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# Orphan Drug Pricing

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## Background

A rare disease is one that affects fewer than 200,000 Americans at any given time.



An orphan drug is a pharmaceutical agent for a medical condition so rare that it would not be profitable to produce without government assistance.

- The median annual cost for an orphan drug in 2016 was over \$32,000. Some are ridiculously expensive.
  - Glybera, treating familial lipoprotein lipase deficiency, is priced at \$1.2 million per dosage. It is approved in Europe where this illness affects 1,200 people. It is not approved in the United States.

The Orphan Drug Act of 1983 was written to spur innovation in rare disease treatment through incentives: 7-year market exclusivity, tax benefits, clinical trial exemptions, and exemptions for application fees by the Food and Drug Administration.

Orphan drug development is complex, especially for orphan drugs. The supply chain from manufacturers, to pharmacies – and the payment chain from payers, including Pharmacy Benefit Managers and insurance companies – is complex with arrangements largely hidden from consumers. It is difficult to determine the relationship between drug prices and costs.

Explicitly, companies tell a story about their pricing – a narrative. A typology of pricing was developed to match with narratives.

## A Typology of Pricing Narratives

Price	Narrative
Low Price, Justified	<ul style="list-style-type: none"> <li>Drug can be used for other diseases (1)</li> <li>Low cost of research and development (R&amp;D) (2)</li> <li>Success in development; few development failures (3)</li> <li>Low cost production after R&amp;D (4)</li> <li>Competition among drugs (5)</li> </ul>
Low Price, Unjustified	<ul style="list-style-type: none"> <li>Pricing below cost to eliminate competition, referred to as predatory pricing, illegal (6)</li> </ul>
High Price, Justified	<ul style="list-style-type: none"> <li>High R&amp;D cost (7)</li> <li>Prices support continuing R&amp;D (8)</li> <li>Acquiring company increases price to support continuing R&amp;D and new drug development (9)</li> <li>Acquiring company brings drug to market through cost-spreading (10)</li> </ul>
High Price, Unjustified	<ul style="list-style-type: none"> <li>Low R&amp;D costs and low prices changed with acquisition (11)</li> <li>Not even priced justifiably to begin with sometimes due to small drug companies selling to large markets increasing the price between sales (12)</li> </ul>

## Findings

- Dysport**, for Cerebral Palsy, experienced sales declines due to importation issues in Brazil. Prices increased when Ipsen and Medicus found additional indications; blepharospasms, aesthetic and other. (1)
- Rifaximi**, for hepatic encephalopathy, is losing money in production towards the orphan disease, though profitable due to indications for irritable bowel syndrome. (1)
- Deflazacort**, for Duchenne muscular dystrophy, was developed by Marathon and criticized for its list price despite it being \$20 or less out-of-pocket for consumers. The controversy forced the sale to a larger drug company that sold the drug for a similar price despite having more flexibility. (10)
- Eloctate**, for hemophilia A, acquired by a larger company, though still not making a profit even at the exuberant marketed price of \$20,000 due to competitors. (10)

## Findings, continued

- Ravicti**, for urea cycle disorders, is priced at \$1,000 for 25mL, the developing company was able to use the orphan drug pricing research and developing costs since there was no generic available. This original developing company, Hyperion, even had an assistance program in place and provided 80% off to customers that did not have insurance or chose not to use a government-sponsored drug plan. The company was then acquired by a larger company, Horizon Pharma providing it with more means to produce more and be capable to lower the price in the long haul which were stated as its intentions when the price was analyzed. (9)

## Conclusions

A key finding is that some drugs fit justified typologies; however, there are also a few that fit typologies for unjustified high prices. Therefore, we need policy changes. Transparency may be necessary to hold companies accountable and allow the public to learn when prices compromise accessibility. Other reforms include prohibiting "pay for delay" tactics that stagnate the introduction of generics into the market and limiting patent extensions strictly due to an improvement or additional benefit in an altered form of a drug. Defining what constitutes a rare disease more rigidly and decreasing coverage under the Orphan Drug Act could also result in lower prices. The ultimate solution to inaccessibility of high price drugs is unclear due to the complexity of rare disease pricing. However, there are steps that could be taken to improve the situation.

Much work remains to identify the narratives for orphan drug prices and to apply the typology.

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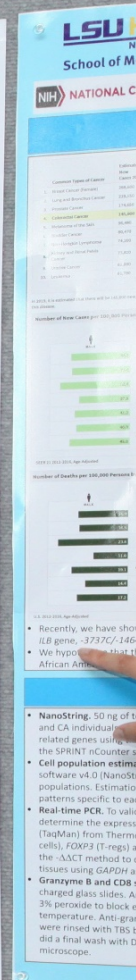
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This research was supported by the Entergy Workforce Training Grant.



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### Introduction

**Figure 1.** Estimated New Cases and Deaths from Colorectal Cancer in 2019 in the U.S.



**Figure 2.** Number of New Cases per 100,000 persons by Race/Ethnicity and Sex: Colorectal Cancer



**Figure 3.** Number of Deaths per 100,000 Persons by Race/Ethnicity and Sex: Colorectal Cancer



Recently, we have shown that African ancestry and pro-inflammatory haplotype the IL1B gene, -3737C/-3464G/-3117/-31C, increases the risk of colorectal cancer (CRC). We hypothesize that the degree and type of immune infiltration in CRC tissues of African American (AA) individuals is different from those of Caucasian American (CA) individuals.

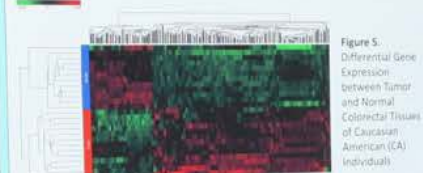
### Methods

- NanoString:** 50 ng of total RNA from tumor and adjacent non-tumor tissues from AA and CA individuals with CRC were used for the unbiased detection of 770 immune-related genes using the CancerImmune panel (NanoString). Detection was done in the SPRINT nCounter system using the Translational Genomics Core.
- Cell population estimation:** Using the "cell type profiling" algorithm in the tSovon software v4.0 (NanoString), we determined the infiltration of different immune cell populations. Estimation of cell population is based on the expression of gene patterns specific to each population.
- Real-time PCR:** To validate the immune cell infiltration, we used real-time PCR to determine the expression of their marker genes. We used primer-probes sets from ThermoFisher to detect S100A2 (PMN), GZMB (exhausted CD8 T[ac]M[em]T), FOXP3 (T-reg) and IL16 as a marker of Th1 inflammatory responses. We used the  $\Delta\Delta CT$  method to determine the relative expression of those genes in tumor tissues using GAPDH as housekeeping gene.
- Granzyme B and CD8 staining:** 5  $\mu$ m slices of tumor tissues of CRC were mounted on tissues using GADPAH as housekeeping gene. Antigen retrieval was performed followed by an immersion in charged glass slides. Endogenous peroxidase and background block at room temperature. Anti-granzyme B and anti-CD8 antibodies were then added. The slides were rinsed with TBS buffer and MACH 4 polymer. We then added a chromogen then a final wash with DI water and 1X TBS buffer. Imaging was captured on a Leica microscope.

### Results



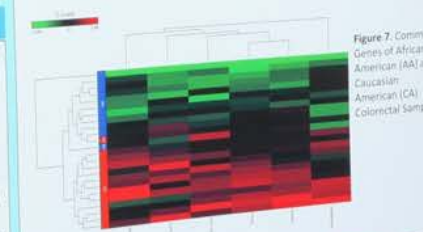
**Figure 4.** Differential Gene Expression between Tumor and Normal Colorectal Tissues of African American (AA) Individuals



**Figure 5.** Differential Gene Expression between Tumor and Normal Colorectal Tissues of Caucasian American (CA) Individuals

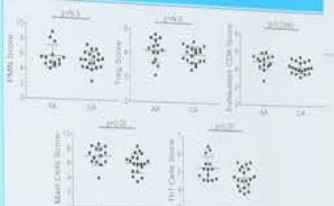


**Figure 6.** Distribution of Significant Genes between African American (AA) and Caucasian American (CA) Colorectal Samples

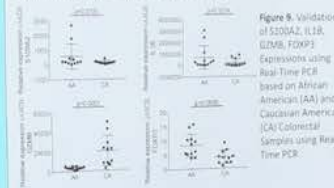


**Figure 7.** Common Genes of African American (AA) and Caucasian American (CA) Colorectal Samples

### Results (cont.)



**Figure 8.** Validation of the Differential Gene Expression between Tumor and Normal Colorectal Tissues of African American (AA) and Caucasian American (CA) Individuals using qSolver software v4.0 (NanoString)



**Figure 9.** Validation of S100A2, IL1B, GZMB, FOXP3 Expressions using Real-Time PCR based on African American (AA) and Caucasian American (CA) Colorectal Samples using Real-Time PCR



**Figure 10.** Immunohistochemistry (IHC) of Colorectal Tissues with staining for CD8 on the top row and Granzyme B on the bottom row

### Conclusions

- Our hypothesis was validated seen in not only clear variations of the common of the AA and CA genes but also in their immune expressions.
- Our work highlights the importance of the immune response in CRC tissues and suggest a differential involvement of immune cell populations associated with AA and CA.
- Additional analysis, including IHC staining on colorectal tissue samples for CD8, granzyme B, and other targets are warranted.

This research project was supported by the National Institutes of Health (NIH), National Cancer Institute (NCI).  
Jenny Paredes, M.S.: NCI Diversity Supplement 5P20CA192994-02S2, Dr. Laura Martello-Rooney and Dr. Jennie Williams: 1P20CA192994-01A1, 3P20CA192994-01A1  
Dr. Jovanny Zabaleta: supported in part by P30GM114732, 1P20GM121288-01, 1P20CA201922-01A1.



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### "Evaluation of the Efficacy of Various Types of Tourniquets Utilizing an Exsanguinating Limb Simulator Model"

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 \*Louisiana State University College of Science  
 \*Tulane University Health Science Center, Department of Surgery



**Background**

**Proximal Model**

**Results**

**Conclusions**



## Evaluation of Makeshift Tourniquet Efficacy on a Simulated Model of an Exsanguinating Limb

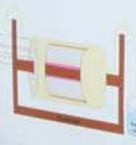
Sara Beaulieu, Rimi Mandal, Patrick Greiffenstein MD  
 \*Louisiana State University College of Science  
 \*Tulane University Health Science Center, Department of Surgery

### Background

Tourniquets are a critical device used in emergency situations to stop bleeding. However, the efficacy of various types of tourniquets is not well understood. This study evaluated the efficacy of various types of tourniquets on a simulated model of an exsanguinating limb.

### Methods and Materials

To test the efficacy of various types of tourniquets, a simulated limb model was used. The model was set up to simulate an exsanguinating limb. The efficacy of various types of tourniquets was evaluated by measuring the amount of blood collected and the time to stop the bleeding.



### Validating the ELS

ELS (Exsanguinating Limb Simulator) was validated by comparing the amount of blood collected and the time to stop the bleeding to that of a real limb. The results showed that the ELS model accurately simulated a real limb.



Figure 1. (A) ELS model with CAT; (B) ELS model with MET; (C) ELS IV bag and pump

**Procedure and Testing for Efficacy**

The procedure for testing the efficacy of the tourniquets was as follows: 1. The ELS model was set up to simulate an exsanguinating limb. 2. A tourniquet was applied to the limb. 3. The amount of blood collected and the time to stop the bleeding were recorded.

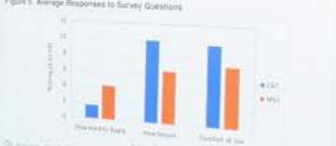


Figure 2. Subject being of tourniquet on the ELS Model

### Results



On average, the MET took 2.5x longer to apply than the CAT. However, the average volume collected for the CAT was 1.5x higher than the MET. The IV bag emptied during the tourniquet application for the simulated limb as the results for subject wearing a MET application were discarded. A trend was indicated for the time until CDF for the CAT and MET. A trend was also indicated for the volume collected for the CAT and the MET. The collected P value for the time until CDF was P=0.00000. The uncollected P value for the volume collected was P=0.00000. The CAT had a 100% success rate and the MET had an 80% success rate. Two subjects failed to do so by the MET.



On average, the CAT was 2.5 times easier to apply than the MET and 1.5 times more secure. The subjects were 1.5 times more comfortable in applying the CAT than the MET. All subjects gave the CAT the highest rating in security.

### Conclusions

- Makeshift tourniquets have an 80% success rate of stopping blood flow but have a 2.5x longer time to apply than the CAT.
- The CAT was 1.5x more secure in stopping blood flow than the MET.
- The CAT was 1.5x more comfortable in applying the CAT than the MET.
- All subjects gave the CAT the highest rating in security.
- The MET had a 100% success rate and the MET had an 80% success rate.
- The CAT had a 100% success rate and the MET had an 80% success rate.
- The CAT was 1.5x more secure in stopping blood flow than the MET.
- The CAT was 1.5x more comfortable in applying the CAT than the MET.
- All subjects gave the CAT the highest rating in security.

This research project was supported through the LSU Health Sciences Center, School of Medicine

# Role of 4E-BP1 and the Unfolded Protein Response in Triple Negative Breast Cancer Cell Survival



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### Introduction

Triple negative breast cancer (TNBC) is an aggressive form of breast cancer. It is characterized by the absence of estrogen receptor (ER), progesterone receptor (PR), and human epidermal growth factor receptor 2 (HER2). TNBC accounts for approximately 15-20% of all breast cancer diagnoses. The prognosis for TNBC is generally poor, with a high rate of relapse and a high mortality rate. The underlying mechanisms of TNBC are not fully understood, but it is thought to be a highly heterogeneous disease. The unfolded protein response (UPR) is a cellular stress response that is activated in response to endoplasmic reticulum (ER) stress. The UPR is a complex signaling pathway that involves several key proteins, including 4E-BP1. 4E-BP1 is a protein that binds to eIF4E, a component of the eIF4F complex, and inhibits its function. This leads to a decrease in the translation of mRNAs that are dependent on eIF4E for their translation. The UPR is thought to play a role in the survival of TNBC cells, and it is hypothesized that 4E-BP1 may be involved in this process.



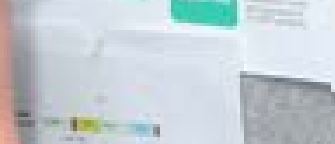
Figure 1: Schematic of the UPR pathway and its connection to 4E-BP1. The UPR pathway involves the phosphorylation of eIF2α, which leads to the inhibition of the eIF4F complex. This inhibition leads to the inhibition of 4E-BP1, which in turn leads to the regulation of the UPR pathway. The diagram also shows that 4E-BP1 is involved in the regulation of the UPR pathway.

### Hypothesis

4E-BP1 controls eIF2α expression through UPR and endoplasmic reticulum stress, ensuring survival of TNBC cells under stress.

### Methods

Cell Culture: TNBC cell lines (MDA-MB-231, BT20, and Hs5787) were cultured in DMEM/F12 medium supplemented with 5% fetal bovine serum (FBS). Cells were treated with thapsigargin (10 μM) for 24 hours to induce ER stress. 4E-BP1 expression was measured by Western blot analysis using anti-4E-BP1 antibody. eIF2α phosphorylation was measured by Western blot analysis using anti-phospho-eIF2α antibody. Cell viability was measured by MTT assay.



### Results

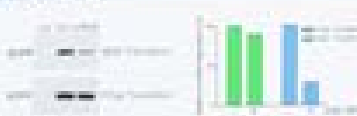


Figure 2: Western blot analysis of 4E-BP1 phosphorylation. TNBC cell lines (MDA-MB-231, BT20, Hs5787) were treated with thapsigargin (10 μM) for 24 hours. The blot shows a significant increase in 4E-BP1 phosphorylation in all cell lines compared to control. A bar graph to the right shows the quantification of 4E-BP1 phosphorylation, indicating a significant increase in all cell lines.

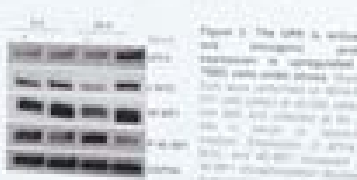


Figure 3: Western blot analysis of eIF2α phosphorylation. TNBC cell lines (MDA-MB-231, BT20, Hs5787) were treated with thapsigargin (10 μM) for 24 hours. The blot shows a significant increase in eIF2α phosphorylation in all cell lines compared to control. A bar graph to the right shows the quantification of eIF2α phosphorylation, indicating a significant increase in all cell lines.



Figure 4: Western blot analysis of 4E-BP1 expression. TNBC cell lines (MDA-MB-231, BT20, Hs5787) were treated with thapsigargin (10 μM) for 24 hours. The blot shows a significant increase in 4E-BP1 expression in all cell lines compared to control. A bar graph to the right shows the quantification of 4E-BP1 expression, indicating a significant increase in all cell lines.



Figure 5: Western blot analysis of 4E-BP1 expression in TNBC cell lines. TNBC cell lines (MDA-MB-231, BT20, Hs5787) were treated with thapsigargin (10 μM) for 24 hours. The blot shows a significant increase in 4E-BP1 expression in all cell lines compared to control. A bar graph to the right shows the quantification of 4E-BP1 expression, indicating a significant increase in all cell lines.



Figure 6: Proposed 4E-BP1 mechanism in TNBC cell survival. ER stress leads to the activation of the UPR pathway, which leads to the phosphorylation of eIF2α. This phosphorylation leads to the inhibition of the eIF4F complex, which in turn leads to the inhibition of 4E-BP1. The inhibition of 4E-BP1 leads to the regulation of the UPR pathway, which in turn leads to the regulation of cell survival.

### Conclusions

- 4E-BP1 is a key component of the UPR pathway and is involved in the regulation of cell survival in TNBC.
- 4E-BP1 controls eIF2α expression through UPR and endoplasmic reticulum stress, ensuring survival of TNBC cells under stress.
- 4E-BP1 is a potential therapeutic target in TNBC.

This research project is supported by NCI P30 CA116126 and P30 CA116122

# Folded Protein Breast Cancer

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Figure 5: Knockdown of 4E-BP1 decreases pS6 and p4E-BP1 levels in MCF-7 cells. Western blot analysis of MCF-7 cells treated with vehicle control (C), 4E-BP1 siRNA (4E-BP1), or 4E-BP1 siRNA + rapamycin (4E-BP1+R). Blots were probed for pS6, p4E-BP1, and GAPDH.



Figure 6: Proposed 4E-BP1 involvement in cellular stress response. ATF4 (UPR) is induced by cellular stress (Hypoxia, No Serum) and leads to decreased p-MYC and p-S6 levels.

## Conclusions

- TNBC cells under cellular stress (hypoxia, no serum) show increased 4E-BP1 mediated production of p-MYC and p-S6.
- 4E-BP1 is involved in normal and tumor cell growth.
- 4E-BP1 is involved in cellular stress response.



# Health Behavior Differences between African-American and White Breast Cancer Survivors

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## Background

- Breast cancer is the most commonly diagnosed cancer among women in the United States.
- It is the second most common cause of cancer deaths in women.
- African-American women with breast cancer suffer worse clinical outcomes than White women with breast cancer.
- Compared to White women, African-American women have a lower median age at breast cancer diagnosis and higher death rates from breast cancer.
- In order to reduce the racial disparity of breast cancer mortality, it is essential to understand demographic and health behavior differences between different racial groups.
- Objective: To evaluate differences in selected health behaviors (physical activity, alcohol consumption, and smoking status) among African-American and White breast cancer survivors.

