**Name(s)**  
Jessica M. Cronin

**Project Number**  
S1301

**Project Title**  
The Effects of Electrolyte Supplements on Endurance Racing

**Objectives/Goals**  
The goal of this project was to determine whether electrolyte supplements had an effect on the performance of the horses at the 2016 Cooley Ranch Ride. The performance was measured by which horses were removed from the race at checkpoints and after the race, and which horses finished. I predicted that the horses' bodies would regulate electrolytes on their own, and that any supplements would only stress the kidneys, therefore the horses that were given electrolyte supplements would perform just as well as the horses that were not.

**Methods/Materials**  
Because this section of the research that was being conducted at the Cooley Ranch Ride was essentially a survey, the only materials that were required were those necessary for recording answers and performing the Chi Square test used to analyze the data.

**Results**  
The Chi Square test told me that there was no statistical difference in my data, therefore there was no significant statistical difference between horses that were given electrolyte supplements and horses that weren't.

**Conclusions/Discussion**  
These results supported my hypothesis, though there is no way of knowing why there was no significant difference, or if more stress was placed on the kidneys and other organs used in electrolyte regulation. The data in my project was taken as part of a research study that was attempting to determine if there was a way of predicting which horses would get sick before they actually did. This is vital to the world of endurance racing because there are a significant amount of horses that get seriously ill after a race due to overworking and dehydration, some horses even die.

**Summary Statement**  
My project represents data collected, at an endurance ride, on electrolyte supplements used, and whether the horses were fit to continue the race.

**Help Received**  
I gathered the information myself, though I was part of a research team lead by Gerry Gillespie, Ph.D., DVM. Gaea Reid and Erin Vaccaro helped me decide how to analyze and present the data, but I performed both Chi Square calculations myself.
# Project Title

**The Effect of Macronutrient Splits on the Loss of Visceral Fat**

## Objectives/Goals
The objective is to determine if there is a difference in visceral fat lost through three different macronutrient splits: normal (40% carbohydrates/30% fats/30% proteins), high carbohydrate (55% carbohydrates/15% fats/30% proteins), and high fat (25% carbohydrates/35% fats/30% proteins.)

## Methods/Materials
Materials required were 15 human subjects, a scale, a computer, and access to Avatar Nutrition. We weighed each subject and found their maintenance calories and fiber requirements. Each of the subjects were split into the 3 splits and told them to maintain their activity level. After 3 weeks of dieting, the subjects were weighed again. After the results, we ran a T-test.

## Results
The average visceral fat lost was .72% through all groups. The normal group lost 1.06%, the high carbohydrate group lost 2.2%, and the high-fat group lost 0% visceral fat on average. The difference between the high carbohydrate and high fat group was the only test proven to be significant through a T-test.

## Conclusions/Discussion
The Normal Diet proved to be in the middle of the other two diets due to it being based off of the average unhealthy American’s diet. The visceral fat loss was just as we expected. In the High fat split, the subjects lost weight as expected but lost more subcutaneous fat than visceral fat. This split did not work for losing much visceral fat because lipids go through a longer process to convert into glucose so it became Visceral fat. The High Carbohydrate diet led to a larger percent in visceral fat loss. In addition, less subcutaneous fat was lost. This could have been because of the fast process of using Carbohydrates as energy. A T-Test was conducted to see if these results were significant but all except one lacked enough evidence to prove significance. This was due to the lack of subjects and not being able to fulfill the Central Limit Theorem. The results still hold value to them and show that the high carbohydrate split led to the greatest loss in visceral fat.

## Summary Statement
Our project found that the high carbohydrate split was the best way to lose the fat held around organs which can help solve many of the health drawbacks that are associated with visceral fat, such as Heart disease and Type 2 Diabetes.

## Help Received
My biology teacher helped us conduct a t-test on the subjects. Layne Norton, PhD in nutrition, reviewed our methods and suggests helpful changes to the experiment so that we could improve it.
# Break a Leg! A Study of the Correlation between Bone Mineral Density and Compressive Strength of Various Mammalian Bones

**Abstract**

All animals within the Vertebrata subphylum develop a skeleton that is essential to life. Unfortunately, bones can fracture under heavy pressure or forceful hits. This project was designed in order to examine whether a higher body weight of a mammal correlates with stronger bones & higher bone mineral density (BMD). If mammalian bones are tested for compressive strength and bone mineral density, then mammals with higher body weights will have stronger and denser bones than mammals with lower body weights.

