

ENCOURAGE.

RECOGNIZE.

CELEBRATE.



Nevada State Undergraduate Research Journal 2023-2024

Volume 9 Issue 1 Fall 2023

Editors: Izzy Dalla, Alina Evans, Anders Hoover

Director: Dr. Tanya M. Kelley

Published online at scholarworks.unr.edu by the Associated Students of the University of Nevada (ASUN) 3rd Floor Joe Crowley Student Union Mailstop 058 University of Nevada, Reno, Reno, NV 89557

DOI: http://dx.doi.org/10.15629/6.7.8.7.5

ISSN: 2375-4826



Table of Contents

Volume 9 Issue 1 Fall 2023

From the Editors	. 3
Letter from the Editors	
Testimonials	4
Testimonials from NSURJ Edition 8 Alumni	
NSURJ Graphs of Distribution	. 5
NSURJ Submission Demographics	
Research Articles	. 6
ApoE4-mCherry2 Fusions Enabled Visual Tracking of Protein Production During Expression in E. coli	
By Kirun Teinen, Jaeden Tedsen, Dr. James Davis, and Dr. Jeffrey Harper	. 6
Virus-Infected Caterpillars Perform Better on Low Protein Compared to High Protein Diets	
By Anna Dlabalova and Angela Smilanich	17
Accounting for Long-Term Drought in the Development Process in the Truckee Meado Watershed	
By Jacob Rosenbaum and Sameer Bhattarai	25
Disability and the Campus-Built Environment: Enhancing Mobility for UNR Students with Disabilities	
By Lea Gifford, Scott Kelley, Ph.D., Kerri Jean Ormerod, Ph.D.	34
Understanding Jealousy Among Individuals with Minoritized Identities By Courtney Brothers and Sarah Mitchell, Ph.D., CFLE	54
Acknowledgments	79
Acknowledgments	

LETTER FROM THE EDITORS

NSURJ is excited to present:

Volume 9 Issue 1 Winter 2023

On the ten-year anniversary of the Nevada State Undergraduate Research Journal's publication, we as editors passionately reassert the incredible value of its existence.

Founded with humble beginnings in 2014 by the Associated Students of the University of Nevada, the Nevada State Undergraduate Research Journal (NSURJ) is intended to highlight those outstanding accomplishments of undergraduates, whose work is of the same caliber as that of graduate students and yet is rarely featured in peer-reviewed journals.

As three editors of vastly different intellectual backgrounds, this work speaks to us in different ways. Each of us has benefitted from the unique opportunity of conducting undergraduate research that the University of Nevada, Reno provides. The chance to learn from other students' work and to help them revise it is profoundly rewarding to us - Not only have we honed our scientific writing skills, but we also have learned so much about ground-breaking work in our own fields.

Although NSURJ is interdisciplinary, we often find that arts, humanities, and business students are hesitant to submit their work. We argue that "research" refers to any project that generates knowledge, that all students' work has a place here, and that everyone can benefit from undergoing the peer-review process. And in the coming years, we will continue to strive to feature an even proportion of work from students of all fields.

It is with immense pride that we present the ninth volume of the Nevada State Undergraduate Research Journal. We hope that students who have not yet published with us will read this iteration of our journal and be inspired to pursue research on campus.

To our readers, thank you for supporting us through ten amazing years of publications.

Sincerely,

Izzy Dalla, Alina Evans, and Anders Hoover



Testimonials from Edition 8 Alumni

"By submitting a paper to NSURJ, I gained a stronger understanding of how to explain my research as I learned which parts needed to be explicitly explained in more detail, and which parts I could reasonably expect the audience to already understand without explanation. Most people are not going to be super familiar with your exact area of research, so understanding how to explain your research well to someone unfamiliar with it is very important, and submitting to NSURJ helped me to develop this skill."

—Kurtis Bertauche, author of "Application of Machine Learning in Clear Cell Renal Cell Carcinoma Prediction"

"Being an interdisciplinary journal, publishing with the NSURJ was great for learning to explain complex discipline-specific subjects to a general academic audience. I now have a second paper published with a larger journal and my experience with NSURJ helped prepare me for this. It has also been great to get my foot in the door of academic research and this has led to other opportunities for me. I am now in a graduate program where they are selective about allowing students to do a thesis. Having a proven track record of publishing research helped me to get my thesis approval. I can thank the NSURJ for contributing to this success."

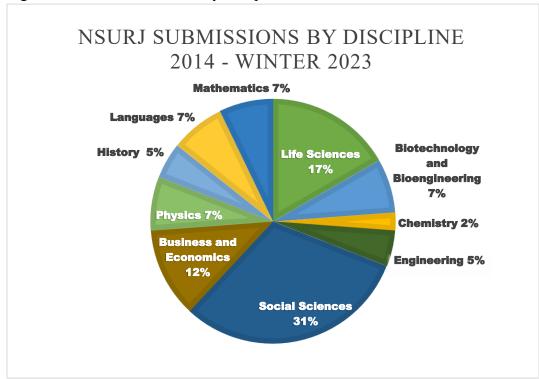
—Alexandra Zoellner, author of "Practices and Beliefs of Superstitions in Nevada's Scottish Diaspora"

"Working with the editors and traversing the peer-review process over the course of the next several months completely changed this, opening my eyes to the rigors and satisfaction of science communication and the publishing system. Overall, the NSURJ is an incredibly beneficial tool for young scientists at Nevada in their pursuit of excellence, and I would encourage anyone who is interested or has written a research paper – even if just for a class – to submit and see where it takes you!"

—Riley Gilis, author of "Predicted Major Annual Pathological Consequences of Obesity — Encompassing Cardiovascular Disease with Respective Mortality, Cancer with Respective Mortality, Type II Diabetes Mellitus, And Depression with Related Suicide"

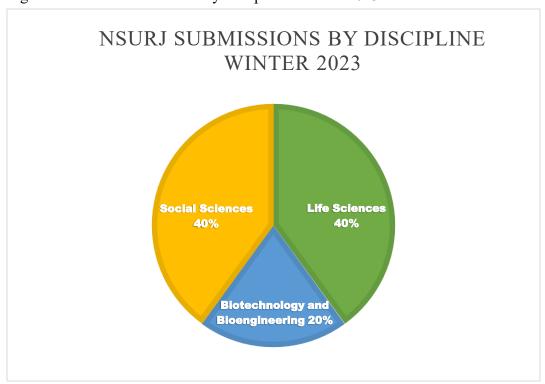
NSURJ Submission Demographics

Figure 1: NSURJ submissions by discipline – all time



Pi chart of NSURJ submissions from its inception in 2014 through this winter 2023 edition.

Figure 2: NSURJ submissions by discipline – winter 2023



Pie chart of NSURJ submissions for the current winter 2023 edition.

ApoE4-mCherry2 Fusions Enabled Visual Tracking of Protein Production During Expression in *E. coli*

Kirun Treinen, Jaeden Tedsen, Dr. James Davis, and Dr. Jeffrey Harper¹.

Department of Biochemistry & Molecular Biology, University of Nevada, Reno

Abstract

Human apolipoprotein E4 (ApoE4) has been proposed to exhibit the same properties as the ApoLp III lipoprotein, which forms lipid nanodiscs when sonicated in the presence of membrane lipids and is secreted extracellularly as a full-length protein by E. coli. These unique properties could lend ApoE4 to be a highly useful delivery system for proteins. In order for this utility to be exploited, ApoE4 first has to be produced efficiently in E. coli as well as be secreted as a full-length protein with the addition of a protein-coding region of interest. To evaluate this, mCherry2 was fused to either side of ApoE4 to produce two alternative orientations of recombinant protein, PelB-ApoE4-mCherry2-6xHis and PelB-mCherry2-ApoE4-6xHis. The addition of mCherry2 enabled for visual tracking of the protein during protein production steps, and easily confirmed if the protein was being secreted into the supernatant. Proteins were then isolated using 6xHis tag purification. The recombinant proteins were successfully purified, but were not secreted as full-length proteins as confirmed by SDS-PAGE gel analysis. The addition of 3% ethanol to the induction media was found to improve protein yield. The majority of recombinant protein was found to be located in the lysed bacterial pellet. Further optimization of protein production methods is necessary to produce full-length recombinant protein for downstream utilization of lipid nanodisc delivery. The construct was suspected to contain a cleavage site that caused degradation. Once the cause of degradation is diagnosed and fixed, the work can concentrate on the delivery potential of ApoE4 and the creation of recombinant proteins with therapeutic applications.

Keywords: Lipid nanodisc, ApoE4, nanodisc-forming, recombinant protein, E. coli

Introduction

Natural proteins can be engineered to have diverse functions, such as specificity for a biochemical target or the ability to deliver drug molecules. These characteristics are often combined when designing recombinant proteins, making them exciting platforms for targeted delivery. Developing proteins with improved secretion from hosts is another important aspect to select for when designing recombinant proteins. When designed successfully, recombinant proteins can be applied to producing antibodies and biosimilars, or drugs that mimic the structure of a natural biological product (Sørensen et al., 2005). One of the most well-known examples of a recombinant protein being used as a human therapy, is the production of human insulin in E. coli. Before the sequence of insulin was determined by Dr. Frederick Sanger, insulin had to be harvested from porcine or bovine pancreases. Insulin derived from these organisms had many issues and routinely induced host rejection in diabetic patients, which was why producing insulin from recombinant DNA was so revolutionary (Landgraf & Sandow, 2016). Producing any recombinant protein consists of introducing foreign genetic material into a host organism and employing its cellular machinery to produce the desired protein. The goal of this research was to fuse mCherry2 to the apolipoprotein, ApoE4, and demonstrate that the entire recombinant protein can be secreted by E. coli.

Apolipoproteins play a key biological role of transporting lipids in the body, such as cholesterol. Apolipoproteins are composed of proteins and phospholipids and assume a spherical shape that makes them optimal transporters of medication to specific targets (Baumann & Pham-dinh, D., 2002). The addition of mCherry2 fluorescent protein to ApoE4 evaluated whether fusing a protein-coding region to ApoE4 would affect its secretion as a full-length protein.

MCherry2 was also selected to enable easy visual tracking. Demonstrating that ApoE4 can be secreted with the addition of mCherry will enable future projects to focus on attaching alternative protein-coding regions to ApoE4. These could include ferritin or dectin coding regions, which could be applied to developing medications, bioremediation tools, or food additives.

The PelB leader sequence has been identified as an important translocation signal peptide when expressed in E. coli (Shi et al., 2022). This sequence has been shown to translocate proteins into the periplasmic space of *E. coli* cells, which is a requisite step for eventually expressing proteins extracellularly for purification (Rosano et al., 2014). Human ApoE4 was included because a similar apolipoprotein called ApoLP III (derived from locusts) was shown to be secreted into the cell-free supernatant by E. coli (Ryan et al., 1995). ApoLP III exhibited the unique quality to form lipid nanodiscs when sonicated in the presence of membrane lipids, an adequate confirmation for delivery (Lethkoe et al., 2021). MCherry2 was included due to its fluorescent properties that are emitted upon oligomerization (Karimi et al., 2016). The red color displayed by mCherry2 enabled visual tracking during protein production and heavily aided in protein purification steps. The method used to purify proteins of interest was 6xHis tag purification. A 6xHis tag was included on the N-terminal domain of the recombinant protein to facilitate this step. E. coli was chosen as the host organism to express recombinant proteins due to its quick generation time, inexpensiveness, and fast high-density cultivation. However, some

issues may arise when working with *E. coli* where proteins are not fully secreted, resulting in low yields.

The subject of my research was to develop a novel recombinant protein that could be expressed in T7 Express strain E. coli cells, and measure expression of PelB-ApoE4 with the addition of an mCherry2 coding region. The goal of this research was to determine that an additional coding region can be added to the PelB-ApoE4 expression system and not interfere with the properties of the lipoprotein. One application for a potential recombinant protein could include adding a ferritin coding region to develop a recombinant protein for biomedical applications. Ferritin oxidizes free Fe II to Fe III, which is used to bind oxygen in heme in the body (U.S. Department of Health and Human Services, 2023). Individuals with iron deficiency anemia have issues with iron uptake and meet their body's iron needs by adopting a high-iron diet or taking iron supplements (Pruthi, 2022). These methods can be improved upon due to high-iron diets eliciting their own side-effects in patients, and commercial iron supplements not increasing iron bioavailability in the body equally (Christides et al., 2014). Adding a dectin coding region would be of interest, due to dectin's innate ability to recognize fungal structural motifs (Brown, 2006). Producing a recombinant protein with dectin and apolipoprotein could be used to develop a targeted antifungal drug, where the apolipoprotein delivers medication specifically to fungi. In order to achieve these goals, the addition of mCherry2 to the PelB-ApoE4 protein had to first be demonstrated to be successfully secreted by

E. coli. The recombinant protein contained a PelB leader sequence, ApoE4 coding region, and mCherry2 coding region.

Methods

Design of Recombinant Constructs

The PelB-ApoE4 clone was generously donated by the Ryan lab located in the Department of Biochemistry and Molecular Biology at the University of Nevada, Reno for research purposes. Subcloning was conducted to add the 6xHis tag. Two orientations of the recombinant protein were

assessed for differences in expression of full-length protein. The orientations tested were PelB-mCherry2-ApoE4-6xHis and PelB-ApoE4-mCherry2-6xHis, where mCherry2 was added either on the N-terminal side or C-terminal side of ApoE4. The PelB-mCherry2-ApoE4-6xHis orientation was digested with the restriction enzyme MscI. MCherry2 was amplified from base pair position 3,397 using

oligonucleotides 3068a & 3068br and stitched using NEB builder (New England Biolabs, Ipswich, MA, USA). The PelB-ApoE4-mCherry2-6xHis orientation was digested with AscI/NotI. MCherry2 was amplified from base pair position 3,397 using oligonucleotides 3070a & 3070br and stitched using NEB builder. Oligo sequences are given in the table below.

Table 1. Oligo Sequence Information The information provided lists the sequences used to design the recombinant constructs.

	T
Oligo number	Oligo sequence
3068a	cgctgcccagccggcgat ggccaTGATGGTT TCTAAGGGAG
3068br	cacegcetgttccactttca TACCGGATCCCT TATACAATTC
3070a	cgtgaacgtctgggcccg ggcgcgccCATGGT TTCTAAGGGAG AG
3070br	tggtggtggtgctcgagtg cggccgcCTTATAC AATTCATCCATG C

Ligation Reaction & Electroporation Table 2. Ligation Salt Master Mix for 1 ReactionThe information provided describes the master mix used in the construction of the plasmid DNA that was later transformed into T7 *E. coli* cells.

Component	Volumetric Amount
Ligase salts/buffer	1 μL
T4 DNA ligase	0.2 μL
Water	4 μL
Total	5 μL

Table 3. DNA Mix for each Ligation Reaction The information listed describes how the protein-coding genes of interest (insert DNA) were introduced into the plasmid (backbone DNA) for each individual ligation reaction by way of PCR.

Component	Volumetric Amount
Insert DNA	x μL
Backbone DNA	yμL
Water	zμL
Total	5 μL

5 μL of ligase salt master mix was added to each DNA ligation reaction. Once combined, these were allowed to incubate on the bench for 1 hour or at 16°C overnight. 1 μL of ligation mixture was transformed into 50 μL of T7 electrocompetent E. coli cells on ice. The cell & DNA mixture was allowed to incubate on ice for 1 to 10 minutes. The cell & DNA mixture was then added to a pre-chilled electroporation cuvette. The cuvette(s) were pulsed under the given conditionsL 2,000 V, 50 uF, 125 Ω . 900 µL of 2XYT was immediately added. Cuvette contents were then transferred to an Eppendorf tube and allowed to incubate at room temperature for 30-60 minutes. Eppendorf tubes were spun for 30 seconds at 10,000 rpm to pellet cells. 750 µL of supernatant was removed, and cells were resuspended in the remaining liquid. T7 cells were then plated onto appropriate antibiotic plates. Plates were placed in the 37°C incubator overnight.

Growing E. coli Cells

20 μL of T7 Express strain E. coli cells (New England Biolabs, Ipswich, MA, USA) were grown in 40 mL of 2XYT (Sigma-Aldrich, Burlington, MA, USA) and antibiotic selection. These cells were grown overnight at 37°C in a shaking incubator set to 200 rpm. 400 mL 2XYT and ampicillin antibiotic selection was inoculated by pouring in the culture, as a 1:10 dilution, to a working ampicillin concentration of 50 μg/mL. These were allowed to grow for approximately 3 hours at 37°C until optical density was approximately 0.6 at 600 nm. 1 M IPTG (Sigma-Aldrich, Burlington, MA, USA) was added to a final concentration of 0.5 mM. Cells were allowed to grow for 2.5 hours at 37°C. Cells received 3% ethanol in addition to 0.4 mM IPTG at the time of inoculation in accordance with the method written by Chhetri et al.

Cell Pelleting

Cell collection was done by pouring the expressed *E. coli* cell mixture into batches. Batches were spun for 30 minutes at 6,000 rpm at 4°C. Lysozyme was dissolved (Thermo Fisher Scientific, Waltham, MA, USA) on ice during spin to 0.1 mM. Pellets were collected and allowed to store for at least overnight at -20 C. All samples were kept on ice prior to lysis.

Lysis of Cells

Each pellet was resuspended in 15 mL of cold lysis buffer (1.5 mg/mL lysozyme in 6xHis Binding Buffer [20 mM Tris, 500 mM NaCl, 10% glycerol, pH 8.0]) (Thermo Fisher Scientific, Waltham, MA, USA). and 100 mM PMSF (Thermo Fisher Scientific, Waltham, MA, USA). The pellets sat for 15

minutes at room temperature and were frozen overnight at -20°C. Pellets were thawed in approximately 5 minutes under hot water and PMSF was added for a final concentration of 1 mM. 10% Triton x100 was added (Sigma-Aldrich, Burlington, MA, USA) to lysate for 0.4% final concentration and thoroughly mixed. The lysis process was finished by sonication (Rochester Industrial Service Inc., Rochester, NY, USA), where duty cycle was set to 30% and output was set to 3. Sonication occurred for 30 seconds, with rest periods of 45 seconds following sonication. Sonication was repeated until the viscosity of the sample was reduced to a water-like consistency. Samples were stored on ice until the entirety of the sample was sonicated. Lysate was spun for 30 minutes at 20,000 X g at 4°C. The supernatant was collected following this spin cycle.

6xHis Purification of Recombinant Proteins

Purification was done by resuspending 400 μL of Ni-NTA bead (Qiagen) 50% slurry. Beads were rinsed with 1.5 mL 6xHis Binding Buffer [20 mM Tris, 500 mM NaCl, 10% glycerol, pH 8.0] 3 times. E. coli cell lysate was added and rocked for 45 minutes at 4°C then spun at 1,000 rpm for 5 minutes. Beads were transferred for batch washing by pelleting at 1,000 rpm for 1 minute at 4°C. Beads were then washed with 1.5 mL 6xHis Binding Buffer and spun at 1,000 rpm for 1 minute at 4°C. Chromatography columns (Bio-Rad, Hercules, CA, USA) submerged in ice were used to separate beads and elution. Beads were resuspended in 1 mL 6xHis Binding Buffer and transferred onto columns. 1 mL of 6xHis Wash Buffer (20

mM Tris, 100 mM NaCl, 10 mM imidazole, pH 8.0) was added to columns 4 times. After the final wash, 100 µL of 6xHis Elution Buffer (20 mM Tris, 100 mM NaCl, 300 mM imidazole, pH 8.0) was added onto the column 4 times, after exchanging collection tubes. After final wash, samples were spun at 1xg under cold room conditions. Binding Buffer was used to bind 6xHis-tagged proteins to the column. Wash Buffer was used to remove excessive and unbound proteins from the column and Elution Buffer was used to remove 6xHis-tagged recombinant proteins for collection.

Storage

Elution was transferred into a Pierce Concentrator concentration filter of molecular weight cut off of 10K and a maximum volume capacity of 500 μL (Thermo Fisher Scientific, Waltham, MA, USA). Samples were concentrated and spun at 15,000 X g for 30 minutes at 4°C using a SS-34 centrifuge. The concentrated protein was transferred into a 1.7 mL tube and the final volume was recorded. Samples were stored in a -20°C freezer with sterile 80% glycerol, with the final concentration of 80% glycerol being calculated by dividing the final sample volume by 0.6.

