



CABRINI UNIVERSITY

**These students' works were presented
at the 2017 and 2018 Arts, Research
and Scholarship Symposiums**



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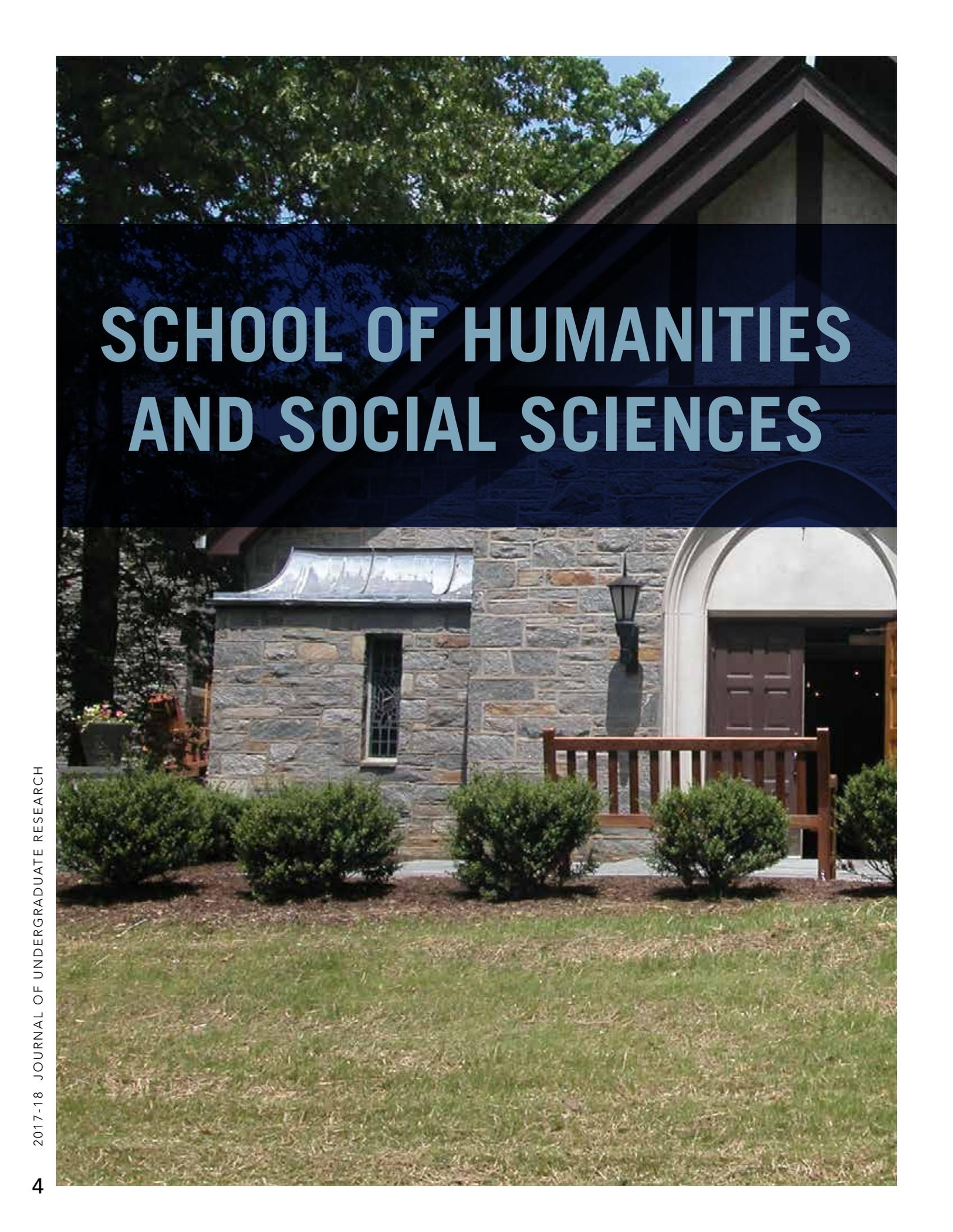
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SCHOOL OF HUMANITIES AND SOCIAL SCIENCES