**Methods/Materials**

Six moose metacarpals, deer metacarpals, coyote tibiae, coyote fibulae, and coyote tibia/fibula articulated pairs were ordered from an online company in Idaho. The mass & anterior surface area of each bone was measured, and BMD was calculated by dividing the mass by the area. Horizontal & vertical compressive strength tests were then performed three times for each set. In the horizontal test, bones were placed flat on a plywood sheet, and weights were stacked & balanced on each bone. 595 lbs. of weight plates were provided for use throughout these tests. For the vertical test, a crusher was constructed to balance weights on the bone. Due to safety limitations, only 552 lbs. could be used in the vertical test. Weight placement ended when an open fracture was observed or the maximum weight was reached.

**Results**

The average BMD from greatest to least were moose metacarpals (3.66g/cm^2), whitetail deer metacarpals (1.76g/cm^2), coyote tibiae (1.10g/cm^2), coyote tibia/fibula pairs (0.96g/cm^2), & coyote fibulae (0.21g/cm^2). The vertical tests matched this order exactly with the averages being 552 lbs., 552 lbs., 492 lbs., 473 lbs., & 6.7 lbs. respectively. In the horizontal tests, the averages were 595 lbs., 420 lbs., 565 lbs., 595 lbs. & 82 lbs. respectively. The average body weight of moose is 800-1,000 lbs. Whitetail deer weigh 155-203 lbs., and coyotes weigh 24-39 lbs.

**Conclusions/Discussion**

The data partially supported the hypothesis, as there were two horizontal discrepancies that occurred due to the anatomy of the bones. Thus, the anatomy of certain bones definitely has an effect on the strength of bones at different areas and angles of pressure. Ultimately, the correlation found between BMD and average body weight can be linked to multiple studies regarding osteoporosis, as a significantly low body weight may be a risk factor for osteoporosis in anatomically susceptible areas of bone.

**Summary Statement**

Bone mineral densities and compressive strengths of various mammalian lower limb bones were found in order to determine a correlation between the two values and confirm that higher body weights are associated with denser & stronger bones.

**Help Received**

Home Depot Staff cut wood for the crusher; Edwards AFB Gym Staff provided 595 pounds of weight plates for use throughout the project; Mrs. Lewis, my Anatomy teacher, helped me brainstorm through the engineering process of the vertical testing; & my family assisted in stacking weights on the bones.
Investigation of a Novel Saccade-Based Diagnostic for Rapid Concussion Detection

Objectives/Goals
In the United States, athletes suffer roughly 3.8 million concussions per year. Unfortunately, many current concussion diagnostics are subjective or easily memorized, which can place athletes in potentially dangerous situations. This research addresses the need for a rapid and accurate method to quantify concussion symptoms by evaluating the ability of a custom-built, field-deployable electrooculogram to detect saccadic eye movements. This study takes the first step in validating the electrooculogram as a potential concussion screening tool by determining whether it has the capacity to accurately identify saccades during a previously validated concussion diagnostic, the King-Devick Test.

Methods/Materials
Based off a design from Matsuzaka et al., an electrooculogram was constructed with a bio-amplifier consisting of four stages: an input stage, a broad-band amplifier, a gain controller, and a final band pass amplifier. Electrodes placed on the subjects' temples fed impulses through the bio-amplifier and into an oscilloscope, which transferred the data to a computer for post-processing. Ten healthy adult subjects were asked to complete two trials of the King-Devick Test while monitored by the electrooculogram. Time to test completion and error rate were recorded during the test, and saccadic amplitude and total number of saccades were isolated post-processing in Matlab r2016b.

Results
The average number of oblique saccades (7.291 ± 0.622) and total saccades (41.958 ± 4.546) recorded among test subjects during each King-Devick Test card correlated with the expected number of oblique saccades and total saccades. The average voltage of the oblique saccades was 0.820 ± 0.318 V, while the average voltage of the horizontal saccades was -0.285 ± 1.117 V.

Conclusions/Discussion
The electrooculogram was able to sensitively detect both the directionality and differentiating amplitude of oblique and horizontal saccades. Additionally, the number of saccades recorded corresponded with the number of expected saccades. These results indicate that the electrooculogram is sensitive enough to accurately detect saccades, and therefore has the potential to act as a concussion diagnostic.

Summary Statement
This study takes the first step in validating a novel concussion screening device, the electrooculogram, by examining the saccadic eye movement of individuals during the King-Devick Test.