Protein Analysis

SDS-PAGE gels analysis was conducted with PelB-ApoE4-mCherry-6xHis and PelB-mCherry2-ApoE4-6xHis. Gels were stained for approximately 90 minutes and ran at 200 V and were then allowed to de-stain overnight. A Bradford assay was conducted with 175, 500, and 1000 ng/μL standards measured at OD600 to generate a standard curve in Microsoft Excel. Raw protein

concentrations were calculated from this data and then used to generate adjusted protein concentration values in ng/µl. These values were used to determine what volume of protein to load onto the protein gel.

Results

In order to examine if an additional coding region could be fused to ApoE4, preliminary data had to be obtained. Before comparing the two alternative orientations of the recombinant protein, the appropriate weight at which to load the recombinant proteins onto the gel had to be determined. This initial experiment was performed to verify if recombinant protein expression was greater when higher concentrations of IPTG were used during induction. The appropriate weight of protein to load onto the gel was found to be 2 μg. Increasing concentrations of IPTG were found to have little to no effect on the expression of the recombinant protein (Fig. 1). Due to this finding, 0.4 mM IPTG was used for following gel analysis experiments. Very little full-length protein was seen, but distinct bands were found to be present that were ~40 KDa, ~27 KDa, and ~21 KDa in size. When plasmid map calculations were made to determine what these bands could represent, no conclusive explanation could explain the ~40 KDa band. The plasmid map calculations supported the idea that the ~27 KDa band represented mCherry2 and that the ~21 KDa band represented ApoE4.

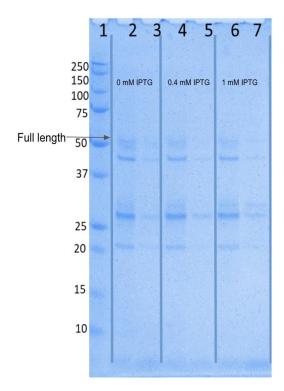


Figure 1 PelB-ApoE4-mCherry-6xHis Induced with Increasing Concentrations of IPTG Proteins were run on an SDS-PAGE gradient gel after 6xHis tag purification and stained with coomassie. Lane 1 shows the Precision Plus Protein All Blue ladder (Bio-Rad, Hercules, CA, USA), which is labeled in KDa. Lanes 2-7 display the PelB-ApoE4-mCherry2-6xHis recombinant protein. Each sample was run as a pair, with the first lane containing 2 ug and the second lane containing 0.5 ug total protein, pairs are delineated in the gel image. All lanes have a band at the bottom past the 10 KDa marker representative of the tracking dye used.

A second SDS-PAGE gel was run to compare the two alternative orientations of the recombinant protein, the location, and the effect of ethanol on expression. The same banding pattern seen for the PelB-ApoE4-mCherry2-6xHis recombinant protein was seen on this gel (Fig. 2). However, this banding pattern was not found to be consistent for the alternative orientation, where mCherry2 was fused on the N-terminal side of ApoE4. Instead, the

bands for PelB-mCherry2-ApoE4-6xHis were discovered to be larger. Expression of the full-length protein was found to be higher when mCherry2 was fused to the N-terminal side of ApoE4. Location and ethanol treatment were not shown to have a significant effect on expression for the alternative orientation where this data was present.

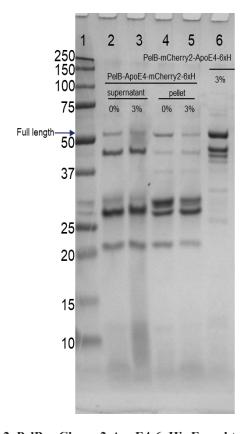


Figure 2. PelB-mCherry2-ApoE4-6xHis Found to Secrete more Full Length Protein than Alternative Orientation Proteins were run on a SDS-PAGE gradient gel after 6xHis tag purification and stained with coomassie blue. Lane 1 displays the Precision Plus Protein All Blue ladder labeled in KDa. The gel was stained for ~1 hour and ran at 200V until bands reached the bottom. Lane 2 displays PelB-ApoE4-mCherry2-6xHis obtained from the supernatant induced with 0% EtOH. Lane 3 displays the same construct obtained from the supernatant induced with 3% EtOH. Lane 4 displays the same construct obtained from the pellet induced with 0% EtOH. Lane 5 displays the same construct obtained from the

pellet induced with 3% EtOH. **Lanes 6** displays PelB-mCherry2-ApoE4 from the pellet induced with 3% EtOH.

A Bradford assay was used to quantify the protein yield based on location and presence of 3% ethanol during induction for PelB-ApoE4-mCherry2-6xHis. These data were used to generate a bar graph where location and presence of 3% ethanol were listed on the X-axis and ng of protein was listed on the Y-axis. It was found that 3% ethanol increased overall yield regardless of location. Fold changes were calculated to be 2.4 for the supernatant and 4.7 for the pellet. Despite the measures taken to release the recombinant protein into the supernatant, the majority of the protein persisted in the pellet after cell lysis.

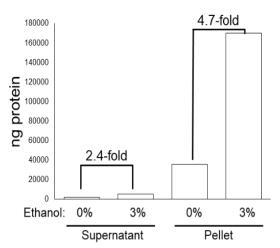


Figure 3. ApoE4-mCherry2 Fusions were Retained in the Insoluble Fraction PelB-ApoE4-mCherry2-6xHis yields and fold changes were calculated from Bradford assay data. Bars are labeled by location. Left hand bars received 0% ethanol during induction and right hand bars received 3% ethanol during induction. Induction methods were optimized by including 3% ethanol in the induction media, which elevated protein yield significantly.

Discussion

The experimental results established that ApoE4 can be successfully expressed in E. coli with the addition of a mCherry2 coding region at high yields. It was discovered through SDS-PAGE gel analysis, that when mCherry2 was on the N-terminal side of ApoE4, overall protein expression was greater than when it was attached on the C-terminal side of ApoE4. This finding was unexpected, and future work will have to be conducted to understand definitively why this occurred. SDS-PAGE gel analysis also revealed that there was a large amount of degradation present for both alternative orientations of the recombinant protein. There could be several factors causing this, but since the band patterns differed between the two orientations, it was suspected that a cleavage site was present in the mCherry2 coding region. If a cleavage site is located in the mCherry2 coding region, it could explain the unique band patterns seen for PelB-ApoE4-mCherry2-6xHis and PelBmCherry2-ApoE4-6xHis, which are present in the comparative SDS-PAGE gel. Visual tracking of the recombinant protein during protein production signaled that the majority of the protein was retained by the insoluble fraction after centrifugation and cell lysis steps. The cause of why the recombinant protein persisted in the insoluble fraction was not clear, as cells were exposed to detergent and sonicated. One hypothesis posed is that the route taken by the recombinant protein to escape from E. coli has a size-exclusivity limit, where mCherry is too large to be fully secreted. Cell lysis was expected to release the recombinant protein into the supernatant, but was

unsuccessful. Visual tracking and a Bradford assay clearly showed that the majority of protein yield was not present in the supernatant. This could be the case because of the above hypothesis or due to the protein becoming denatured during sonication or from high temperatures (Rosano et al., 2014).

Previous publications have not used a purification method to obtain recombinant proteins containing ApoE4. This was because a 6xHis tag was not incorporated into the recombinant proteins used in previous work. The method seen in this paper, sought to ameliorate this through 6xHis tag purification, in order to verify if the protein being produced was the expected full-length protein. ApoE4 was shown to escape from a bioreactor as a foamate, which contained some amounts of PelB-ApoE4-mCherry (Lethcoe et al., 2021). This research exemplified the nanodisc-forming property of the ApoE4 lipoprotein, which this work hopes to exploit at later stages. Further studies will have to be conducted to discern if degradation of recombinant protein during production in E. coli can be avoided. This can be achieved by strictly controlling sonication duration, temperature, and experimentally testing the sizeexclusivity hypothesis. Future studies could repeat SDS-PAGE analysis using a different lipoprotein and fluorophore and explain if degradation was unique to the PelB-ApoE4mCherry2-6xHis and PelB-mCherry2-ApoE4-6xHis recombinant proteins. More protease inhibitors can be included during purification steps in an effort to reduce degradation. If degradation persists after

these changes have been made, then the exact cleavage sites in the recombinant protein construct will have to be identified and corrected via site-directed mutagenesis.

Once it can be demonstrated that full-length proteins can be reliably expressed, future work can focus on exploiting the nanodisc-forming property of ApoE4 to produce a targeted delivery system. An exciting finding of this work was that the PelB-mCherry2-ApoE4-6xHis resulted in higher expression of full-length protein. Using this information will govern how future recombinant proteins are constructed. Future recombinant proteins will be constructed where additional coding regions are fused to an apolipoprotein to offer solutions to real-world problems. This has been exemplified by the work of Khumsupan et al., where curcumin was able to be encased inside of a lipid nanodisc. Another study demonstrated that lipid nanodiscs can be highly specific in their delivery, enabling smaller doses of medication to be administered (Tanaka et al., 2019). The mRNA COVID vaccine also utilizes lipid particles to stabilize mRNA and ensure that it does not get degraded by host cells (Hou et al., 2021). Applications for future recombinant proteins include developing an iron supplement from plant ferritin, developing a food additive for vegan meat products from plant ferritin, and developing an antifungal medication from dectin.

References

- Baumann, N., & Pham-dinh, D., (2002). Astrocytes. *Encyclopedia of the Human Brain*, 252-268. https://www.sciencedirect.com/science/article/abs/pii/B012227210200042X
- Brown G. D. (2006). Dectin-1: a signaling non-TLR pattern-recognition receptor. *Nature reviews. Immunology*, 6(1), 33–43. https://doi.org/10.1038/nri1745
- Chhetri, G., Kalita, P., & Tripathi, T. (2015). An efficient protocol to enhance recombinant protein expression using ethanol in Escherichia coli. *MethodsX*, 2, 385–391. https://doi.org/10.1016/j.mex.2015.09.005
- Christides, T., Wray, D., McBride, R., Fairweather, R., & Sharp, P. (2015). Iron bioavailability from commercially available iron supplements. *European journal of nutrition*, *54*(8), 1345–1352. https://doi.org/10.1007/s00394-014-0815-8
- Hou, X., Zaks, T., Langer, R., & Dong, Y. (2021). Lipid nanoparticles for mRNA delivery. *Nature Reviews Materials*, 6(12), 1078–1094. https://doi.org/10.1038/s41578-021-00358-0
- Karimi, S., Ahl, D., Vågesjö, E., Holm, L., Phillipson, M., Jonsson, H., & Roos, S. (2016). In Vivo and In Vitro Detection of Luminescent and Fluorescent Lactobacillus reuteri and Application of Red Fluorescent mCherry for Assessing Plasmid Persistence. *PloS one*, 11(3), e0151969. https://doi.org/10.1371/journal.pone.0151969
- Khumsupan, P., Ramirez, R., Khumsupan, D., & Narayanaswami, V. (2011). Apolipoprotein E LDL receptor-binding domain-containing high-density lipoprotein: a nanovehicle to transport curcumin, an antioxidant and anti-amyloid bioflavonoid. *Biochimica et biophysica acta*, 1808(1), 352–359. https://doi.org/10.1016/j.bbamem.2010.09.007
- Landgraf, W., & Sandow, J. (2016). Recombinant Human Insulins Clinical Efficacy and Safety in Diabetes Therapy. *European endocrinology*, 12(1), 12–17. https://doi.org/10.17925/EE.2016.12.01.12
- Lethcoe, K., Fox, C. A., & Ryan, R. O. (2022). Foam fractionation of a recombinant biosurfactant apolipoprotein. *Journal of biotechnology*, 343, 25–31. https://doi.org/10.1016/j.jbiotec.2021.11.004
- Mayo Clinic. (2022, January). *Iron deficiency anemia*. Mayoclinic.org. https://www.mayoclinic.org/diseases-conditions/iron-deficiency-anemia/symptoms-causes/syc-20355034
- NIH Office of Dietary Supplements. (n.d.). Strengthening knowledge and understanding of dietary supplements. https://ods.od.nih.gov/factsheets/Iron-Consumer
- Rosano GL and Ceccarelli EA (2014) Recombinant protein expression in Escherichia coli: advances and challenges. *Front. Microbiol*, *5:172*. doi: 10.3389/fmicb.2014.00172
- Ryan, R. O., Schieve, D., Wientzek, M., Narayanaswami, V., Oikawa, K., Kay, C. M., & Agellon, L. B. (1995). Bacterial expression and site-directed mutagenesis of a functional recombinant apolipoprotein. *Journal of lipid research*, *36*(5), 1066–1072.
- Shi, L., Liu, H., Gao, S., Weng, Y., & Zhu, L. (2021). Enhanced Extracellular Production of *Is*PETase in *Escherichia coli* via Engineering of the pelB Signal Peptide. *Journal of agricultural and food chemistry*, 69(7), 2245–2252. https://doi.org/10.1021/acs.jafc.0c07469

- Song, X., Zheng, Y., Liu, Y., Meng, H., Yu, R., & Zhang, C. (2022). Conversion of recombinant human ferritin light chain inclusion bodies into uniform nanoparticles in *Escherichia coli* for facile production. *Engineering in life sciences*, 22(6), 453–463. https://doi.org/10.1002/elsc.202100164
- Sørensen, Hans & Mortensen, Kim. (2005). Soluble expression of recombinant proteins in the cytoplasm of Escherichia coli. *Microbial cell factories*. 4. 1. 10.1186/1475-2859-4-1.
- Tanaka, M., Hasegawa, M., Yoshimoto, N., Hoshikawa, K., & Mukai, T. (2019). Preparation of Lipid Nanodisks Containing Apolipoprotein E-Derived Synthetic Peptides for Biocompatible Delivery Vehicles Targeting Low-Density Lipoprotein Receptor. *Biological & pharmaceutical bulletin*, 42(8), 1376–1383. https://doi.org/10.1248/bpb.b19-00287

Virus-Infected Caterpillars Perform Better on Low Protein Compared to High Protein Diets

Anna Dlabalova, Dr. Angela Smilanich

Department of Biology, University of Nevada-Reno

Abstract

Junonia coenia densovirus (JcDV) is a single-stranded DNA virus from the *Parvoviridae* family that offers potential to be engineered for pest biocontrol, namely lepidopterans as they are the primary host. However, the effect of dietary metabolites on JcDV infection is poorly understood. Understanding this will offer strategies for controlling pest populations, especially those that are susceptible to JcDV. Prior studies with a different family of viruses (*Baculoviridae*) have found that infected caterpillars had higher survival when reared on a protein-rich diet vs. a low-protein diet. Our objective was to test whether a similar pattern would be found with JcDV, namely if a high protein diet enhanced performance, survival, and immunity in JcDV-infected painted lady caterpillars (*Vanessa cardui*: Nymphalidae). Immunity was measured by quantifying the activity of the phenoloxidase enzyme. Overall performance was measured as development time from 3rd to 5th instar and survival to pupation. We found that, contrary to previous research on a different family of viruses, the development time and survival to pupation was best for the infected group reared on a low-protein diet. We conclude that JcDV has a different effect on host metabolism compared to baculoviruses. In particular, there may be assimilation costs for the high protein diet.

Keywords: pest biocontrol, JcDV, dietary metabolites, Lepidoptera, fitness

Introduction

The United Nations predicted in 2017 that the world's human population will reach 9.7 billion people by 2050 (Melorose et al, 2015; Janssen et al, 2017). With an exponentially increasing human population (United Nations, 2022), it is crucial to have reliable crops that will sustain such growth. However, the productivity of crops is endangered by insect pests. A concrete example includes the Lepidoptera species *Spodoptera frugiperda* (Noctuidae), commonly known as the fall armyworm, whose populations are increasing at alarming rates (Matthews, 2018). The Junonia coenia densovirus

(Protoambidensovirus lepidopteran1) has good potential to be engineered for use in insect pest biocontrol, and specifically Lepidoptera, as they are the primary host of this virus (Labadie et al, 2021). This potential has been supported by both field trials and laboratory engineering work (Bergoin & Tijssen, 1998; Carlson et al, 2006; Labadie et al, 2021). Its specificity for lepidopterans has the potential to be engineered to achieve a much more precise and effective elimination of pests than in chemical products (Hussain et al, 2022). To help achieve the effectiveness and precision of such engineering, it is necessary to understand the effect of the virus on other non-pest insects, including other

Lepidoptera such as the widely distributed *Vanessa cardui* used in this experiment, and the effect JcDV has on them under different nutritional scenarios. The effect of JcDV on Lepidoptera is just beginning to be investigated (Smilanich et al. 2018, Muchoney et al. 2022, 2023), and the effects of primary metabolites from dietary sources is completely unknown. Primary metabolites are molecules that are directly involved in metabolism, such as carbohydrates, proteins, and lipids. The objective of our experiment was to investigate the effect of the primary metabolite protein on the fitness of JcDV infected non-pest species *Vanessa cardui*.

Studies with the nucleopolyhedrovirus (NPV) from a different family of viruses (Baculoviridae), have shown that infected caterpillars have better survival when consuming a proteinrich diet (Lee et al, 2005). In addition, the immune response as measured by the activity of the phenoloxidase enzyme of caterpillars reared on a high-protein diet showed higher activity than that of caterpillars reared on a low-protein diet. The phenoloxidase (PO) cascade is used routinely to determine immune system activity in caterpillars (Lee et al, 2005, Resnik and Smilanich 2020; see Wilson et al. 2001 for details on its relevance to the immune system). The phenoloxidase cascade plays a major role in the lepidopteran immune system; it is based on the specific chain reaction of enzymes which culminates in the melanization of the pathogen or parasite. Since activation of the immune system relies on many complex enzymes, it is costly in terms of protein. It has been hypothesized that the high-protein diet replenishes caterpillars with protein needed for the PO cascade immune response

(Lee et al, 2005). Thus, in this experiment, we measured the immune system activity through PO immune assays. We also measured fitness by examining survival into pupal stage. Another measure of fitness was development time; the faster adulthood is reached, the higher the chances are of mating and producing a new generation onto which genes are passed ("slow-growth-highmortality" hypothesis; Benrey & Denno, 1997).

Methods

Insect rearing

The lepidopteran species used for this experiment was Vanessa cardui, the painted lady butterfly. It is one of the most widespread butterfly species and has one of the widest diet breadths of all butterflies (Zhang et al, 2021), making it a good model to simulate non-pest lepidoptera response to JcDV infection in various nutritional scenarios. Given the time-constrained nature of our research, laboratory-bred Vanessa cardui were ordered from Carolina Biological Supplies in the form of 8 "batches" (maternal lines could not be traced) of 30 eggs each, totaling 240 eggs. Individuals were reared in climatecontrolled incubators (Percival growth chambers) (in separate control vs virus chambers later in the experiment) at the University of Nevada, Reno campus at a "standard" rearing temperature and diurnal cycle of 15 hour day at 25°C, and 9 hour night at 20°C (same rearing conditions as used in closely related research, see Muchoney 2022; Muchoney 2023, differing only by one hour in diurnal cycle due to incubator technicalities). Individuals were reared on a general painted lady rearing diet provided by Carolina Biological until the

end of second instar (each instar is separated by molting) in their respective "batches" in 5 ounce plastic soufflé cups. At third instar, each caterpillar was placed in an individual 2 ounce plastic soufflé cup and was systematically assigned to one of four treatment groups based on treatment and diet: control/ low protein, control/ high protein, virus/ low protein, virus/ high protein. Individuals were reared on their respective low protein/high protein diets up to death or pupation. The two artificial diets (based on Mason, Smilanich, & Singer, 2014) mimicked a "normal" plant-based diet of a caterpillar and differed by protein (casein) to carbohydrate (sucrose) ratio at 15P:25C (low protein) and 35P:5C (high protein). This ratio was prepared to equal 16g dry weight. Thus, the low-protein diet contained 6.0g of casein and 10.0g of sucrose, and the high protein diet contained 14.0 g of casein and 2.0g of sucrose. In addition, macro and micronutrients essential to the growth of the caterpillar in respective concentrations from previous research (Smilanich 2014) were added. Namely, for both diets, another macronutrient added was cellulose (22.27g), mimicking the main carbohydrate source intake from plants that a caterpillar would ingest in a natural environment. Micronutrients essential to the growth of the caterpillar were added, namely: Wesson's salt mixture (0.96g), linoleic acid (0.2g), cholesterol (0.2g), ascorbic acid (0.12g), methyl paraben (0.25g), vitamin mix (0.21mL), choline chloride (0.3mL). Finally, to achieve a solid and homogeneous consistency of the artificial diet, agar (5.12g) and boiling water (160mL) were added to both diet mixtures, mixed in a blender and left to cool until a solid consistency was reached. Diets were administered ad libitum via diet cubes.

Individuals were monitored daily for mortality and development.

