be kept in a locked cabinet in room A640 Nichol Hall, Loma Linda University.

COSTS/COMPENSATION

There is no cost for participating in this study; you will receive monetary compensation for participation in the amount of a $25 gift card.

IMPARTIAL THIRD PARTY

If you wish to contact a third party not associated with the study for any questions or a complaint, you may contact the Office of Patient Relations at Loma Linda University, Loma Linda University Medical Center, Loma Linda, California 92354. Phone (909) 558-4647.

_____Initial
_____Date

Loma Linda University
Institutional Review Board
Approved 7/3/12 Valid to 1/15/2012
#5110305

"Does therapeutic humor enhance the learning, recall, and recognition of short term memory in elderly and age matched elderly Type 2 diabetics?"

INFORMED CONSENT STATEMENT

I have read the contents of the consent form and have listened to the verbal explanation given by the investigator. My questions regarding the study have been answered to my satisfaction. I hereby give voluntary consent to participate in the study described here. Signing this form does not waive my rights nor does it release the responsibilities of the investigator, Gurinder Bains or Loma Linda University of their responsibilities. I may call Gurinder Bains during routine office hours at (909) 558 7274 or leave a voice mail message at this number during non office hours.

_____ _____

Signature of subject Date

INVESTIGATOR'S STATEMENT

I have reviewed the contents of the consent form with the person signing above. I have explained potential risks and benefits of the study.

Signature of investigator _____

Phone Number _____ Date _____

Loma Linda University
Adventist Health Sciences Center
Institutional Review Board
Approved 7/3/12 Valid to 11/15/2012
#5110305 Chair

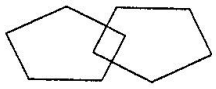
APPENDIX B

MINI MENTAL STATE EXAM

Mini-Mental State Examination (MMSE)

Patient's Name: _____ Date: _____

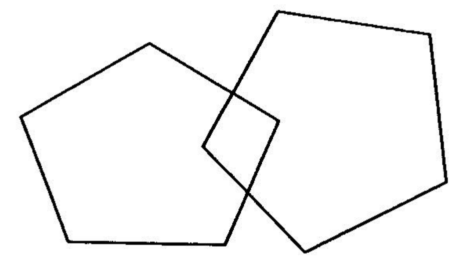
Instructions: ***Ask the questions in the order listed. Score one point for each correct response within each question or activity.***

Maximum Score	Patient's Score	Questions
5		"What is the year? Season? Date? Day of the week? Month?"
5		"Where are we now: State? County? Town/city? Hospital? Floor?"
3		The examiner names three unrelated objects clearly and slowly, then asks the patient to name all three of them. The patient's response is used for scoring. The examiner repeats them until patient learns all of them, if possible. Number of trials: _____
5		"I would like you to count backward from 100 by sevens." (93, 86, 79, 72, 65, ...) Stop after five answers. Alternative: "Spell WORLD backwards." (D-L-R-O-W)
3		"Earlier I told you the names of three things. Can you tell me what those were?"
2		Show the patient two simple objects, such as a wristwatch and a pencil, and ask the patient to name them.
1		"Repeat the phrase: 'No ifs, ands, or buts.'"
3		"Take the paper in your right hand, fold it in half, and put it on the floor." (The examiner gives the patient a piece of blank paper.)
1		"Please read this and do what it says." (Written instruction is "Close your eyes.")
1		"Make up and write a sentence about anything." (This sentence must contain a noun and a verb.)
1		"Please copy this picture." (The examiner gives the patient a blank piece of paper and asks him/her to draw the symbol below. All 10 angles must be present and two must intersect.) 
30		TOTAL

(Adapted from Rovner & Folstein, 1987)

Source: www.medicine.uiowa.edu/igec/tools/cognitive/MMSE.pdf

CLOSE YOUR EYES



(sentence)

T	R	D