Help Received
Lyndia Wu and Calvin Kuo provided supervision as I tested human subjects and advice as I tackled unfamiliar subject areas. Dr. David Camarillo allowed me to conduct trials in his lab.
**Name(s)**
Austin Hartman; Ed van Bruggen

**Project Number**
S1305

**Project Title**
**Know Your AGE: Measuring Advanced Glycation End-products in Your Skin**

**Objectives/Goals**
Advanced glycation end-products (AGEs) are lipids or proteins that become glycated due to exposure to sugars which build up in cells due to aging and life style. They can be predictive markers of degenerative diseases such as diabetes or cardiovascular disease and can be used to differentiate between chronological and biological age. For example, Hemoglobin becomes glycated and is used by doctors to monitor diabetes, but this test requires taking blood and so cannot be performed at home. It was our objective to build a small, affordable, and non invasive device which can be easily used to measure AGEs in the user's skin, and thus predict degenerative diseases, by measuring a specific AGE fluorescence on exposure to ultraviolet light.

**Methods/Materials**
In order to measure AGEs in a user's skin we constructed a lightweight device which uses an ultraviolet LED and a raspberry pi computer to record the autofluorescence of a subject's forearm. The UV excitation had a narrow range of 370 nm and was contained in a light-tight box. A python script on the pi inside triggers the LED to turn on via a basic circuit and proceeds to take a picture with the camera module attached under an opening where the user places their forearm. The image is analyzed on the pi itself for the AGE specific 450 nm excitation. The results are displayed on a screen connected to the device.

**Results**
Our raspberry pi powered device was able to make accurate measurements of the excited AGE molecules in a user's skin which can be used as early predictors to diabetes and cardiovascular disease. We compared old and young volunteers to determine that the skin autofluorescence was reflective of age.

**Conclusions/Discussion**
We were able to achieve our goal of building a small affordable device which can detect AGEs in vivo. The results show that the degree of autofluorescence intensity for skin at 370 nm can be measured and, with more studies, this measurement could be used as a biologic marker of cellular aging in vivo and assess the risk of degenerative diseases.

**Summary Statement**
We designed, built, and tested a simple and affordable device capable of assessing advanced glycation end-products in the skin of human volunteers as a way to determine the risk of age related illnesses such as diabetes.

**Help Received**
No help was received, we designed and tested the device ourselves.
**Project Title**

The Effect of Surface Temperature on Fingerprint Clarity

**Objectives/Goals**

The point of this experiment was to find out if fingerprints are clearer when they are lifted from a hotter or colder surface temperature. It was expected that if a fingerprint is obtained from a surface temperature of 122 degrees Fahrenheit, then it will be clearer on a scale of 1-6: 6 being the most clear and 1 being the least, than a fingerprint obtained from a surface temperature of 32 degrees Fahrenheit.

**Methods/Materials**

The main materials used in this experiment were 6 drinking glasses, a stainless steel bowl, ice, the hot bottom of a coffee maker, a thermometer, powdered sugar, and tape. Three of the drinking glasses were set to cold temperatures (32, 50, and 68 degrees Fahrenheit) and the other three were set to warm/hot temperatures (86, 104, and 122 degrees Fahrenheit), and then fingerprints were lifted off of each glass and their clarity was judged. Then, the same thing was done to a stainless steel bowl, but only one was used unlike the six drinking glasses.

**Results**

The fingerprints lifted off of the cold surface temperatures were clearer. Two of the colder surface temperatures (32 and 50 degrees Fahrenheit) had a higher average clarity rating than the hotter temperatures. These results are the same on both glass and stainless steel.

**Conclusions/Discussion**

The conclusion is that the fingerprints lifted off of the colder surface temperatures were clearer than the ones lifted off of the hotter temperatures because the heat of the hotter temperatures evaporates the oil and water in the fingerprints. This information can be used by officials when obtaining fingerprints at a crime scene.

**Summary Statement**

This project tested how surface temperature affects the clarity of a fingerprint.

**Help Received**

None. I conducted the experiment and did the research by myself.
Krish Kapadia; Anjay Saklecha

Salivary Occludin: A Novel Biomarker for Concussion

Objectives/Goals
Traumatic brain injury (TBI) damages cerebral cells leading to debilitating neurological symptoms. A concussion is mild TBI affecting over 2.5 million people/year. The annual cost of TBI is $76.5 billion as per CDC. Yet, concussion remains a subjective diagnosis, as CT scans are mostly normal. Current research supports blood-based biomarkers as early prognostic indicators of TBI. Saliva is a new, noninvasive diagnostic medium; however, TBI biomarkers in saliva remain unexplored.