Administration of JcDV

JcDV infects Lepidoptera species via ingestion of viral particles; the virus that is on/in the diet is ingested and passes through the midgut to further replicate in tracheae, haemocytes, visceral muscles and epidermis (Muchoney et al, 2023; Wang et al, 2013). Thus, the virus treatment groups were experimentally infected with JcDV via respective low/high protein artificial diet disc ingestion inoculated with 1*10⁷ viral particles/μL JcDV. Individuals were given 24 hours to ingest the disc, after which they were re-administered their respective low/high protein diets via diet cubes.

Immune assays

During 5th instar individuals with weight equal or greater to 0.2800 g were assayed to assess the strength of their immune response by measuring the activity of the phenoloxidase (PO) enzyme. To conduct this, hemolymph (i.e. insect "blood") was extracted from individuals (n=137), immediately after which a spectrophotometric assay was performed based on the protocol of Smilanich et al. (2018).

For each assayed individual, hemolymph was extracted by gently piercing the cuticle of the caterpillar's final abdominal proleg with a fine entomological needle sterilized with 70% ethanol. Using a pipette, 5 μ L of the individual's hemolymph was transferred into chilled Eppendorf tubes containing 250 μ L of ice-cold phosphate buffered saline (prepared as a mixture of one tablet of PBS (Sigma-Aldrich) dissolved in 100mL of DI water) and vortexed. A 100 μ l aliquot of the solution obtained was

transferred onto a single well into a 96-well microplate (BioRad iMark). For each immune assay run, one of the wells served as a "blank" containing 100 µl of PBS. Immediately after transferring all individuals' aliquots and the PBS "blank" into the microplate, 200 µL of 5 mM Ldopamine (prepared as a mixture of 30mL DI water and 0.0229 L-Dopamine powder (Sigma Aldrich) was added to each microplate well. Dopamine is the substrate needed for the reaction to occur between it and the phenoloxidase enzyme, if the latter is present in the aliquot. Phenoloxidase activity is positively related to darkness of aliquot and absorbance (optical density, OD). Colorimetric measurements were recorded using the iMark Microplate Absorbance Reader (Bio-Rad), which measured absorbance at 490 nm every 30 seconds for 45 minutes. PO activity was calculated as the slope (OD 490/min) over the 45-min period, during which time the enzymatic reaction remained in the linear phase. PO activity measurements which had values of less than zero (n=90) were excluded from statistical analysis.

Results

All analyses were performed using SAS software (version 9.4). Contrary to results from research on a different family of viruses (Lee et al. 2006), we found that survival and development time were best for individuals that were infected with JcDV and reared on the low protein diet.

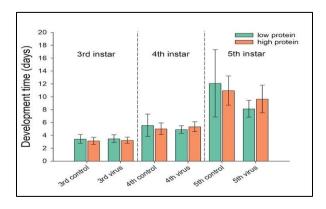


Figure 1: Development time for instars 3, 4, and 5. We found that the development times were similar across treatment and diet for 3^{rd} and 4^{th} instars. However, at the 5^{th} instar, development time was significantly faster for infected individuals in the low protein group (MANOVA, treatment: $F_{1,230}$ =45.36, P<0.001; diet: $F_{1,230}$ =1.07, P=0.314).

Development time of individuals in instars 3, 4 and 5 was analyzed using MANOVA (**Figure 1:** treatment: $F_{1,230}$ =45.36, P<0.001; diet: $F_{1,230}$ =1.07, P=0.314). Development times were similar across treatment and diet for 3^{rd} and 4^{th} instars. However, at the 5^{th} instar, development time was significantly faster for infected individuals reared on a low protein group.

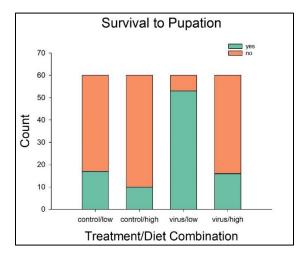


Figure 2: The highest survival to pupation was found for individuals that were infected and reared on the low protein diet (Chi-square = 46.68, P<0.001).

In a similar pattern, we observed that the highest survival to pupation, measured as a yes/no variable count, was found for individuals that were infected and reared on the low protein diet (Figure 2: Chi-square = 46.68, P<0.001). Survival to pupation was otherwise generally low for the other three groups (low and high protein control groups and virus high protein groups). It is interesting to note that the control high protein group had the lowest survival to pupation, which is the opposite of what has been observed in NPV research, where the control group reared on the diet with the highest amount of protein (vs. carbohydrate) had highest survival to pupation (Lee et al. 2006).

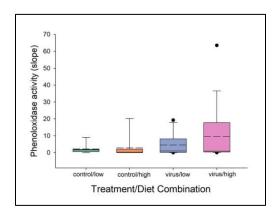


Figure 3: Immune response as measured by PO enzymatic activity was highest in infected individuals reared on the high protein diet, although not a significant effect (2-way ANOVA, treatment: $F_{1,47}$ =1.68, P=0.201; diet: $F_{1,47}$ =0.63, P=0.4311; treatment x diet: $F_{1,47}$ =0.38,P=0.539).

Overall, the immune response as measured by PO enzymatic activity was highest in infected individuals reared on the high protein diet, although we did not see a significant effect (**Figure 3:** 2-way ANOVA, treatment: $F_{1,47}$ =1.68, P=0.201; diet: $F_{1,47}$ =0.63, P=0.4311; treatment x diet: $F_{1,47}$ =0.38, P=0.539). Overall, the PO enzymatic activity was low, which is mostly likely due to the artificial diet that was used for rearing compared to a plant-based diet (Smilanich per. observation). It is possible that the artificial diet did not contain all the necessary micronutrients/metabolites to support a strong immune response.'

Conclusion: Discussion of results and directions for future research

The larger picture of this experiment is to understand under which nutritional scenarios JcDV is the least lethal to a nonpest species (Vanessa cardui), that is, in a low or high protein crop. To mimic a reallife nutritional scenario, we used artificial diets with a varying protein to carbohydrate ratio. We found that overall, there is the highest performance in development time and survival to pupation in the virus low protein group, which may indicate that this is where the JcDV virus is least lethal to a vastly distributed non-pest lepidoptera species, Vanessa cardui. It is important to note however that aspects of performance might differ from species to species, including the strength of the immune response. Future research on how different amounts of protein affect the performance of other non-pest lepidopteran species should be a priority for better insight into viral-host interactions.

Though it might seem counterintuitive that the individuals reared on a low protein diet had better survival and faster development time, a similar trend was found by Mason et al. (2014) with the generalist caterpillar, Grammia incorrupta (Erebidae), where individuals that were immune challenged preferred a high carbohydrate diet versus a high protein diet. We speculate that this trend is due to the carbohydrate component of the diet ratios. In fact, the low protein artificial diet had a high carbohydrate content, and vice versa. Similar to human bodies, the excess carbohydrates in caterpillars may be stored in the form of fat in the fat body, which is the site of production for many caterpillar immune system precursors (Mason et al, 2014; Beckage, 2008). Thus, it is interesting to note that the carbohydrate component might be of more interest than protein in similar future studies.

Although we saw a similar trend to that in previous research with NPV in immune activity, with the high protein groups displaying higher immune activity as compared to low protein groups, this was not a significant effect across treatment (P=0.201), diet (P=0.431) and treatment and diet crossed (P=0.539). The lack of significant effect was most likely due to the small sample sizes per each group, which was caused by the elimination of the vast majority of assayed individuals from analysis given they had immune assay slope values of less than zero. We attribute the low PO activity to the defined diets that the caterpillars were reared on, which may have lacked the necessary micronutrients to support a strong PO response. For future

studies, we encourage doing a similar study but with a pest species such as Spodoptora furgiperda rearing it on plant based diets of agricultural crops that would significantly differ in protein to carbohydrate nutritional values. This could give good insight as to in which nutritional scenario JcDV can be the most effective for controlling pest populations. We would also encourage doing a similar experiment with a different virus from a different family of viruses and compare it to that of the trends seen with JcDV (Parvoviridae) and NPV (Baculoviridae) to understand how insect species perform on low protein compared to high protein diets.

Acknowledgments

We would like to thank Nadya Muchoney for valuable technical support, training and advice. We would also like to thank Victoria Peechatt for insight and assistance. This work was supported by the Nevada Undergraduate Research Award.

References

- Armbruster, P., & Hutchinson, R. A. (2002). Pupal mass and wing length as indicators of fecundity in *aedes albopictus* and *aedes geniculatus* (diptera: Culicidae). *Journal of Medical Entomology*, *39*(4), 699–704. https://doi.org/10.1603/0022-2585-39.4.699
- Benrey, B., & Denno, R. F. (1997). The slow-growth--high-mortality hypothesis: A test using the cabbage butterfly. *Ecology*, 78(4), 987. https://doi.org/10.2307/2265852
- Bergoin, M., & Tijssen, P. (1998). Biological and molecular properties of Densoviruses and their use in protein expression and biological control. *The Insect Viruses*, 141–169. https://doi.org/10.1007/978-1-4615-5341-0 6
- Carlson, J., Suchman, E., & Buchatsky, L. (2006). Densoviruses for control and genetic manipulation of mosquitoes. *Advances in Virus Research*, 361–392. https://doi.org/10.1016/s0065-3527(06)68010-x
- Hussain, A. G., Wennmann, J. T., Goergen, G., Bryon, A., & Ros, V. I. D. (2021). Viruses of the fall armyworm spodoptera frugiperda: A review with prospects for biological control. *Viruses*, *13*(11), [2220]. https://doi.org/10.3390/v13112220
- Janssen, R. H., Lakemond, C. M., Fogliano, V., Renzone, G., Scaloni, A., & Vincken, J.-P. (2017). Involvement of phenoloxidase in browning during grinding of Tenebrio molitor larvae. *PLOS ONE*, 12(12). https://doi.org/10.1371/journal.pone.0189685
- Labadie, T., Garcia, D., Mutuel, D., Ogliastro, M., & Cambray, G. (2021). Capsid proteins are necessary for replication of a parvovirus. *Journal of Virology*, 95(17). https://doi.org/10.1128/jvi.00523-21
- Lee, K. P., Cory, J. S., Wilson, K., Raubenheimer, D., & Simpson, S. J. (2005). Flexible diet choice offsets protein costs of pathogen resistance in a Caterpillar. *Proceedings of the Royal Society B: Biological Sciences*, 273(1588), 823–829. https://doi.org/10.1098/rspb.2005.3385
- Mason, P. A., Smilanich, A. M., & Singer, M. S. (2014). Reduced consumption of protein-rich foods follows immune challenge in a polyphagous Caterpillar. *Journal of Experimental Biology*. https://doi.org/10.1242/jeb.093716
- Matthews, G. (2018). The spread of Fall Armyworm (FAW) *spodotera frugiperda*. *Outlooks on Pest Management*, 29(5), 213–214. https://doi.org/10.1564/v29 oct 07
- Melorose J, Perroy R, Careas S. World population prospects. *United Nations*. 2015;1: 587–92.
- Muchoney, N. D., Bowers, M. D., Carper, A. L., Teglas, M. B., & Smilanich, A. M. (2023). Use of an exotic host plant reduces viral burden in a native insect herbivore. *Ecology Letters*, 26(3), 425–436. https://doi.org/10.1111/ele.14162
- Muchoney, N. D., Bowers, M. D., Carper, A. L., Mason, P. A., Teglas, M. B., & Smilanich, A. M. (2022). Use of an exotic host plant shifts immunity, chemical defense, and viral burden in wild populations of a specialist insect herbivore. *Ecology and Evolution*, 12(3). https://doi.org/10.1002/ece3.8723
- Smilanich, A. M., Langus, T. C., Doan, L., Dyer, L. A., Harrison, J. G., Hsueh, J., & Teglas, M. B. (2018). Host plant associated enhancement of immunity and survival in virus infected caterpillars. *Journal of Invertebrate Pathology*, *151*, 102–112. https://doi.org/10.1016/j.jip.2017.11.006
- United Nations. (n.d.). *World Population Prospects 2022: Summary of results* | *Population Division*. United Nations. https://www.un.org/development/desa/pd/content/World-Population-Prospects-2022

- Wang, Y., Gosselin Grenet, A. S., Castelli, I., Cermenati, G., Ravallec, M., Fiandra, L., Debaisieux, S., Multeau, C., Lautredou, N., Dupressoir, T., Li, Y., Casartelli, M., & Ogliastro, M. (2013). Densovirus crosses the insect midgut by transcytosis and disturbs the epithelial barrier function. *Journal of Virology*, 87(22), 12380–12391. https://doi.org/10.1128/jvi.01396-13
- Wilson, K., Cotter, S. C., Reeson, A. F., & Pell, J. K. (2001). Melanism and disease resistance in insects. *Ecology Letters*, 4(6), 637–649.https://doi.org/10.1046/j.1461-0248.2001.00279.x
- Zhang, L., Steward, R. A., Wheat, C. W., & Reed, R. D. (2021). High-quality genome assembly and comprehensive transcriptome of the painted lady butterfly *Vanessa Cardui*. *Genome Biology and Evolution*, *13*(7). https://doi.org/10.1093/gbe/evab145

Accounting for Long-Term Drought in the Development Process in the Truckee Meadows Watershed

Jacob Rosenbaum, A.S., Environmental Science, University of Nevada, Reno Sameer Bhattarai, Ph.D., Truckee Meadows Community College

Author Note

Correspondence concerning this article should be addressed to JRosenbaum@nevada.unr.edu.

Abstract

Snow-melt dominated watersheds are very susceptible to the impacts of drought and climate change. This is exacerbated in areas such as the Truckee Meadows in northern Nevada that are undergoing growth, and where water resources are presently fully- or over-allocated. Surface water rights in the Truckee Meadows watershed are fully allocated, and growth is currently accommodated by the purchase of existing water rights from agricultural users. Ground water resources are over-allocated, with typical annual withdrawals above the perennial yield of the region's aquifers. Results of climate change models indicate that water concerns will increase over the next few decades. Bringing all water users into the water allocation process and enacting an offset program so that developers must dedicate more water resources than the anticipated water use for a project would help mitigate likely water scarcity.

Keywords: Watershed, Truckee Meadows, Development

Introduction

Drought is generally defined as a prolonged period of abnormally dry weather that results in ecological, social, or economic impacts. Drought is a recurrent problem in many western states, including Nevada, and the frequency and duration of drought appears to be increasing (Biondi & Meka, 2019). Water managers must account for anticipated drought in their regional plans. However, in a snow-melt dominated hydrological region such as the Truckee Meadows of northern Nevada, it can be difficult to plan very far ahead. The sources of water in the Truckee Meadows watershed include groundwater as well as surface waters. The water is used to meet tribal requirements, agriculture, cities and

industries, mining and geothermal, water recreation, and wetlands restoration needs. The primary surface water source is the Truckee River, which passes through the Reno-Sparks Metropolitan area, as well as a variety of smaller communities; it has long been used to meet the water needs of these regions. Supplemented by additional groundwater in dry years, this worked well for much of the region's existence.

The multitudinous uses of the watershed must be balanced within the confines of the available, sustainable water resources. The Truckee Meadows Water Authority (TMWA), jointly operated by Reno, Sparks, and Washoe County, is the primary water provider to the region, with water provided in outlying regions by

General Improvement Districts (GIDs), such as Canyon GID in Lovelock, NV. Those not serviced by the water providers obtain water directly from surface water diversions and/or individual wells. There is enough water for about 500,000 people in the region for residential uses, together with industry and agricultural uses (TMWA, 2015). However, the population in this area has been growing rapidly in recent decades (Figure 1). Projections of this trend indicate that the population currently approaches 500,000, and will soon exceed that figure. Concomitant with this population growth is an increase in assorted business and industry. As the population in the Truckee Meadows grows, water demands increase, leading to potential shortfalls, particularly in times of drought.

Like many western states, Nevada uses a water rights system. Under Nevada Revised Statutes (NRS) §278.377 and §533-534, regulatory agencies require that developers buy and dedicate water rights to meet the needs of proposed development, although there is an exception in NRS §534.180 for domestic wells where there is no public water provider. The dedication of water rights was intended to achieve and maintain sustainable water use. As populations have grown in the past few decades, water use has shifted from agricultural to municipal uses, but has generally remained at relatively sustainable rates. However, results from various climate change models indicate that global climate change will significantly impact water resources in this region (e.g., Kitlasten et al., 2021). While the total amount of precipitation is not expected to vary much,

the form will be drastically different, with substantially reduced winter snowpack, likely causing reduced groundwater recharge rates as well as a substantial decrease in summer-time surface water availability.

The purpose of this paper is to find an effective approach to accommodate likely limitations in water resources that result from rapid population growth and the anticipated effects of climate change.

Methods

The Truckee Meadows watershed is an enclosed snow-melt dominated basin that originates at Lake Tahoe in the Sierra Nevada mountains and terminates in Pyramid Lake (Figure 2). The watershed is located primarily in Nevada; the western portion is in California. Surface water flows primarily northeast via the Truckee River.

Various small streams and creeks drain into the Truckee River along its 140 mile length. The watershed encompasses 3120 square miles of variable terrain, from the forested mountains at the western edge of the watershed to range or scrub at the eastern border. The major population center, the Reno/Sparks area, is centrally located in the watershed. The majority of agriculture in the area is somewhat southeast of Wadsworth, NV, where part of the Truckee River is diverted to join the Carson Basin to irrigate the Newlands Project. While that area is not in the Truckee Meadows watershed per se, it does utilize substantial water from the Truckee River, and thus is included in many studies of the region.

Water resources data for the Truckee Meadows watershed were collected from various governmental and non-governmental agencies. Surface water resources data were primarily derived from the U.S. Geological Survey database, which operates streamflow gages throughout the country. Streamflow data are available in the region for the last 100+ years. Current ground water resources and allocations were obtained from the Nevada Division of Water Resources, while surface water allocations were obtained from TMWA. Water rights requirements associated with development were obtained from the Nevada Revised Statutes, while population figures for the region were obtained from the U.S. Census Bureau database.

The results of various climate change models were used to investigate the likely changes in water resources in terms of form, quantity, and timing of precipitation and of water availability in the region due to the anticipated effects of climate change (e.g., Kitlasten *et al.*, 2021; Seager *et al.*, 2012; Biondi & Meko, 2019).

Results

Allocating water resources in the West is often contentious. After extensive legal action, Truckee River water was fully allocated under the 1944 Orr Ditch Decree to a variety of users, primarily agricultural. This Decree set "the number of water rights by priority, by owner, and by quantity" (TMWA. 2015, p. 66). It allows the transfer of usage from one category to another, but the total amount of allocable Truckee River and tributary water (224,000 annual acrefeet) does not change. Over time, many of these agricultural surface water rights were sold and redirected to municipal and industrial uses (Figure 3). Currently, about half the water is allocated to TMWA for municipal and industrial uses, and as

municipal demands continue to grow, agricultural allocations are expected to decline. If the Truckee River and its tributaries do not have sufficient streamflow to deliver all allocated water, the amount delivered is reduced. The amount of water delivered in that situation considers seniority of water rights, but can be adjusted for the greatest perceived benefit by the U.S. Water Master and/or the State Water Engineer.

Groundwater resources are allocated to a variety of uses in the watershed (Figure 4). Unfortunately, as is often the case in the western states, the amount allocated is greater than the sustainable yield (Deacon *et al.*, 2007). The total committed groundwater resources of nearly 44,000 acre-feet per year are substantially above the perennial yield of 27,000 acre-feet per year (Nevada Division of Water Resources, 2021). In fact, the municipal, domestic, and domestic well uses together sum to slightly more than the perennial yield.

As the impacts of global climate change increase, droughts are expected to become more frequent and more severe (e.g., Biondi & Meko, 2019). Even in a typical water year, warmer temperatures are anticipated to result in reduced water availability during critical summer months (e.g., Kitlasten et al., 2021). In a 3°C temperature increase scenario, total water volumes are expected to remain relatively unchanged, but streamflow in the winter and early spring is expected to be substantially higher, whereas late spring and summer streamflow is likely to be significantly reduced (Figure 5).

Discussion

By 1944, all available surface water was fully allocated under the Orr Ditch Decree. Most of the water was allocated for agricultural purposes. Over time, the majority of those allocations have shifted to municipal-type uses, but the total amount allocated has not changed. However, the allocation for agricultural purposes is now only around 13 percent of the total surface water allocations (Figure 3), so these shifts to support population growth cannot continue indefinitely.