## Methods

- Among breast cancer survivors, African-American are more likely than White women to have lower levels of physical activity, lower education levels, lower income levels, higher alcohol consumption, higher BMI, and a higher likelihood of being a current or former smoker.
- For this study, we analyzed data from African-American and White breast cancer survivors over the age of 18 in the National Health Interview Survey (NHIS) dataset from 2012-2017. NHIS is a national dataset that routinely investigates a broad range of health topics through personal household interviews.
- Health behaviors of interest included physical activity (sedentary, vigorous), alcohol consumption (never, current light/moderate, or heavy), current or former smoker.
- Other clinical factors included age at breast cancer diagnosis, BMI, education level, and income.
- We examine differences between African-American and White breast cancer survivors in age at interview, age at breast cancer diagnosis, and age at cancer diagnosis.

## National Health Interview Survey



### What is the NHIS?

- The NHIS is the primary source of information on the nation's health.
- As the country's largest continuous household health survey, the NHIS has been taking the pulse of the nation since 1957.
- The Trusted Gold Standard NHIS data are used to:
  - Monitor progress towards national health objectives
  - Develop health policies and programs
  - Track changes in health behaviors and health care use

### Cover Key Topics of National Importance

- Health insurance
- Quality of life goals
- Preventive services and other health behaviors

## Results

### Alcohol Consumption



### Physical Activity



### Smoking



### BMI



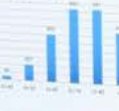
### Income Status



### Education



### Age at Interview



### Age at Breast Cancer Diagnosis



## Results (continued)

Table 1. Chi square analysis for variables of interest

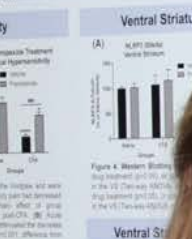
Variable	African-American (n=11,492)	White (n=11,492)	p-value
Physical Activity	65.0%	65.0%	0.999
Alcohol Consumption	45.0%	45.0%	0.999
Smoking Status	45.0%	45.0%	0.999
BMI	50.0%	50.0%	0.999
Income Status	30.0%	30.0%	0.999
Education Level	40.0%	40.0%	0.999
Age at Interview	55.0%	55.0%	0.999
Age at Breast Cancer Diagnosis	55.0%	55.0%	0.999

## Conclusion

- African-American women were more likely than White women to have significantly lower levels of physical activity, alcohol consumption, education, and income.
- African-American women were more likely than White women to have higher BMI.
- Differences in smoking status were significant.
- African-American women were younger than White women.
- At the time of the interview, the average age for African-American women was 56. The average age for White women was 59, so White women tended to be older.
- There was no statistically significant differences in age at breast cancer diagnosis.
- Our study findings showed that African-American breast cancer survivors tended to have worse health behaviors and lower socioeconomic status than White breast cancer survivors.
- Understanding the factors that are associated with breast cancer health disparities are necessary to decrease the morbidity rate in African-American women.

This research was supported by the Entergy Workforce Training Grant.

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**LSU Health**  
NEW ORLEANS  
School of Medicine

# Characterization of Human Amniotic Fluid Stem Cells (hAFSCs)

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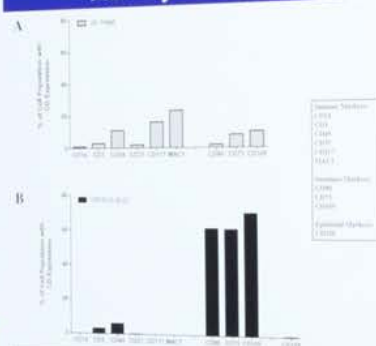
## Introduction

- Amniotic fluid (AF) is an underexplored and readily obtainable source of adult stem cells.
- Human amniotic fluid stem cells (hAFSCs) are now considered a new source for therapy because of their ability to differentiate into multiple cell lineages.
- Stem cells can be isolated from amniotic fluid collected in term from clinical laboratory sections.
- Stemness is determined by adhering to a surface forming colonies, the ability to divide for multiple passages, and the ability to differentiate into multiple cell types.

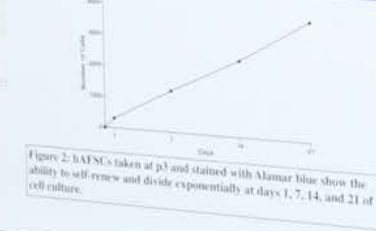
## Methodology



## Flow Cytometry



## Proliferation



## Results

### Colony Forming Unit Assay (CFU)

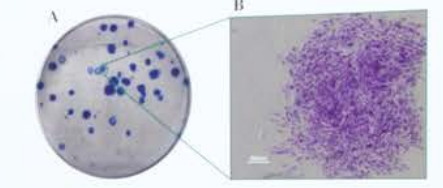


Figure 3: hAFSCs at p3 for CFU. A. Plate shows colonies stained with crystal violet at day 14. B. Morphology of a colony exhibits fibroblast-like cellular morphology.

## Conclusions

- The AF provides a novel source of stem cells with potential use in cellular therapy and regenerative medicine:
- Immune markers decrease with successive passages
  - Stemness markers increase with sequential passages
  - Cells have a fibroblast morphology
  - Proliferation assay shows hAFSCs grow exponentially over a 21 day period

## Future Direction

The continued goal of this study is to confirm stemness by differentiating hAFSCs into microblasts, adipocytes, and chondrocytes.

## Acknowledgments

This research was supported by the Entergy Workforce Training Grant. Thanks to Katelynn Montgomery and team in Dr. Bruce Bunnell's lab.

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# Dopamine Signaling as a Non-Opioid Analgesic Strategy for Chronic Pain

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### Introduction

Precise events in a single neuron maintain chronic pain. However, neurons within the CNS are not only responsive to pain, but also to other stimuli. Dopamine is a neurotransmitter that has been shown to be involved in chronic pain. We used a rodent model of chronic pain to investigate the effects of dopamine on pain. We used a rodent model of chronic pain to investigate the effects of dopamine on pain. We used a rodent model of chronic pain to investigate the effects of dopamine on pain.

### Methods

Animals were divided into saline and CFA groups. Mechanical hypersensitivity was assessed using von Frey filaments. Western blotting was used to measure pTH40, pGluR1, and pNR2B in the VS. Statistical significance was determined using Two-way ANOVA.

### Results

Chronic inflammation (CFA) increased mechanical hypersensitivity. Dopamine treatment (DOPAM) significantly reduced mechanical hypersensitivity. Dopamine treatment also significantly reduced pTH40 and pGluR1 levels in the VS.

### Conclusions

Dopamine signaling may be a potential target for chronic pain treatment. Dopamine treatment significantly reduced mechanical hypersensitivity and pTH40/pGluR1 levels in the VS.

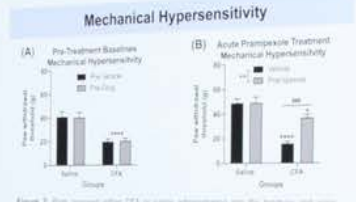


Figure 2: Mechanical Hypersensitivity. (A) Baseline hypersensitivity. (B) Effect of acute pramoxene treatment on hypersensitivity.

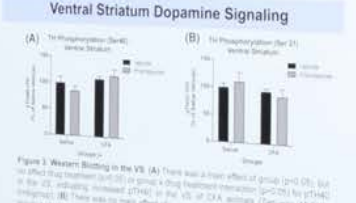


Figure 3: Dopamine Signaling. (A) pTH40 phosphorylation. (B) pGluR1 phosphorylation.

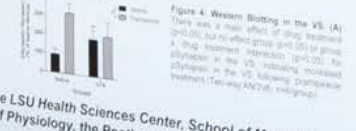


Figure 4: Pre-Synaptic Signaling. pGluR1 phosphorylation in the VS.

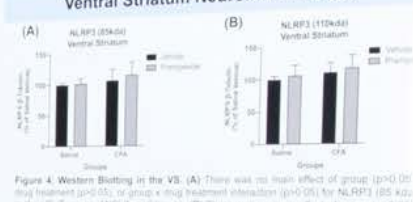


Figure 4: Neuroinflammation. (A) NLRP3 (26kDa). (B) NLRP3 (110kDa).

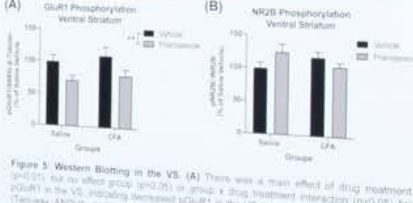


Figure 5: Post-Synaptic Signaling. (A) GluR1 phosphorylation. (B) NR2B phosphorylation.

### Conclusions and Future Directions

- Chronic inflammation increases mechanical hypersensitivity and pTH40/pGluR1 levels in the VS.
- Dopamine treatment significantly reduces mechanical hypersensitivity and pTH40/pGluR1 levels in the VS.
- Future studies should investigate the role of dopamine in pain signaling and the potential for dopamine-based analgesics.

This work was supported through the LSU Health Sciences Center, School of Medicine, and the National Institutes of Health (NIH). We would like to thank the LSUHSC Department of Physiology, the Postbaccalaureate Research Education Program (PREP), and the Edwards lab.

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# The Effects of Adolescent Alcohol Exposure on Stress-Related Pathways and Behaviors



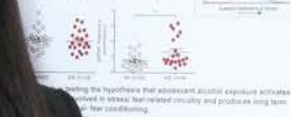
Taylor Collins, Ben Litchfield, Chelsea Kasten, Eleanor Holmgren, Tiffany Wills.  
Louisiana State University Health Sciences Center, Department of Cell Biology and Anatomy, New Orleans, LA.

## Introduction

Individuals who consume alcohol as adolescents have an increased risk of developing alcohol use disorder, but the mechanisms leading to increased vulnerability are unknown.

The BNST is involved in stress and negative affect related alcohol intake.

Previous work using a mouse model of adolescent alcohol exposure (AIE) demonstrated that adolescent alcohol exposure activates glutamate release and plasticity in the bed nucleus of the stria terminalis (BNST).



Supporting the hypothesis that adolescent alcohol exposure activates pathways involved in stress, fear-related circuitry and produces long-term fear conditioning.

## Methods

LSU mice were exposed to two four-day cycles of 16 mg/kg alcohol (AIE) or saline (SAL) in a 2x2 factorial design with 24h withdrawal, separated by 14-day intervals.

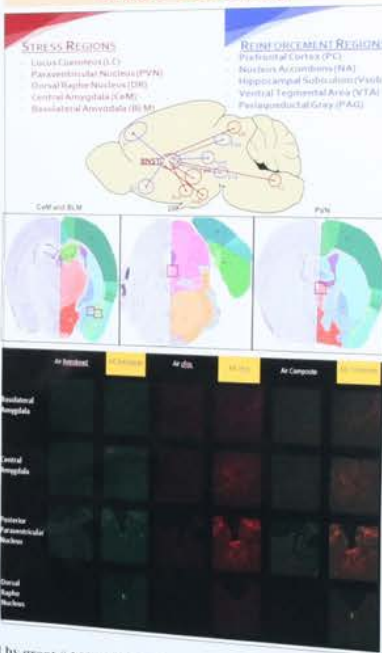
Behavior of mice was recorded with Open Field (OF) and Elevated Plus Maze (EPM) 24 hrs after acute withdrawal from AIE. Anxiety was assessed by recording the number of entries into open and closed arms of the maze.

Behavior of mice was also recorded in contextual fear conditioning (CFC) 24 hrs after acute withdrawal from AIE. Mice were exposed to AIE and then re-exposed to the context (contextual fear conditioning) 24 hrs after acute withdrawal from AIE.

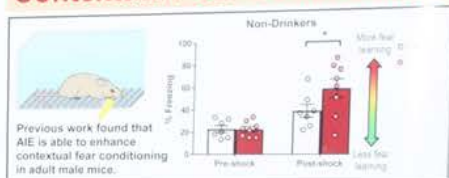
Behavioral data were analyzed using a 2x2 factorial ANOVA. The amount of time spent in each arm of the maze, as well as anxiety-like activity (number of entries into open arms) was recorded.



## BNST Circuitry

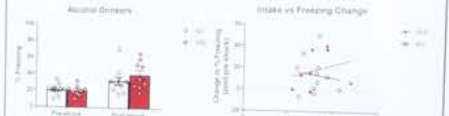


## Contextual Fear Conditioning

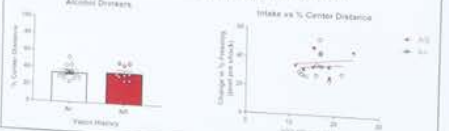


Previous work found that AIE is able to enhance contextual fear conditioning in adult male mice.

Voluntary alcohol drinking following AIE eliminates the enhanced fear conditioning seen in previous AIE experiments. The % freezing was not correlated with voluntary ethanol intake.



AIE does not alter basal anxiety levels. The level of anxiety activity was not correlated with voluntary ethanol intake.



## Discussion

- Retrieval was only strongly expressed in the dorsal BNST nucleus
- This largely serotonergic-activating region has been implicated in depression & negative affect
- Lack of retrieval in other regions may be due to the use of a small, unilateral saline
- AIE mice had extensive Fos in stress regions during withdrawal
- Fos activation activity during withdrawal may contribute to a heightened fear response
- Enhanced fear activity is normalized by voluntary ethanol intake
- Voluntary ethanol intake following AIE withdrawal may normalize increased activity in regions associated with stress and fear

Research project was supported by grant # 1659752 through the National Science Foundation (NSF) and NIAAA (K99/R00 AA022651).  
Research Experiences for Undergraduates (REU) Program





# A heterogeneous population of ventral tegmental area neurons project to the central amygdala

Michael Constans, Elizabeth Avegno, Lucas Albrechet-Souza, Nicholas Gilpin  
Department of Physiology, Louisiana State University Health Sciences Center, New Orleans, LA.

## Introduction

- 16 million Americans are diagnosed with Alcohol Use Disorder (AUD), according to the NIAAA;
- We believe that the circuit between the ventral tegmental area (VTA) and the central amygdala (CeA) plays a role in alcohol dependence.
- The CeA is a brain region associated with stress and is known to play an important role in alcohol dependence associated behaviors.
- The VTA is implicated in alcohol reward.
- One subpopulation of VTA neurons that projects to the CeA has been shown to become activated during alcohol withdrawal, indicating that this circuit plays a role in dependence.
- However, this circuit remains largely under-characterized.
- The goals of this research are to better characterize the expression profiles of these CeA-projecting VTA neurons.



## Hypothesis

- Since early work in our lab has established that only about 30% of CeA-projecting VTA neurons are dopaminergic, it is hypothesized that a substantial amount of glutamatergic CeA-projecting VTA neurons will be observed.

## Methods



Adult male Long Evans rats undergo surgery in which CTxB is injected into the CeA

Brain is sectioned, CeA injection site verified

RNAscope protocol is performed

Images are analyzed on fluorescent microscope

## Mixed populations of VTA neurons project to CeA of naïve rats



Figure 1. 4x image of CTxB (green) in central amygdala (CeA). Nuclei are stained with DAPI (blue). BLA, basolateral amygdala.



Figure 2. 40x representative image of VTA neurons. Cholera toxin (CTxB) containing neurons on right, vesicular glutamate transporter 2 (vGluT2) in middle, tyrosine hydroxylase (TH) on right. Arrow denotes cell containing all three.

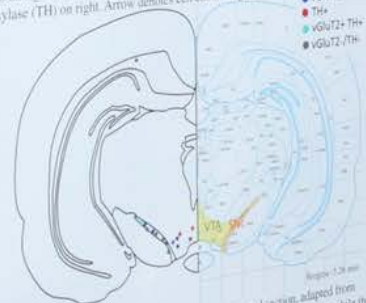


Figure 3. Map of the midbrain-containing coronal section, adapted from Paxinos and Watson (2004). The VTA is highlighted by the yellow while the substantia nigra pars compacta (SNc), which also sends projections to the CeA, is denoted by the orange. Left shows the representative position of CeA-projecting neurons. Data averaged from 1 section per rat, 3 rats.

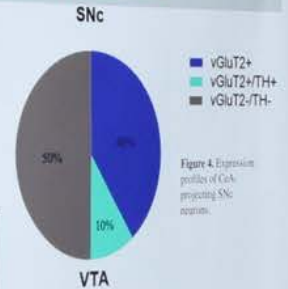


Figure 4. Expression profiles of CeA-projecting SNc neurons.

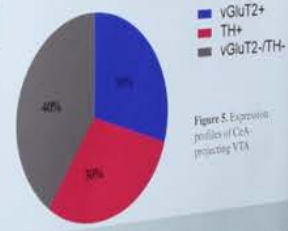
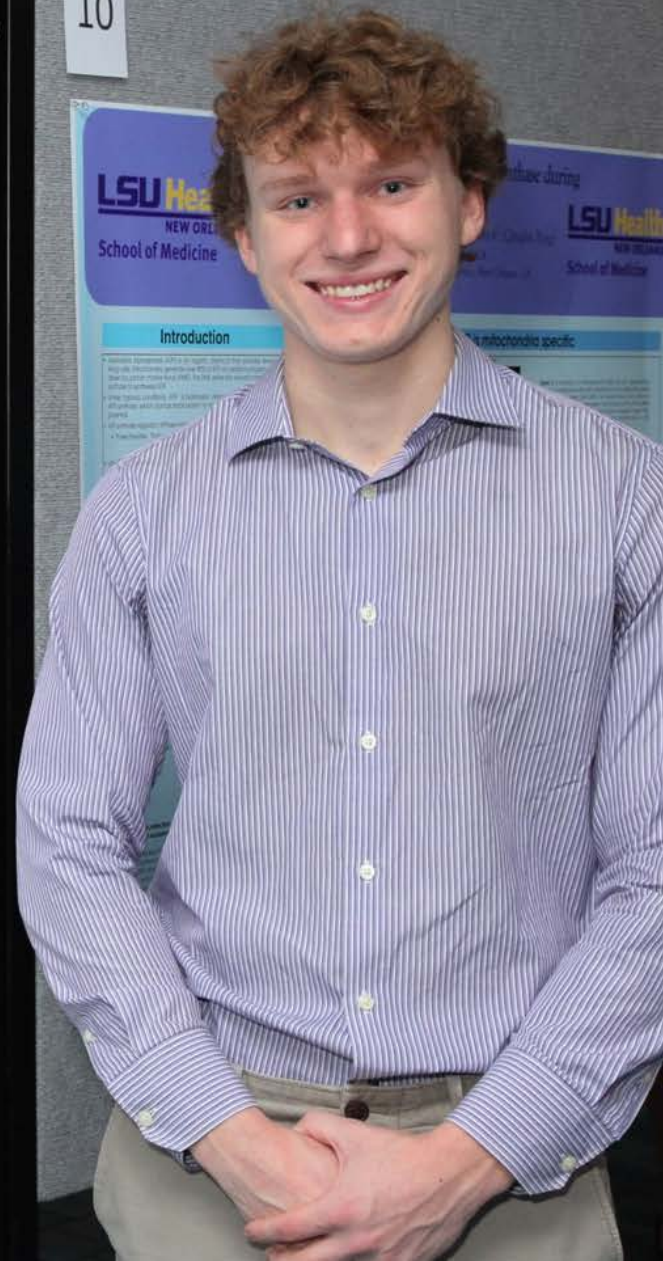


Figure 5. Expression profiles of CeA-projecting VTA neurons.

## Conclusions

- A relatively large number of CeA-projecting neurons were found to be neither glutamatergic or dopaminergic. These are most likely GABAergic.
- There were no CeA projecting VTA neurons that were both glutamatergic and dopaminergic. This was different from expectations as previous studies have shown that around 50-80% of CeA-projecting dopamine neurons also express vGluT2. This could possibly be explained by the small sample sizes.
- As expected, around 30% of CeA-projecting VTA neurons were TH+.
- Future research will involve looking for GABA or additional TH and vGluT2 as well as comparing naïve and dependent rats.
- These findings help better characterize the expression profile of these CeA-projecting neurons and how they affect the CeA downstream.