We identified Occludin (OCLN), a tight junction protein that maintains the endothelial blood brain barrier (BBB), as an optimal biomarker with high serum concentration and a broad temporal profile. We sought to determine if OCLN levels in saliva correlated with severity of TBI.

Methods/Materials
Saliva was collected within 24 hours of head injury from 10 adult ER patients receiving CT scans and 10 non-injured controls. ER doctors categorized TBI severity. Samples were processed using Aviva ELISA protocol. Spectral analysis was used to measure absorbance.

Results
Median OCLN concentration in saliva of the control group, mild TBI group, and moderate/severe TBI group was 121 pg/mL, 113 pg/mL, and 474 pg/mL respectively. Correspondingly, mean OCLN concentrations were 120 pg/mL ±11%, 115 pg/mL ±8%, and 370 pg/mL ±37%. The moderate/severe TBI group showed a 4-fold increase in the median and a 3-fold increase in the mean OCLN concentration as compared to the mild TBI group.

Conclusions/Discussion
TBI results in disruption of tight junctions of the BBB, and OCLN levels reflect its functional integrity. Measuring OCLN may provide an early indication of the physiological alteration in the brain before changes are visible on CT scan, and can offer an opportunity for earlier intervention. Saliva is easier to obtain than blood or CSF, and tests for OCLN can be repeated more often and at the site of trauma.

This is the first study to show an increase of OCLN in saliva taken from TBI patients. In our limited data set, OCLN clearly separates mild TBI from moderate/severe TBI. While low OCLN level may help avoid overuse of CT scan, elevated OCLN in patients with negative CT scans may indicate the need for close observation. Repeating the study with a bigger cohort will be necessary to help determine statistical significance. Thus, salivary OCLN may serve as an objective decision-making tool for ER physicians to

Summary Statement
This is the first study to use saliva to show that increased occludin can differentiate moderate/severe traumatic brain injury from concussion and has the potential to become the first noninvasive biomarker test of disrupted brain function.

Help Received
We met with ER doctors to understand how concussion patients are evaluated, performed a literature search of biomarkers, curated potential biomarkers, and analyzed our data. We worked with Dr. Podoly (SJ BioCube) to do the experiments and with Dr. Feldman (Good Samaritan ER) to interpret our results.
**Name(s)**
Morgan M. Kopecky

**Project Number**
S1308

### Project Title
**Two Quantitative Colorimetric Screening Tests for the Noninvasive Detection of Type II Diabetes**

### Abstract
The objective of this study was to create a simple test for the early diagnosis of Type II Diabetes using a noninvasive sample. Over 30 million Americans have diabetes and 86 million have prediabetes, while 28% and 89% remain undiagnosed respectively.

### Objectives/Goals
Objectives/Goals

### Methods/Materials
Methods/Materials
Sodium concentration in sweat was identified as a novel biomarker for the detection of diabetes. Two methods to determine the levels of sodium in sweat were devised in this study. The compatibility of sodium assay kit reagents was tested with the stabilizing reagent Trehalose dihydrate. In one detection method, a sodium assay kit was miniaturized to require 40 µl of reagents, a transfer pipette and smartphone. In the second detection method, wax printing was utilized to fabricate a paper sensor. Reagents were stabilized and embedded in the test zones of the sensor to create a semi-automated paper-based screening test. Both screening tests were tested for accuracy and precision with samples containing a range of sodium concentrations. The resulting color change was quantified using ImageJ.

### Results
Results
Sodium assay kit reagents were compatible with Trehalose dihydrate. Wax printing was an inexpensive and rapid method to produce paper sensors. The paper was able to filter out contaminants from samples and the sensor consistently absorbed 7 µl of sample. Reagents stabilized with Trehalose dihydrate maintained activity and can be stored on paper and in aqueous form. ImageJ proved to be an accurate tool to quantitatively analyze the colorimetric results of both detection methods.

### Conclusions/Discussion
Conclusions/Discussion
The first detection method has a standard curve with an R2 value of .9817, costs $1.54, requires a 5 µl sample, and generates results in under an hour. The paper sensor detection method has a standard curve with an R2 value of .9926, requires a 3 µl sample, costs $0.25 and generates results in under one hour.