When there is a shortage of surface water in a drought situation of sufficient duration or intensity that reservoir storage is depleted, surface water allocations may be curtailed. In a low water year, water flows are about 20% of what they are in the typical water year, and are about half of the total allocated river waters (Bureau of Reclamation, 2016).

Unlike surface water allocations, ground water resources are allocated at levels over 60 percent above the sustainable yield (Figure 4). Further, when there is a shortage of surface water, many turn to groundwater to make up the shortfall. The resulting depletion in the groundwater table has been observed for well over half a century, and until relatively recently, public opinion seemed on the side of treating groundwater as a nonrenewable resource to be used until fully depleted, rather than attempting to keep withdrawals to a sustainable rate (Thomas, 1955). This has led to a declining water table, especially in the southwestern portions of the watershed (Figure 6).

This situation could be addressed with artificial aquifer recharge throughout

the watershed during wet years. However, under anticipated climate change scenarios, wet years will become rarer over the coming decades, with anticipated increased frequency, severity, and duration of drought conditions. Even in an average year, however, the Truckee Meadows watershed is a snow-melt fed system so anything that affects snowfall affects water resources year round. Winter storms in the Truckee Meadows come as atmospheric river events, depositing large amounts of snow in a short timeframe. This snow then melts slowly in the spring, delivering snow-melt into the Truckee River often well into July. The temperatures during snowfall are modest, such that a few degrees increase in temperature may result in the precipitation falling as rain rather than snow. Unlike snow that is stored on the ground for long periods, the rainwater quickly enters the river (Figure 5). Unfortunately, the irrigation season coincides with the reduced water availability timeframe. Thus, even though the river may deliver the same amount of annual water, the water will flow before it is needed for irrigation, and be gone by the time the crops need to be irrigated (Kitlasten et al., 2021). The reduced surface water availability during times of peak irrigation results in an increase in groundwater pumping; a substantial decrease in the water table of up to 28 feet near irrigation wells that are supplementing surface water delivery shortfalls are anticipated (Kitlasten et al., 2021). In addition to agricultural uses, municipal water demands are also higher in the summer months, exacerbating the situation. Significant depletion of area aguifers also leads to land fissures and

sinking as well as reduced water availability (Deacon *et al.*, 2007; Zektser *et al.*, 2005). The combined effects of climate change and population growth can easily lead to serious issues with water availability in the future.

There are a number of ways planners might address this. Each approach has advantages and disadvantages, and some approaches will be easier to implement than others. One method is to rework the water rights allocation system, perhaps using a market-based approach for trading water rights from lower to higher valued uses, as is done in other arid countries such as Australia (Productivity Commission, 2003). Such an approach would be challenging to accomplish, especially during the transition period while the value of the water is established. People would potentially see their water bills increase substantially in the short term while the market settles, but the approach would be effective in the long term. Another approach is to discount all water rights equally to account for potential reduced availability. This might be difficult to plan for, as water years are highly variable, but is similar to the approach taken in drought years, except that those reductions may or may not be equal. Currently, the Water Master and State Water Engineer determine how to distribute limited surface and ground water in a drought situation based on their assessment of the most beneficial use of the limited water. The current approach to accommodate population growth by purchasing water rights from agricultural users reduces the agricultural output of the watershed. The approach can be used to offset climate change impacts for a time, but once all the

available agricultural water has been acquired, growth will need to be accommodated in a different way.

Another approach would utilize changes to the current development approval process. The exemption to the acquisition of water rights for development using (nonreplacement) domestic wells should be eliminated. Groundwater is already overallocated, and new domestic wells automatically allow for further overallocation. Allowing individuals outside of the TMWA service area to purchase water rights from TMWA would enhance efficiency and ensure that the purchased water rights were acceptable. Further, while these wells are provided a standard allotment of two acre-feet per year, the wells are not metered so actual withdrawals are not known. Wells should be metered so that excess water usage can be curtailed.

Such changes for domestic wells are important, but most development in the Truckee Meadows watershed occurs in areas that are serviced by water providers such as TMWA. Because water resources are fully and/or over-allocated, and because anticipated global climate change is likely to reduce the available water, rights acquired for new development should be discounted. Currently, developers of a project must purchase and dedicate to the water provider water rights equal to the expected water use of the project. If we consider that the anticipated streamflow changes under a moderate climate change scenario such as that presented by Kitlasten et al. (2021) show a 30% reduction in summer streamflow, less water would be available in the summer. Therefore, we believe that at a

minimum, 1.3 acre-feet of water rights should be required for a project that is anticipated to require one annual acre-foot of water usage. For a private well, if users agree to an allocation of only one acre-foot of water rights, this should count as the offset requirement. The excess dedicated water rights will either reduce overallocation, or allow for aquifer replenishment.

Thus, to effect the suggested changes for domestic wells, we recommend revisions to NRS §534.180 to require that domestic wells be metered and that new non-replacement domestic wells be limited to 1 acre-foot of water rights per year unless

additional water rights are obtained. We further recommend that Washoe County Development Code be amended so that approval for development in the Truckee Meadows watershed requires 1.3 acre-feet of water rights be dedicated for each acre-foot of anticipated water use.

Acknowledgments

This work was funded through the National Science Foundation and Nevada Established Program to Stimulate Competitive Research (EPSCoR) Undergraduate Research Opportunity Program.

References

- Biondi, F., & Meko, D.M. (2019). Long-term hydroclimatic patterns in the Truckee-Carson Basin of the Eastern Sierra Nevada, USA. *Water Resources Research*, Vol. 55, pp. 5559–5574. https://doi.org/10.1029/2019WR024735
- Bureau of Reclamation (2016). Reclamation: Managing water in the West. Chapter 9: Truckee River Basin. 2016 SECURE Water Act Report to Congress.
- Deacon, J.E., Williams, A.E., Williams, C.D., & Williams, J.E. (2007). Fueling population growth in Las Vegas: How large-scale groundwater withdrawal could burn regional biodiversity. *Bioscience*. September. Vol. 57, No. 8, pp 88-98. doi.org/10.1641/B570809
- Kitlasten, W., Morway, E.D., Niswonger, R.G., Gardner, M., White, J.T., Triana, E., & Selkowitz, D. (2021). Integrated hydrology and operations modeling to evaluate climate change impacts in an agricultural valley irrigated with snowmelt runoff. *Water Resources Research*, Vol. 57, e2020WR027924. https://doi.org/10.1029/2020WR027924
- Nevada Division of Water Resources (2021). Groundwater commitments and availability. http://water.nv.gov/displayhydrographicmannerofusecommitment.aspx?basin=087. Retrieved 6/11/2021.
- Productivity Commission (2003). *Water rights arrangements in Australia and overseas*. Commission Research Paper, Productivity Commission, Melbourne.
- Seager, R., Ting, M., Li, C., Naik, N., Cook, B., Nakamura, J. & Liu, H. (2012). Projections of declining surfacewater availability for the southwestern United States. *Nature Climate Change*. DOI: 10.1038/NCLIMATE1787
- Thomas, H.E. (1955). Water rights in areas of ground-water mining. USGS Circular 347. U.S. Geological Survey, Washington DC. doi.org/10.3133/cir347
- Thomas, K.A. (2009). Organic compounds in Truckee River water used for public supply near Reno, Nevada, 2002-05: U.S. Geological Survey Fact Sheet 2009-3100. pubs.usgs.gov/fs/2009/3100/
- (TMWA) Truckee Meadows Water Authority (2015). 2016 2035 Water Resources Plan, Volume II, Chapters 1-6. www.tmwa.com/wp-content/uploads/docs/your_water/2035WRP/_2035_WRP_VolumeII_chptr_1-6 Final.pdf.
- (TMWA) Truckee Meadows Water Authority (2015b). 2016 2035 Water Resources Plan, Volume III, Appendix 2-9, Basin summaries. https://tmwa.com/wp-content/uploads/docs/your water/2035WRP/ 2035 WRP VolumeIII Appendices.pdf
- (TMWA) Truckee Meadows Water Authority (2020). Water rights. https://tmwa.com/wp-content/uploads/2020/12/Water_Rights_tp_20201223.pdf
- Zektser, S., Loáiciga, H.A., & Wolf, J.T. (2005). Environmental impacts of groundwater overdraft: selected case studies in the southwestern United States. *Environmental Geology*. Vol. 47, pp 396-404. DOI 10.1007/s00254-004-1164-

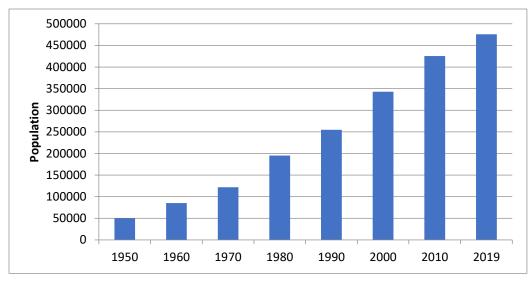


Figure 1.

Population of the Reno-Sparks Metropolitan Area. Data obtained from U.S. Census Bureau.

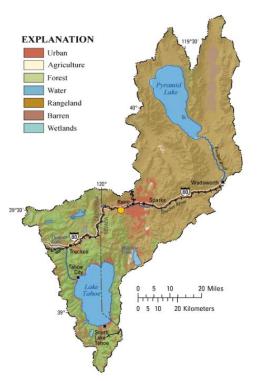


Figure 2. *The Truckee River Watershed*. Adapted from Thomas (2009).

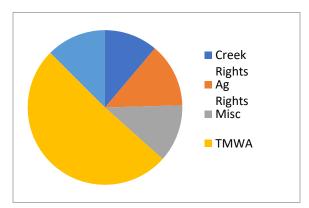


Figure 3. Surface water allocations in the Truckee River Watershed. Data derived from TMWA (2020).

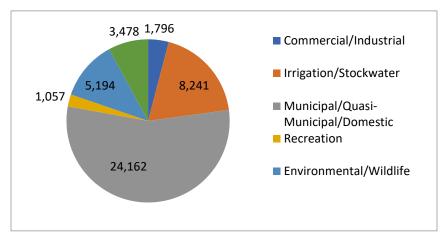


Figure 4. Committed groundwater resources in the Truckee River Watershed. Data from Nevada Division of Water Resources (2021).

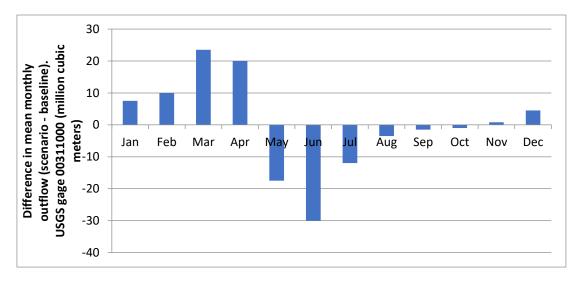


Figure 5. The difference in mean monthly outflow (scenario - historical waterflow baseline) in millions of cubic meters under a 3°C climate increase scenario. Adapted from Kitlasten et al. (2021).

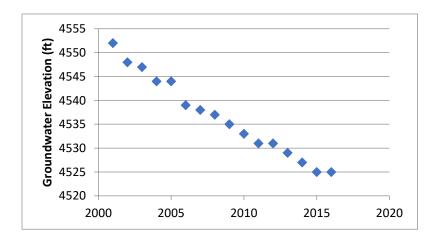


Figure 6. Groundwater elevation in a well in the southwest alluvial fan aquifer. Adapted from TMWA (2015b).

Disability and the Campus-Built Environment: Enhancing Mobility for UNR Students with Disabilities

Lea Gifford, Scott Kelley, Ph.D., Kerri Jean Ormerod, Ph.D.

Abstract

Navigating a university campus for people with disabilities can be helped or hindered by the built environment. This study investigates how buildings and pathways on the University of Nevada, Reno (UNR) campus could be improved to enhance mobility for students with disabilities. Using an online survey distributed through the Disability Resource Center (DRC), this research solicited feedback from students with disabilities to identify the most significant physical barriers to disabled students' ability to traverse buildings and pathways. The purpose of this study is to provide recommendations to increase access to equal opportunity for students with disabilities by identifying issues with and generating solutions for the built environment of the UNR campus.

Keywords: Disability, Accessibility, Mobility, Urban Built Environment, University Campus

Introduction

Under the social model of disability, the economic, physical, and social environment, rather than an individual's medical condition, results in the, "loss or limitation of opportunities to take part in the life of the community on an equal level with others," (Burchardt, 2004). The Americans with Disabilities Act (ADA) of 1990 along with the subsequent Accessibility Standards (2010) intended to address such environmental inequities, particularly those of the urban built environment. However, equal opportunity has not been achieved. Institutionalized discrimination against people with disabilities is perpetuated by inaccessible built environments (Imrie and Wells, 1993). Environments remain inaccessible due to the exclusion of disabled people from every step of ADA implementation, from policy creation to planning to evaluation to even research.

Current literature calls for increased involvement and participation from this community in all of these areas (Green, 2011; Nykiforuk et al., 2021; Sherman and Sherman, 2012).

Those in charge of implementing ADA standards do the bare minimum to meet compliance because they do not understand the true needs of people with disabilities (Sherman and Sherman, 2012). For example, rather than constructing ramps to every entrance of a building, only one is constructed for a back or side door. Past work demonstrates that the bare minimum is not enough to create the ADA's desired equity of opportunity for people with disabilities because an inaccessible built environment remains a barrier to participation in life activities, including work and education (Clarke et al., 2008; Nykiforuk et al., 2021; Sherman and Sherman, 2012; Green, 2011). A 2017 study

of the accessibility of six university campuses in Ghana concluded that, "...on a daily basis, students with disabilities have to struggle with the challenges posed by the nature of the built environment in order to pursue a degree in higher education," and therefore universities must construct inclusive buildings and facilities to provide disabled students with equal access to higher education (Tudzi et al., p. 286).

UNR Facilities Services conducted an evaluation of campus barriers in 2007. From this evaluation, barriers were prioritized in a 2008 transition plan, which was reviewed by the DRC Plan Review Committee. The plan was most recently updated in 2019 and identifies 1,485 inaccessible items. Top priority barriers include accessible parking and building entrances (2021 Annual Campus Accessibility Forum, 2021). However, this evaluation emphasized ADA compliance and did not solicit student feedback on accessibility.

In order to provide recommendations for how the UNR campus can be altered to increase mobility for disabled students, and thereby improve equal access to opportunity for these students, this research looks to discover which buildings and pathways on the UNR campus are most in need of attention and improvement, and identify which aspects of commonly-noted buildings and pathways are most problematic. Using an online survey (Appendix A), this study solicits feedback on what aspects of campus buildings and pathways, elements of UNR's urban built environment, limit mobility and impede equal access to opportunity for students with disabilities. This project

contributes to filling two research gaps. First, little research has been done that specifically evaluates access on university campuses, or school campuses of any kind, especially in the United States. Second, the survey method for collecting data allows a large number of people with disabilities who must traverse UNR's campus to participate in the study. This adds a uniquely disabled perspective to a larger conversation concerned with creating equitable urban built environments.

Methods

To conduct our survey, we designed an online questionnaire using Qualtrics. The survey contained a mix of open-ended, multiple choice and Likert scale questions. We asked students to provide information on the type of disability they have and whether or not they use a mobility aid. Students were then asked to name two of the least accessible buildings on the UNR campus based on their personal experience. Students were asked to identify what made these buildings difficult to use from a pregenerated list of options, with the opportunity for an open-ended response. Students were also asked to name two of the most accessible buildings on the campus and identify what made these buildings easy to use. Ease and difficulty of use were used as indicators of accessibility and inaccessibility. At the end, students were asked to provide any other information they would like about the physical accessibility of buildings and pathways on campus. Prior to the survey's advertisement and distribution, the instrument and recruitment materials were reviewed and approved

(deemed exempt) by UNR's Institutional Review Board.

We pretested the survey instrument with five disabled people and one nondisabled person. These individuals did not take the final version of the survey. Based on their feedback, we made minor changes to the survey. The survey was then sent to the DRC who distributed it via email to the nearly 3,000 students registered with them initially in March 2022, with a reminder email sent three days later. The survey was open for one week and we received 198 responses, for a response rate of 6.6%. In our data analysis, we focused on the 36 responses that indicated they had a mobility impairment. We coded and analyzed this data in Excel, compiling summary statistics and making charts, histograms, and tables. To compare responses for specific buildings on campus, we subtracted the number of times a building was identified as inaccessible from the number of times a building was identified as accessible, providing a general "accessibility score" for each building on campus that received responses. Based on these findings we produced maps to show variations in how respondents perceived the accessibility of buildings across campus. To do so, we appended survey responses about specific buildings on campus to a shapefile produced by UNR Facilities Services Department and created choropleth maps of responses to questions regarding building accessibility in ESRI's ArcGIS Pro 2.9.

Results

1.1 Campus Buildings

As shown in Figures 1 and 2, the 36 participants identified 28 different buildings as the least accessible to them. Edmund J. Cain Hall (EJCH) was most frequently cited as the least accessible, with seven participants identifying this building. This was followed by the William J. Raggio Building (WRB) which was named six times. The other 26 identified buildings were named fewer than four times each, with 13 buildings identified only once.

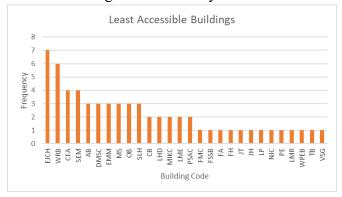


Figure 1. The 28 buildings identified as one of the least accessible on the UNR campus.

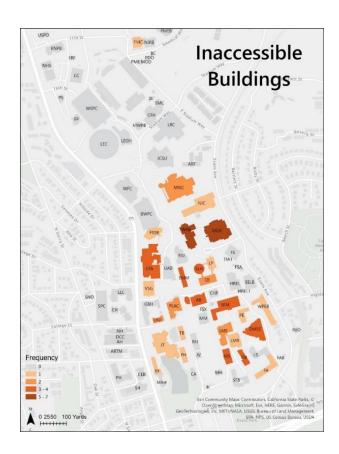


Figure 2. Map depicting how frequently participants identified buildings on the UNR campus as inaccessible.

Of the 10 physical features participants were asked to evaluate for the buildings they identified, Table 1 shows that all 10 potential barriers were identified as difficult to use for at least five of the buildings. Physical barriers were the greatest obstacles to accessibility. Doors were identified as difficult to use for 18 of the buildings, closely followed by stairs. Buildings that were more frequently identified as the least accessible usually had six to nine inaccessible features. Buildings only named once as the least accessible usually had one to four inaccessible features

Table 1. The frequency with which students identified building features as difficult to use with a breakdown for the top five least accessible buildings.

D '11'	T . 1	Top 5 Least Accessible Buildings					A11 O/1
Building Feature	Total Frequency	Edmund J. Cain Hall (EJCH)	William J. Raggio Building (WRB)	Church Fine Arts (CFA)	Scrugham Engineering and Mines (SEM)	Ansari Business Building (AB)	All Other 'Buildings
Doors	29	2	4	2	2	1	18
Stairs	28	6	2	2	3	1	14
Elevators	22	3	2	4	1	1	11
Sidewalks	21	0	1	0	1	2	17
Ramps	17	0	2	0	2	2	11

Rooms	15	0	3	0	1	1	10
Bathrooms	12	0	1	1	1	0	9
Hallways	8	2	1	0	1	0	4
Maps	7	2	2	0	0	0	3
Signs	5	1	0	0	0	0	4

Figures 3 and 4 show that participants identified 10 buildings as the most accessible to them, substantially fewer than those identified as least accessible. William N. Pennington Student Achievement Center (PSAC) was most frequently identified as the most accessible building on campus with 11 participants naming this building. The PSAC was followed by the Joe Crowley Student Union (JCSU), which was named 10 times, and the Mathewson-IGT Knowledge Center (MIKC), which was named nine times. Half of the most accessible buildings were named only once.

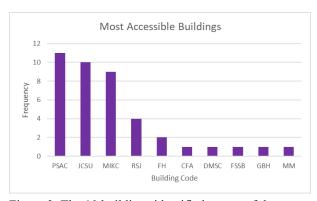


Figure 3. The 10 buildings identified as one of the most accessible on the UNR campus.

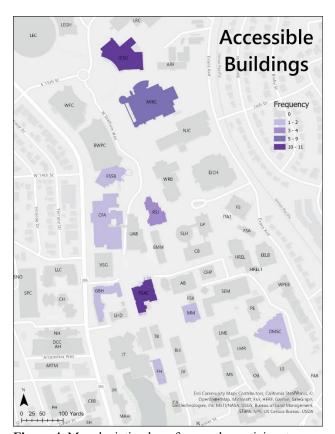


Figure 4. Map depicting how frequently participants identified buildings on the UNR campus as accessible.