This research project was supported through the LSU Health Sciences Center, School of Medicine.



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# Direct evidence of IF1 preserves ATP synthase during hypoxia



Dr. John Hendry T. Datta<sup>1</sup>, Luther Lamberbeck<sup>1</sup>, Qinglin Yang<sup>2</sup>  
<sup>1</sup>Tulane University, New Orleans, LA  
<sup>2</sup>LSUHSC, Cadmus Center of Excellence, New Orleans, LA



## Introduction

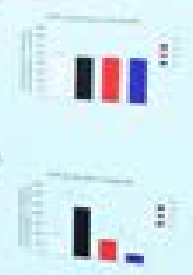
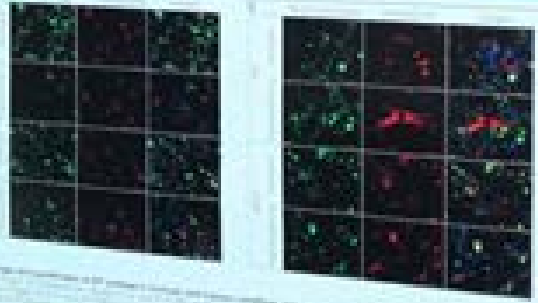
Introduction text describing the background of the research, including the role of ATP synthase and the effects of hypoxia on mitochondrial function.

## mitoMaLionR is mitochondria specific



Figure 1. Mitochondria-specific localization of mitoMaLionR. MitoMaLionR (green) and mitochondrial marker (red) were co-expressed in MEFs. Scale bar, 10 μm.

## ATP production in MEF under normoxia and hypoxia



## Methods



## Summary and Conclusions

Summary and Conclusions text summarizing the key findings of the study, including the role of IF1 in preserving ATP synthase during hypoxia.

This research project was supported through the LSU Health Sciences Center, School of Medicine.

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# ALCOHOL-MEDIATED DYSREGULATION OF MITOCHONDRIAL PROTEIN EXPRESSION IN SKELETAL MUSCLE OF SIV-INFECTED FEMALE RHESUS MACAQUES



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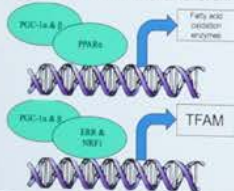
JE Elnaggar, DE Levitt, PE Molina, L Simon

Department of Physiology, Louisiana State University Health Sciences Center, New Orleans, LA

## Background

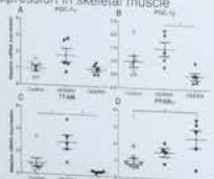
- Rates of heavy drinking in people living with human immunodeficiency virus (PLWH) is almost twice that in the non-HIV-infected population
- At-risk alcohol use contributes to skeletal muscle dysregulation
- Skeletal muscle is a highly metabolic tissue needed to regulate whole-body energy homeostasis
- Mitochondria are essential for skeletal muscle metabolic health
- People are living longer with HIV due to antiretroviral therapy, increasing risk for age-related comorbidities
- Skeletal muscle mitochondrial dysfunction with chronic at-risk alcohol could contribute to metabolic comorbidities in PLWH

Proteins implicated in mitochondrial function:



## Preliminary Data

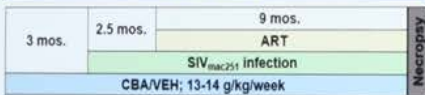
- Chronic binge alcohol dysregulates mitochondrial gene expression in skeletal muscle



## Hypothesis

Chronic binge alcohol alters mitochondrial-related protein expression in skeletal muscle from SIV-infected, antiretroviral therapy-treated female rhesus macaques.

## Methods



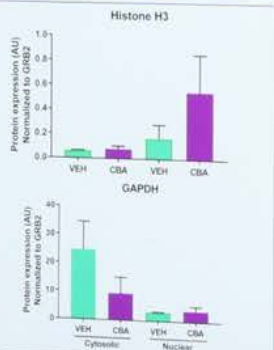
Study design: N=10 female macaques

- Homogenize skeletal muscle
- extract proteins
- Measure protein concentration
- Western blot for PGC-1α & β, PPARα, & TFAM
- Normalize to GRB2

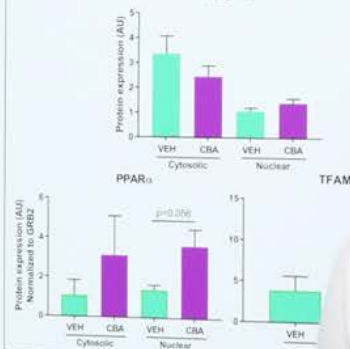
## Cytosolic and Nuclear Fractions

This indicates cytosolic and nuclear-enriched protein fractions

- Histone H3 found in the nucleus
- GAPDH found in the cytosol



## Protein Expression



## Conclusion

- CBA did not alter PGC-1α, PPARα, or TFAM expression
- Trend for CBA to increase PPARα in nuclear fractions
- Future studies include:
  - Measuring mitochondrial function including utilization
  - Measuring expression of proteins down and TFAM
  - Assessing post-translational modifications

This research was supported by the Entergy Workforce Training Grant and by NIH/NIAAA P60AA009803 (PM)



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# The Effects of Traumatic Stress on Reactivity to Acoustic Stimuli in Rats with a History of Alcohol Consumption

Tyvon M. Eugene, Connor Schratz, Lucas Albrecht-Souza, Nicholas W. Gilpin  
Louisiana State University Health Sciences Center, Department of Physiology



## Introduction

- Post-traumatic stress disorder (PTSD) is marked by symptoms of avoidance to experiencing, and hyperarousal that develop subsequent to traumatic events.
- Not only are human beings at risk for developing PTSD, but they also experience altered cognitive and emotional responses with PTSD.
- Acute and Chronic Alcohol (A/C) is commonly comorbid with PTSD. Approximately 40% of people with PTSD also meet the criteria for A/C. PTSD-A/C comorbidity is also associated with a decreased response to treatment, as well as a poorer prognosis when compared to individuals with only one of either of the disorders.

## Objective

- The aim of this study is to evaluate a history of alcohol drinking effects reactivity to acoustic stimuli in male and female rats exposed to trauma.

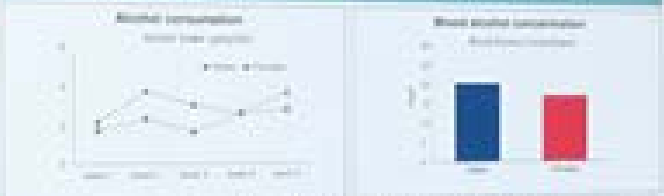
## Methods

- 2-Week Chronic Male and female rats were exposed to alcohol consumption over a period of 2 weeks using an automated two-bottle choice model. During this period, 20% w/vol alcohol was given on Monday, Wednesday, and Friday for a period of 24 h, and restricted along with water consumption.
- Controlled Place Avoidance (CPA) tests were then conducted in the CPA apparatus (Pavlovian CPA) and classified as Avoidant or Non-Avoidant based on proactive versus latency and classified as Avoidant or Non-Avoidant based on their sleep reactivity as measured by the activity of the posture over 24 hours post-avoidance period.

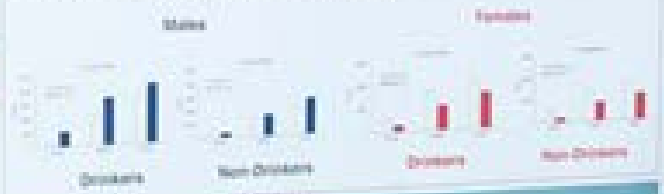


- Acoustic Startle Response (ASR): Two days post-CPA, rats were tested for responsiveness to acoustic stimuli through the use of the ASR. There were 20 trials in which the acoustic stimulus was randomly played at 80, 100, and 115 dB with 30 second intervals. Each decision hour was played a total of 10 times.

## Results



Reactivity to acoustic stimuli in male and female rats exposed to postnatal saline with and without a history of alcohol drinking



## Conclusions

- Males and females who exhibited a tendency to avoid reactivity to the acoustic stimuli were found to be less reactivity to acoustic stimuli when compared to their respective control groups in both sex and conditions, and exhibit comparatively a history of alcohol consumption.
- Male rats with a history of alcohol consumption exhibited greater reactivity to the acoustic stimuli.
- Increased male rats ASR in terms of active avoidance responses greater avoidance of stimuli in response to the sound stimuli in comparison to male rats without a history of alcohol consumption.
- These findings further indicate the existence of significant differences in the expression of male and female in response to stimuli, as well as suggest that alcohol consumption does affect reactivity to acoustic stimuli, despite by changing the magnitude of the response.
- Future Research: Investigate the neurobiology underlying the differences in ASR reactivity between the groups.

This research was supported by the Energy Workforce Training Grant.



# Characterizing Drug Resistant Virus in SIV-Infected Rhesus Macaque Treated with ART

Emma Freeman, Nedra Lacour, Spencer Robichaux, Liz Simon PhD, Angela Amedee PhD.  
 Department of Microbiology, Immunology, and Parasitology and  
 Comprehensive Alcohol Research Center, LSUHSC, New Orleans, LA



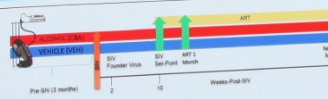
## Background and Rationale

- The SIV-infected rhesus macaque exposed to chronic binge alcohol (CBA) has proven to be a highly useful model for elucidating the effects of alcohol misuse on HIV disease.
- The use of antiretroviral therapy (ART) has significantly reduced the morbidity and mortality from HIV infections and now triple drug therapy is commonly used for treatment of people chronically infected with HIV.
- Drug resistant virus has been observed in ART treated SIV-infected animals with persistent viremia.
- Characterizing and understanding drug resistance (DR) to ART in the SIV-infected CBA macaque is important for the refinement of our model.

## Objective

- The objective of this study was to evaluate viral expression in SIV-infected macaque model fed a Western diet and to compare the efficacy and development of drug resistance with ART-treated macaques from previous studies.

## Study Design



### Animal Cohorts Evaluated

Well-studied Model	Pilot Study Model
21 females (11 CBA; 10 VEH) and 14 males (7 CBA; 7 VEH)	5 females (3 CBA; 2 VEH)
Typical Macaque Diet (5% fat)	Western Diet (20% fat and 27% sucrose)
2 Drug ART Regimen- both reverse transcriptase inhibitors	3 Drug ART Regimen- 2 reverse transcriptase inhibitors and an integrase inhibitor
Injectable weight-adjusted dose: 20 mg/kg ZDV and 30mg/kg FTC	Oral fixed dose (1/2 pill): 25 mg BIC, 100 mg FTC, and 12.5 mg TAF

## Methods

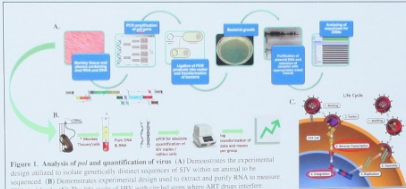


Figure 1. Analysis of pool and quantification of virus. (A) Demonstrates the experimental design utilized to isolate genetically diverse sequences of SIV within an animal in the experiment. (B) Demonstrates experimental design used to reverse and purify RNA to measure SIV viral loads. (C) The life cycle of HIV with critical steps where ART drugs interfere.

## ART Viral Load

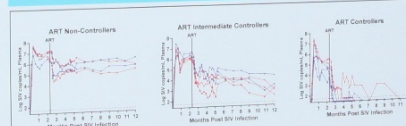


Figure 2. Mean plasma SIV RNA levels over the course of the main study in 7 SIV (black) and 7 CBA-treated macaques (red). Initiation of ART is indicated by vertical line. Level of SIV detection of 50 SIV copies per ml of plasma is indicated by dashed line. Plasma SIV RNA levels over time for individual controller animals, intermediate controller animals, and non-controller controls.

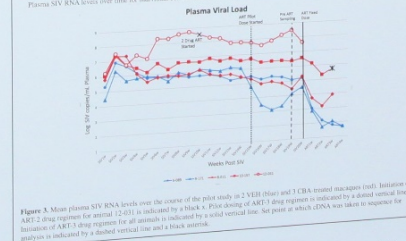


Figure 3. Mean plasma SIV RNA levels over the course of the pilot study in 2 VEH (blue) and 3 CBA-treated macaques (red). Initiation of ART 2 drug regimen for animal 12-01 is indicated by a black x. Prior dosing of ART 1 drug regimen is indicated by a solid vertical line. Initiation of ART 2 drug regimen for all animals is indicated by a solid vertical line. Set point at which cDNA was taken to sequence for analysis is indicated by a dashed vertical line and a black asterisk.

## Drug Resistant Mutations

Table 1. Mutation Analysis of 14 Male Study

Animal	Group	Resistance	Sample	Genotype	ART	Resistant	Prevalence
12-01	VEH	None	12-01-01	CRF01_A1	None	None	0%
12-02	VEH	None	12-02-01	CRF01_A1	None	None	0%
12-03	VEH	None	12-03-01	CRF01_A1	None	None	0%
12-04	VEH	None	12-04-01	CRF01_A1	None	None	0%
12-05	VEH	None	12-05-01	CRF01_A1	None	None	0%
12-06	VEH	None	12-06-01	CRF01_A1	None	None	0%
12-07	VEH	None	12-07-01	CRF01_A1	None	None	0%
12-08	VEH	None	12-08-01	CRF01_A1	None	None	0%
12-09	VEH	None	12-09-01	CRF01_A1	None	None	0%
12-10	VEH	None	12-10-01	CRF01_A1	None	None	0%
12-11	VEH	None	12-11-01	CRF01_A1	None	None	0%
12-12	VEH	None	12-12-01	CRF01_A1	None	None	0%
12-13	VEH	None	12-13-01	CRF01_A1	None	None	0%
12-14	VEH	None	12-14-01	CRF01_A1	None	None	0%
12-15	VEH	None	12-15-01	CRF01_A1	None	None	0%

Table 2. Mutation Analysis of Pilot 5 Female Study

Animal	Group	Resistance	Sample	Genotype	ART	Resistant	Prevalence
12-16	VEH	None	12-16-01	CRF01_A1	None	None	0%
12-17	VEH	None	12-17-01	CRF01_A1	None	None	0%
12-18	VEH	None	12-18-01	CRF01_A1	None	None	0%
12-19	VEH	None	12-19-01	CRF01_A1	None	None	0%
12-20	VEH	None	12-20-01	CRF01_A1	None	None	0%

## Comparison of W...



## Conclusions and

- In our 14 male study (and our 21 animal female study) drug resistant virus was observed.
- Intermediate and non-controllers all harbored drug resistant virus during ART was due to drug resistant virus. Not all resistant mutations.
- Our pilot study also showed a range of responses demonstrating poor control even after ART initiation. Our preliminary analysis has not shown that drug resistant mutations that were previously reported were also seen in our female (2-03) in the pilot study, presumably due to high viral load.
- Comparisons of viral loads in our female and male study have shown that intermediate and non-controller animals have succeeded viral loads.
- Future studies will continue to monitor viral loads in our study to determine whether mutations mapping to determine whether drug resistance response to ART.

This research project was supported through the LSU Health Sciences Center, School of Medicine and Comprehensive Alcohol Research Center.

The Effects of Traumatic Stress in Rats with PTSD

**LSU Health NEW ORLEANS School of Medicine**

Stimuli in Rats with PTSD

Tivon M. Eugene, Connor S. Tivon, Louisiana State University

**Introduction**

Post-traumatic stress disorder (PTSD) is marked by symptoms of avoidance, hyperarousal, and hyperarousal that develop subsequent to traumatic events. PTSD is more prevalent in women than men, but they also experience different symptoms and comorbidities associated with PTSD. PTSD is commonly comorbid with PTSD; approximately 50% of people with PTSD also meet the criteria for AUD. PTSD-AUD comorbidity is also associated with a decreased response to treatment, as well as a poorer prognosis when compared to individuals with only one of either disorder.

**Objective**

The objective of this study was to evaluate the effects of a history of alcohol-drinking affects on the behavior of male Wistar rats exposed to trauma.

**Conclusions and**

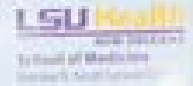
- Stressed rats showed increased alcohol consumption.
- Stressed rats showed increased water consumption.
- Stressed rats showed increased CPA.
- Stressed rats showed increased CPA for Non-Avoiders based on the CPA test.
- Stressed rats showed increased CPA for the predator odor (i.e. fear response).

This research was supported by the National Institute of Mental Health (NIMH) grants R01MH119010, R01MH119015, and R01MH119018.

# Pharmacokinetics of ASO therapy in a mouse model of Usner syndrome

Mark Fisher<sup>1</sup>, Keturik Riddick<sup>1</sup>, Marlene Hallsaeny<sup>1</sup>, Jeremy Dubois<sup>1</sup>, Bakyya Jun<sup>1</sup>, Nicolas G. Beaudry<sup>1</sup>

<sup>1</sup>Translational Center of Excellence and Hersey S. Reid Cancer Center, LSU Health, New Orleans, LA



## Introduction

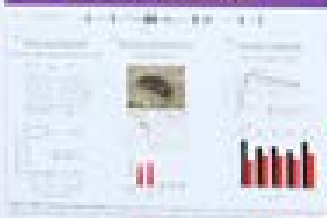
Introduction text describing the study's background and objectives.

## Background

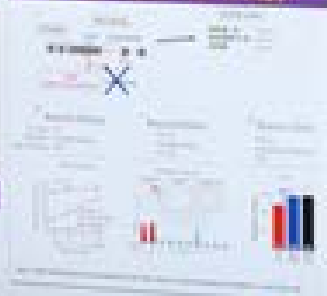
### 1. Acute Usher syndrome



### 2. Usher1C mouse model



### 3. STBA-targeted ASO Therapy



## Results

### 4. ASOs improve Ush1c splitting



### 5. LCMS analysis of ASO



### 5. ASO Quantification/Toxic response



## Conclusions

Conclusions text summarizing the findings of the study.

## Acknowledgements

Acknowledgements text listing funding sources and contributors.

# Estrogenic Regulation of Lysyl Oxidase in Cardiac Fibroblasts

Tierra Foley, Nicholas Fried, Dr. Jason Gardner

Department of Physiology, Louisiana State University Health Sciences Center



## Background

According to the American Heart Association (AHA), cardiovascular disease remains the leading cause of death in the United States, accounting for approximately 650,000 deaths in 2010 and 2011. The AHA reports that approximately 45% of deaths are due to heart disease, which is a leading cause of death in both developed and developing countries. The AHA also reports that heart disease is the leading cause of death in women, accounting for approximately 40% of deaths. The AHA also reports that heart disease is the leading cause of death in men, accounting for approximately 50% of deaths. The AHA also reports that heart disease is the leading cause of death in children, accounting for approximately 10% of deaths. The AHA also reports that heart disease is the leading cause of death in infants, accounting for approximately 5% of deaths. The AHA also reports that heart disease is the leading cause of death in adolescents, accounting for approximately 2% of deaths. The AHA also reports that heart disease is the leading cause of death in young adults, accounting for approximately 1% of deaths. The AHA also reports that heart disease is the leading cause of death in middle-aged adults, accounting for approximately 3% of deaths. The AHA also reports that heart disease is the leading cause of death in older adults, accounting for approximately 15% of deaths. The AHA also reports that heart disease is the leading cause of death in the elderly, accounting for approximately 30% of deaths. The AHA also reports that heart disease is the leading cause of death in the very elderly, accounting for approximately 50% of deaths.



## Hypothesis

Estrogen treatment leads to an increase in Lysyl Oxidase (LOX) activity and expression through the ERα pathway.