### Summary Statement
Summary Statement
This study devised two quantitative screening tests for the early detection of Type II Diabetes through the novel use of sweat sodium concentration as a noninvasive biomarker.

### Help Received
Help Received
Ph.D. student at UC Irvine, Neha Garg, supervised use of wax printer. Dr. Robert Edwards allowed use of his laboratory at UCI to store materials and access scientific equipment. Dr. Andres Martinez from California Polytechnic University provided guidance on paper sensor methodology and design.
**Name(s)**  
Nithya Krishnamurthy

**Project Number**  
S1309

<table>
<thead>
<tr>
<th>Project Title</th>
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<tr>
<td><strong>Hippocampal Theta Phase Precession Adapts to Changing Place Fields: Implications for Episodic Memory</strong></td>
<td><strong>Abstract</strong></td>
</tr>
<tr>
<td><strong>Objectives/Goals</strong></td>
<td>The Hippocampus is critical for the formation of new episodic memories in rats and humans. O Keefe's Nobel Prize winning work demonstrated that &quot;place cells&quot; in the CA1 region of the Hippocampus exhibit a theta rhythm with a frequency range of 4-12 Hz. As an animal traverses a cell's firing field, action potentials shift to earlier phases of the theta oscillation, exhibiting phase precession. This study is the first of its kind to assess whether the spiking activity of Hippocampal neurons is consistent with the irregularities in time that exist in human episodic memory. Using a data-set with single neurons with multiple fields and fields of different lengths, I used the phase precession phenomenon to test this hypothesis.</td>
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<tr>
<td><strong>Methods/Materials</strong></td>
<td>I obtained files of data set of animal firing pattern recordings which included the position, and LFP's (local field potential). I filtered the LFP file using MATLAB to isolate the frequency range of 5-10 Hz and wrote a script to calculate the theta phase. I assigned &quot;phases&quot; to each LFP data point, using the fraction of the wavelength at that point. I then created spiral maze with a sample rat's repeated firing fields from a single recorded neuron. Then, I created a script in MATLAB to define each loop and provide an image of the rat's neuronal firing. Subsequently, I calculated the Pearson correlation between theta phase in polar coordinates and the fraction of the field length in linear coordinates. Finally, I ran Kruskal-Wallis statistical test on loop length versus loop number and phase precession versus loop number.</td>
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<tr>
<td><strong>Results</strong></td>
<td>There is strong correlation between loop length and loop number across all the neurons. The Phase precession versus fraction of field length (Loop 4, Loop 3, Loop 2, Loop 1) shows significant correlation between phase precession and loop length and similar correlation across loops.</td>
</tr>
<tr>
<td><strong>Conclusions/Discussion</strong></td>
<td>The strength of phase precession is the same despite the field length which leads to the conclusion that the rate of phase precession adapts to the field lengths. This means that the Hippocampus is capable of compressing memories as they are being laid down. This is a novel finding that sheds light on the fundamental process of memory formation. This work has significant clinical implications, as this form of memory is most susceptible to injury and is the first symptom of dementia such as in Alzheimer's disease.</td>
</tr>
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**Summary Statement**  
Similar experiences of different durations are laid down in memory as if they were the same.

**Help Received**  
I received the data set of the recordings of firing neurons from Douglas Nitz, PhD, Professor, Cognitive Neuroscience at University of California at San Diego. He also provided guidance on implications and reviewed my work which was done at home and my school.
Environmental Enrichment Can Have a Positive or Negative Effect on Long Term Memory

Abstract

Objectives/Goals
The objective of this study is to examine if Environmental Enrichment (EE) encourages learning in FMR1-KO mice, a model for human autism. Another objective is to see if the memory consolidation process of WT mice, mice without defects, can be disrupted via EE.

Methods/Materials
Approximately 30-40 mice total, home cages, Enriched Environment containment, 30x30 white chambers, small clear glass funnels, Ethovision NX, NOT Program. FMR1-KOs and WTs were placed in EE and were then trained and tested in the Object Location Memory (OLM) task. WT mice were put in OLM training, EE, then testing to determine memory disruption and consolidation. Measurements were recorded in how long mice interacted with displaced and stationary glass funnels during OLM in seconds. Both measurements were inputted into the discrimination index formula (100*(displaced time - stationary time)/(total exploration time)) to determine learning scores.