Table 2 depicts the frequency with which specific building features were identified as easy to use in the most accessible buildings. All 10 of the potential barriers were identified as easy to use for at least three of these buildings. Eight out of 10

of the features were identified as easy to use for at least half of the named buildings. Buildings most frequently identified as accessible usually had eight to 10 accessible features and those that were only named once as the most accessible usually had one to two accessible features. Overall, elevators and doors were most often identified as easy to use in the most accessible buildings.

Table 2. The frequency with which students identified building features as easy to use with a breakdown for the top five most accessible buildings.

Building Feature	Total Frequency						All Other Buildings
		William N. Pennington Student Achievement Center (PSAC)	Joe Crowley Student Union (JCSU)	Mathewson IGT- Knowledge Center (MIKC)	Reynolds School of Journalism (RSJ)	Frandsen Humanities (FH)	Ü
Elevators	30	8	8	7	4	1	2
Doors	27	9	8	6	3	0	1
Bathrooms	24	7	7	5	3	0	2
Hallways	21	7	4	6	1	1	2
Rooms	15	7	2	2	2	0	2
Ramps	13	3	3	3	3	0	1
Sidewalks	12	4	3	2	2	0	1
Signs	7	3	1	1	1	0	1
Stairs	7	3	1	0	2	0	1
Maps	5	2	2	0	0	0	1

Overall, respondents reported that doors, stairs, elevators, and sidewalks hinder accessibility. For example, EJCH's stairs were the biggest concern for respondents (Figure 5). Figure 6 shows that this building's stairs are steep and have many

steps, which respondents said make them difficult to use. Elevators and doors were also a concern for EJCH. The most accessible buildings are noted for having easy to use doors and elevators. For example, these were both the most

accessible features identified for the JCSU (Figure 7). Figure 8 shows that JCSU's entranceway doors are wide and automatic, which respondents said make them easy to use.

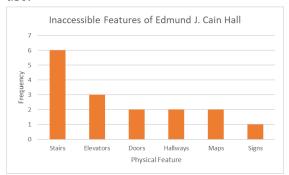


Figure 5. Bar graph depicting the number of times a building feature was identified as inaccessible in EJCH by participants.



Figure 6. EJCH stairs.

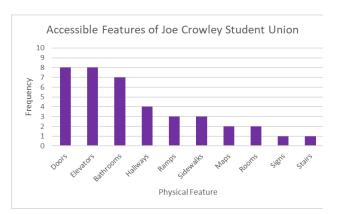


Figure 7. Bar graph depicting the number of times a building feature was identified as accessible in JCSU by participants.



Figure 8. JCSU doors.

The frequency with which buildings were identified as most and least accessible by research participants is shown in Figure 9. 28 buildings were identified as inaccessible while 10 were identified as accessible with 32 buildings named in total. Six of the 32 buildings (19%) were named by participants as one of the least and most accessible buildings on campus.

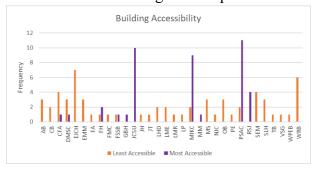


Figure 9. Buildings identified as most or least accessible.

Figure 10 illustrates the geographic distribution of building-level accessibility scores across the UNR campus. Negative scores (orange) indicate inaccessibility and positive scores (purple) indicate accessibility. JCSU earned the highest accessibility score of 10 and EJCH earned the lowest score of -7. Most buildings listed by participants in the survey are located in

the southern half of the campus, and buildings with negative accessibility scores are more frequent than those with positive scores.



Figure 10. Map depicting the accessibility scores of the most and least accessible buildings on the UNR campus. Positive scores denote accessibility. Negative scores denote inaccessibility.

We next consider the relationship between building age and accessibility scores. Taking just the top 10 buildings identified as the most accessible and the top 10 buildings identified as the least accessible, Figure 11 illustrates that eight out of the 11 buildings with negative accessibility scores were constructed prior to 1990, when ADA legally changed accessibility standards. This is compared to five out of the seven buildings with positive accessibility scores that were constructed after 1990. Church Fine Arts (CFA) and

Davidson Math and Science Center (DMSC) appear on both top 10 lists.



Figure 11. Timeline of when the top 10 least accessible and top 10 most accessible buildings were constructed with two repeated buildings. Dates from UNR's *Around campus* (n.d.).

1.2 Accessibility by Disability and Mobility Aid

The majority of the 36 participants with a mobility impairment identified as physically disabled (n=20, 56%) and/or chronically ill (n= 13, 36%). As Table 3 illustrates, those with physical disabilities identified 22 buildings as inaccessible, with EJCH, WRB, Ansari Business Building (AB), and Effie Mona Mack (EMM) appearing most frequently. Three participants in total identified AB as inaccessible and all three identified as physically disabled. This was the same for EMM. This group also identified nine of the 10 most accessible buildings, with the JCSU and PSAC most frequently named. As Table 4 shows, those with chronic illnesses named 14 buildings as difficult to use. CFA was named most frequently within this group. In total, CFA was named as one of the least accessible buildings by four participants and all four identified as chronically ill. Participants with chronic illnesses were also three of the seven people who identified EJCH as inaccessible. This group also named six buildings as easy to use, with JCSU and MIKC both named five times each.

Inaccessible Building	Frequency	Accessible Building	Frequency
AB	3	JCSU	5
ЕЈСН	3	PSAC	5
EMM	3	MIKC	3
WRB	3	RSJ	3
CFA	2	CFA	1
MIKC	2	FH	1
SEM	2	FSSB	1
СВ	1	GBH	1
DMSC	1	MM	1
FMC	1		
FSSB	1		
FA	1		
JT	1		
LHD	1		
LME	1		
LMR	1		
LP	1		
NJC	1		
OB	1		
PSAC	1		
SLH	1		
	1		

VSG	1	

Tables 4. Table depicts the frequency with which people identifying as chronically ill noted a building to be inaccessible (left) or accessible (right).

Inaccessible Building	Frequency	Accessible Building	Frequency
CFA	4	JCSU	5
ЕЈСН	3	MIKC	5
DMSC	2	DMSC	1
MS	2	FH	1
OB	2	MM	1
SLH	2	PSAC	1
SEM	2		
СВ	1		
EMM	1		
ЛН	1		
JT	1		
MIKC	1		
WPEB	1		
WRB	1		

Canes were the most used mobility aid for participants with 10 people indicating they used one at least some of the time while on the UNR campus. Table 5 shows that cane users identified 15 inaccessible buildings and only three of these buildings, Orvis Building (OB), Scrugham Engineering and Mines (SEM), and WRB, were named more than one time. This group made up the

majority of individuals who found PSAC to be the most accessible building, with six of the 10 cane users naming it as one of their most accessible. They also found JCSU, MIKC and Mackay Mines (MM) to be accessible. The three participants who indicated that they use a manual wheelchair all agreed JCSU was one of the most accessible buildings (Table 6).

Table 5. Table depicts the frequency with which people who use canes noted a building to be inaccessible (left) or accessible (right).

Inaccessible Building	Frequency	Accessible Building	Frequency
ОВ	2	PSAC	6
SEM	2	JCSU	2
WRB	2	MIKC	2
AB	1	MM	1
СВ	1		
CFA	1		
DMSC	1		
EJCH	1		
EMM	1		
FMC	1		
LHD	1		
MIKC	1		
NJC	1		
SLH	1		
ТВ	1		

Table 6. Table depicts the frequency with which people who use manual wheelchairs noted a building to be inaccessible (left) or accessible (right).

Inaccessible Building	Frequency	Accessible Building	Frequency
CFA	1	JCSU	3
ЕЈСН	1	MIKC	1
EMM	1		

JT	1	
LP	1	
MS	1	

In reviewing the open-ended responses that participants provided, we note that the most frequently identified topics pertained to the locations of buildings and disabled person parking. Three participants found it difficult to move between buildings because of the lack of nearby disabled person parking. Elevation differences created problems for four participants. Three participants found the distance between buildings to be too long and the paths to be indirect.

Discussion

The finding that newer buildings tended to score higher with respondents indicates that ADA standards did increase the accessibility of newly constructed buildings which had to be designed accordingly. In contrast, older buildings that needed to be retrofitted to meet standards have struggled to actually become accessible. This study did not examine how well ADA standards have been met in buildings so these older buildings may not actually meet accessibility standards or they have done so in a way that meets standards on paper while the accommodations remain difficult to access and utilize in practice. Also, given that three buildings with negative accessibility scores were constructed after 1990, this demonstrates that simply meeting ADA accessibility

standards is not alone sufficient to create fully accessible and equitable spaces.

The inaccessibility frequency findings can offer a guide for how to prioritize addressing modifications and upgrades to campus buildings to improve accessibility. The findings about building characteristics likewise offer a way to prioritize fixes by feature. For example, since EJCH was most frequently identified as the least accessible building, and its stairs were the biggest concern for participants who identified the building, addressing this issue should be the top priority. This issue could be resolved by replacing the stairs with a switchback ramp, for example. EJCH's elevators and doors were the next biggest concern for students. Since these features were noted as easy to use in the most accessible buildings, alterations to these features in EJCH and other inaccessible buildings, can be modeled after these features in accessible buildings like JCSU. The inaccessible doors in EJCH could be replaced with automatic doors like the ones found in JCSU.

While changes like these would be large and expensive projects, there are also smaller issues that can be more easily addressed. For example, participants wrote that the push buttons to open doors in EMM and the Jot Travis Building (JT) are broken or only function some of the time. Other participants indicated that the elevators in

CFA, EMM, and Paul Laxalt Mineral Research (LMR) are broken or work inconsistently. In these cases, the necessary accessibility features exist and they simply require maintenance. These issues require fewer resources to resolve and can notably improve the accessibility of these buildings.

The issues related to building locations and disabled person parking can be resolved in a couple of ways. More disabled person parking could be added to existing lots and more parking lots could be added around buildings currently lacking them. The DRC could also expand their services to provide rides for students with disabilities and assist them in moving between buildings that are far apart from each other or at notable elevation differences. A more environmentally friendly option could be to establish a bike-share program as other universities have done (Kellstedt et al., 2019). To make the program accessible, it should also include electric mobility scooters. A program like this at UNR could benefit all disabled and non-disabled students and staff who must travel significant distances and elevations across campus, especially those like the two participants who noted that they needed mobility assistance but were ineligible for disabled person parking.

Conclusions

This study collected the opinions of University of Nevada, Reno students registered with the DRC in the Spring 2022, finding that students identified 28 inaccessible buildings and 10 accessible

buildings, with six buildings falling into both categories. The lack of accessibility takes many forms; however, doors, stairs, elevators and sidewalks were the most commonly reported inaccessible features in our study. While UNR's newer buildings are notably more accessible than older buildings, the newer ones still have inaccessible features that need to be addressed. While the older buildings were retrofitted to meet ADA standards, they remain difficult to use and more work must be done on them to make them accessible for students with disabilities. Future research should repeat this study as the student body changes periodically. The study could be expanded to include disabled staff as well as students with disabilities who are not registered with the DRC. The survey could also be distributed within the larger Reno area or on other university campuses. Since this study also focused on only two areas of physical accessibility, buildings and pathways, future research should examine other elements of the built environment, such as accessible parking provisions, distance between buildings, and elevation changes between buildings. Other research should expand beyond physical disability to focus on neurodiversity and built environment accessibility, as well.

Acknowledgments

This research was funded by the Nevada Undergraduate Research Award. Thank you to the UNR Disability Resource Center, Dr. Randall Owen, Dr. Marney White and the research participants.

Appendix A - Survey Instrument

I invite you to participate in a short, confidential survey about the physical accessibility of the UNR campus for people with disabilities. This research is being funded by the UNR Office of Undergraduate Research. My goal is to identify which physical elements on the University of Nevada Reno (UNR) campus could be improved to enhance mobility for students with disabilities. The survey should take about 10 minutes to complete. I'm reaching out to you because you are registered with the Disability Resource Center (DRC) and we believe you could provide valuable feedback that will help us better understand barriers to physical accessibility on the UNR campus. This can help inform future recommendations for on-campus improvements to facilitate better accessibility.

This research is considered to be minimal risk of harm. This means the risks of your participation in the research are similar in type or intensity to what you encounter during your daily activities. UNR researchers will treat the information collected about you with professional standards of confidentiality and protect it to the extent allowed by law. You will not be personally identified in any reports or publications that may result from this research. The US Department of Health and Human Services, UNR's Research Integrity Office, and the Institutional Review Board may look at your study records.

Your participation in this survey is voluntary. You may stop at any time. Declining to participate or stopping will not

have any negative effects on you or impact your accommodations with the DRC. If you have questions about your participation, please contact Lea Gifford at leagif@nevada.unr.edu or 775-781-2066. If you have concerns or complaints about this research, you may report them (anonymously if you so choose) by contacting Dr. Kerri Jean Ormerod at 775-784-6347 or kormerod@unr.edu, Dr. Scott Kelley at 775-784-6705 or scottkelley@unr.edu, or the University of Nevada, Reno Research Integrity Office at 775-327-2368.

By clicking the "next" arrow, you agree that:

- You have read the above information
- You voluntarily agree to participate
- You are a student at UNR
- You are at least 18 years of age

Do you have an impairment that impacts your mobility? (Choose one)

- Yes
- No
- Unsure

Which category(ies) describe you? (Check all that apply)

- Amputee
- Blind/Visually Impaired
- Chronically Ill
- Deaf/Hard of Hearing
- Paraplegic
- Physically Disabled
- Undiagnosed
- Other

Do you use a mobility aid while on the UNR campus? (Choose one)
• All the time
Most of the time
• Some of the time
• Never
Prefer not to say
What kind of mobility aid(s) do you use? (Check all that apply)
• Cane
• Crutch(es)
Manual Wheelchair
Power Chair
• Prosthetic(s)
• Scooter
Service Animal
• Other
In your experience, which two (2) buildings on the UNR campus are the LEAST accessible to you? (The next set of questions are about your experience with these two buildings.)
Building 1Building 2
Thinking about [Building 1/2], which of the following are difficult to use? (Check all that apply)
• Bathrooms
• Doors
• Elevators
Hallways
• Maps
±

• Ramps (Outdoor and/or Indoor)

• Rooms (Classrooms, Offices, Labs, etc.)

- Sidewalks (Around Building)
- Signs
- Stairs (Outdoor and/or Indoor)
- Other _____

(For any of the above that are selected by a respondent, they would see any of the corresponding follow-up questions, otherwise these next questions are skipped)

Why are bathrooms difficult to use in [Building 1/2]? (Check all that apply)

- Difficult to find
- Too small
- Lighting is too bright
- Lighting is too dim

•	No or bad handrails Other
T 71	
Why ar	e doors difficult to use in [Building 1/2]? (Check all that apply)
•	Difficult to find
•	No push buttons
•	Push buttons are difficult to find
•	Don't stay open long enough
•	Too narrow
•	Too heavy
•	Other
Why ar	e elevators difficult to use in [Building 1/2]? (Check all that apply)
•	Difficult to find
•	Buttons are difficult to find
•	Too small
•	Too far from entrances
•	Lighting is too bright
•	Lighting is too dim
•	Feel unsafe
•	Other
Whv ar	e hallways difficult to use in [Building 1/2]? (Check all that apply)
•	Too narrow
•	Too long
•	Cluttered
•	Uneven surface
•	Slippery surface
•	Lighting is too bright
•	Lighting is too dim
•	Feel unsafe
•	Other
M/hrr om	a mana difficult to was in [Dwilding 1/2]? (Charle all that annly)
wny ar	e maps difficult to use in [Building 1/2]? (Check all that apply)
•	Difficult to find
•	Too small
•	Too high
•	Difficult to understand
•	Out of date
•	Other
-	e ramps (outdoor and/or indoor) difficult to use for [Building 1/2]? all that apply)

- Difficult to find
- Too steep
- Too narrow
- Too long

•	Uneven surface
•	Other
Why are	e rooms (classrooms, offices, labs, etc.) difficult to use in [Building 1/2]? (Check
all that	apply)
•	Difficult to find
•	Too small
•	Too narrow
•	Cluttered
•	Lighting is too bright
•	Lighting is too dim
•	Furniture is poorly arranged
•	Furniture can't be moved
•	Other
·	
Why ar	e sidewalks (around building) difficult to use for [Building 1/2]?
	all that apply)
•	Too steep
•	Too narrow
•	Too long
•	Uneven surface
•	Not enough curb cuts
•	Other
Why or	e signs difficult to use in [Building 1/2]? (Check all that apply)
·	Difficult to find
•	
•	Too small
•	Too high
•	No or unreadable Braille
•	Other
Why ar	e stairs (outdoor and/or indoor) difficult to use for [Building 1/2]? (Check all that
apply)	
•	Difficult to find
•	Too steep
•	Too narrow
•	Too wide
•	Too many steps
•	No or bad handrails
•	I can't use stairs
•	Other
-	
In your experience, which two (2)	buildings on the UNR campus are the MOST accessible to you? (The next set of
questions are about your experience	
,	5 /
Building 1	
Building 2	-
<u> </u>	

• Too short

•	bout [Building 1/2], which of the following are easy to use? (Check all that apply) Bathrooms			
•	Doors			
•	Elevators			
•	Hallways			
•	Maps			
•	Ramps (Outdoor and/or Indoor)			
•	Rooms (Classrooms, Offices, Labs, etc.)			
 Sidewalks (Around building) 				
•	Signs			
•	Stairs (Outdoor and/or Indoor)			
•	Other			
	Why are bathrooms easy to use in [Building 1/2]? (Check all that apply)			
	 Easy to find 			
	• They are large			
	 Lighting is just right or adjustable 			
	 Good handrails 			
	• Other			
	Why are doors easy to use in [Building 1/2]? (Check all that apply)			
	 Easy to find 			
	 Many push buttons 			
	 Push buttons are easy to find 			
	 Stay open for a long time 			
	 Automatic doors 			
	They are wide			
	They are wide			
	They are light			
	• They are light			
	They are lightOther			
	 They are light Other Why are elevators easy to use in [Building 1/2]? (Check all that apply) 			
	 They are light Other Why are elevators easy to use in [Building 1/2]? (Check all that apply) Easy to find 			
	 They are light Other Why are elevators easy to use in [Building 1/2]? (Check all that apply) Easy to find Buttons are easy to find 			
	 They are light Other Why are elevators easy to use in [Building 1/2]? (Check all that apply) Easy to find Buttons are easy to find They are large 			
	 They are light Other Why are elevators easy to use in [Building 1/2]? (Check all that apply) Easy to find Buttons are easy to find They are large Near entrances 			

- They are wide
- They are short
- Clear of clutter

•	Even surface
•	Surface is not slippery
•	Lighting is just right or adjustable
•	Feel safe
•	Other
Why or	e maps easy to use in [Building 1/2]? (Check all that apply)
• •	Easy to find
•	They are large
•	Height is just right
•	Easy to understand
•	Up to date
•	Other
•	e ramps (outdoor and/or indoor) easy to use for [Building 1/2]? (Check all that
apply)	
•	Easy to find
•	Slope is just right
•	They are wide
•	Length is just right
•	Even surface
•	Other
Why ar	e rooms (classrooms, offices, labs, etc.) easy to use in [Building
•	Check all that apply)
•	Easy to find
•	They are large
•	Clear of clutter
•	Lighting is just right or adjustable
•	Furniture is easy to navigate
•	Furniture can be moved
•	Other
•	e sidewalks (around building) easy to use for Building 1/2? (Check all that apply)
•	Slope is just right
•	They are wide
•	Length is just right
•	Even surface
•	Lots of curb cuts
•	Other
Whv ar	e signs easy to use in [Building 1/2]? (Check all that apply)
., 11y a1v	Easy to find
-	

- They are large
- Height is just right
- Readable Braille

•	Other				
---	-------	--	--	--	--

Why are stairs (outdoor and/or indoor) easy to use for Building 1/2? (Check all that apply)

- Easy to find
- Not too steep
- Width is just right
- No or few steps
- Good handrails

Is there anything else you would like to tell us about the accessibility of buildings and/or pathways on the UNR campus?