## Methods

The cardiac fibroblast cells were collected from 10- to 12-week-old female Sprague-Dawley rats. After collecting the cells were treated by Dulbecco's Modified Eagle Medium (DMEM). The cells were detached, plated, and passaged until they reach 75% confluency. In this point, the cells were divided into 4 treatment groups: estrogen (E2) at 10<sup>-8</sup> M, 10<sup>-7</sup> M, 10<sup>-6</sup> M, and vehicle (ethanol). Cells were plated into a new well-bottomed plates, each plate contained between 100,000 and 200,000 cells. After 24 hours, the cells were collected.

Samples were treated the cells for LOX activity by using a LOX substrate that were provided as LOX substrate. The rate of color change is detected using the Lysyl Oxidase substrate and enzyme only increasing fluorescence that is measured in a fluorescence reader.



## Interpretation

In the present article data we have shown that there is a significant increase in LOX activity with estrogen treatment. Specific agonists of alpha and beta estrogen receptors, PPE and BPE respectively demonstrate that LOX regulation by estrogen is via the alpha estrogen receptor. With the introduction of estrogen, LOX activity increases 50% (p < 0.001) in comparison to the control (ethanol).

## Ongoing Experiments



The cardiac fibroblast cells were collected from 10- to 12-week-old female Sprague-Dawley rats. After collecting the cells were treated by Dulbecco's Modified Eagle Medium (DMEM). The cells were detached, plated, and passaged until they reach 75% confluency. In this point, the cells were divided into 4 treatment groups: estrogen (E2), estrogen + ERα, estrogen + ERβ, estrogen + ERα/β, and vehicle (ethanol). Cells were plated into six well plates with a seeding density of 75,000 cells/plate. ERα and ERβ antagonists had a treatment concentration of 10<sup>-8</sup> M, while estrogen had a treatment concentration of 10<sup>-8</sup> M. After 24 hours, the cells were harvested, fixed, and the media was collected.

The fixed cells will be tested using qPCR analysis of fibroblast marker for estrogen regulation of mRNA. The transcription gene BDNF is used as a housekeeping gene.

## Conclusion

LOX activity is elevated with estrogen treatment, and this elevation is specifically mediated by alpha estrogen receptor agonism. Our hypothesis will be rejected either in part or in totality pending qPCR results of LOX expression.

This research project was supported by grant # 1424902 through the National Science Foundation (NSF), Research Experiences for Undergraduates (REU) Program



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# Effect of norepinephrine on REST expression and subcellular localization in cerebellar interneurons

**LSU Health**  
NEW ORLEANS  
School of Medicine

Jordyn Fong, Jessica Fawcett-Patel, and Sigiong June Liu  
Department of Cell Biology and Anatomy, LSUHSC New Orleans, LA

## INTRODUCTION

REST is a transcription factor that is expressed in a variety of tissues and is known to be involved in the regulation of gene expression. In the brain, REST is expressed in neurons and is thought to play a role in neuroprotection and neurodegeneration. The aim of this study was to investigate the effect of norepinephrine (NE) on REST expression and subcellular localization in cerebellar interneurons. We found that NE treatment significantly increased REST expression and caused it to translocate from the nucleus to the cytoplasm. This suggests that a second messenger pathway is involved in this process.

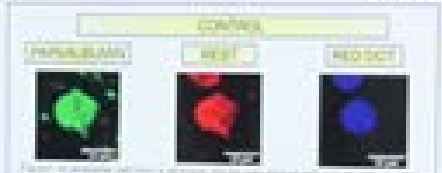
## METHODS

Cerebellar slices were prepared from wild-type mice and treated with NE (100 nM) for 30 minutes. The slices were then fixed and stained for REST (green), DAPI (blue), and a nuclear marker (red). Fluorescence microscopy was used to visualize the subcellular localization of REST. Western blot analysis was performed to quantify REST protein levels. Statistical significance was determined using a t-test.

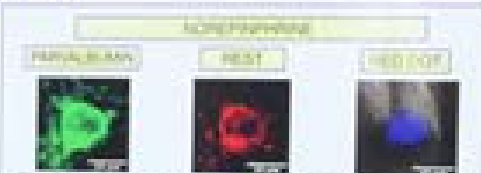
## REFERENCES

1. Fong J, Fawcett-Patel J, Liu SJ. Effect of norepinephrine on REST expression and subcellular localization in cerebellar interneurons. *J Neurosci*. 2018;38(12):3123-3132.

## RESULTS



Fluorescence microscopy images showing the subcellular localization of REST in cerebellar interneurons under control conditions. REST is localized in the nucleus (red) and co-localizes with PAPALBAMIN (green). DAPI (blue) stains the nucleus.



Fluorescence microscopy images showing the subcellular localization of REST in cerebellar interneurons after treatment with norepinephrine (NE). REST (red) has translocated from the nucleus to the cytoplasm. PAPALBAMIN (green) remains in the nucleus. DAPI (blue) stains the nucleus.



## SUMMARY

REST is a transcription factor that is expressed in a variety of tissues and is known to be involved in the regulation of gene expression. In the brain, REST is expressed in neurons and is thought to play a role in neuroprotection and neurodegeneration. The aim of this study was to investigate the effect of norepinephrine (NE) on REST expression and subcellular localization in cerebellar interneurons. We found that NE treatment significantly increased REST expression and caused it to translocate from the nucleus to the cytoplasm. This suggests that a second messenger pathway is involved in this process.

This research project was supported through the LSU Health Sciences Center, School of Medicine.



## Construction of a short-lived fluorescent protein genetic reporter

Hassan A. Hassan<sup>1</sup>, Li Shen MD, PhD<sup>2</sup>

<sup>1</sup>Rivers University of Louisiana, New Orleans, LA  
<sup>2</sup>Department of Microbiology, Immunology, & Parasitology, LSU Health Science Center, New Orleans, LA

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### Abstract

Fluorescent proteins (FPs) have become an essential tool for visualizing biological processes in living cells. However, the traditional GFP-based reporter systems often suffer from long maturation times and high photobleaching rates. To address these issues, we have developed a novel short-lived fluorescent protein genetic reporter system. This system utilizes a genetic circuit that produces a short-lived FP, allowing for real-time monitoring of cellular processes with high temporal resolution. The genetic circuit consists of a promoter driving the expression of a short-lived FP gene, which is then rapidly degraded by a specific protease. This design ensures that the FP signal is transient and accurately reflects the activity of the promoter at the time of measurement. We have demonstrated that this system can be used to study the dynamics of gene expression in response to various stimuli, including chemical and physical signals. The short-lived FP system offers several advantages over traditional GFP-based reporters, including faster maturation times, reduced photobleaching, and the ability to track dynamic changes in gene expression over time. This system represents a significant advancement in the field of genetic reporting and will be valuable for studying a wide range of biological processes.

### References

1. [1] [2] [3] [4] [5] [6] [7] [8] [9] [10] [11] [12] [13] [14] [15] [16] [17] [18] [19] [20] [21] [22] [23] [24] [25] [26] [27] [28] [29] [30] [31] [32] [33] [34] [35] [36] [37] [38] [39] [40] [41] [42] [43] [44] [45] [46] [47] [48] [49] [50] [51] [52] [53] [54] [55] [56] [57] [58] [59] [60] [61] [62] [63] [64] [65] [66] [67] [68] [69] [70] [71] [72] [73] [74] [75] [76] [77] [78] [79] [80] [81] [82] [83] [84] [85] [86] [87] [88] [89] [90] [91] [92] [93] [94] [95] [96] [97] [98] [99] [100]

### Strains and plasmids used

Strain/Plasmid	Description
Strain 1	[Detailed description]
Strain 2	[Detailed description]
Strain 3	[Detailed description]
Plasmid 1	[Detailed description]
Plasmid 2	[Detailed description]
Plasmid 3	[Detailed description]

### Cloning Strategy



### Map of plasmid

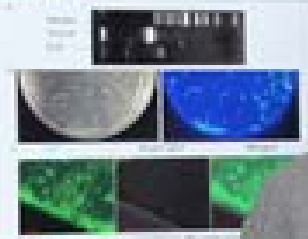


### Agarose gel electrophoresis

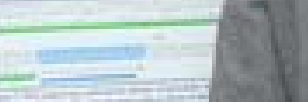


This research project was supported by grant # 5R01NS086568 from the National Institutes of Health (NIH), Research Experience for Undergraduates (REU) Program and NSF grant # 1245479.

### Results



### Sequence of the C-terminus



### Conclusion and future work

The short-lived FP system described here provides a powerful tool for studying dynamic changes in gene expression. Future work will focus on optimizing the system for use in a wider range of cell types and tissues, as well as developing new genetic circuits that utilize the short-lived FP system.

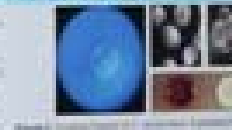
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## Identification of Isoform-Specific Human Pathogenic Fungus Cryptosporidium

Haibo Hill, Ping Wu  
 Department of Microbiology, Immunology, and Parasitology, Louisiana State University Health Science Center, New Orleans, LA, USA

### Introduction

### III. Virulence Factors



### IV. Hematology of C6v1 with ITSP



### V. Construction of C6v1-L and C6v1-H



The C6v1-L and C6v1-H cell lines were constructed by introducing specific genetic modifications into the C6v1 cell line. These modifications were designed to study the role of specific virulence factors in the pathogenesis of Cryptosporidium.

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# Identification of Isoform-Specific Intersectin Mutants in Human Pathogenic Fungus *Cryptococcus neoformans*.



Haley Hill, Ping Wang.

Department of Microbiology, Immunology, and Parasitology, Louisiana State University Health Sciences Center, New Orleans, LA, USA

### Introduction

*Cryptococcus neoformans* is a pathogenic fungus that causes meningoencephalitis and highly infectious lung infections. It varies the evolutionary obtained genes to generate the virulence factors. However, the mechanisms underlying the ability to survive in the host immune system are poorly understood. Intersectin (ITSN1) is a conserved protein that is essential for the survival of the host immune system. It is a conserved protein that is essential for the survival of the host immune system. It is a conserved protein that is essential for the survival of the host immune system.

### III. Virulence Factors

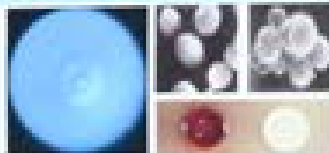


Figure 3. Virulence Factors of *C. neoformans*. A pathogenicity island (PIL) is a cluster of genes that are involved in the virulence of the fungus.

### IV. Homology of Cin1 with ITSN1



Figure 4. Cin1 shows high sequence homology with human ITSN1 protein.

### V. Construction of Cin1-L and Cin1-S

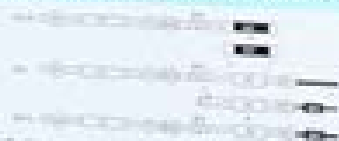


Figure 5. A schematic representation of Cin1-L and Cin1-S construction. Cin1-S was previously obtained by an insertion of the 5'UTR in the 1st Ex. A 116 bp fragment with a CAG/TA-CAT in the 5'UTR was inserted into the 5'UTR of the Cin1 gene to generate Cin1-L. These mutants were confirmed by PCR and sequencing.

### VI. Gel Electrophoresis

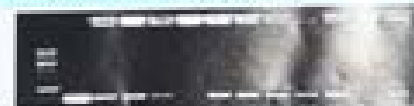


Figure 6. Obtained PCR fragments following cloning PCR of 17 JEC21 transformants. 15 JEC21 transformants were obtained and only obtained.

### VII. DNA Chromatogram



Figure 7. DNA sequencing of JEC21 transformants in 1000bp. First mutation. The diagram on the left is DNA sequence from a transformant that displayed the wild-type sequence. The diagram on the right is DNA sequence from a transformant that displayed the wild-type and Cin1-L gene.

### Results

- The total 17 screened JEC21 transformants displayed either the wild-type or both the wild-type and Cin1-L allele.
- None of the transformants displayed only the Cin1-L allele.
- The remaining 17 JEC21 transformants are being processed.

### Conclusion and Future Directions

- 17 JEC21 transformants are identified amongst the 17 transformants will be obtained and screened.
- Functional impairment of JEC21 mutants, the Cin1-L mutants will be limited only the loss of this.
- A mutant model will be searched for the comparison of Cin1-L to Cin1-S.
- Detailed comparison of the Cin1-L to Cin1-S will yield an improved understanding of the pathogenesis mechanisms of *C. neoformans* and aid in the development of the existing treatment options.

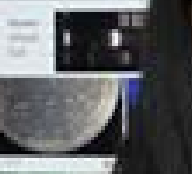
Hill H, Wang P. Phenotypic function of intersectin homologous Cin1 of *Cryptococcus neoformans*. *Mol Microbiol*. 2011; 80:222-30.  
 Wang P, Hill H. The intersectin-like protein of pathogenic lung-infecting yeast and genetic pathway. *Mol Microbiol*. 2011; 80:222-30.

This research project was supported by grant # 0449732 through the National Science Foundation (NSF), Research Experiences for Undergraduates (REU) Program

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Center, New Orleans, LA

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# T. cruzi-induced Changes in Cardiac Endothelial Cells

**LSU Health**  
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School of Medicine

Rebecca Hinojosa<sup>1</sup>, Douglas Johnston<sup>1</sup>  
<sup>1</sup>University of Alabama, Tuscaloosa; <sup>2</sup>LSUHSC New Orleans MP



## Introduction

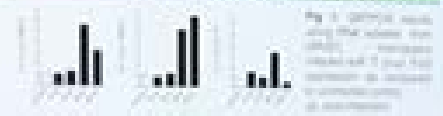
Chagas Disease is a parasitic infection caused by the protozoan *Trypanosoma cruzi* and is a leading infectious cause of heart disease and death. It is estimated that approximately 10 million people are infected worldwide, with 30 million currently at risk. In the United States, approximately 300,000 people are infected, with 10,000 deaths annually. The disease is caused by the parasite's ability to invade and damage heart muscle, leading to heart failure, stroke, and death. The parasite enters the body through contact with the feces of the insect vector, *Triatoma dimembris*, or through blood transfusion, organ transplantation, or congenitally. The parasite enters the heart through the coronary arteries, where it can cause myocarditis and eventually lead to heart failure. The parasite also enters the heart through the coronary veins, where it can cause thrombosis and eventually lead to heart failure. The parasite enters the heart through the coronary arteries, where it can cause myocarditis and eventually lead to heart failure. The parasite also enters the heart through the coronary veins, where it can cause thrombosis and eventually lead to heart failure.



Our lab is working to identify a comprehensive set of markers that can be used to identify and monitor the progression of Chagas disease in the heart. We are using a systems biology approach to identify and monitor the progression of Chagas disease in the heart. We are using a systems biology approach to identify and monitor the progression of Chagas disease in the heart. We are using a systems biology approach to identify and monitor the progression of Chagas disease in the heart.



## T. cruzi induces classic EndMT regulators



## Cell-specific/EndMT marker expression

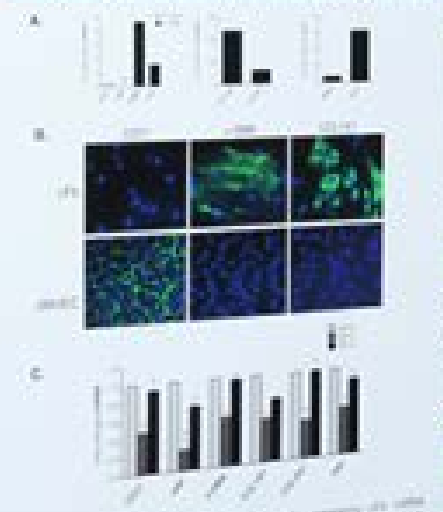


Fig. 4. (A) *T. cruzi* induces the expression of classic EndMT markers (MMP2, MMP9, MMP13, MMP14, MMP15, MMP17, MMP19, MMP20, MMP21, MMP23, MMP24, MMP25, MMP26, MMP27, MMP28, MMP29, MMP30, MMP31, MMP32, MMP33, MMP34, MMP35, MMP36, MMP37, MMP38, MMP39, MMP40, MMP41, MMP42, MMP43, MMP44, MMP45, MMP46, MMP47, MMP48, MMP49, MMP50, MMP51, MMP52, MMP53, MMP54, MMP55, MMP56, MMP57, MMP58, MMP59, MMP60, MMP61, MMP62, MMP63, MMP64, MMP65, MMP66, MMP67, MMP68, MMP69, MMP70, MMP71, MMP72, MMP73, MMP74, MMP75, MMP76, MMP77, MMP78, MMP79, MMP80, MMP81, MMP82, MMP83, MMP84, MMP85, MMP86, MMP87, MMP88, MMP89, MMP90, MMP91, MMP92, MMP93, MMP94, MMP95, MMP96, MMP97, MMP98, MMP99, MMP100) in response to *T. cruzi* infection.

## Co-Culture Infection

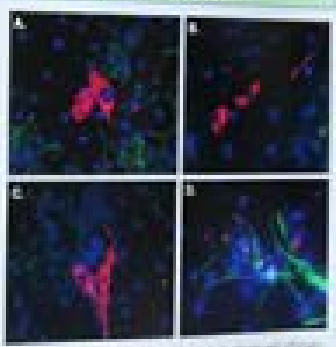


Fig. 5. Co-culture infection of cardiac endothelial cells with *T. cruzi*. (A) Co-culture of *T. cruzi* with cardiac endothelial cells. (B) Co-culture of *T. cruzi* with cardiac endothelial cells. (C) Co-culture of *T. cruzi* with cardiac endothelial cells. (D) Co-culture of *T. cruzi* with cardiac endothelial cells.

## Conclusions

- *T. cruzi* induces the expression of classic EndMT markers.
- *T. cruzi* induces the expression of cell-specific/EndMT markers.
- *T. cruzi* induces the expression of cell-specific/EndMT markers.
- *T. cruzi* induces the expression of cell-specific/EndMT markers.
- *T. cruzi* induces the expression of cell-specific/EndMT markers.

## Acknowledgements

This research was supported by the Energy Workforce Training Grant.

This research was supported by the Energy Workforce Training Grant.

# Arginine-170 is Important in Stabilizing the Active Parkin Oligomer

Kariza Hossain, Jennifer Klein, Virginia Ronchi, Oygul Mirzalieva, and Arthur Haas

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## Introduction

The ubiquitin-proteasome system (UPS) is a major pathway for protein degradation in eukaryotic cells. Ubiquitin is a small, highly conserved protein that is covalently attached to substrate proteins, marking them for degradation by the 26S proteasome. The ubiquitin-proteasome system is involved in a wide range of cellular processes, including cell cycle regulation, signal transduction, and protein quality control. Mutations in ubiquitin ligase genes, such as Parkin, can lead to neurodegenerative diseases, including Parkinson's disease.



## Materials and Methods

Protein expression and purification were carried out using standard protocols. The ubiquitin-proteasome system was assayed using a ubiquitin-amyloid precursor protein (UAP1) substrate. The ubiquitination assay was performed in the presence of different concentrations of Parkin and ubiquitin. The ubiquitinated substrate was analyzed by SDS-PAGE and immunoblotting with anti-ubiquitin antibody.



## Results

Figure 1. Activation of Parkin by PINK1

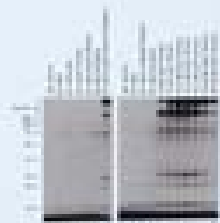


Fig. 1. (A) Western blot analysis of Parkin phosphorylation by PINK1. (B) Bar graph showing the relative intensity of phosphorylated Parkin.

Figure 2. Substrate ubiquitination reveals the E2 ubiquitin binding site



Fig. 2. (A) Line graph showing substrate ubiquitination levels. (B) 3D model of the E2 ubiquitin binding site.

Figure 3. Site-specific inhibition of Parkin activity by Ubiquitin 88-1



Fig. 3. (A) Inhibition of Parkin activity by Ubiquitin 88-1. (B) Inhibition of Parkin phosphorylation by Ubiquitin 88-1.