Results
Several mice were trained and tested in the OLM task, with EE placed at different times. FMR1-KO mice put in EE prior were able to perform the OLM task. WT mice weren't able to learn the task if exposed to EE (1, 6 hours) post-training, but could learn if exposed 24 hours post-training.

Conclusions/Discussion
FMR1-KO mice exposed to EE learn effectively as WT mice. It emphasizes that environments can impact learning capabilities. Further studies could include researching the effects of EE on dendritic spine morphology of Fragile X mice. We would predict that EE fixed the dendritic protrusion malformation in KO mice, encouraging cognition. It is also concluded that mice need more uninterrupted time for memory consolidation.

Applying EE to autistic subjects could serve as treatment. Applying EE to subjects with no mental deficits may enhance their learning ability and act as prevention towards mental disorders such as Alzheimer's disease. Students can gauge how much time is needed after studying for consolidation.

Summary Statement
I showed that Environmental Enrichment can increase cognition, but can also be a part of memory disruption dependent on when mice are placed in EE.

Help Received
Lab Technician Aliza Le in the Department of Anatomy & Neurobiology in UC Irvine assisted in handling the mice subjects. I also received advice and discussed results with Dr. Christine Gall from the same institution.
## CALIFORNIA STATE SCIENCE FAIR
### 2017 PROJECT SUMMARY

### Name(s)
Miranda Li

### Project Number
S1311

### Project Title
Impact of Substance Use on Cytokine Levels in Healthy Female Individuals

### Abstract

Objectives/Goals
Substance (tobacco, alcohol) use and various inflammatory diseases (periodontitis, oral lichen planus, leukoplakia) induce chronic inflammation, which is a mechanism for head and neck squamous cell carcinoma (HNSCC). Analysis of salivary cytokine levels reveals abnormal cytokine production, which, if detected early enough, could improve treatment and survival rates for HNSCC. The correlation between substances (tobacco, alcohol, and marijuana) and cytokine levels has not been well researched. Measuring the cytokine levels in saliva samples of healthy, substance using individuals may identify biomarkers of individuals at risk of cancer. This study aims to investigate the correlation between substance use (tobacco, alcohol, marijuana) and cytokine levels (IFN-a, IL-10, IL-12, IL-13, MIP-1a, TNF-a, IL-4, IL-6, IL-8, IL-1a, IL-1b) in healthy female individuals and to determine if there is a role for saliva as a risk factor analytic medium for head and neck cancers.

### Methods/Materials

Samples of saliva, collected from healthy female individuals, were obtained for a previous, unrelated study and stored in a -80°C freezer. Luminex-based multi-analyte MILLIPLEX MAP Human Cytokine/Chemokine Magnetic Bead Kits (Millipore Corp., Billerica, MA) and MAGPIX imaging technology was used to analyze the saliva samples (group 1, controls, n=24; group 2, tobacco and light alcohol use, n=21; group 3, tobacco and heavy alcohol use, n=18; group 4, marijuana and light tobacco/alcohol use, n=8).

### Results

There was a statistically significant difference in cytokine IL-1B levels between the control group (n=24, SD=23.72) and the tobacco/light alcohol user group (n=21, SD=50.97), p<0.05. There was also a statistically significant difference in cytokine interleukin (IL)-8 levels between the control group (n=22, SD=242.71) and the tobacco/heavy alcohol user group (n=18, SD=422.88), p<0.01.

### Conclusions/Discussion

The results of this pilot study suggest that young women who use tobacco and alcohol heavily are already showing signs of chronic inflammation that make them at risk for head and neck cancer later on. These findings also indicate that a saliva-based test could be a cost-effective tool in assisting early diagnosis of head and neck cancer through promising associations of substance use and cytokines.

### Summary Statement

By measuring salivary cytokine levels in healthy female individuals, I identified possible biomarkers for people at risk of head and neck cancers.

### Help Received

The saliva samples and the equipment I used were provided by Dr. Moscicki, the PI of the lab at UCLA where I conducted my research. I designed the experiment, processed the samples, and analyzed the data on my own. Dr. Moscicki and Hazel Huang, the project adviser, also reviewed my work upon completion.
Alexander B. Vu

The Effect of Probiotics on the Passage of Allergens through the Intestinal Epithelium

Objectives/Goals
The incidence of allergies across the U.S. is increasing at an alarming rate. Recent research has suggested the existence of an important connection between allergies and the state of the affected individual's intestinal microbiome. People with allergies are thought to have a more permeable epithelium that permits more allergens to cross the barrier. Probiotics, however, have been shown to decrease the permeability of the epithelial tight junctions. The objective of this study is to determine whether or not probiotic bacteria can decrease the permeability of the intestinal epithelial barrier to allergenic proteins.