References

2021 annual campus accessibility forum. (2021, December 8). University of Nevada,

Reno. https://www.unr.edu/accessibility

Americans With Disabilities Act of 1990, 42 U.S.C § Subchapter III (1990).

https://www.ada.gov/pubs/adastatute08.htm

Americans with Disabilities Act: Accessibility standards. (2010). U.S. Access Board. https://www.accessboard.gov/ada/

Around campus. (n.d.). University of Nevada, Reno. https://www.unr.edu/around-campus

- Burchardt, T. (2004). Capabilities and disability: The capability framework and the social model of disability. *Disability & Society*, 19(7), 735-751. https://doi.org/10.1080/0968759042000284213
- Clarke, P., Ailshire, J. A., Bader, M., Morenoff, J. D., & House, J. S. (2008). Mobility disability and the urban built environment. *American Journal of Epidemiology*, 168(5), 506-513. doi: 10.1093/aje/kwn185
- Green, R. J. (2011). An introductory theoretical and methodological framework for a Universal Mobility Index (UMI) to quantify, compare, and longitudinally track equity of access across the built environment. *Journal of Disability Policy Studies*, 21(4), 219-229. doi: 10.1177/1044207310384998
- Imrie, R. F., & Wells, P. E. (1993). Disablism, Planning, and the Built Environment. *Environment and Planning C: Government and Policy*, 11(2), 213–231. https://doi.org/10.1068/c110213
- Kellstedt, D., Spengler, J. O., Bradley, K., & Maddock, J. E. (2019). Evaluation of free-floating bike-share on a university campus using a multi-method approach. *Preventive Medicine Reports*, *16*, 100981–100981. https://doi.org/10.1016/j.pmedr.2019.100981
- Nykiforuk, C. I. J., Glenn, N. M., Hosler, I., Craig, H., Reynard, D., Molner, B., Candlish, J., & Lowe, S. (2021). Understanding urban accessibility: A community-engaged pilot study of entrance features. *Social Science & Medicine*, 273, 1-5. https://doi.org/10.1016/j.socscimed.2021.113775
- Sherman, S., & Sherman, J. (2012). Design professionals and the built environment: Encountering boundaries 20 years after the Americans with Disabilities Act. *Disability & Society*, 27(1), 51-64. https://doi.org/10.1080/09687599.2012.631797
- Tudzi, E. P., Bugri, J. T., & Danso, A. K. (2017). Human rights of students with disabilities in Ghana: Accessibility of the university built environment. *Nordic Journal of Human Rights*, *35*(3), 275-294. https://doi.org/10.1080/18918131.2017.1348678

Understanding Jealousy Among Individuals with Minoritized Identities

Courtney Brothers, B.A., and Sarah Mitchell, Ph.D., CFLE

University of Nevada, Reno

Abstract

Past research on romantic jealousy has focused on measuring the aspects of jealousy in predominantly cis-gender, White, straight, college-aged participants using forced choice design and, often, hypothetical scenarios. These studies have yielded conflicting results in relation to sex-based differences in the experience of romantic jealousy. Research using more diverse participants and prioritizing lived experiences over hypothetical scenarios with forced choice design could reveal more about romantic jealousy in a real-world context, specifically in relation to precursors and reactions to jealousy as well as the outcomes. This qualitative study on romantic jealousy utilized semi-structured interviews from six participants with multiple minoritized identities. Three main themes resulted: 1) The conceptualization of jealousy, 2) Antecedents - behaviors - consequences (ABCs), and 3) Gender, race and ethnicity, and the intersection of identities. The first and third themes revealed that participants' intersecting identities appeared to have a significant impact on how they conceptualized jealousy and how they reacted to it. Under the second theme, participants identified similar antecedents, yet described a variety of different behaviors in response to them. Furthermore, even with this diversity in reported behavior, consequences tended to be relatively similar. A discussion of the results highlights the nuances of understanding previous, current, and future ABCs related to romantic jealousy, particularly for those with intersecting minoritized identities. Research implications and suggestions for practice are also discussed.

Keywords: Romantic jealousy, intersectionality, antecedents, behaviors, consequences

Understanding Jealousy Among Individuals with Minoritized Identities

Jealousy is a concept that affects everyone and has been defined in various contexts within the academic community. Romantic jealousy is a specific type of jealousy that can have serious consequences for individual partners as well as entire family units. Jealousy can become a form of control over a partner which can result in domestic abuse (Turell et al., 2018).

Conversely, emotional jealousy can strengthen a relationship in the respect that it shows the jealous partner values the relationship and it also opens the door for affective communication (Pfieffer & Wong, 1998; Rubinsky, 2019). Affective communication is "a spoken exchange of thoughts, opinions and feelings, which has the potential to arouse emotion in individuals engaged in the exchange" (Rhee et al., 2020, p. 4). This type of communication relies on nonverbal cues such as body language which helps open

emotional expression and vulnerability, which is vital to intimacy.

Past research on jealousy has focused on differences based on sex or gender. These studies are limited in their methodology where forced-choice paradigms are often used, that is people are presented a set number of options to choose from. This can often lead people to choose the closest option, or even a random option, if their true response is not represented among the options. Additionally, study samples are mostly White, cis-gender, heterosexual, college-age students, with some not specifying the sexuality of participants, often implying that they are straight (Broemer & Diehl, 2004; Buunk & Hupka, 1987; Demirtaş & Dönmez, 2006; Pines & Friedman, 1998). The findings of such studies are potentially exaggerated when compared to a study using "nationally representative survey of American households" which concluded that "significant sex differences only emerged for forced choice measures and not for continuous measures" where participants may place themselves on a sliding scale, indicating more accurate representations of themselves (Zengel et al., 2013, p. 47). There is very limited research on romantic jealousy in minoritized groups including those who have a minoritized sexuality, gender identity, race, or ethnicity. The experiences and perceptions of these underresearched populations may be different from White, cisgender, heterosexual populations, as minoritized communities experience higher levels of stress and face discrimination and stereotype threats (Balsam et al., 2011). There is a clear need

for research with more diverse and representative populations to provide greater insight into the experiences of romantic jealousy and impact on self-perception.

This study aims to highlight how jealousy is defined, experienced, and coped with by people with minoritized identities. Minoritized groups are underrepresented and understudied in many social sciences. Additionally, this is alarming given the replication crisis, or lack of studies being repeated to ensure the methodologies of studies are sound and the conclusions are valid (Schmidt, 2016). In retrospect, scholars can examine the flaws and limitations of past research, but there is no pressure to replicate these studies with improved methodologies or more representative samples. These past studies are the foundation of knowledge on a given subject and need to be clarified and contextualized. By using qualitative methods in addition to recruiting more diverse participants, this study can produce a more nuanced understanding of experiences, perceptions, and consequences of romantic jealousy for diverse populations.

Definitions and Types of Jealousy

Thorough research on jealousy did not appear until the 1980s and although the research has continued, it has been limited and sporadic. The definition of jealousy is complicated to pinpoint, as it may seem similar to envy; "Whether there is a distinction between envy and jealousy has been the cause for much philosophical debate" (Salovey & Rodin, 1984, p. 780). However, for romantic jealousy specifically, there is a generally agreed-upon definition in

that romantic jealousy is a reaction to a perceived or real threat to a valued relationship (Buunk & Hupka, 1987; Demirtas & Dönmez, 2006; Pfeiffer & Wong, 1998; Pines & Friedman, 1998; Steis et al., 2019; White, 1981a). This reaction has many triggers and involves emotions, thought processes and cognitions, behavioral reactions, and coping mechanisms (Pfeiffer & Wong, 1998). There are also several aspects that impact the expression of these reactions, including self-esteem, social comparison, and mate retention desire (Broemer & Diehl, 2004; Walsh et al., 2019; White, 1981a). Research has indicated that cognitive and behavioral jealousy may be more pathological (where thinking too much about jealousy or exhibiting a behavioral reaction can be harmful to relationships); conversely, a small amount of emotional jealousy is typically normal (Deans & Bhogal, 2019; Pfiffer & Wong, 1998). In fact, a reasonable amount of emotional jealousy can function to preserve the relationship by showing that a partner places value in the relationship and has been found to be positively related to love (Pfeiffer & Wong, 1998).

Theories on Jealousy

Researchers have developed several theories on jealousy. The one that has received the most focus is the evolutionary (or biological) theory of jealousy, which claims that men and women developed different jealousy mechanisms due to different needs (Basset, 2005; Pines & Friedman, 1998; Walsh et al., 2019). Under this view, there are two main types of infidelity; sexual and emotional. Men are more upset over sexual infidelity because

they would not be able to know if a child was theirs or not. Women are more upset over emotional infidelity because it would mean a man pulling his resources away from her and the child. This theory accounts for sex differences in types of infidelity. However, recent research has found sex differences to be less significant than originally claimed and cannot account for our evolving understanding of the spectrum of sexuality (de Almeida et al., 2018; Harris, 2013; Pines & Friedman, 1998).

Salovey and Rodin (1986) described how social comparison theory addresses jealousy in general but can also be applied to jealousy within the context of romantic relationships. The authors posit that social comparison revolves around self-evaluation; people want to be more like superior others and less like inferior others. The two main types of threats in a romantic social comparison dynamic are a rival with similar qualities that are valued by the partner and an inferior rival with unique qualities that may be valued by the partner. Romantic jealousy under this theory also differs in the respect of emotions as compared to jealousy overall. Anger and depression were greater in situations evoking romantic jealousy. Additionally, rivals were perceived more negatively in romantic situations rather than in social comparison situations (Salovey & Rodin, 1986).

Lastly, attribution theory focuses on control, causality, and intention. If a person experiences jealousy and they interpret their partner's actions to be intentional, within the partner's control, and of an internal causality, they will be more jealous (Bauerle

& Hupka, 2002). Intentionally flirting with another person or engaging in jealousytriggering behavior may lead to selfevaluation on the individual's part, potentially leading to social comparison. The interpretation of the behavior, action, or situation must come first. Attachment theory in adult relationships comes from John Bowlby's theory of infant attachment. Individuals incorporate their infant attachment styles (secure, anxious, avoidant, or disorganized) into their peer attachment styles growing up and then incorporate them again into their romantic relationships (secure, preoccupied or anxious/ambivalent, and dismissing or anxious/avoidant; Crowell & Waters, 1994). For example, people with anxious attachment styles may have more intense reactions to jealousy and cope in harmful ways (Kim et al., 2018; Madey & Roberts, 2009). Attachment styles are also predictive of whether a relationship will be successful or unsuccessful (Madey & Roberts, 2009). If a person has a secure attachment style, that is reflected in how they lean on their partner in times of stress rather than isolate, which an anxiously attached person is more likely to do.

Gender Differences, Sexuality, and Race

Although there are some findings that suggest a difference in intensity of jealousy as well as types of coping mechanisms between genders, some studies have found no significant difference (Broemer & Diehl, 2004; Buunk & Hupka, 1987; Demirtaş & Dönmez, 2006; Harris, 2002; Pines & Friedman, 1998; Salovey & Rodin, 1986; White, 1981b). As for the few articles addressing differences in jealousy in

gay and lesbian individuals, the findings supported that both groups would find potential emotional infidelity more upsetting than potential sexual infidelity (Harris, 2002; Guitar et al., 2015). However, only a handful of studies looking at sexual orientations other than heterosexual exist. Bisexuality, polyamory, and other minoritized sexualities and relationship configurations are being studied to a higher degree (Deri, 2011; Dijkstra et al., 2001; Rubinsky, 2019; Turell et al., 2018). However, most of the studies have a participant group consisting of predominantly cis-gender, heterosexual, and White couples.

For the bisexual population in particular, many misperceptions about promiscuity and infidelity affect how they are viewed by others, including their partners. Some studies have focused on these previously discussed detriments of binegativity - which comprises negative perceptions about people who are bisexual (e.g., that they are more promiscuous or unfaithful, that bisexuality is not a "true" sexuality in that a bisexual individual has simply not decided if they are straight or gay; Yost & Thomas, 2012). However, there is a lack of studies aimed at understanding how populations with intersecting minoritized identities experience and understand jealousy and how these experiences impact their lives.

One study was done with the expressed purpose of examining race, and it has limitations. This study used hypothetical situations and forced-choice measures to examine vague differences in sexual versus

emotional partner infidelity depending upon if a rival was of the same race of the partner or of a different race (Basset, 2005). The race of the rival was not specified beyond either matching or not matching the target's race. Although the author used a scale to measure anti-Black sentiment, no other measures were used to gauge other racist sentiments or beliefs (Basset, 2005). One study that focused on bisexual individuals noted jealousy is often related to intimate partner violence and control dynamics. Furthermore, they found that Black men and bisexual men experienced higher victimization of intimate partner violence with jealousy of the partner as a potential contributing factor (Turell et al., 2018).

The Present Study

There are many overlapping factors contributing to romantic jealousy, but the measures and the groups that have been used in previous studies do not provide a nuanced and contextualized understanding of the topic. There are currently five main jealousy measures that are commonly used in romantic jealousy research. These include the Romantic Jealousy Questionnaire, Chronic Jealousy Scale, and Relationship Jealousy Scale, which have about the same reliability when compared to each other (White, 1984). The Multidimensional Jealousy Scale (MJS) developed by Pfeiffer & Wong, is another that breaks down jealousy into emotional, cognitive, and behavioral categories and measures the intensity of each one. Lastly, a newer scale separates jealousy into subtypes; obsessive, depressive, separation anxiety, and paranoid (Marazziti et al., 2010). Although these scales can be used in researching jealousy in

more diverse groups, they only offer several choices written in formal language, lacking the ability to accurately document full feelings and experiences of romantic jealousy through the participant's own descriptive language.

The goal of this study is to examine the lived jealousy experiences of people with minoritized identities by analyzing personal experiences of jealousy through a psychological framework of antecedentbehavior-consequence (ABC). ABC gives structure to individual differences in behavior so these differences can be studied more systematically (Dubie & Pratt, 2008). This form of behavior analysis is a foundational psychological data collection method. Examining the role of individuals' backgrounds, past experiences, and identities of individuals in groups that have been underrepresented in research can expose the mechanisms, triggers, and results of romantic jealousy. Qualitative analysis yields a more in-depth and contextualized examination of potential factors of romantic jealousy and falls in line with potential prevention and intervention strategies. Given study aims, the research questions were as follows:

- 1. How do those with minoritized identities understand jealousy?
- 2. How do those with minoritized identities conceptualize the antecedent behavior consequence of experienced and expressed jealousy?
- 3. How is jealousy experienced by those with multiple and

intersecting minoritized identities?

Methods

Sample

The sample consisted of six participants. Each participant had at least one minoritized identity in the demographics of race and ethnicity, gender orientation, sexual orientation, and relationship type; three participants had two minoritized identities (see Table 1). Participants were young adults ranging in age from 19 to 33 and resided in the U.S. Most subjects were undergraduate students at a Western public university. Two identified as White, one as Asian, and three as multiracial. All participants identified as cis gender, with two men and four women. Two participants identified as straight/heterosexual, one as lesbian, one as bisexual, one as demisexual (being sexually attracted to people that one has a strong emotional bond with), and one as gay. Five out of six participants were in monogamous relationships; one was in a polyamorous relationship. The authors use mono and poly to describe the participants' relationship status in the results section.

Recruitment

A survey from a larger study on intersecting identities and relationship experiences was advertised at a Western University through flyers, social media outreach, and class announcements or presentations. The survey was also published multiple times over social media sites like Reddit, Instagram, and Twitter. The flyer and other adverts for the survey included information about IRB approval,

the purpose of the study, what kind of participants were needed, estimated time to complete the survey, and contact information for the researchers. Adverts indicated that the researchers were interested in learning about the experiences of people with minoritized identities (e.g., LGBTQ+, disability, neurodivergence, being a person of color, being polyamorous, having a nonprivileged religious identity, etc.). There was also information explaining that participants could provide contact information (an email and/or phone number) to be entered into a raffle for a \$10 Amazon gift code and to possibly be contacted for interviews. Interviewees would be compensated with a \$10 Amazon gift code per interview.

Survey

Interested participants accessed the survey link and were directed to an instruction page explaining the survey and asking for their consent to start the survey. On this same page, they were given the option to provide contact information for potential interviews. They then filled out open-ended and forced choice demographic questions assessing gender, sexuality, race, ethnicity, age, religiosity, income, geographic location, education, and relationship status. They also answered quantitative and open-ended questions on the topics of labeling, jealousy, relationships, and experiences around having marginalized and minoritized identities. After they finished the survey, they were given another opportunity to enter contact information and enter the raffle.

Interviews

The data from the surveys were then screened to identify people willing to be contacted for semi-structured interviews. For purposes of this study, participants who identified themselves under at least one minoritized identity, had at least one partner, and completed the jealousy section were then contacted to schedule an initial interview. Six people responded to requests for a Zoom interview. Once scheduled, participants were emailed a Zoom link. First participants were shown a consent form and asked to indicate willingness to continue the interview. After participants consented and picked a pseudonym, audio recording of the interview commenced. Participants were asked how they defined jealousy in general and then romantically. They were also asked to explain intimacy, romance, and sexual activities and the intersection. Participants elaborated upon personal experiences with romantic and general jealousy as well as hypothetical scenarios under a socialcomparison framework. They were also asked questions about their boundaries with their partner and relationship, and sexual satisfaction as well as their partner's views on similar topics. Five participants were contacted for a follow-up interview to either continue the semi-structured interview questions or to address follow-up questions.

Analysis

Qualitative analysis of the semistructured interviews gives priority to the everyday experiences and realities of participants. This allows for an understanding of romantic jealousy itself, rather than measurement of aspects of the concept. The semi-structured interviews

were framed by a nuanced explorative reflexive approach (Binder & Holgerson, 2012). This allowed the interviewer to explore the topics while also recognizing the "otherness of the participant's world" (Binder & Holgerson, 2012, p. 108). This approach acknowledges that the interviewer brings their own preexisting notions about a topic, usually unconsciously and must reflect on their own background as they begin to interpret the words of the interviewee. By confronting those preexisting ideas about romantic jealousy, the first author then coded the transcripts with an inductive approach, "a process of coding the data without trying to fit it into a pre-existing coding frame, or the researcher's analytic preconceptions" (Bruan & Clark, 2012, p. 68). "Memoing" or journaling after every interview was one tool that was used by the researchers to reflect on their positionality or background and preconceived biases. The researchers also met biweekly to discuss positionality and overarching topics that came up in interviews.

Following the final interview, the first author reviewed transcripts line by line and coded for ideas as it pertained to study research questions. She then established codes and subcodes that were applied to the transcripts of all participants and were used to build the latent themes. The coding was supported by critical realism, which recognizes that individuals make meaning out of their experiences in various ways and these meanings connect back to a greater social context (Braun & Clark, 2012). The first author coded three of the transcripts using Dedoose and then consulted with the

second author. Several rounds of discussion on the resulting themes and subthemes were had, until consensus was reached. The rest of the transcripts were coded using the established codes and subcodes. Newly discovered codes were discussed among the authors and were either integrated into the existing paradigm or were amended to fit an existing theme or subtheme. Following data analysis, exemplary quotes were then selected from the transcripts to represent the concluding, latent themes of this study.

Results

In discussing experiences of and perceptions of jealousy among individuals with multiple minoritized identities, three main themes resulted: 1) The conceptualization of jealousy, 2)
Antecedents - behaviors – consequences (ABCs), and 3) Gender, race and ethnicity, and the intersection of identities.

Theme 1 – The Conception of Jealousy

Participants had varied ideas around the conceptualization of jealousy. This theme highlighted the numerous factors contributing to romantic jealousy for these individuals. Although there is a generally agreed upon definition in the fields of psychology and human development, the definitions that people live their everyday lives around are quite different, and contain more emotion and subtext. Most participants defined romantic jealousy in the context of real life, with Joseph (23, multiracial, straight, mono) presenting it as, "It can be small, once, or it could be extremes of complete lack of trust. I don't feel that jealousy is something that should be completely eliminated from our lives, but I

do understand how damaging it can be." This provided a much broader picture of romantic jealousy. Sue (19, multiracial, bisexual, mono) offered a more pointed illustration of how it can be spurred. She explained that jealousy can happen because of "sexual or romantic relationships that can get you kind of satisfied, but it's not exactly what you want." All of the definitions tended to be very thorough and based on personal contexts, setting the foundation for how romantic jealousy was experienced in an individual's life.

Offering an even more specific example of the scale jealousy can exist on, Brittney (33, White, demisexual, poly) said "it can be as subtle as a primary partner reaching for their other partners hand and grabbing it and I've seen it be as explicit as a knock down drag out fight." Tamara (21, Asian, heterosexual, mono) began her definition by saying "jealousy could be defined as almost feeling possessive about someone... like feeling strong negative emotions towards your partner or towards other people in general." In this quote, Tamara alludes to a social comparison framework which was further supported by later parts of her interview. Helena (20, multiracial, lesbian, mono) similarly noted that romantic jealousy "is more specific to someone you think can replace you. Like someone who you think your partner would be attracted to. So, jealousy would be integrated with that fear of losing your partner to someone else." Although closely aligned with the definition for romantic jealousy that is commonly used in research, the idea of being replaced tells a more contextualized story.