## Conclusions

Our results demonstrate that Arginine-170 is important for stabilizing the active Parkin oligomer. The ubiquitination assay revealed the E2 ubiquitin binding site, which is essential for Parkin activity. Site-specific inhibition of Parkin activity by Ubiquitin 88-1 suggests that this region is involved in the ubiquitination process.

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3. Ronchi, V. et al. (2012) Substrate ubiquitination reveals the E2 ubiquitin binding site. *Biochem Biophys Res Commun* 421, 123-130.
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This research project was supported by grant # 1R01NS081113 through the National Institutes of Health (NIH). Research Experiment for Learning Initiative (RELI) Program.

and Chlamydia  
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**LSU Health**  
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School of Medicine

# The Role of HPV & EBV in the Detection of Biopsy-Proven Cervical Dysplasia in HIV+ Patients

Phalyn LaBranche, Amber Trauth, MPH,  
Annie Talbot, Michael Hagensee, MD, PhD.

Spelman College  
A College for Women

LSU Health School of Medicine  
**SPIRIT-CHD**  
National Partnership for Promoting Diversity  
& Training in Cancer Health Disparities

## Introduction

A study conducted from 2010-2012 in New Orleans, Louisiana, found that 25% of HIV+ patients had abnormal Pap smears. The prevalence of abnormal Pap smears was significantly higher in HIV+ patients with HPV+ and EBV+ status compared to HIV+ patients with HPV- and EBV- status. The prevalence of abnormal Pap smears was significantly higher in HIV+ patients with HPV+ and EBV+ status compared to HIV+ patients with HPV- and EBV- status.



Figure 1. The presence of HPV greatly reduces the likelihood of being abnormal Pap smear. The presence of EBV in addition to HPV increases the likelihood of being abnormal Pap smear results for HPV+ as compared to HPV+ who were not sexually significant partners.

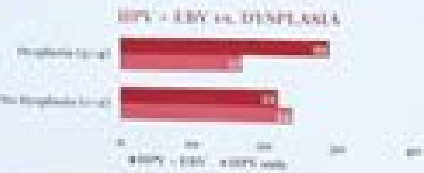


Figure 2. There is a higher likelihood of HPV+ EBV+ patients who also have HPV+ and EBV+ positive Pap smears. There is a higher likelihood of HPV+ EBV+ patients who also have HPV+ and EBV+ positive Pap smears. There is a higher likelihood of HPV+ EBV+ patients who also have HPV+ and EBV+ positive Pap smears.

Worst PAP Smear (out of 148 patients)	Worst Biopsy (out of 148 patients)
Negative: 20 (13.5%)	Negative: 24 (16.2%)
ASCUS: 26 (17.6%)	Mild: 62 (41.9%)
LAI: 44 (29.7%)	Severe: 18 (12.2%)
HSIL: 54 (36.6%)	

## NORMAL BIOPSY VS. ABNORMAL BIOPSY



Figure 3. The presence of HPV+ EBV+ status had more abnormal Biopsies. There is a higher likelihood of HPV+ EBV+ patients who also have HPV+ and EBV+ positive Pap smears.

## DEGREE OF DYSPLASIA



Figure 4. There is a higher likelihood of HPV+ EBV+ patients who also have HPV+ and EBV+ positive Pap smears. There is a higher likelihood of HPV+ EBV+ patients who also have HPV+ and EBV+ positive Pap smears.

## Conclusions

There is a higher likelihood of HPV+ EBV+ patients who also have HPV+ and EBV+ positive Pap smears. There is a higher likelihood of HPV+ EBV+ patients who also have HPV+ and EBV+ positive Pap smears. There is a higher likelihood of HPV+ EBV+ patients who also have HPV+ and EBV+ positive Pap smears.

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This project was supported by the National Institutes of Health (NIH), National Cancer Institute (NCI).

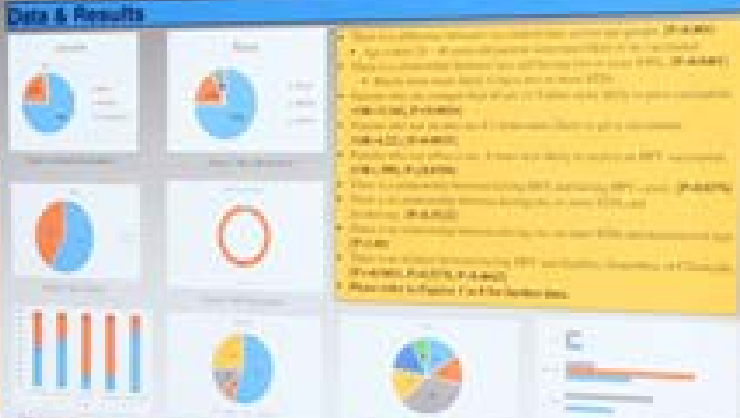
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# HPV-related Cancers, HSV, Syphilis, Gonorrhea and Chlamydia Infections among HIV-Positive Patients at the Emergency Department of University Medical Center New Orleans.

Kelsey Lain, Victoria Lulich, Evrim Oral, PhD, Chieoba Ogbuefi, Zion Rouege, Raj Patel, Keyana Yamada MD, Michael Okonriwo MD, Stacy Rhodes MD, Kanayo Okeke-Eweni MBBS, MPH, Lisa Moreno-Walton MD, MS, MSCR, FAAEM  
USCNC Emergency Department, LSUHSC School of Medicine

### Introduction

HPV-related cancers, HSV, Syphilis, Gonorrhea and Chlamydia infections are common among HIV-positive patients. The Emergency Department (ED) is a critical point of care for these patients. This study aims to determine the prevalence of these infections among HIV-positive patients presenting to the ED at the University Medical Center New Orleans.



### Discussion

The findings of this study indicate that HIV-positive patients present to the ED with a high prevalence of HPV-related cancers, HSV, Syphilis, Gonorrhea, and Chlamydia infections. This highlights the need for comprehensive screening and management of these infections in the ED setting.

Project supported by the National Institutes of Health (NIH), National Cancer Institute (NCI).

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LSU

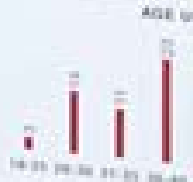
School of Medicine

NATIONAL

Intro

Introduction text describing the study's background and objectives.

Demo







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### Ethanol dysregulates chondrocyte differentiation via different sources of reactive oxygen species in chondrocyte ATDC5 cells

Jonathan Lewis<sup>1</sup>, James Watt<sup>2</sup> and Martin Ronis<sup>2</sup>  
<sup>1</sup>University of New Orleans, <sup>2</sup>Department of Pharmacology, LSU Health Sciences Center

#### Introduction

Chondrocyte differentiation is a tightly regulated process involving a complex network of signaling pathways. Ethanol, a common environmental factor, has been shown to disrupt this process, leading to altered chondrocyte differentiation and increased risk of osteoarthritis. This study investigates the mechanisms by which ethanol dysregulates chondrocyte differentiation via different sources of reactive oxygen species (ROS) in chondrocyte ATDC5 cells.

#### Background

Chondrocyte differentiation is a tightly regulated process involving a complex network of signaling pathways. Ethanol, a common environmental factor, has been shown to disrupt this process, leading to altered chondrocyte differentiation and increased risk of osteoarthritis. This study investigates the mechanisms by which ethanol dysregulates chondrocyte differentiation via different sources of reactive oxygen species (ROS) in chondrocyte ATDC5 cells.

#### Methods

ATDC5 cells were cultured in the presence of ethanol (0, 1, 5, 10, 20, 40, 80, 160 mM) for 24 hours. Cell viability was assessed using a CellTiter-Glo assay. ROS levels were measured using a ROS assay kit. Gene expression levels were determined using quantitative real-time PCR. Western blot analysis was performed to determine protein levels of key signaling molecules.

#### Results




Figure 1: Ethanol treatment leads to a dose-dependent increase in ROS production in ATDC5 cells. ROS levels were significantly higher in cells treated with 10, 20, 40, 80, and 160 mM ethanol compared to the control (0 mM).




Figure 2: Cell viability was significantly reduced in a dose-dependent manner by ethanol treatment. Viability decreased significantly at 10, 20, 40, 80, and 160 mM ethanol concentrations compared to the control.




Figure 3: Gene expression levels for various markers were significantly increased in a dose-dependent manner by ethanol treatment. Expression levels of key markers increased significantly at 10, 20, 40, 80, and 160 mM ethanol concentrations compared to the control.




Figure 4: Western blot analysis showed that protein levels of key signaling molecules were significantly increased in a dose-dependent manner by ethanol treatment. Protein levels of key signaling molecules increased significantly at 10, 20, 40, 80, and 160 mM ethanol concentrations compared to the control.

#### Gene Expression

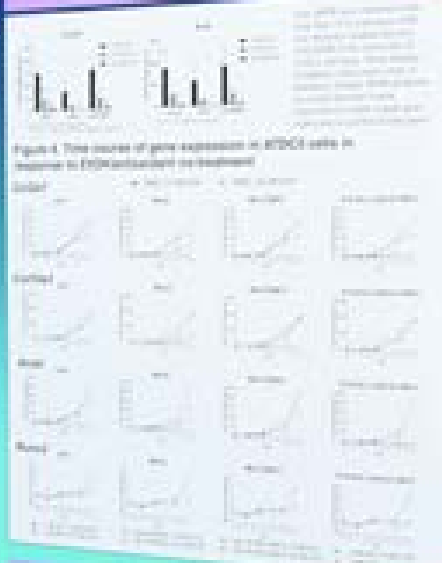


Figure 5: The time course of gene expression in ATDC5 cells in response to ethanol treatment. Gene expression levels were significantly increased in a dose-dependent manner by ethanol treatment. Expression levels of key markers increased significantly at 10, 20, 40, 80, and 160 mM ethanol concentrations compared to the control.

#### Conclusion

Ethanol treatment leads to a dose-dependent increase in ROS production in ATDC5 cells, which in turn dysregulates chondrocyte differentiation via different sources of ROS. This study provides new insights into the mechanisms by which ethanol dysregulates chondrocyte differentiation and highlights the importance of ROS in this process.

This research project was supported by grant # 1407071 through the National Science Foundation (NSF), Research Experience for Undergraduates (REU), Program and NIAAA R21-DA036262 (NLR).

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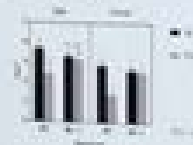
**Background**

Polychlorinated biphenyls (PCBs) are a class of synthetic chemicals that are used in a wide variety of applications. PCBs are known to be toxic to many organisms, including humans. PCBs are known to be endocrine-disrupting chemicals, and they have been shown to interfere with the function of many hormones, including thyroid hormone. PCBs are also known to be neurotoxic, and they have been shown to cause developmental delays in children. PCBs are also known to be carcinogenic, and they have been shown to cause cancer in many animals, including humans.



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**Aim & Hypothesis**

- We hypothesized that Indian Hedgehog (IHH) signaling in the growth plate and marrow regulates bone formation. IHH is upregulated in the marrow from PCB exposure and is essential for PCB-induced bone loss.
- Agonist IHH levels are downregulated and inhibited the IHH signaling pathway, and PCB-induced bone loss is reversed.

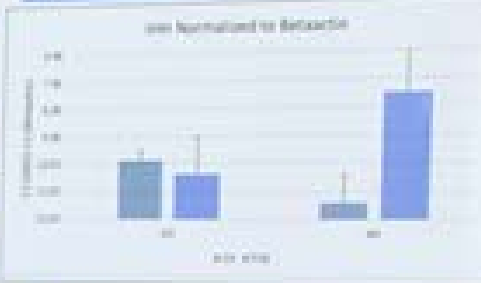
**Experimental Design**



**Figure 1: PCR Primer Product Test**



**Figure 2: IHH real-time PCR Data of Shaft**



**Conclusions**

- From this data, we can see that in the bone samples treated with PCBs (26), Indian Hedgehog is expressed more than control treated with oil.
- We suggest this induction to play a role in the overall smaller bone size seen in PCB-treated animals.
- There is also protection against the induction of IHH by Aryl Hydrocarbon Receptor Knockout mice, showing this is a AHR-mediated process.

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**Acknowledgments**

- LSUHSC, New Orleans
- Dr. Martin Ronis
- Dr. James Watt
- Dr. Kim Peterson
- LSUHSC, Physiology Department
- LSUHSC PIRP
- National Institutes of Health (NIH) R37 AA 018282 (MR)

This research project was supported through the LSU Health Sciences Center, School of Medicine, R28GM112189, National Institutes of Health (NIH) R37 AA 018282 (MR)



Division of the Study of Human Reproductive Function  
Environmental Health Sciences Institute

Adrian Williams, Dr. Jason Park  
LSUHLS, Department of Pharmacology

### Sex & Hypertension

Sex-based differences in the pathogenesis of hypertension

### Experimental Design

Randomized, controlled trial

## Voluntary Ethanol Consumption and Alterations in Reward Circuitry in Ethanol Exposed Adolescent Male Mice

Ben Crawford, Taylor Collins, Deanna Hoangtran, Chelsea Kellum, Tiffany Smith  
LSU Health Shreveport Cancer Department of Cell Biology and Anatomy, Shreveport, Louisiana, LA

### Background

Adolescent ethanol exposure (AEE) is associated with increased risk for substance use disorders and mental health issues. The brain's reward circuitry, including the nucleus accumbens (NAc), is highly sensitive to ethanol exposure during this developmental period.

### Two-Bottle Choice

Adolescent ethanol-exposed (AEE) mice are significantly more likely to consume ethanol than controls.

### Conclusions

AEE increases voluntary ethanol consumption and alters the brain's reward circuitry, including the NAc. These findings suggest that AEE may contribute to the development of alcohol use disorders.

The research project was supported through the LSU Health Shreveport Cancer, School of Medicine and NIAAA (R01 AA022814)

# "Evaluation of the Efficacy of Various Types of Tourniquets Utilizing an Exsanguinating Limb Simulator Model"

<sup>1</sup>Rimi Mandal, <sup>2</sup>Sara Beaulieu, <sup>3</sup>Patrick Greiffenstein  
<sup>1</sup>Tulane University, <sup>2</sup>LSU, LSU Health Sciences Center, School of Medicine, Department of Surgery UMC, Section of Trauma/Critical Care Surgery



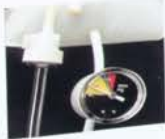
## Background

- Tourniquets are devices that occlude a proximal limb or torso and prevent the blood flow by tightening around major arteries and veins.
- Tourniquets have been in use for centuries and proven effective in hemorrhage control in battlefields and modern intraoperative applications.
- Different types of tourniquets are:
  - The Military Emergency Tourniquet (MCT)
  - Combat Application Tourniquet (CAT)
  - Israeli Silcock Tourniquet (IST)
  - SWAT Tourniquets
  - Special Operations Forces Tourniquet (SOF-T)
- An ideal tourniquet design is supposed to achieve effective circumferential compression of a limb to achieve cessation of distal blood flow.
- New national efforts such as the "Stop the Bleed Campaign" helps provide training opportunity to learn the proper usage of this technique and educate lay people.
- However, testing the efficacy usage of different tourniquets, specifically the CAT and IST, in the lay population has not yet been analyzed.
- The purpose of this study was to evaluate the efficacy of two types of commercially available tourniquets including the CAT and IST tourniquet on an exsanguinating limb simulator (ELS).

## Materials and Methods

The following materials were used to build the ELS model

- Flex Foam IT
- An oak wood long base
- Full containers to mold muscle
- Creativity milk foam sheet
- Zip ties
- Eggsperware
- Aluminum foil
- Tubing to act as veins
- IV pump
- Escoflex 0020
- Escoflex 0050
- Excellite clear cast tubing
- HD Alaris Pump infusion set
- Legs Everyday pantyhose
- Red fabric
- Wooden sticks



- After the model was built, the study participants (N=10) were simula and data was collected on:
  - time it took to tighten the tourniquet on the ELS model until cessation of flow
  - speed of application of tourniquets on ELS
  - subjective ease of use of the CAT and IST
- The systolic blood pressure was set to 150 (mmHg) to the IV bag and kept constant throughout the experiment.

## Preliminary Model



Figure 1. Above is the preliminary model that is first made out of silicone rubber and foam sheets to test out the materials that mimic the exsanguinating limb simulator model. The goal was to replicate the same thickness and texture in muscle groups to the thigh.

## Experimental Design of the ELS model



Figure 2. To the left there is a picture of the CAT tourniquet, Israeli Tourniquet, and the IV pump that was used to provide a constant flow. The picture to the right shows the interior design of the thigh model made from commercially available materials.

## Description of the ELS model

Validating the ELS model as a human limb

- In order to validate the simulated model, three subjects were asked to test out the tourniquets
- Before starting the process, we made sure that each of the participants knew about the project, have previous knowledge of applying a tourniquet, the purpose, and the data we were trying to collect

Figure 3. Final product of the ELS. Attached to the model is the CAT tourniquet and IV pump.



Figure 4. The results for the validation of the ELS model. Subjects were asked to test the CAT and IST. The times from the point they picked up the tourniquet until cessation of flow.

## Results

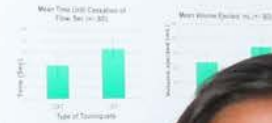


Figure 4. Mean times for cessation of flow on CAT and IST.

Table 1. Demographics of the Study Participants

Age (years)	Range 20-30
Gender	50% Male, 50% Female
Occupation	50% Student, 50% Professional

## Conclusion

Based on the results of the preliminary model, we performed a study on the efficacy of the CAT and IST. All of the participants were able to apply the CAT and IST. The CAT was found to be more effective in stopping the flow of blood than the IST. The IST was found to be more difficult to apply than the CAT. The results of this study suggest that the CAT is a more effective tourniquet than the IST. The IST is a more difficult to apply than the CAT. The results of this study suggest that the CAT is a more effective tourniquet than the IST. The IST is a more difficult to apply than the CAT.

This research was supported by the Entergy Workforce Training Grant.

# Evaluation of Makeshift Tourniquet Efficacy Model of an Exsanguinating Limb

<sup>1</sup>Sara Beaulieu, <sup>2</sup>Rimi Mandal, <sup>3</sup>Patrick Greiffenstein MD  
<sup>1</sup>Louisiana State University College of Science  
<sup>2</sup>Tulane University College of Science  
<sup>3</sup>Louisiana State University Health Science Center, Department of Surgery

## Background

Tourniquets are a device used in emergency medicine to stop and occlude blood vessels. The compression device that has and proven most effective is the Israeli Silcock Tourniquet (IST). However, there are several other types of tourniquets available. These include the Combat Application Tourniquet (CAT), the Special Operations Forces Tourniquet (SOF-T), and the SWAT Tourniquet. The purpose of this study was to evaluate the efficacy of two types of commercially available tourniquets including the CAT and IST tourniquet on an exsanguinating limb simulator (ELS).

## Methods and Materials

The makeshift tourniquets were simple to use. The CAT or makeshift tourniquet was applied to the ELS model. The model included a pump that provided a constant flow of blood to the limb. The time from the point they picked up the tourniquet until cessation of flow was recorded. The results of this study suggest that the CAT is a more effective tourniquet than the IST. The IST is a more difficult to apply than the CAT.

## Procedure and Testing for Efficacy

To address the question of whether the ELS model was a valid model for testing the efficacy of the CAT and IST, we performed a validation study. Three subjects were asked to test out the tourniquets. Before starting the process, we made sure that each of the participants knew about the project, have previous knowledge of applying a tourniquet, the purpose, and the data we were trying to collect.

## Validating the ELS



Figure 3. Subject testing of tourniquet on the ELS Model



Figure 4. The results for the validation of the ELS model.

This research project was supported through the LSU Health Sciences Center, School of Medicine.