Methods/Materials
A caco-2/goblet cell (HT29-MTX) monolayer co-culture (purchase from Admecell, Inc.) grown in transwell inserts was used as a model system for the intestinal epithelium. Lactobacillus acidophilus (Carolina Biological) and Bifidobacterium infantis (commercially available capsules) were the probiotics tested, and a fragment of soybean beta-conglycinin was used as a representative peptide. I labeled the peptide with a fluorescent dye to measure its concentration with a microplate reader. The cell monolayers were exposed to the bacteria for 2 hours, and the transepithelial electrical resistance (TEER) of the monolayers was measured before and after the assay.

Results
I was unable to assess the effects of the probiotics. The TEER of all cell monolayers at the end of the assay was below 40 ohms*cm^2, the minimum resistance of an intact monolayer. The monolayers lost integrity due to cell lysis, likely resulting from the inadvertent addition of a hypotonic buffer solution to the cells. Additionally, the undiluted labeled peptide showed no measurable fluorescence above background noise, indicating that a measurement method at least 1000x more sensitive is needed.

Conclusions/Discussion
In future experiments, the assay buffers need to be properly prepared, another method of measuring peptide concentration must be developed, and various concentrations of bacteria should be tested. However, this model system still shows promise as an in vitro simulation of how probiotics affect the intestinal barrier. Overall, if probiotics such as these are helpful in decreasing intestinal permeability to allergens, they can be used by patients with symptoms of minor food allergies and taken as a preventative measure in case of contact with allergens.

Summary Statement
I evaluated an experimental in vitro model system for studying the effects of two probiotics on the permeability of the intestinal epithelial barrier to a soybean allergen peptide.

Help Received
My scientific research teacher, Dr. Huff, reviewed my research plans and experimental design. I used laboratory facilities available at my high school under her supervision. Materials were purchased by my high school.
Characterization of the Osteochondral Interface by Digital Volumetric Imaging: A 3-D Study of Osteoarthritis

Objectives/Goals
The Osteochondral (OC) interface is composed of the subchondral plate (ScP), the attached articular cartilage (AC), and the calcified cartilage between. Histology has suggested that pores in the ScP are vascular channels or resorption pits, evidence of osteoarthritis (OA) pathogenesis. Previous tests, however, lack sufficient resolution and ability to image in 3-D, which would enable detailed analysis of these features. This study aimed to characterize the OC interface in both normal and OA tissue with Digital Volumetric Imaging (DVI) methods. It is hypothesized that OC interfaces have distinctive features, apparent in DVI, that are distinguishable in OA tissue and indicative of OA pathogenesis.

Methods/Materials
Knee tissue, with three normal (NL) and three clinical OA, were obtained (~4mm³ cube, 50% bone, 50% cartilage around the ScP). Samples were fixed in paraformaldehyde, decalcified in EDTA, stained with Acridine Orange and Eosin Y, and embedded with Spurr resin. Prepared samples were sectioned and imaged through DVI at 10X magnification with a (0.89 μm)³/voxel resolution. The resulting images were processed and then analyzed based on activity, size, and bony cap behavior.

Results
Vascular channels, in addition to the overall structure of the osteochondral interface, were clearly delineated in DVI. Despite similar overall quantity of vascular channels, the OA sample exhibited higher ratio of active to inactive channels, higher incidence of missing/open bony caps, and greater penetration of vascular channels compared to normal osteochondral tissue.

Conclusions/Discussion
This study, with procedures designed and performed by the author and under the surveillance of lab supervisors, elucidates the structural changes in the osteochondral interface that occur during the pathogenesis of osteoarthritis. With 3-D, features such as vascular channels are visible that were previously omitted in 2-D staining methods. With a structural backbone and well-developed understanding of osteoarthritis development, treatments for progressing OA can be pursued.

Summary Statement
This study elucidates the structural changes in the osteochondral interface that occur during the pathogenesis of osteoarthritis through the use of novel imaging techniques and methods.

Help Received
Van W. Wong provided safety training as well as training in standard operating procedures. Neil Chang provided assistance with the Digital Volumetric Imaging machine, and Robert L. Sah facilitated the project as the head of the lab.