Other participants aligned romantic jealousy with envy. Eric (25, White, gay, mono) claimed that romantic jealousy is:

... a malicious envy. Something that you wish you had or wish happened to you. [You can be] jealous of what two other people have or what someone has for someone else other than you... you desire something that you don't have or, again, that someone else has that actively gets in the way.

This type of envy – that someone might feel in relation to something they perceive they do not have or that someone else has – is more closely related to a framework centered around control than it is to social comparison theory. Joseph (23, multiracial, straight, mono) supported a similar definition focused on control, explaining, "I think jealousy would just be the idea that somebody or something in your world is not the way you'd like it to be and that makes you upset or uncomfortable." For these participants, control was not a harmful or dangerous thing. They described when jealousy could reach these extreme levels, but the control Eric and Joseph connected to their definitions was more about trust than it was about controlling another person. Brittney (33, White, demisexual, poly) supported this more wishful definition by saying "Jealousy is somewhere in the middle between fantastic and realistic." She had experienced this in her relationship with her ex-husband who would dismiss her when she brought up fears that he was cheating on her with "well, you're just jealous." The conception of jealousy, in this case, was

more cyclical; based on feelings and observations but then altered by the feedback from the partner.

Some participants made connections to romance and sex, emphasizing the link to intimacy. Tamara (21, Asian, heterosexual, mono) presented romantic jealousy as "something that you thought only belonged to the two of you now isn't that," or a violation of intimacy. This violation of intimacy can be viewed as a break of trust too. Joseph (23, multiracial, straight, mono) illustrated the relational effects that can come from this in saying, "if it's something large where you're feeling a lack of trust or you're feeling like a lack of, again, communication, if we're talking relations, then it can feel very painful almost like you've lost a loved one." In his second interview, Joseph continued this thought:

The minute you lose that intimacy kind of creates a tough fight or flight response; that's where the jealousy comes in. Because intimacy is something that takes so long to really develop or so ingrained in your system, but the minute you lose it, it just kind of feels like abandonment and so it's irrational behavior.

All of the participants put an emphasis on intimacy as a casualty of romantic jealousy but also mentioned communication as a way to maintain intimacy and combat unhealthy romantic jealousy.

Theme 2 – Antecedent, Behaviors, and Consequences (ABCs)

The specific examples of antecedents, behaviors, and consequences as

well as the justification for the behaviors present a methodical approach to understanding the process of jealousy while also considering the unique identities and experiences of people with minoritized identities. Participants were asked about things that preceded feelings of jealousy for them (antecedents). To assess what behaviors would result from jealousy, participants were asked to give generalized examples of what jealousy might look like in themselves and to describe how they reacted to a specific situation in which they felt jealous. Finally, participants were asked to discuss what came of these specific situations (consequences).

General Antecedents

Common antecedents reported by the participants revolved around their own mental health, their partner's expressed or implied interest in or attraction to others, and a lack of communication. Joseph (23, multiracial, straight, mono) offered a connection to mental well-being: "Usually [it's] the mental health. But if I'm having some sort of emotional episode, I feel a lot more jealousy and at those moments I feel more alone because I'm jealous." Helena (20, multiracial, lesbian, mono) described an antecedent related to a partner's behavior. She explained, "I get jealous when [romantic interests] spend time with another person and then they don't try to include me at all, or there's another person in their life where they don't tell me about them." Similarly, Tamara (21, Asian, heterosexual, mono) described when her boyfriend "followed a lot of beautiful local women on Instagram" which made her jealous. Joseph discussed an antecedent related to

communication, explaining that "when there's no communication, is when I feel jealous. Just from past experiences, typically when there's no communication and then I find out something that [normally] wouldn't make me jealous... then I started to have a lack of trust."

Brittney (33, White, demisexual, poly) offered a more unique set of antecedents. She was in a consensual polyamorous relationship with a married man at the time of the first interview and had also been going on individual dates with other people. She expressed that although she never felt jealousy about the man's wife, she did sometimes envy the activities the husband and wife did together that the agreed upon boundaries did not allow her to do. Watching this married couple, Brittney said "I need someone who is stable and who can be there, not only mentally and emotionally on the weekends, but who can be there mentally and emotionally for me and my children during the week."

Brittney also mentioned jealousy related to the partner of a very close friend. About her relationship with her friend's partner she explained, "I don't like her. I've tried to be friendly to her. It's oil and vinegar." She clarified that seeing this woman, or even hearing her friend mention her can elicit feelings of jealousy for her. One thing she contributed to this jealousy was her history with her friend; "I wish we were both single at the same time, so we can kind of explore how not good together we would be."

General Behaviors

The interviewees then discussed general behaviors in which they would likely engage, in response to those previously mentioned antecedents. Helena (20, multiracial, lesbian, mono) suggested that she might engage in "prying into someone's life and wanting to know more information and trying to keep an eye out for that person" or acting "passive aggressive and a little hostile." Joseph (23, multiracial, straight, mono) recounted, "I think the thing I do most often is, I just vent to my friends about my discomfort and just [say], 'Oh, I bet they're doing this' or whatever. And I'll just be like 'that just kind of sucks."" Mostly, he would reach out to friends if he was having an "emotional episode" related to his mental health that he recognized was exaggerating his feelings of jealousy. Tamara (21, Asian, heterosexual, mono) expanded on the concern of her partner showing interest in other potential partners; "even [before we] met each other he followed a lot of beautiful local women on Instagram and going into our relationship, I told him I was insecure about that." She decided to convey existing antecedents to her jealousy before encountering an antecedent to romantic jealousy in this relationship.

Specific Behaviors and Consequences

When asked about specific instances of behaviors and consequences in response to jealousy, the participants recounted various differing behaviors, sometimes to the same category of antecedent. For example, Helena (20, multiracial, lesbian, mono) and Joseph (23, multiracial, straight, mono) behaved quite differently when they

sensed a lack of communication in their relationships. Helena relayed how a potential partner would "keep on canceling on me or keep on like avoiding my texts." With this antecedent of lack of communication, Helena stated that,

In the response, it was just like a tit for tat where they wouldn't respond to me for a few days so when they finally did respond, I would wait a few days to respond and then I wouldn't be very open about my life.

This was a long-distance relationship, and the consequences were, "we kind of just left each other alone for long enough. Once we got back together and hey, oh, we're calling [each other], [and] jealousy just kind of isn't important anymore." For Helena, giving her partner space and not directly addressing the jealousy resulted in the ultimate continuation of the relationship.

Joseph's behaviors in response to lacking communication were quite different. During a past relationship, Joseph noticed, "for the first few months there and then slowly, I noticed that like the texts sort of getting a lot more space in between, even though there were no major life changes." This antecedent was a lack of communication coming from the partner. He stated that his reaction, or behavior, was to:

...reach out and say, "Hey, what are you up to?", and then they'll say something like, "oh, I'm out with this person." And that's when I started to get jealous because I feel like I'm out of the loop and there's no

communication. So, I have no idea what's going on.

Joseph communicated more in order to gain more information about the state of his partner. The partner in turn revealed that they were out with others, and this reinforced the feeling of jealousy. The consequence of this specific instance was that Joseph ended the relationship after the communication of the partner became less frequent and they began to hang out with other people and potential partners. After that relationship ended, he experienced a similar situation with a subsequent partner: "We would talk a couple days after and then there'd be radio silence, and then I would see, typically, we'll have like Snapchat or something else, and I'd see them out with another person." This time Joseph communicated more directly to this partner about his concerns around the lack of communication and said, "Hey, you know, if you felt this way about this person [another romantic interest], you could have just told me." This slightly different approach of increased, direct, communication (behavior) also resulted in the dissolution of the relationship.

Tamara (21, Asian, heterosexual, mono) also shared her behaviors and consequences of two specific instances of jealousy. She described that her boyfriend "followed a lot of beautiful local women on Instagram" which made her jealous. She perceived that her partner was interested in someone else. Her initial behavior was to open a line of communication and attempt to set a boundary within the relationship; "I told him I was insecure about that." Her

boyfriend reacted by saying "oh just tell me who I need to unfollow and then I'll do it" but Tamara ultimately denied the offer. A consequence of this behavior was that the boyfriend continued to like other women's photos. So later, she experienced another antecedent, and explained, "my boyfriend liked this girl's photo of her wearing a maid outfit, like the sexy maid outfit and the girl's caption was personal maid." This time her behaviors were different. Tamara continued. "so I screenshotted that to him, and I was like 'Oh, do you want to go be with your personal maid then? Fine, go ahead." This reaction was more intense than the first behavioral reaction, possibly because there was a repetition of a previously discussed issue. However, she followed this behavior by expressing more directly how the situation made her feel. This fostered affective communication which resulted in a better consequence. She described her boyfriend's reaction: "'yeah, I completely understand why you would feel that way, because if you followed a lot of like cute guys and stuff like that, I would feel insecure and I would feel upset about that." The partner's behavior changed to alleviate one specific antecedent for Tamara's jealousy.

Brittney (33, White, demisexual, poly) also used communication skills to reduce her discomfort, but not necessarily resolve those feelings of jealousy. She mentioned a time when they all three sat down together to try and resolve some tension. The friend's partner asked if Brittney and the friend had ever slept together. Brittney admitted to having a sexual relationship with him about 10 years

prior to the start of the woman's relationship with him, something the friend had not divulged to his current partner. After that, she simply told her friend "I don't want to hear about her." The cycle continues where Brittney is at a gathering where the friend and his partner are, she feels jealous, and then avoids conversation about the person. In place of continuing a conversation about her jealousy towards the partner and her own romantic feelings towards her friend, she revealed that she often thinks about the possibility of a relationship between her and her friend in the future; "If you are single, while I'm single... [and] he asked me out, absolutely. Let's make it work."

The participants discussed many specific ABC occurrences. Participants mentioned some similar antecedents, such as the lack of communication from a partner and a perceived or observable interest a partner takes in another potential partner. However, based on their individual contexts. their behaviors were different and led to different consequences. Helena (20, multiracial, lesbian, mono) and Brittney (33, White, demisexual, poly) both avoided talking about the antecedent to their jealousy, but Brittney set a clear boundary of not wanting to discuss a certain person whereas Helena waited until communication was brought up by the potential partner to either discuss the issue or move on. This resulted in Brittney continuing to feel jealous when antecedents were present and hoping that there was a possibility of a romantic relationship with her friend in the future. In Helena's case, it resulted in the continuation of a friendly relationship but no discussion of changing the antecedents or

nature of the relationship. For Joseph (23, multiracial, straight, mono) and Tamara (21, Asian, heterosexual, mono), they experienced jealousy when they perceived their partner was interested in another potential partner. They both communicated their feelings and placed the expectation of communication from the partner too. However, Tamara expressed having difficulty not only understanding the underlying competition she may experience but also with communicating the aspects of her jealousy effectively. Ultimately, the participants detailed their individual experiences with the antecedents, behaviors, and consequences related to jealousy and even with some similarities between participants, similar antecedents resulted in different behaviors and yet similar outcomes.

Participants' Reflections and Perceptions of Their ABCs

Beyond recounting the antecedents, behaviors, and consequences of specific instances of romantic jealousy, participants were asked to elaborate on their thought process behind their behavioral and cognitive reactions to the situation as well as the resulting outcomes. Regarding the instance of lack of communication from her long-distance romantic interest, Helena (20, multiracial, lesbian, mono) described "the thought process would be like, 'oh, they don't care about me as much anymore.' So, I'm not going to care about them and that's why I'm just going to try to stop fostering that relationship as much." Joseph (23, multiracial, straight, mono) had a different thought process: "I try my best to make sure that there's like a benefit of the doubt, like

maybe that person is busy or maybe something else is going on," which supports his behavioral reaction to continue communication. Specific to Tamara's (21, Asian, heterosexual, mono) experience with romantic jealousy related to her partner interacting with other women over social media, she explained that her first reaction was, "Well I don't necessarily want you unfollowing people' [just] because I had a difficult time processing what proper boundaries were and what I felt comfortable with." She was conflicted and said, "to an extent that made me feel insecure, but it also was physical proof that he was looking at other women." This internal conflict may have contributed to the more intense behavioral reaction in the second situation.

As one of the last explorations of the interview, participants were asked to discuss how their perceptions and experiences of romantic jealousy have changed over time. Most participants connected various aspects of jealousy to the concept of maturity. Helena (20, multiracial, lesbian, mono) referred to jealousy itself as something negative that "becomes a little bit more shameful" as she gets older. More pointedly, she discussed the behavior-consequence cycle of her own experiences: "It totally has to do with maturity and thinking that like jealousy is kind of childish and the lack of communication is kind of childish." Joseph (23, multiracial, straight, mono), Sue (19, multiracial, bisexual, mono), and Tamara (21, Asian, heterosexual, mono) all supported the idea that maturity matters with respect to behavioral reactions. Tamara described it as:

Some people think about jealousy based on maturity level and I wouldn't really say those feelings are based on maturity level, but it's how you respond to those feelings [that] could be based on maturity level and the level of understanding of your own emotions.

Tamara followed her introspection with, "the way I've been trying to approach it now is just having an open conversation about it because usually I feel like I'm really used to bundling up my feelings and not saying anything." She implies that it's not solely an issue of maturity but also a conscious choice to notice the antecedents, behaviors, and consequences and shape healthy habits when faced with romantic jealousy. Similarly, Sue (19, multiracial, bisexual, mono) asserted, "I've learned to deal with my jealousy in appropriate ways as I've gotten older." For her, this meant proactive and affective communication. Joseph (23, multiracial, straight, mono) took a more philosophical approach to the questions. He said, "As I've grown older, I've just realized to kind of expect that people really do have the best intentions in mind and that things happen." This mirrors his thought process that involved giving partners or potential partners the benefit of the doubt. He followed this with, "People will do what they're going to do. And I really can't control that. I'm just going to live my life and that'll just be the way it is."

The various portions of the interviews revealed that many of the participants experienced jealousy through some sort of social comparison framework.

Joseph referred back to his experiences with a partner that decreased communication while spending time with other potential partners. He compared himself to the people his partner was hanging out with and explained that in that situation, "usually, that [lack of communication] makes me feel like second place or discarded without rationale, like less than human."

Instead of reflecting on how she might compare herself to her partner's acquaintances, Helena (20, multiracial, lesbian, mono) discussed how she compared her reactions to jealousy to the ways others may react. She explained,

When I see people [showing] that kind of petty behavior that I can recognize in myself, that's kind of what changes my attitude because if I don't like what they're doing, then I have to take that negativity back to my own actions.

Tamara (21, Asian, heterosexual, mono) also described a form of social comparison that was less centered on a rival and more so on how an individual may process jealousy. She said, "I've had this conversation with my roommate and if her boyfriend follows or likes another girl's stuff, it's not a big deal [for her]...what might be comfortable to someone else might not be comfortable to you." She went on to explain how social comparison is impacted by the opinion of her male partner, "If I were to perceive a woman as attractive [but] my boyfriend indicated that he was attracted to her, I'd probably be insecure...I would be attacking her within my head; I'd try to find things wrong with her." Tamara felt the

internalized male gaze impacted her experiences with jealousy. The foundation of the internalized male gaze develops "because of the sociocultural practice of men looking at women for sexual purposes, women become socialized to view themselves from an outsider's viewpoint" (Yilmaz & Bozo, 2019, p. 2). In this process, "[women] check for themselves whether they are acceptable to others" (p. 2). In other words, they internalize the gazes upon their bodies and practice self-objectification (Yilmaz & Bozo, 2019). Tamara explained,

I viewed other women he followed as competition and [it] made me feel insecure...but if I perceive them just as women and not as a competition for my relationship, I'd obviously be like, "Oh yeah, these women are amazing. They're beautiful."

Conclusions

All participants detailed either general or specific examples of antecedents, behaviors, and consequences related to romantic jealousy. There were common antecedents, like communication whether that be a lack thereof or a misunderstanding. Some participants engaged in different behaviors as a reaction to the antecedents. Some attempted to open up communication while others remained silent. Those behaviors affected the consequence; whether the jealousy was reduced or resolved, or even if the relationship itself continued or ended. In their explanations, there were clear echoes that past experiences and intersections of their identities were reflected in what they perceived as an

antecedent, what behaviors they then engaged in, and the ultimate consequence of that behavior.

Theme 3 – Gender, Race and Ethnicity, and the Intersection of Identities

The impact of identities was a major theme throughout the interviews. First, one of the identities that was discussed by participants was gender identity. Tamara (21, Asian, heterosexual, mono) applied a feminist framework to romantic jealousy when she said, "I feel like there's also that kind of taboo around jealousy, and especially for women...if you're jealous, you're a crazy bitch or you're possessive and all those negative terms." She also commented on the social dynamics between men and women in society. She continued her conception of jealousy: "I think it also came from my identity as a woman and what we're taught as women to prioritize relationships with men." This viewpoint outlined the varying but traditional dynamics existing between men and women in society, a dynamic that is tied to heteronormativity. "Heteronormativity points at the everyday and mundane ways in which heterosexuality is privileged and taken for granted, that is, normalized and naturalized" and has expanded in feminist thought by drawing attention to the societal structure that maintains men as people of power and authority over women (Herz & Johansson, 2015, p. 3).

Connections to the gender identity of a partner were also identified by Tamara who said, "[my partner] is trans and I think that gives him a deeper understanding of certain societal things [so] he can see things from my perspective and understand more things from my perspective than cis-guys would." In this situation, Tamara felt that her partner's previous experiences before transitioning gave him a better understanding of her concept and feelings of jealousy. This understanding opened the doorway for developing greater affective communication about jealousy in their relationship. In this situation, Tamara felt that her partner's previous experiences before transitioning gave him a more direct understanding of her perspective which built more effective communication.

Secondly, participants discussed racial, ethnic, and cultural identities as being influential to their relationships and understanding of jealousy. For example, Sue (19, multiracial, bisexual, mono) recognized differences between their and their partner's racial/ethnic identities. She said:

Being biracial has always been very confusing for me to think about because it's not as concrete or as definite...my partner is just a White man; he never has to think about that or think about if how he grew up was different from other White Americans.

Sue described a type of jealousy that was broader and not centered around romance or romantic rivals, but rather an ease of being understood as an individual in any context, but especially when communicating. This does not necessarily diminish communication but can make it more arduous. She further explained:

We had a discussion recently that was just about family households and family structures in Japan and oftentimes we'll compare it with American culture. That discussion was very difficult for me because I couldn't comfortably say that I grew up in an American culture an American household. But I also really can't confidently say that I grew up in a Japanese household with Japanese culture either.

She concluded by commenting on how difficult it was to express the culmination of cultural identity she experienced, even with people she is close with, which can present a barrier when communicating in her romantic relationship.

Lastly, some participants discussed the influence of the intersection of identities on their understanding and experiences of jealousy. For example, Tamara (21, Asian, heterosexual, mono) highlighted gender expectations as a function of race and culture in her analysis of her perceptions of jealousy. She said, "fair skinned or East Asian people are glorified as dolls and Southeast Asian people are less favored and [seen] more as housekeepers and sex workers." She then added that the over exaggeration of jealousy in the portrayal of Asian women in the media led to the potential fetishization of Asian people. It also reaffirmed that women are seen to experience jealousy more intensely and behave more irrationally than men. Tamara further discussed the ways both East Asian and Southeast Asian women can be fetishized, usually by straight, white men,

but then reflected upon other stereotypes that relate more to romance and intimacy than sex. She shared:

When it comes to the subservient thing, I see that in my mom, and I feel like that's had an impression on me. [My dad's] cheated on her before and they're still together... ultimately that gave me a twisted perception of relationships and jealousy.

Some discussed how their gender identity intersected with their sexual orientation. Helena stated, "I know I have very, very close relationships to my female friends. An outsider would easily see as romantic, but I don't see as romantic." She continued:

[romantic jealousy] is going to probably happen with a partner. That they're going to have a very close female friend where it's normal to hand hold and that kind of intimacy without romance. So, I think that might spur more jealousy, than if I had a male partner who doesn't generally do those things with their close male friends.