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# Sex Differences in Fat Signaling in Obesity Prone and Obesity Resistant Rats




Corinne Martin & Stefany Primeaux, PhD

Department of Physiology, Louisiana State University Health Sciences Center, New Orleans

## Introduction

- Obesity affects 1 in every 4 Americans
- Insulin and the leptin receptor are among the most widely studied candidates for obesity therapy
- OB-R is the precursor for both receptor found in the hypothalamus and adipose tissue, and is thought to be the primary signaling system for the brain to monitor for the body's nutritional and energy status
- OB-R is a cell surface receptor which promotes acute eating
- OB-R is expressed in select regions of the hypothalamus for neurons eating (i.e., orexinergic and arousal nucleus of the hypothalamus (OBH/ARC))
- OB-R's receptor (OB-R2) is expressed in the brainstem (OBH/ARC), a brainstem center of the brain
- The goal of this current study was determine sex differences for both OB-R expression in males and females in both hypothalamus and brainstem



## Hypotheses

- OB-R will be expressed in both males and females in both regions of the brain (OBH/ARC and OBH/ARC)
- OB-R will be expressed in both males and females in both regions of the brain (OBH/ARC and OBH/ARC)
- OB-R will be expressed in both males and females in both regions of the brain (OBH/ARC and OBH/ARC)

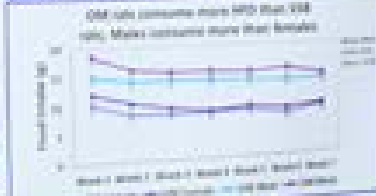
## Methods and Materials



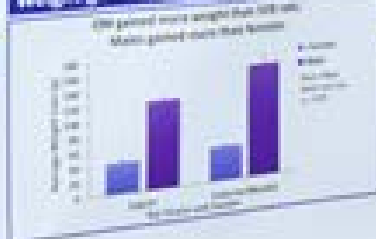
## Methods and Materials



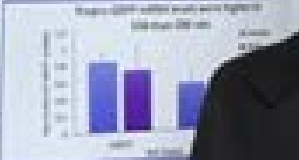
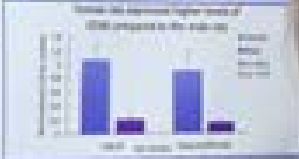
## Food Intake in OM and SR Males & Females



## Weight gain in OM and SR rats



## Gene Expression in OM and SR rats



## Summary & Conclusions

OB-R is expressed in both males and females in both regions of the brain (OBH/ARC and OBH/ARC). OB-R is expressed in both males and females in both regions of the brain (OBH/ARC and OBH/ARC). OB-R is expressed in both males and females in both regions of the brain (OBH/ARC and OBH/ARC).



LSU Health School of Medicine  
Scientist Sulfide's Approach to Human Color  
Samuel Martin, Chae Nique PhD

## Proposed Mechanism



## Results



# Sulindac Sulfide's Apoptotic and Anti-proliferative Effects on Human Colon Cancer Cells

Samuel Martin, Ches'Nique Phillips PhD, Yaguang Xi MD PhD.

Department of Genetics, Stanley S. Scott Cancer Center,  
Louisiana State University Health Sciences Center, New Orleans, LA



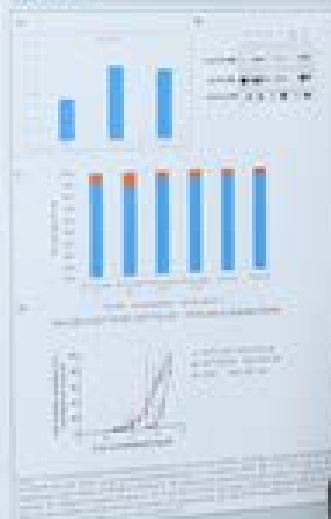
## Introduction

Colorectal cancer is the second leading cause of cancer death in the United States. Sulindac, a non-steroidal anti-inflammatory drug (NSAID), has been shown to have chemopreventive and anticancer effects in various animal models and human cell lines. The present study aims to investigate the apoptotic and anti-proliferative effects of Sulindac Sulfide on human colon cancer cells.

## Proposed Mechanism



## Results



## Results



## Conclusions

Sulindac Sulfide induces apoptosis and inhibits proliferation in human colon cancer cells. The proposed mechanism involves p53 activation and Bcl-2 downregulation.

This research project was supported through the LSU Health Sciences Center, School of Medicine, Stanley S. Scott Cancer Center.





# Workplace Wellness and Cancer Screening



Alina Mohiuddin<sup>1</sup>, Douglas LeBlanc<sup>2</sup>, MPH, Mikal Giancola<sup>2</sup>, MPH, Donna L Williams<sup>2</sup>, DrPH  
Loyola University New Orleans<sup>1</sup>, LSUHSC-NO School of Public Health<sup>2</sup>

## Background

Approximately 1.5 million people die of preventable cancers each year in Louisiana, the number of cancer is especially high. These numbers include breast cancer, cervical cancer, and colon/rectal cancer. Cancer is a leading cause of death among Louisiana residents. For example, there were 1259 cases of breast cancer per 100,000 women in 2019 making it the second leading cause of death among women in Louisiana. The United States suffered from 61,487 deaths due to breast cancer in 2019 alone. Barriers to care include getting lost at work, not having able to make an appointment, or even fear. One of the most powerful tools for preventing cancer are screenings. In 2019, breast cancer screening rates in Louisiana were 78.7-81.8%, while colorectal cancer screening was 64.1%, and cervical cancer was 84%. The *CA: A Cancer Journal for Clinicians*, states that 40% of all cancer-related deaths can be prevented with proper screening.

## Description

For the World Health Organization, employee wellness is a policy which results in the making and implementing of a healthy environment and thoughts for its workers. Through analyzing employee wellness initiatives of other institutions, this project provides recommendations to Louisiana State University Health Sciences Center (LSUHSC).

## Methods

Best practices from the University of Alabama at Birmingham (UAB), University of Maryland, and the Louisiana Department of Health were utilized during this project. UAB focuses on the promotion of collective cancer screening and prevention while the University of Maryland focused on completion of health activities by all employees throughout the year. UAB uses initiatives such as sending the message through flyers, incentive systems, and creating campaigns. A list of recommendations from these institutions was then compiled for administration at LSUHSC. Finally, about 5000 and 1000 were created to support the project.

General Statistics: General Population of Louisiana

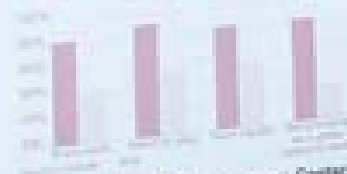


State of Current Employees of LSUHSC



## Engaging wellness

Crucial factors include offering, supporting, and promoting a safe and healthy work environment.



## Results

LSUHSC was able to increase their screening rates for colorectal cancer from 48.7% to 51.3% in just one year. This increase is the equivalent of 2,887 people. The pie charts on the left analyze the demographics of the employees at the university. Currently, there are 1,080 employees that are 65 years or older in Louisiana. Based on data from other institutions, we tried to see if we could increase their 1.5% screening rates to see how it would affect work.

## Discussion

We utilized a mix of the research evidence to determine factors that may affect the health and well-being of the employees at LSUHSC. We then used this evidence to create a healthy workplace culture where there is an emphasis on providing a safe and sound health care for our employees. As a part of the new start in the LSUHSC workplace wellness is also creating a culture change that empowers individuals to take control of their own health. This includes the change systems including creating a safe working environment, then developing the following program to be implemented. These initiatives will address programs to create an environment for questions. These initiatives will address programs to create an environment for questions. These initiatives will address programs to create an environment for questions. These initiatives will address programs to create an environment for questions.

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This research project was supported through the LSU Health Sciences Center, School of Medicine, Stanley E. Scott Cancer Center

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Alzaxia Mouse Model



Energy



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# Electrophysiological Techniques to Study Neurological Activity in the CA1 Region of the Hippocampus



Samantha Morin<sup>1</sup>, Katelyn Gurley<sup>2</sup>, Theodore Weyand<sup>2</sup>  
<sup>1</sup>Tulane University Department of Neuroscience,  
<sup>2</sup>Louisiana State University Health Sciences Center Department of Cell Biology and Anatomy

## Introduction

## Electrode Fabrication

## Surgical Implantation

## Behavioral Task



## Data Analysis



## Conclusion

## Data Recording



## Results



This research project was supported through the LSU Health Sciences Center, School of Medicine.

# Of Mice and Many Repeats: Tissue Specific Expansion in a Friedreich Ataxia Mouse Model

Na'ija S. Nelson, Jennie C. L. Roy, Caroline Burroughs, Ashley Henderson, and Ed Graczyk  
Department of Genetics, Louisiana State University Health Sciences Center



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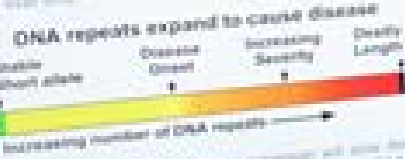
### Introduction

#### Friedreich Ataxia

Friedreich ataxia is the most common inherited cerebellar ataxia affecting children and young adults. Friedreich ataxia (FRAXA) neurodegeneration and ataxia is the result of a mutation located on chromosome 9q33, consisting of an expanded GAA triplet repeat in the first exon of the FXN gene. FXN encodes frataxin, a mitochondrial protein. FXN deficiency leads to iron accumulation in the mitochondria, oxidative stress, and ultimately to cell death.

#### Repeat Expansion

FRAXA repeats expand in a tissue-specific manner. The rate of expansion is highest in the brain and lowest in the heart. The expansion of GAA repeats is associated with increasing severity and disease length.



We hypothesized that limiting repeat expansion will slow disease progression. We have developed a strategy to slow GAA-TCG repeat expansion in a FRAXA patient cell line and will now use our strategy in a FRAXA mouse model. Our goal is to slow the expansion of GAA repeats and improve quality of life for patients.

#### FRAXA Mouse Model

The YGS mouse model that we are using to test our strategy to slow GAA-TCG repeat expansion contains a human FXN gene (4735) with a total number of GAA-TCG repeats. The goal of this project is to determine the rate at which expanded repeats expand and at what ages. Once we have this information, we can test the total expansion in various tissues.

### They Gain in the Brain

#### YGS Mouse Model Tissues Expand at Various Rates



FRAXA and images of GAA-TCG repeat expansion in brain tissue. FRAXA mice aged at 8 weeks, 8 months, and 23 days. Images show GAA-TCG repeats in brain tissue. Each row represents a different age group. The number of GAA-TCG repeats is shown in the top right corner of each image.

### Ear Today, Growing Tomorrow?

#### Ears Expand Dramatically



FRAXA mice aged at 8 weeks, 8 months, and 23 days. Images show GAA-TCG repeats in ear tissue. Each row represents a different age group. The number of GAA-TCG repeats is shown in the top right corner of each image.

#### Discussion

##### What We Found

- Repeat expansion in the brain. The expansion rate was similar in all tissues.
- Expansion rate was similar in all tissues.
- Expansion rate was similar in all tissues.
- Expansion rate was similar in all tissues.

This research was supported by the Energy Workstation Training Grant.

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### Electrophysiological Techniques Neurological Activity in the CA1 Hippocampus

Samantha Martin, Katelyn Dunlop, The  
Troy University, Department of  
Louisiana State University Health Sciences Center  
New Orleans

#### Behavioral Task



#### Data Analysis



Electrophysiological techniques were used to measure neurological activity in the CA1 hippocampus. The results show that there is a significant increase in activity during the behavioral task.



# Expression of Nicotinic Acetylcholine Receptors in Pulmonary Artery Endothelial and Smooth Muscle Cells

Riley Nguyen, David Woods, Kinping Yau  
Department of Physiology, LSU Health Sciences Center



## Abstract

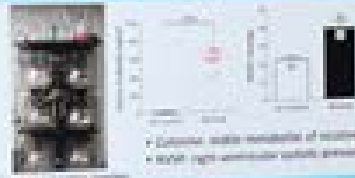
Nicotinic acetylcholine receptors (nAChRs) mediate the body's response to nicotine, regardless of their location. In the brain and organ-specific, response to nicotine. Previous studies from our lab show that in many chronic disease processes leads to extensive cellular remodeling and increased right ventricular output pressure. The aim of the current study was identify the receptor that are expressed by the pulmonary vasculature, including endothelial cells (ECs) and smooth muscle cells (SMCs), using immunohistochemical techniques. Pulmonary artery ECs (PAECs) and SMCs are known to express nAChRs. These receptors were shown to be of  $\alpha 5 \beta 2$  type. PAECs and SMCs were shown to be responsive to nicotine and all have been shown to be involved in many signaling pathways. The presence of nAChRs in these cells suggests that they are involved in nicotine-induced pulmonary vascular remodeling. In addition, we used immunohistochemistry (IHC) to locate lung tissue sections to detect nAChR expression. IHC results showed that nAChR immunoreactivity of  $\alpha 5 \beta 2$  type was present in PAECs and SMCs. PAECs immunoreactivity was followed by  $\alpha 7$  subtype. IHC on lung tissue sections showed that  $\alpha 5 \beta 2$  is present in the smooth muscle cells, and epithelial cells. Nicotine expression in the endothelial cells is unique. Our future direction is to examine the functional role of  $\alpha 5 \beta 2$  in PAECs, SMCs, and the role of both  $\alpha 5 \beta 2$  and  $\alpha 7$  in nAChR-mediated vascular remodeling in vitro and in vivo.

## Introduction

- About 60 million US adults smoke cigarettes and a 2 billion people and high school students use at least one tobacco product.
- Nicotine is the addictive component of all tobacco products. However, the effects of nicotine outside of the central nervous system (for example cardiovascular system) is not well understood.
- Nicotine signals through various subunit types (nAChRs) which are  $\alpha 7$ ,  $\alpha 3 \beta 4$ ,  $\alpha 4 \beta 2$ ,  $\alpha 5 \beta 2$ .
- Nicotine has been shown to activate multiple signaling pathways including calcium-mediated signaling pathway.



## Preliminary Data



## Methods



## Results

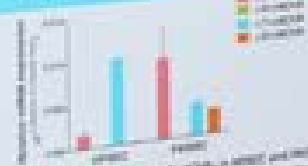
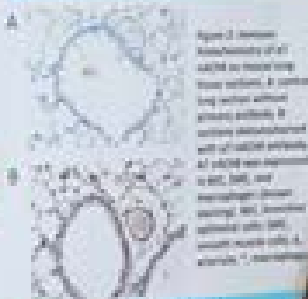


Figure 1: Immunohistochemical analysis of nAChRs in PAECs and SMCs. PAECs, human pulmonary artery endothelial cells; SMCs, pulmonary artery smooth muscle cells. Top, left: low magnification; Top, right: high magnification.

## Results



## Summary

- In PAECs, both  $\alpha 5 \beta 2$  and  $\alpha 7$  subunits are expressed.
- In SMCs,  $\alpha 5 \beta 2$  subunit is expressed, but  $\alpha 7$  subunit is not expressed.
- In PAECs,  $\alpha 5 \beta 2$  and  $\alpha 7$  subunits are expressed, but  $\alpha 3 \beta 4$  subunit is not expressed.
- In SMCs,  $\alpha 5 \beta 2$  subunit is expressed, but  $\alpha 3 \beta 4$  and  $\alpha 7$  subunits are not expressed.
- In PAECs,  $\alpha 5 \beta 2$  and  $\alpha 7$  subunits are expressed, but  $\alpha 3 \beta 4$  subunit is not expressed.
- In SMCs,  $\alpha 5 \beta 2$  subunit is expressed, but  $\alpha 3 \beta 4$  and  $\alpha 7$  subunits are not expressed.

## Future Directions

- Examine the functional role of nAChRs in PAECs and SMCs.
- Examine the functional role of both  $\alpha 5 \beta 2$  and  $\alpha 7$  subunits in PAECs and SMCs.
- Examine the effects of nicotine on PAECs and SMCs.

# Of Mice and Men: Tissue Specific Expansion in a Fried

Nahe E. Nelson, Jennie C. Li, Amy Caroline Burroughs  
Department of Genetics, Louisiana State University

## Introduction

Proteinase 3 (PR3) is a neutrophil serine protease that is highly expressed in the lungs, where it plays a role in the pathogenesis of ANCA-associated vasculitis. PR3 is also expressed in the liver, where it plays a role in the pathogenesis of liver disease. The aim of the current study was to examine the expression of PR3 in the liver and lungs of mice with ANCA-associated vasculitis.

## They Gain in the





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# Application of Machine Learning to Biomarker Discovery and Outcome Prediction in Colon Cancer using Genomic Data

<sup>1</sup>Ethan Nicklow, <sup>2</sup>Tarun Mamidi, <sup>3</sup>Dr. Jiande Wu, <sup>4</sup>Dr. Chindo Hicks  
<sup>1</sup>Department of Biomedical Engineering, Duke University, Durham, NC, 27708, <sup>2</sup>Department of Genetics, Louisiana State University Health Sciences Center, New Orleans, LA, 70012.



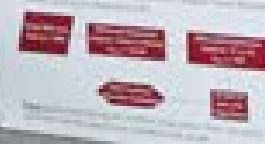
## Introduction

Colorectal cancer is the second leading cause of cancer death in the United States. The identification of biomarkers that predict patient outcomes is a critical step in the development of personalized medicine. Machine learning (ML) offers a powerful tool for analyzing large-scale genomic data to identify such biomarkers.

In this study, we applied ML to a dataset of genomic data from colon cancer patients to identify biomarkers associated with patient outcomes. We used a combination of feature selection and classification algorithms to identify a set of biomarkers that predicted patient survival.

## Materials and Methods

- 1. Data Collection: Genomic data was collected from a cohort of colon cancer patients.
- 2. Feature Selection: A set of features was selected based on their association with patient outcomes.
- 3. Model Training: A machine learning model was trained on the selected features to predict patient survival.
- 4. Validation: The model was validated on a separate set of data to assess its performance.



## Tumor vs Normal

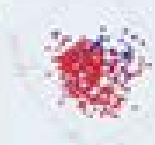


## Results from Tumor vs Normal

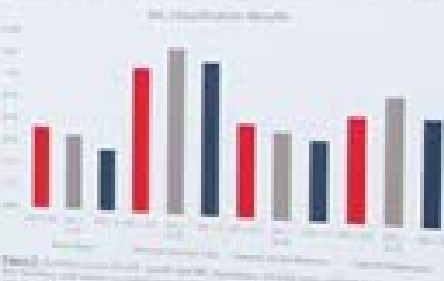
- We identified a set of genes that were significantly upregulated in the tumor compared to normal tissue.
- The expression of these genes was correlated with patient survival.
- Our findings suggest that these genes may be potential biomarkers for colon cancer.

## Patient Risk Classification

	Training	Testing
Accuracy	0.88	0.82
Specificity	0.95	0.85
Sensitivity	0.80	0.78



The ML model was trained on a dataset of genomic data from colon cancer patients. The model was able to accurately predict patient survival based on the expression levels of a small number of genes.



## Conclusions

- Our findings suggest that machine learning can be used to identify biomarkers for colon cancer.
- The identification of these biomarkers may lead to the development of personalized medicine for colon cancer patients.
- Further research is needed to validate these findings in a larger cohort of patients.

This research was supported by the Entergy Mathews Training Fund.

# Low Clinic Attendance Rates for Hepatitis C Appointments at UMCNO

Chizoba Ogbuefi, Kanayo Okeke-Eweni MBBS, MPH, Michael Okoronkwo MD, Stacey Rhodes MD, Evrim Oral PhD, Lisa Moreno-Walton MD, MS, MSCR, FAAEM  
UMCNO Emergency Department, LSUHSC School of Medicine



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## Introduction

Hepatitis C virus (HCV) is a single-stranded RNA virus that is transmitted through contact with infected blood. Each year, 70,000 new cases of HCV are reported in the United States. The infection is often asymptomatic, but can lead to liver disease, cirrhosis, and liver cancer. The Centers for Disease Control and Prevention (CDC) estimates that 3% of the U.S. population is infected with HCV. The majority of the population with HCV are unaware of their infection and therefore, 90% of the HCV-infected population do not receive treatment. This study was conducted to determine the reasons for low clinic attendance rates for HCV appointments at UMCNO.

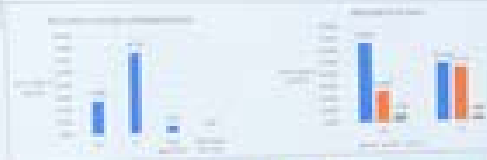
## Purpose

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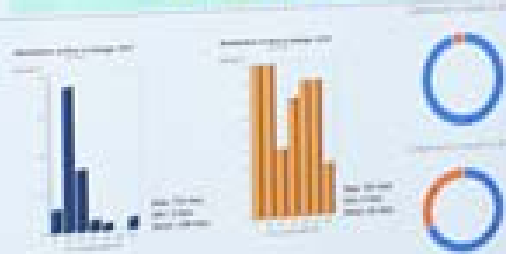
## Methods

We conducted a cross-sectional study of 100 patients who were scheduled for HCV appointments at UMCNO in the Emergency Department from 2017 to 2019. We collected data on patient demographics and clinic attendance. We conducted interviews with patients who did not attend their appointments to determine the reasons for low clinic attendance. We also conducted interviews with patients who attended their appointments to determine the reasons for high clinic attendance. We analyzed the data using descriptive statistics and logistic regression.

## Demographics



## Linkage to Care



## Results

The study found that 70% of patients who were scheduled for HCV appointments at UMCNO in the Emergency Department from 2017 to 2019 did not attend their appointments. The reasons for not attending were transportation (35%), cost (25%), distance (15%), and other (25%). The study also found that 30% of patients who were scheduled for HCV appointments at UMCNO in the Emergency Department from 2017 to 2019 did attend their appointments. The reasons for attending were transportation (15%), cost (15%), distance (15%), and other (55%).

## Conclusion

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This research project was supported through the LSU Health Sciences Center, School of Medicine.

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# Neighborhood Concentrated Disadvantage and Smoking among Young Adults in Greater New Orleans

Shawna Robinson, Stephen Kambou, MD, Ryan Kiser, MD, Denise Dancos, PhD  
Yusuf Choudhry, MD, Stephen Kambou, MD, Denise Dancos, PhD  
Yusuf Choudhry, MD, Stephen Kambou, MD, Denise Dancos, PhD  
Yusuf Choudhry, MD, Stephen Kambou, MD, Denise Dancos, PhD



## Introduction

Neighborhood concentrated disadvantage (NCD) is a measure of the social and economic conditions in a neighborhood. It is defined as the percentage of the population in a neighborhood that is in the lowest quartile of the distribution of each of the following variables: low education, low income, low home ownership, low employment, and high unemployment. NCD is a measure of the social and economic conditions in a neighborhood. It is defined as the percentage of the population in a neighborhood that is in the lowest quartile of the distribution of each of the following variables: low education, low income, low home ownership, low employment, and high unemployment.

## Objective

The objective of this study is to examine the relationship between NCD and smoking among young adults in Greater New Orleans. We hypothesized that young adults living in neighborhoods with high NCD would have higher rates of smoking.

## Data and Methods

We used data from the 2010 Behavioral Risk Factor Surveillance System (BRFSS) to examine the relationship between NCD and smoking among young adults in Greater New Orleans. We used a multivariate logistic regression model to estimate the odds ratios for smoking among young adults living in neighborhoods with high NCD, adjusting for age, sex, race, and education.

## Results

We found that young adults living in neighborhoods with high NCD had higher rates of smoking compared to those living in neighborhoods with low NCD.



- Young adults living in neighborhoods with high NCD had higher rates of smoking compared to those living in neighborhoods with low NCD.
- The relationship between NCD and smoking was stronger among young adults with lower education and lower income.
- The relationship between NCD and smoking was stronger among young adults who were not employed.
- The relationship between NCD and smoking was stronger among young adults who were not homeowners.
- The relationship between NCD and smoking was stronger among young adults who were not married.

Table 1. Characteristics of young adults living in neighborhoods with high and low NCD.

Characteristic	High NCD	Low NCD
Age (mean)	22.5	22.5
Sex (male)	45%	45%
Race (white)	15%	15%
Race (black)	75%	75%
Race (hispanic)	10%	10%
Race (other)	0%	0%
Education (high school or less)	65%	35%
Income (less than \$10,000)	55%	25%
Home ownership (renter)	85%	45%
Employment (unemployed)	45%	25%
Married	15%	15%
Smoking (current)	25%	15%

## Results

Table 2. Odds ratios for smoking among young adults living in neighborhoods with high NCD, adjusting for age, sex, race, and education.

Characteristic	Odds Ratio	95% CI
High NCD	1.5	1.2 - 1.8
Age (per year)	0.95	0.95 - 0.95
Sex (male)	1.05	0.95 - 1.15
Race (white)	0.8	0.6 - 1.0
Race (black)	1.0	0.9 - 1.1
Race (hispanic)	1.0	0.9 - 1.1
Race (other)	1.0	0.9 - 1.1
Education (high school or less)	1.5	1.3 - 1.7
Income (less than \$10,000)	1.2	1.0 - 1.4
Home ownership (renter)	1.0	0.9 - 1.1
Employment (unemployed)	1.5	1.3 - 1.7
Married	1.0	0.9 - 1.1

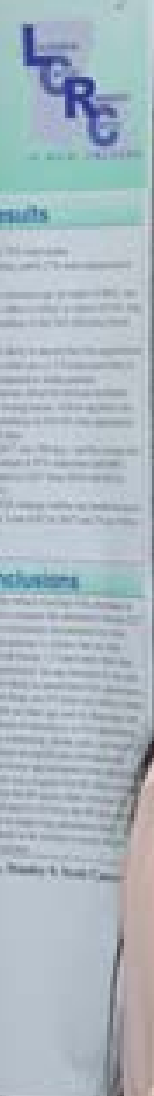
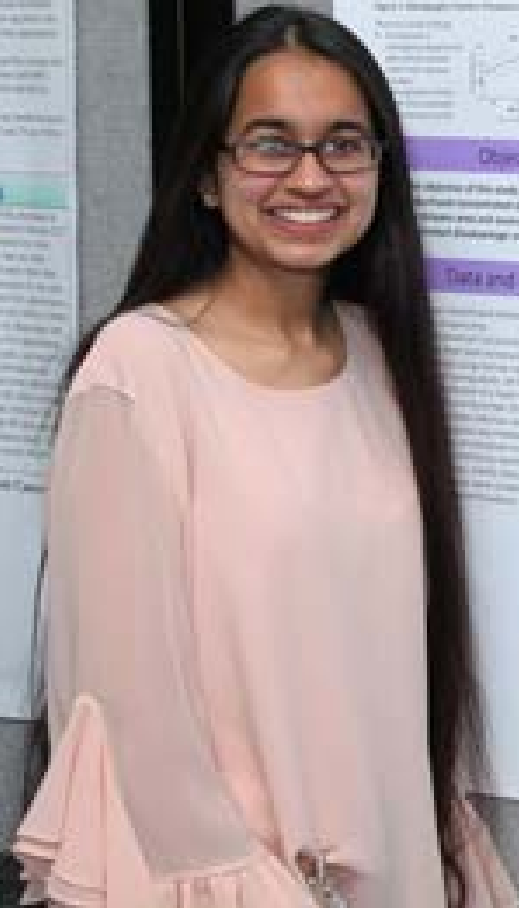


## Discussion

The findings of this study suggest that young adults living in neighborhoods with high NCD have higher rates of smoking compared to those living in neighborhoods with low NCD. This relationship was stronger among young adults with lower education and lower income, and among young adults who were not employed and not homeowners. These findings suggest that NCD is an important determinant of smoking among young adults in Greater New Orleans.

**Conclusions:** Young adults living in neighborhoods with high NCD have higher rates of smoking compared to those living in neighborhoods with low NCD. This relationship was stronger among young adults with lower education and lower income, and among young adults who were not employed and not homeowners.

This research was supported by the Entergy Workforce Training Grant.



# PLA2G6 and $\alpha$ -synuclein Interaction in Human RPE Cells

Catherine Rockwell<sup>1</sup>, Sophia Marathonitis<sup>2</sup>, Sayantani Bhattacharjee<sup>1</sup>, Jorgelina Calandria<sup>3</sup>.

Louisiana State University<sup>1</sup>, Tulane University<sup>2</sup>, Louisiana State University Health Sciences Center, Neuroscience Center of Excellence<sup>3</sup>.

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## Introduction

The loss of lysosomal function due to mutations in PLA2G6 is associated with Parkinson's disease (PD). PLA2G6 is a phospholipase A2 that hydrolyzes phospholipids into diacylglycerol and fatty acids. It is located in the lysosome and is involved in the degradation of phospholipids. Mutations in PLA2G6 lead to a deficiency of this enzyme, which results in the accumulation of phospholipids in the lysosome. This accumulation is thought to be the cause of the neurodegeneration seen in PLA2G6-deficient patients.

PLA2G6 is a member of the PLA2 family of enzymes. There are four main types of PLA2: PLA2G1, PLA2G2, PLA2G3, and PLA2G6. PLA2G6 is unique in that it is located in the lysosome and is involved in the degradation of phospholipids. Mutations in PLA2G6 lead to a deficiency of this enzyme, which results in the accumulation of phospholipids in the lysosome.

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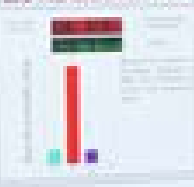
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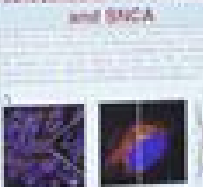
## MD-ATG1 increased the expression of PLA2G6 and PLA2G4A mRNAs



## MD-ATG2 increases L/S PLA2G6/PLA2G6



## MD-ATG1 induces the colocalization of PLA2G6 and SACA



## Dysregulation of PLA2G6 activity affects its colocalization with SACA



## Conclusions

- MD-ATG1, but not MD-ATG2, increases the expression of PLA2G6
- PLA2G6 is affected by both MD-ATG1 and MD-ATG2
- MD-ATG1 increases the frequency of PL-ATG6 cleavage into diacylglycerol and fatty acids
- MD-ATG2 increases the overall abundance of L/S PLA2G6
- MD-ATG2 treated cells had an increased ratio of L/S PLA2G6/PLA2G6
- MD-ATG1 increases the colocalization of PLA2G6 and SACA
- PLA2G6 inhibitors also increased the colocalization of PLA2G6 and SACA

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This research project was supported through the LSU Health Sciences Center, School of Medicine.



# Purification and Functional Analysis of Monoclonal Antibodies Protection Against *C. auris* Invasive Infections

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Department of Microbiology, Immunology, and Parasitology LSUHSC New Orleans, LA



## Introduction

*Candida auris* is a multidrug-resistant fungus, and is a leading cause of bloodstream infections affecting immunocompromised patients in the United States. Despite the effectiveness of antifungal therapies, the mortality rate remains unacceptably high. Due to the high morbidity rate and burden on the healthcare system, novel approaches are needed to improve antifungal therapy. Monoclonal antibodies are considered a promising approach. *C. auris* is a global fungus and has been identified over 100 years in the United States as of July 2019 (20) where it is now on a rising trend because of the high mortality rate (24-26%), and it is a challenging research. The use of monoclonal antibodies has not been established before in other species of *Candida*.

Previously, other studies have identified and isolated a panel of monoclonal antibodies against specific *Candida* and related species, and the studies provide protection in host against bloodstream infections caused by naturally acquired fungal species including *C. auris*.

Goals: The goal of this study was to produce antibodies using a self-inducing antibody system against the *Candida* and related species, and the study provide protection in host against bloodstream infections caused by naturally acquired fungal species including *C. auris*.

## Methods



## Immunoassay Methods

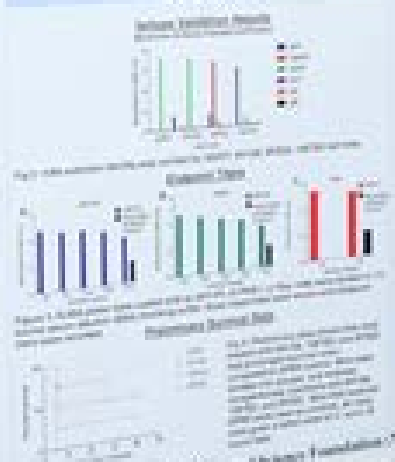


The *C. auris* was used as a target for antibody production and used as an antigen in a dot blot assay. The antibodies were tested for their ability to bind to *C. auris* in a dot blot assay.

## Flow Cytometry and Fluorescent Staining



## Results



## Conclusions

The study demonstrated that the generated monoclonal antibodies show high binding affinity and neutralizing activity against *C. auris*. These findings suggest that monoclonal antibodies may be a promising approach for the treatment of *C. auris* invasive infections.

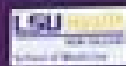



Motor Vehicle Collision (M...)  
 Pediatric Patients  
 ... MD, Evrim Oral PhD, Nick Sa...  
 ... MBS, MPH, Lisa Moreno-Walton M...



# DiETING Apps: What's Under the Hood?

Brayden W. Rohr & Lauri O. Byerley  
 Department of Physiology, LSU Health Sciences Center, New Orleans, LA 70112





### Introduction


- 40% of respondents used a dieting app
- 60% of users used food logging apps, most in a week
- 70% of users used calorie trackers
- 80% of respondents used apps that they had installed on their phone

### Results


#### Apps Selected




#### Databases Utilized for Food Comparison




#### Food Input Features



### Monetization Features



### Other Features



### Conclusion

There are data sources, features, and monetization methods that may contribute to a user's food-log nutrition information being inaccurate.

This research was supported by the Energy Workforce Training Grant.

# Disparities in Motor Vehicle Collision (MVC) among Pediatric Patients

Zion Rouege, Stacey Rhodes MD, Evrim Oral PhD, Nick Sausen MD, Kanayo Okeke-Eweni MBBS, MPH, Lisa Moreno-Walton MD  
UMCNO Emergency Department, LSUHC School of Medicine



## Introduction

Motor vehicle collisions are collisions between a vehicle and a pedestrian, animal, fixed object, or another vehicle. Each year 1.25 million people lose their lives in motor vehicle collisions (MVC). An estimated 4.3 million people were severely injured in a motor vehicle collision in 2015. Motor vehicle collisions are one of the three leading causes of death in pediatric patients in the United States. The goal of this study is to discover trends in demographics and information regarding the use of protective devices, severity of injuries, surgery rates, and length of stay at the University Medical Center New Orleans (UMCNO). The research can potentially reduce disparities in motor vehicle collision rates, length of stay, and trends in injury (MVA, CT scans, and MRIs).

## Methods

The Louisiana Trauma registry was used to obtain information on patients between the ages of zero to 18 years who were in a MVC that met the criteria for a level I trauma activation at UMCNO. The charts for all patients meeting criteria were reviewed in EPIC. Variables collected included race, age, date of admission and discharge, gender, injury severity score (ISS) and use of protective devices. All data was de-identified and then analyzed using Statistical Analysis System (SAS) v.9.4. Relationships between categorical variables were assessed using Pearson's chi-square or Fisher's exact test. We also performed Wilcoxon rank-sum tests to compare independent groups.

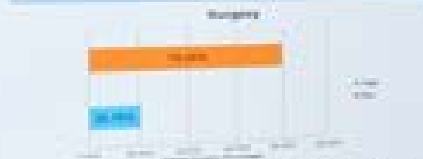
## Objectives

- To assess the severity of injury in pediatric patients involved in a Motor Vehicle Collision (MVC).
- To assess surgery rates across gender, age, and race.
- To assess hospital length of stay across Pediatric IHC patient groups.
- To assess the use of protective devices in pediatric motor vehicle collision patients.

## Demographics



## Surgery Rates



## Protective Devices

Restraint Usage by Race, Gender, and Age



This research was supported by the Eastrix Workforce Training Grant.

## Results

- 250 patients were included in the study...
- White patients had higher rates of surgery...
- Female patients had higher rates of surgery...
- Older patients had higher rates of surgery...
- White patients had higher rates of surgery...
- Female patients had higher rates of surgery...
- Older patients had higher rates of surgery...





# Retinal Sensitivity in Hormonally Modulated *Hyla cinerea* Using Electrophysiological Techniques



<sup>1</sup>Ashley Santana, <sup>1</sup>Whitney Walkowski M.S., <sup>1</sup>Hamilton Farris PhD

<sup>1</sup>Neuroscience Center of Excellence, <sup>2</sup>Department of Cell Biology and Anatomy, LSU Health, New Orleans, LA

### Introduction

Retinal sensitivity is the ability of the retina to respond to light stimuli. This is a critical component of the visual system, allowing organisms to detect and respond to changes in their environment. In this study, we investigate the effects of hormonal modulation on retinal sensitivity in the frog *Hyla cinerea*.



### Retinal Sensitivity



### Retinal T-Log(I) Response Curves



### Discussion and Future Experimentation on Endocrine Modulation

Endocrine modulation of the retina is a complex process involving multiple hormones and receptors. In this study, we have shown that retinal sensitivity is significantly affected by the presence of certain hormones. Future experiments should focus on identifying the specific mechanisms of endocrine modulation and the role of different receptors in this process.



Figure 1: Retinal T-Log(I) response curves for different experimental conditions. The curves show a sigmoidal relationship between response and log intensity, with shifts in the curves indicating changes in retinal sensitivity.

### Conclusion

In conclusion, our study demonstrates that retinal sensitivity in *Hyla cinerea* is modulated by hormones. This modulation occurs through a complex signaling pathway involving multiple receptors and second messengers. Our findings provide a foundation for further research into the endocrine control of the visual system.

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### Acknowledgments

We thank the National Science Foundation (NSF) for their support of this research. We also thank the members of the Farris laboratory for their helpful discussions and assistance throughout the project.



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3. Ashby, J. S., Farris, H. M., & Walkowski, W. (2020). Hormonal modulation of retinal sensitivity in the frog *Hyla cinerea*. *Journal of Experimental Biology*, 233(1), 1-10.



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# Glioblastoma multiforme (GBM): Clinical presentation, experimental animal models and novel experimental treatments

John S. Bauman, Nathan Berman, Emily Swanson, Leland Chickering, Sharon Malmgren, Lindsay Eberhart, Nicolas G. Bazan  
USC Health, School of Medicine, Neuroscience Center of Excellence, Los Angeles, CA, USA

**Abstract**  
Glioblastoma multiforme (GBM) is the most common and aggressive primary brain tumor. It is characterized by rapid growth, local invasion, and resistance to therapy. This study aims to improve our understanding of GBM pathogenesis and to evaluate novel experimental treatments in preclinical models.

**Introduction**  
GBM is a highly heterogeneous disease with diverse molecular profiles. The standard of care includes maximal safe resection, followed by radiotherapy and temozolomide chemotherapy. However, the median survival remains poor, highlighting the need for novel therapeutic approaches.

**Methods**  
We established a series of experimental animal models using orthotopic and syngeneic approaches. These models were used to evaluate the efficacy of novel treatments, including immunomodulatory agents and targeted therapies, in combination with standard of care.

**Results**  
Our results demonstrate that the novel treatments significantly improved survival and reduced tumor burden in the experimental models compared to control groups. These findings suggest that these treatments may have therapeutic potential for GBM patients.

**Conclusions**  
The development of novel experimental treatments for GBM is a high priority. Our preclinical studies provide a strong rationale for further clinical investigation of these promising therapies.