Brittney (33, White, demisexual, poly) also discussed her intersecting identities of gender, sexual orientation, and relationship preference, pointing out how they related to her ex-partner's expression of romantic jealousy. Reflecting on her past relationship with her ex-husband, Brittney remembered "he went in[to the marriage] knowing that I was polyamorous... I couldn't look at someone without him losing his

temper, being jealous." Her ex-husband's jealousy took on a form that mirrored binegativity. The underlying assumptions about promiscuity, infidelity, and instability that comprise binegativity are seen in her experiences with her ex-husband, although nuanced because of her pansexuality. The jealousy escalated over time to the point where her ex-husband became controlling and psychologically and physically abusive. When going over the context of this intimate partner violence, she gave some background. Both she and her husband grew up in a small, rural, generally conservative town. Before getting married, she received advice from her mother; "make sure your husband is always satisfied." She was expected to be a traditional woman and wife; to take on the role of homemaker and to obey her husband. Her ex-husband's romantic jealousy and his behaviors of abuse and control were due to his expectations around her gender, intersecting with her sexual orientation. The experience related to her ex-husband's jealousy had lasting impacts on her conceptualization of jealousy.

The participants expressed how their identities directly connected to their perceptions and experiences of jealousy. Sometimes a singular identity like gender or race and ethnicity was salient in their discussions. Other times, the intersections of identities, particularly the intersections of gender, race and ethnicity, and sexual orientation supplemented their conceptions of jealousy and the related ABCs. Most of the participants gave examples from past relationships or reflected on how romantic jealousy was conceptualized in their families

in connecting to singular or intersecting identities.

Summary of Results

The first main theme, the conception of jealousy, supported the broad range of definitions in actual lived experiences. Participants discussed trust, communication, intimacy, romance, desire, and dissatisfaction in their descriptions of jealousy. All the participants agreed that jealousy ranges in intensity but can be shown through a variety of behaviors. Through the second theme, antecedent, behaviors, and consequences (ABCs), the participants gave context to romantic jealousy. Each person gave examples of antecedents to jealousy, usually rooted in lack of effective communication or a perceived threat to their relationship, particularly a partner's interest in another person. There were several common antecedents, but the behaviors had quite a bit of variation. Some behaviors included prying into the partner's life, venting to friends, communicating passiveaggressively to the partner, or not speaking to the partner at all. Despite the diversity in behaviors, the consequences tended to be the same; the relationship was maintained, or it was ended. The third theme, gender, race and ethnicity, and intersection of identities, gave some context for the first two themes. Past experiences in relation to the participants intersecting identities appeared to have a significant impact on how they conceptualized jealousy on a more personal level and how they reacted to it. Societal expectations or stereotypes related to gender, race, and ethnicity spoke to the positionality of some participants in

romantic relationships. Others' reactions to these identities sometimes predicted jealousy-related cognitions and behaviors like fetishization, control, and in one case, violence.

Discussion

Throughout the interviews, participants spoke to the first theme of how they conceptualized jealousy; what it was, how it may look or feel, and how it can change or have an impact on people's lives. They also broke down some of their own antecedents to their romantic jealousy, described how they reacted to it, and then debriefed on what happens after words, covering the second theme. For the third theme, participants expanded upon how their identities and experiences intersected to influence their experiences of romantic jealousy.

The results contextualize the connection between jealousy and identity in several ways. First, although closely aligned with the definition for romantic jealousy that is commonly used in research, the idea of being replaced, as reported by many participants, tells a more contextualized story. Tamara (21, Asian, heterosexual, mono) spoke about her fear of her partner replacing her for someone with better qualities. Joseph (23, multiracial, straight, mono) and Helena (20, multiracial, lesbian, mono) both expressed a feeling of neglect that sometimes came with jealousy, especially when a partner or potential partner began communicating less or openly spending time with other potential partners. Given potential experiences with discrimination and marginalization (as

discussed by Tamara and Helena, for example), individuals with minoritized identities may conceptualize jealousy more so around neglect or being dismissed — matching some of their more general day-to-day experiences unrelated to romantic relationships.

Additionally, some participants also contributed that a sense of maturity, either connected to personal growth or effective communication had changed how they viewed jealousy and how they handled it in their own personal relationships, as referenced by Joseph, Helena, and Sue (19, multiracial, bisexual, mono). In focusing on communication, growth, and maturity (as did Joseph and Tamara), individuals with minoritized identities may be drawing on resiliency and other skills that are necessary for thriving in a majority society. People with multiple minoritized identities often face additional and unique stressors which can be mitigated by individual-based resilience, which revolves around personal agency, and community-based resilience which involves interpersonal relations and social resources (McConnell. et al., 2018). These results indicate that one or more marginalized identities may be related to perceptions of neglect but also resiliency – components of jealousy reported by the participants, but not often included in traditional conceptualizations of jealousy.

Secondly, participants noted different romantic jealousy ABCs around a singular identity, depending on the other intersecting identities of an individual. For example, one participant felt that women are more likely to have intimate friendships with

one another than men are, and this greater intimacy may present a greater risk for antecedents to jealousy for a woman who is attracted to and in relationships with women. Participants often discussed the challenges of navigating romantic relationships around personal, familial, cultural, and societal expectations; expectations which sometimes conflicted. When considering individuals from multiple, diverse backgrounds, antecedents and reactions to jealousy may not be easily categorized and often require a discussion about the intersection of identities and past experiences given the contexts of family and society.

To summarize, when their own individual antecedents, behaviors, and consequences were parsed out, jealousy went beyond the classic definition related to a real or imagined threat to a valued relationship for these diverse participants. These threats were not only related to their partner's possible interest in another partner, but also mental health, communication, and familial and societal expectations for behavior. Whether it is Joseph's willingness to give partners and others the benefit of the doubt, Tamara's examination of how she perceives other women in a competitive way, or Sue's idea that communication will prevent harm, these approaches to personal relationships cannot be fully understood without an exploration of identity, intersectionality, and marginalization.

Implications for Research

There are several implications for future research inspired by this study. More research is needed to build knowledge

around the unique issues faced by underresearched minoritized groups. The participants from this study indicated that familial, cultural, and societal expectations around identity can create a tension that manifests in romantic relationships. These individual contexts and experiences can impact an individual's antecedents to jealousy, their behaviors and reactions, and then their coping mechanisms related to the experience of romantic jealousy. More studies about the antecedents and experiences of romantic jealousy among diverse populations can contribute to practical public health and educational programs for those individuals. Foundational studies on romantic jealousy should be replicated with more diverse and intersectional participant groups. Additionally, studies should aim to use qualitative methods like interviews, ABC formatting, or open-ended questions about real events as opposed to forced choice paradigms and hypothetical situations.

Suggestions for Practice

The ABC format of jealousy experiences can serve as a tool for practitioners. This study and tool could be ideal for therapists and psychologists working with couples and families, family life educators, and mental health professionals working with victims of domestic violence. The set of interview questions created for the purpose of this study was extensive, but an abbreviated and more targeted set of questions could be used in a clinical or therapeutic setting (see appendix A). The ABC focused questions could be asked in an initial session to establish a firm definition of jealousy for an

individual or couple. This could serve as a guide for both the practitioner and clients to understand previous, current, and future antecedents to jealousy to better establish healthy behavioral reactions, leading to more positive consequences.

A practitioner could also use these questions as a means of assessing shifts in dynamics and triggers of jealousy in subsequent sessions. During the interviews conducted for this study, participants typically disclosed more specific information during the second interview. These questions could be asked during multiple sessions, especially if there are specific contexts warranting more attention (e.g., infidelity, high instances of behavioral or emotional jealousy, violence). In understanding the power of both individualbased resilience and community-based resilience, practitioners can focus on fostering resilience in an individual's relationship with their partner. Of note, is the inclusion of questions around identities and the intersecting influence of those identities on romantic relationships. An intersectional understanding of identity and background along with knowing the ABCs of romantic jealousy, could help practitioners identify targeted therapies or interventions.

Limitations and Conclusions

This study has some limitations and constraints worth noting. First, is the nature of the sample. A larger sample size comprising participants with various minoritized identities would provide more qualitative data for analysis via the ABC framework. There was also a geographical

restriction in this study as most participants resided on the West Coast. Furthermore, a sample with people from a greater variety of geographical areas in the United States would offer more insight on the impact geographical regions have on varying politics, social expectations, demographic concentrations, and perceptions of and experiences of jealousy. A second limitation is the timing of the interviews. Most participants were recalling experiences in which they had felt jealous and were defining jealousy in their current context, exposing the possibility for the person to have their past feelings and experiences altered by their current perception of jealousy. Through this retrospection, some participants gave examples from previous relationships rather than their current or most recent one. Although the authors were interested in the past, present, and future of participant experiences, sometimes, these aspects bled into one another. A shorter interview with a select few questions may be better in allowing participants to focus specifically on romantic jealousy in their current relationships.

Despite these limitations, the study highlighted the value of approaching social phenomena with the goal of understanding the nuances of it rather than simply measuring aspects of it. Allowing participants to express their feelings in their own words reveals more about the nature and process of jealousy than a simple forced-choice measure could. The results from this approach show that practices for addressing romantic jealousy may benefit from being tailored to an individual's antecedents, behaviors, consequences, and

identities. Starting a conversation about identity, relationships, experiences, and changing perceptions of jealousy over time can reveal the philosophy behind how people interact with romantic partners, as well as the specific events that may alter how they interact with their partners. Self-perception is a key component of how people build relationships. Interviews focusing on identity and personal

experiences can guide individuals toward this self-reflection leading to stronger relationships built on a deeper understanding. An examination of jealousy and the intersection of identities like race, ethnicity, gender, and sexuality, can help us better contribute to the well-being of individuals with multiple minoritized identities in their romantic relationships.

References

- Balsam, K. F., Molina, Y., Beadnell, B., Simoni, J., & Walters, K. (2011). Measuring multiple minority stress: The LGBT People of Color Microaggressions Scale. *Cultural Diversity and Ethnic Minority Psychology*, 17(2), 163-174.
- Bassett, J. F. (2005). Sex differences in jealousy in response to a partner's imagined sexual or emotional infidelity with a same or different race other. *North American Journal of Psychology*, 7(1), 71-84.
- Bauerle, S. Y., Amirkhan, J. H., & Hupka, R. B. (2002). An attribution theory analysis of romantic jealousy. *Motivation and Emotion*, 26(4), 297-319.
- Binder, P. E., Holgersen, H., & Moltu, C. (2012). Staying close and reflexive: An explorative and reflexive approach to qualitative research on psychotherapy. *Nordic Psychology*, *64*(2), 103-117.
- Braun, V., & Clarke, V. (2012). Thematic analysis. In H. Cooper, P. M. Camic, D. L. Long, A. T. Panter, D. Rindskopf, & K. J. Sher (Eds), *APA handbook of research methods in psychology, Vol. 2: Research designs: Quantitative, qualitative, neuropsychological, and biological* (pp. 57-71). Washington, DC: American Psychological Association.
- Broemer, P., & Diehl, M. (2004). Romantic jealousy as a social comparison outcome: When similarity stings. *Journal of Experimental Social Psychology*, 40(3), 393-400.
- Buunk, B., & Hupka, R. B. (1987). Cross-cultural differences in the elicitation of sexual jealousy. *Journal of Sex Research*, 23(1), 12-22.
- Crowell, J. A., & Waters, E. (1994). Bowlby's theory grown up: The role of attachment in adult love relationships. *Psychological Inquiry*, *5*(1), 31-34.
- de Almeida, T., de Lima, R. D., & Canezin, P. F. (2018). Homosexuality: Brief history and considerations on the manifestation of jealousy in homosexual relationships. *Open Access Library Journal*, 5(5), 1-11.
- Deans, H., & Bhogal, M. S. (2019). Perpetrating cyber dating abuse: A brief report on the role of aggression, romantic jealousy and gender. *Current Psychology*, 38(5), 1077-1082.
- Demirtaş, H. A., & Dönmez, A. (2006). Jealousy in close relationships (Personal, relational and situational variables). *Turkish Journal of Psychiatry*, 17(3), 181-191.
- Deri, J. (2011). *Polyamory or polyagony? Jealousy in open relationships* [Doctoral dissertation, Simon Fraser University]. Simon Fraser University Library. https://summit.sfu.ca/ flysystem/fedora/sfu migrate/11787/etd6744 JDeri.pdf
- Dijkstra, P., Groothof, H. A., Poel, G. A., LAVERMAN, E. T., Schrier, M., & Buunk, B. P. (2001). Sex differences in the events that elicit jealousy among homosexuals. *Personal Relationships*, 8(1), 41-54.
- Pratt, C., & Dubie, M. (2008). Observing behavior using a-b-c data. The Reporter, 14(1), 1-4.
- Guitar, A. E., Geher, G., Kruger, D. J., Garcia, J. R., Fisher, M. L., & Fitzgerald, C. J. (2017). Defining and distinguishing sexual and emotional infidelity. *Current Psychology*, 36(3), 434-446.
- Harris, C. R. (2002). Sexual and romantic jealousy in heterosexual and homosexual adults. *Psychological Science*, 13(1), 7-12.
- Harris, C. R. (2013). Humans, deer, and sea dragons: How evolutionary psychology has misconstrued human sex differences. *Psychological Inquiry*, 24(3), 195-201.
- Herz, M., & Johansson, T. (2015). The normativity of the concept of heteronormativity. *Journal of homosexuality*, 62(8), 1009-1020.
- Kim, K. J., Feeney, B. C., & Jakubiak, B. K. (2018). Touch reduces romantic jealousy in the anxiously attached. *Journal of Social and Personal Relationships*, 35(7), 1019-1041.
- Madey, S. F., & Rodgers, L. (2009). The Effect of Attachment and Sternberg's Triangular Theory of Love on Relationship Satisfaction. *Individual Differences Research*, 7(2), 76-84.
- Marazziti, D., Sbrana, A., Rucci, P., Cherici, L., Mungai, F., Gonnelli, C., Massimetti, E., Raimondi, F., Doria, M. R., Spagnolli, S., Ravani, L., Consoli, G., & Osso, M. C. D. (2010). Heterogeneity of the jealousy phenomenon in the general population: An Italian study. *CNS Spectrums*, *15*(1), 19-24.

- McConnell, E. A., Janulis, P., Phillips, G. II, Truong, R., & Birkett, M. (2018. Multiple minority stress and LGBT community resilience among sexual minority men. *Psychology of Sexual Orientation and Gender Diversity*, *5*(1), 1–12.
- Pfeiffer, S. M., & Wong, P. T. (1989). Multidimensional jealousy. *Journal of Social and Personal Relationships*, 6(2), 181-196.
- Pines, A. M., & Friedman, A. (1998). Gender differences in romantic jealousy. *The Journal of Social Psychology*, 138(1), 54-71.
- Rhee, S. Y., Park, H., & Bae, J. (2020). Network structure of affective communication and shared emotion in teams. *Behavioral Sciences*, *10*(10), 159-175.
- Rubinsky, V. (2019). Identity gaps and jealousy as predictors of satisfaction in polyamorous relationships. *Southern Communication Journal*, 84(1), 17-29.
- Salovey, P., & Rodin, J. (1984). Some antecedents and consequences of social-comparison jealousy. *Journal of Personality and Social Psychology*, 47(4), 780-792.
- Salovey, P., & Rodin, J. (1986). The differentiation of social-comparison jealousy and romantic jealousy. *Journal of Personality and Social Psychology*, 50(6), 1100-1112.
- Steis, N., Oddo-Sommerfeld, S., Echterhoff, G., Thiel, A., Thiel, J., Briem, K., Ciaramidaro, A., Freitag, C. M., Mecklinger, A., Unterhorst, K., & Stirn, A. (2019). The obsessions of the green-eyed monster: Jealousy and the female brain. *Sexual and Relationship Therapy*, 1-15.
- Turell, S. C., Brown, M., & Herrmann, M. (2018). Disproportionately high: An exploration of intimate partner violence prevalence rates for bisexual people. *Sexual and Relationship Therapy*, 33(1-2), 113-131.
- Walsh, M., Millar, M., & Westfall, R. S. (2019). Sex differences in responses to emotional and sexual infidelity in dating relationships. *Journal of Individual Differences*, 40(2), 63–70.
- White, G. L. (1981a). A model of romantic jealousy. *Motivation and Emotion*, 5(4), 295-310.
- White, G. L. (1981b). Some correlates of romantic jealousy. Journal of Personality, 49(2), 129-145.
- White, G. L. (1984). Comparison of four jealousy scales. Journal of Research in Personality, 18(2), 115-130.
- Yilmaz, T., & Bozo, Ö. (2019). Whose gaze is more objectifying? An experimental study of college women's state self-objectification, body shame, negative mood, and body dissatisfaction. *Mediterranean Journal of Clinical Psychology*, 7(2), 1-23.
- Yost, M. R., & Thomas, G. D. (2012). Gender and binegativity: Men's and women's attitudes toward male and female bisexuals. *Archives of sexual behavior*, 41, 691-702.
- Zengel, B., Edlund, J. E., & Sagarin, B. J. (2013). Sex differences in jealousy in response to infidelity: Evaluation of demographic moderators in a national random sample. *Personality and Individual Differences*, 54(1), 47-51.

Table 1

Participant Demographics

Pseudonym	# of Interviews	Age	Race/Ethnicity	Gender	Sexual Orientation	Relationship Type	# Minoritized Identities
Helena	2	20	Asian, Native Hawaiian/Pacific Islander, and White	Cis woman	Lesbian	Monogamous	2
Joseph	2	23	African American and White	Cis man	Straight/ heterosexual	Monogamous	1
Sue	2	19	Asian and White	Cis woman	Bisexual	Monogamous	2
Tamara	2	21	Asian	Cis woman	Straight/ heterosexual	Monogamous	1
Brittany	2	33	White	Cis woman	Demisexual	Polyamorous	2
Eric	1	25	White	Cis man	Gay	Monogamous	1

Appendix A

Potential Interview Questions for Use by Practitioners

- What do you think the root or roots of jealousy are?
- How do you define jealousy in a romantic/sexual relationship?
 - What does it feel like for you?
 - What does it look like for you?
 - What makes you feel this?
- What does jealousy look like in your relationship(s)?
 - What are the specific triggers or events?
 - How do you react to the feeling of jealousy?
 - What is your thought process?
 - What are your behavioral reactions?
 - What are your emotional reactions?
 - What are the results of your reactions to jealousy?
- What are specific times in any of your current relationship(s) in which you felt jealous? If so, please expand on the circumstances and the intensity of the feeling.
- What do you think has contributed to how you experience and understand jealousy?
- How do you think your labels and identities have contributed to how you understand and experience jealousy?
- How do you think others' perceptions of your identities have contributed to how you understand and experience jealousy?
- How do you think specific experiences have contributed to how you understand and experience jealousy?

ACKNOWLEDGMENTS

Undergraduate Research

NSURJ would like to acknowledge the Undergraduate Research Department for providing admirable opportunities for students to conduct research as undergraduates. Without the NURA, PREP, and Community-based award programs, NSURJ would not have been able to connect with as many intellectually bright undergraduate researchers. The Undergraduate Research department provides many opportunities for students to grow professionally and financially from doing research at the University of Nevada, Reno. Students and faculty are in great benefit from having a program like this. Undergraduate Research has always supported NSURJ in our mission to provide students the opportunity to professionally grow while showcasing their superb education.



Wolf Pack Discoveries

NSURJ would like to extend our gratitude to the premier undergraduate research symposium, held at the University of Nevada, Reno. Wolf Pack Discoveries helps early researchers gain experience presenting their research. This event supports the undergraduate academic community in the Nevada Higher Education System and supports the growth of research in an undergraduate system.

Thank you to all who host the event and to the event for providing the opportunity for NSURJ to connect with young researchers.



Associated Students of the University of Nevada (ASUN)

NSURJ would like to extend thanks to the Associated Students of the University of Nevada (ASUN) for providing support towards the journal's first publication, as well as continuing publications such as this one. ASUN's continued support has allowed NSURJ to grow into the academic stepping stone it is and has continually made it possible for NSURJ to continue its mission. ASUN provides needed support for the journal to grow into an outlet for academic development and success for undergraduate researchers.

University Land Acknowledgement

We acknowledge that the University of Nevada, Reno is situated on the traditional homelands of the Numu (Northern Paiute), Wašiw (Washoe), Newe (Western Shoshone), Nuwu (Southern Paiute) peoples. These lands continue to be a gathering place for Indigenous Peoples and we recognize their deep connections to these places. We extend our appreciation for the opportunity to live and learn on their territory.

Graduate Volunteer Reviewer

We extend our gratitude to Alec Brennan, who has served NSURJ as a volunteer reviewer for this edition. Since our current roster of editors have majors outside of the hard sciences and are undergraduates, Brennan's expertise in chemistry as well as her extensive research experience has been invaluable in ensuring that we can best assist our wonderful contributors.