**References**  
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2. Wang M, et al. (2015) Immunomodulatory agents in glioblastoma: a review. *Cancers* 7:115-125.


**Acknowledgments**  
This work was supported by the USC Health and the Neuroscience Center of Excellence.



# The Protein S LG1+2 Domain Contributes Significantly to Inhibition of Factor IX<sub>a</sub>

Amber Sylvain, Sabyasachi Chatterjee, PhD, and Rinku Majumder, PhD  
LSU Health Science Center.

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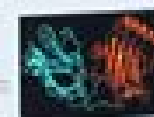

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### Introduction


Factor IX (FIX) is a serine protease that, in its active form (Factor IX<sub>a</sub>), cleaves Factor X to Factor X<sub>a</sub>, which then cleaves prothrombin to thrombin. Factor IX<sub>a</sub> is a key component of the blood coagulation cascade. The protein S LG1+2 domain is a natural inhibitor of Factor IX<sub>a</sub>. The structure of the protein S LG1+2 domain is shown in Figure 1. The structure is a dimer of two monomers, each consisting of a protein S LG1+2 domain and a protein S LG3 domain. The protein S LG1+2 domain is a member of the protein S LG1+2 domain family, which is a member of the protein S LG1+2 domain family.

### Objectives

Which of the two LG1+2 domains of the protein S LG1+2 domain is responsible for binding to and inhibiting Factor IX<sub>a</sub>?





### Circular Dichroism (CD)



**Figure 1.** The protein S LG1+2 domain wild-type and mutant (mutant 1: 133-150 LG1+2 and mutant 2: 133-150 LG1+2) were analyzed by circular dichroism (CD) at 25 °C. The CD spectra were recorded in 10 mM sodium phosphate buffer, pH 7.4, at a protein concentration of 0.1 mg/ml. The CD spectra were recorded at 220, 208, and 192 nm. The CD spectra were recorded at 220, 208, and 192 nm.

### Isothermal Titration Calorimetry (ITC)




**Figure 2.** ITC titration curves showing heat flow vs molar ratio for wild-type and mutant protein S LG1+2 domains. The ITC titration curves were recorded at 25 °C. The ITC titration curves were recorded at 25 °C.

### ITC Data

Condition	$K_d$ (nM)	$\Delta H$ (kcal/mol)
WT	1.2 ± 0.2	-1.2 ± 0.2
Mutant 1	2.5 ± 0.5	-0.8 ± 0.2
Mutant 2	3.5 ± 0.8	-0.5 ± 0.2

**Table 1.** Protein S LG1+2 domain ITC titration data for wild-type and mutant protein S LG1+2 domains at 25 °C.

### Surface Plasmon Resonance (SPR)



**Figure 3.** SPR sensorgrams showing binding of Factor IX<sub>a</sub> to wild-type and mutant protein S LG1+2 domains. The SPR sensorgrams were recorded at 25 °C. The SPR sensorgrams were recorded at 25 °C.

### Conclusion

The protein S LG1+2 domain is a natural inhibitor of Factor IX<sub>a</sub>. The protein S LG1+2 domain is a member of the protein S LG1+2 domain family, which is a member of the protein S LG1+2 domain family. The protein S LG1+2 domain is a member of the protein S LG1+2 domain family, which is a member of the protein S LG1+2 domain family.

This research project was supported by grant # 1039702 through the National Science Foundation (NSF) Research Experiences for Undergraduates (REU) Program



### Neuroadaptations in Estrogen Animal Model of Complex Alcohol Use Disorder

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**Introduction**

Alcohol Use Disorder (AUD) is a complex condition characterized by compulsive alcohol drinking, loss of control over drinking, and continued drinking despite negative consequences. The neurobiology of AUD is complex and involves multiple brain regions and neurotransmitters. The present study aims to investigate neuroadaptations in an animal model of AUD.

**Behavioral Analysis**

The present study used a behavioral analysis to assess the effects of estrogen on alcohol drinking in an animal model of AUD. The results show that estrogen treatment significantly reduced alcohol drinking in the animal model of AUD.



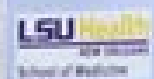
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# Neuroadaptations in Estrogen Signaling in an Animal Model of Complex Regional Pain Syndrome & Alcoholic Neuropathy

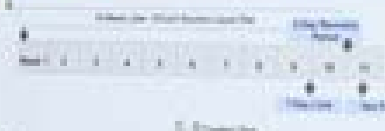


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Department of Physiology, LSU Health Sciences Center - New Orleans  
Tulane University

## Introduction

Complex Regional Pain Syndrome (CRPS) is a chronic pain condition that typically follows an injury.

## Behavioral Analysis



## Alterations in Progesterone Signaling



## Conclusions

- Behavioral data demonstrate that chronic alcohol abuse increases pain sensitivity in our model of CRPS.
- Although the differences in the expression of several estrogen receptors was not statistically significant, there was a general trend for alcohol to increase the expression of total ER and GPER.
- An overall trend did not lead to an increase in progesterone receptor phosphorylation at Serine 294 in the vulvate nodes.

## Future Directions

- Future studies will investigate other pain-related brain regions, including the prefrontal cortex and dorsal amygdala.
- Behavioral pain and motor information model comparisons. Future research will investigate if neuroadaptations to alcohol and CRPS are observed in the motor cortex in accordance with the injured limb.
- To determine associations between pain-like behavior and estrogen/progesterone signaling, we will analyze correlations between pain withdrawal threshold and protein expression.

## Alterations in Estrogen Signaling



Supported in part by LSUC through the National Science Foundation (NSF) Research Experiences for Undergraduates (REU) Program.



# Mechanisms of Nicotine-dependent Activation of Cardiac Fibroblasts

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## Introduction

- Electronic cigarette (e-cig) use has been gaining popularity among adolescents and young adults
- E-cigs are often advertised as "safer" than combustible tobacco products



- While cigarette smoke exposure has been well studied, little is known about exposure to e-cig smoke
- Investment from tobacco companies and availability of a liquid vaporization nicotine has led to understand the physiological effects of nicotine use

## Previous Research

- Embryonic pulmonary fibroblasts exposed to nicotine enhanced fibroblast cell proliferation (Rhee, *Am J Physiol Lung Cell Mol Physiol* 2003)
- Cardiac fibroblast proliferation and collagen production increased due to nicotine exposure, but not when the  $\alpha 7$  nicotinic acetylcholine receptor ( $\alpha 7$ nAChR) was inhibited (Yang, *Am J Physiol Lung Cell Mol Physiol* 2015)

## Hypothesis

In vitro nicotine exposure of cardiac fibroblasts increases the production of collagen I and III, as well as the collagen-crosslinking enzyme, lysyl oxidase (LOX)

## Materials and Methods

- **Culture conditions:** Fibroblasts isolated from cardiac tissue of naive mice were grown in DMEM with 10% FBS, 1% BSA
- Fibroblasts used for functional assays will be cultured in DMEM with 1% FBS, penicillin, streptomycin, and amphotericin 24 minutes before treatment with nicotine
- **Media collection:** Media collected in 24 well dishes will be administered to treatment groups and to each volume of 20  $\mu$ l water will be administered to control groups
- **RT-qPCR:** Fibroblasts that will be isolated and used for a reverse transcription-qPCR to determine changes in total nicotine, alpha smooth muscle actin, and collagen I and III
- **Functional assays:** Wound healing and collagen gel contraction assays will provide functional information about potential nicotine-dependent alterations in the fibroblasts
- **LOX activity assay:** Measure LOX activity with fluorescent assay

## Experimental Workflow



## Expected Results

- Increased total expression of alpha smooth muscle actin, and collagen I and III in non-knockdown fibroblasts
- Faster increase in wound healing rates resulting from increased expression in nicotine-treated fibroblasts

## Future Directions

- Use of electron microscope needed to study the cellular ultra-structure for effects of nicotine smoke
- Search for functional proteins or changes in cardiac function
- Investigate the effects of other e-cig flavors on the cardiovascular

## References

- Rhee K. (2003) Nicotine in a fibroblast cell line. *Am J Physiol Lung Cell Mol Physiol* 285:L1000-L1005
- Yang J. et al. (2015) Nicotine-induced fibroblast proliferation and collagen production is dependent on alpha7nAChR. *Am J Physiol Lung Cell Mol Physiol* 309:L1000-L1005

This research project was supported through the LSU Health Sciences Center, School of Medicine, National Institutes of Health

# Differences in Patient-Provider Communication Between Smokers and Non-smokers

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Michael D. Celestin, Jr., MA, CHES, NCTTS  
Louisiana State University Health Science Center, School of Public Health



## Introduction

- Thinking tobacco users are more of family doctors and less likely to discuss with their GP or pharmacist (general practice)
  - 60% of the number of prescriptions were written by GPs
  - In the US, 20% to 30% of drug users likely are more in primary care
  - Smokers receive less care than non-smokers, with less chronic disease management
  - Although the percentage of smokers has decreased in the United States, this still represents a large number of people
  - Patients who do not receive needed treatment reported a large amount of self-care
  - It may reduce the patient compliance or adherence to treatment in primary care
  - Quality of care affects adherence (compliance) from their doctor or pharmacist
  - Knowledge about problems
  - Communication with treatment when
  - Study their symptoms, and
  - Take medication when needed
- Note:** *Adherence/compliance* is defined as "extent of patient's participation in the management of his or her condition"

## Methods

- 1. **Study Design:** Cross-sectional study using data from the National Health and Medical Research Council (NH&MRC) Australian General Practice Research Network (AGPRN) database
- 2. **Study Population:** All patients aged 15 years and over, who were registered with a general practice in the AGPRN database, and who were prescribed at least one medication during the study period
- 3. **Study Variables:** The primary outcome was the proportion of patients who were prescribed at least one medication during the study period. Other variables included age, sex, and smoking status
- 4. **Statistical Analysis:** Descriptive statistics were used to describe the study population. Chi-square tests were used to compare the proportion of patients who were prescribed at least one medication during the study period, by smoking status



Figure 1. AGPRN collects nationally representative data regarding general practice patients in Australia public, a source of health research information.

## Results



Figure 6: Age of Prescribing Medication



## Results (cont.)

Table 1: Characteristics of patients who were prescribed at least one medication during the study period, by smoking status

Characteristic	Smokers (n=...)	Non-smokers (n=...)
Age (mean)	...	...
Sex (male)	...	...
Prescribed at least one medication	...	...
Number of prescriptions	...	...
Number of visits	...	...

Table 2: Characteristics of patients who were not prescribed any medication during the study period, by smoking status

Characteristic	Smokers (n=...)	Non-smokers (n=...)
Age (mean)	...	...
Sex (male)	...	...
Number of visits	...	...

## Conclusion

- 1. Smoking status was significantly associated with the receipt of at least one medication during the study period.
- 2. The proportion of patients who were prescribed at least one medication during the study period was significantly higher among non-smokers than among smokers.

This research was supported through the LSU Health Sciences Center, School of Medicine.

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# Higher SIV Levels Are Observed in Blood and Tissue Reservoirs of ART-Treated Female Macaques Exposed to Chronic Binge Alcohol



Anita Waye, Nedra Lacour, Angela Amedee, PhD.

Louisiana State University Health Sciences Center Comprehensive Alcohol Research Center and Department of Microbiology, Immunology, & Parasitology.

## INTRODUCTION

Approximately 10% of ART-naïve patients have HIV-1 plasma levels with HIV-1 RNA levels > 1000 copies/mL and/or detectable viral reservoirs. HIV-1 RNA levels are associated with viral reservoirs that affect their quality of life. HIV-1 RNA levels are also associated with immune activation and have been correlated to increased disease progression. HIV-1 RNA levels are also associated with increased HIV-1 RNA levels in tissue reservoirs, although HIV-1 RNA levels are generally lower in these reservoirs.

## HYPOTHESIS

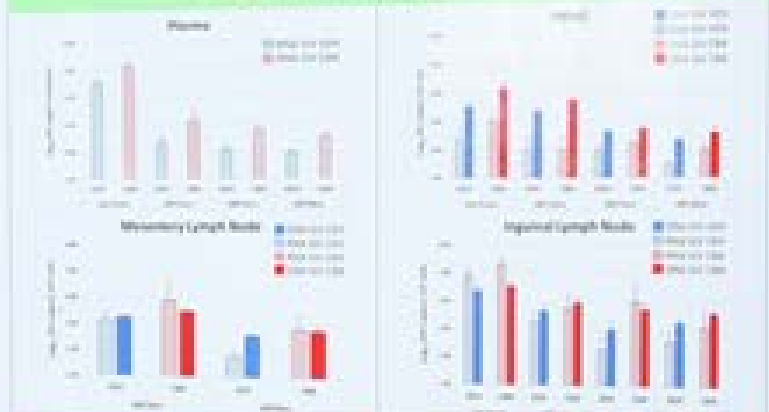
We hypothesize that chronic exposure to binge alcohol (BA) will increase HIV-1 RNA levels in blood and tissue reservoirs, thereby increasing HIV-1 viral reservoirs.

## STUDY DESIGN & METHODS



## RESULTS

### Comparison of mean DNA and RNA Levels



## CONCLUSION & FUTURE DIRECTIONS

Chronic exposure to binge alcohol (BA) increased HIV-1 RNA levels in plasma and tissue reservoirs (lymph nodes) in ART-naïve female macaques. HIV-1 RNA levels were significantly lower in plasma and tissue reservoirs in ART-treated female macaques exposed to binge alcohol. These observations suggest that chronic exposure to binge alcohol increases HIV-1 RNA levels in plasma and tissue reservoirs. HIV-1 RNA levels in plasma and tissue reservoirs were significantly lower in placebo and binge alcohol (BA) groups. HIV-1 RNA levels in plasma and tissue reservoirs were significantly lower in placebo and binge alcohol (BA) groups. HIV-1 RNA levels in plasma and tissue reservoirs were significantly lower in placebo and binge alcohol (BA) groups. HIV-1 RNA levels in plasma and tissue reservoirs were significantly lower in placebo and binge alcohol (BA) groups.

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- HIV-1 RNA levels in plasma and tissue reservoirs were significantly lower in placebo and binge alcohol (BA) groups.

This research project was supported through the LSU Health Sciences Center, School of Medicine, National Institutes of Health (NIH) & LSU HSC CARE.

# Effect of Fasting on the Distribution of Immune Cells In the Mouse Adrenal Gland

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**LSU Health**  
NEW ORLEANS  
School of Medicine

Andrew Williams, Manqi Wang, Matthew Whim  
LSU Health Sciences Center, Department of Cell Biology and Anatomy

## Introduction

- The adrenal gland is a neuroendocrine gland that releases hormones that regulate metabolism including the control of blood glucose levels. The gland has two regions, the outer cortex and the inner medulla. The cortex produces steroid hormones including corticosterone, and the medulla produces the catecholamine hormones, epinephrine and norepinephrine. These hormones are released during the fight or flight response to stress, particularly when the levels of blood glucose fall low as during fasting.
- The adrenal gland also contains a diversity of immune cells that adhere there and respond to the fight or flight response in the brain. The aim of this project was to test the hypothesis that activation of the adrenal gland during fasting would lead to a change in the number or distribution of immune cells.
- To test this idea we used mice that were fed or fasted for 24 hrs. This time was then extended, the adrenal gland removed and immunohistochemical procedures for immunohistochemistry using a fluorescent antibody which labels all leukocytes (antibody against CD45) and antibodies against T lymphocytes and B lymphocytes.

## Materials & Methods

1. Prepare adrenal medullary vein cannulations that bleed 100-200 µl.
  2. Prepare adrenal medullary vein cannulations that bleed 100-200 µl.
  3. Prepare the adrenal medullary vein cannulations that bleed 100-200 µl.
  4. Wash sections in PBS (10 min).
  5. Incubate sections in 2% BSA, for 30 min.
  6. Incubate sections in secondary antibody solution (1 hr).
  7. Wash sections in PBS (10 min).
  8. Mount sections on slides for analysis.
- In some experiments, sections were stained for CD45 (green) and CD3 (red) (1:1000, Abcam) and DAPI.

## Adrenal morphology



**Figure 1. Anatomy of the adrenal gland.**  
The adrenal gland is a neuroendocrine gland that releases hormones that regulate metabolism including the control of blood glucose levels. The gland has two regions, the outer cortex and the inner medulla. The cortex produces steroid hormones including corticosterone, and the medulla produces the catecholamine hormones, epinephrine and norepinephrine. These hormones are released during the fight or flight response to stress, particularly when the levels of blood glucose fall low as during fasting.

## CD45+ cells in the mouse adrenal gland



**Figure 2. CD45+ cells in adrenal glands of fed and fasted mice.**  
The number of CD45+ cells in the adrenal cortex of fed and fasted mice was determined by immunohistochemistry. The number of CD45+ cells per mm² in the adrenal cortex was determined by counting the number of CD45+ cells in the adrenal cortex. The number of CD45+ cells per mm² in the adrenal cortex of fed mice was 40 ± 10 and the number of CD45+ cells per mm² in the adrenal cortex of fasted mice was 60 ± 10.

This research was supported by the Energy Workforce Training Grant.

## CD3+ cells in the mouse adrenal gland



**Figure 3. CD3+ cells in adrenal glands of fed and fasted mice.**

## Conclusion

The number of CD45+ cells in the adrenal cortex of fasted mice was significantly higher than the number of CD45+ cells in the adrenal cortex of fed mice.

Higher STY Levels  
Reservoirs of A...

LSU  
School of Medicine

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NATIONAL LACROSSE INSTITUTE

# "Suppression of Dendritic Cell Maturation by Triple-Negative Breast Cancer Exosomes"

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LSUHC - SPIRIT  
SPIRIT-CHD  
Louisiana Partnership for Improving Research  
& Training in Cancer Health Disparities

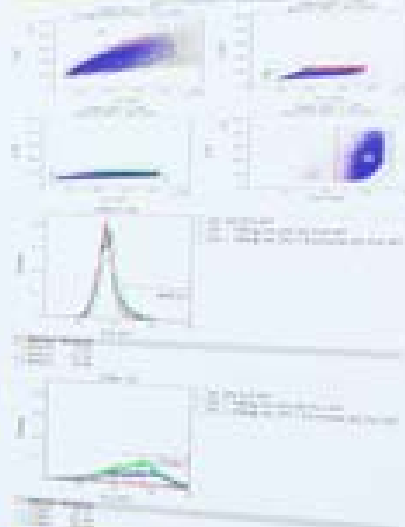
## Introduction

Dendritic cells (DCs) are professional antigen-presenting cells that play a central role in the initiation of an immune response. They are highly motile and migrate from peripheral tissues to lymphoid organs, where they present antigens to T cells. DC maturation is a process that involves the upregulation of surface molecules and the production of cytokines. This process is essential for the activation of T cells and the initiation of an immune response.

## Exosome Isolation

- Ability to bind to DCs
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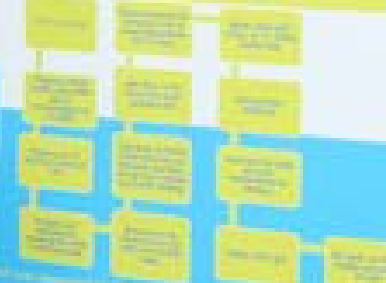
## Results



## Conclusion

Our results demonstrate that TNBC exosomes suppress DC maturation, which may contribute to immune evasion and tumor progression. This finding highlights the importance of studying the mechanisms of immune evasion in cancer and the potential for developing therapies that target these mechanisms.

## Flow Cytometry



## Conclusion

Our results demonstrate that TNBC exosomes suppress DC maturation, which may contribute to immune evasion and tumor progression. This finding highlights the importance of studying the mechanisms of immune evasion in cancer and the potential for developing therapies that target these mechanisms.