Mi Español

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Mi español es mexicano como los chilaquiles y los tamales. Mi español es Poblano como el mole poblano y los pambazos. Mi español no viene de Dora The Explorar. Mi español viene del Chavo del ocho y las novelas mexicanas para niños. Desde el drama hasta las canciones; cada día cantándolas con mucho entusiasmo. Mi tía regañando me porque no me sabía las tablas del 2 y 3 o cómo escribir iglú. Cada día repasando las reglas de mi español; es "a las 8" no "alas 8". Para mi español, Disney es la sirenita y mi película favorita Bichos: Una Aventura en Miniatura. Mi español es Harry Potter y la piedra filosofal. Mi español me enseñó sobre el Cinco de Mayo que era la batalla de Puebla contra los franceses; el Cinco de Mayo no es la independencia de México como muchos me dijeron en los Estados Unidos.

Avancemos rápido, y mi español estaba en tercer grado sin saber el idioma de oro. Decían aprende lo más rápido que puedas si no las oportunidades se van. Lo aprendí y gracias a Dios, mi español se quedó conmigo. Mi español ayudo a mis padres en buenos y malos tiempos mientras el idioma de oro tomo mi mente y me robo los pensamientos de mi español. Mi español estuvo dormido por unos años porque ya no quería que se burlaran de mí. Quería ser como los otros niños y fue así que mi español dejo las novelas y el Chavo del ocho; el idioma de oro agarro mi interés y convirtió mi niñez en Hannah Montana y Harry Potter and the Chamber of Secrets.

Lastimé a mi español, pero lo nutrí poco a poco. Creció otra vez y es más fuerte que nunca. Mi español no es solo un idioma; es una cultura rica y hermosa. Ahora mi español y mi inglés viven en armonía como dos mejores hermanas que piensan igual. Ya no se pelean si es La sirenita o The Little Mermaid si no aman a los dos iguales. Mi español siempre vivirá conmigo hasta que la muerte nos separen.

Gendered Perceptions of Rape Victims:
The Relationship Between Rape Myths, Victim Blaming, and Gender

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Abstract

Rape myth acceptance and victim blaming are common phenomena that exist in modern rape culture. When people ascribe to false perceptions and beliefs about rape, victims are often subjected to secondary victimization as the blame is transferred from the perpetrator to the victim. This study seeks to analyze the relationship between rape myth acceptance, victim blaming, and the gender of both the rape victim and the person perceiving the assault. The results show that men are more likely to accept rape myths and victim blame than women, regardless of the gender of the rape victim.

Introduction

In her memoir *Lucky* (1999), Alice Sebold describes herself as the perfect rape victim: a young white female virgin who was brutally attacked at night by a stranger in a park. She was wearing modest clothing, showed visible signs of a physical altercation, and was not intoxicated. Her attacker was a black man and she reported her assault immediately after it happened. She was described as the “perfect” rape victim, and due in part to her testimony at trial, her rapist was convicted.

Sebold’s story, as she herself admits, is abnormal. For most rape victims, their stories do not end with a conviction or even an arrest (U.S. Department of Justice, 2015). Most sexual assault victims share experiences similar to that of a Canadian woman who was raped over a bathroom sink at a house party in 2014 (Willingham & Hassan, 2016). During the trial, the judge overseeing her case, Robin Camp, asked the woman the following questions: “Why couldn’t you just keep your knees together?” and “Why couldn’t you just sink your bottom down into the basin so he couldn’t penetrate you?” (Willingham & Hassan, 2016). More of Camp’s comments regarding the case include: “Young women want to have sex, particularly if they’re drunk,” and

“Some sex and pain sometimes go together... that’s not necessarily a bad thing” (Willingham & Hassan, 2016).

The views displayed by Judge Camp are stereotypical of someone who believes in rape myths, which are “attitudes and beliefs that are generally false but are widely and persistently held, and that serve to deny and justify male sexual aggression against women” (Lonsway & Fitzgerald, 1994, p. 134). This phenomenon is fueled by a combination of stereotypes and prejudices held by both men and women (Burt, 1970). Rape myths exist in patriarchal societies- that is, societies in which men are the dominant group- to remove the blame from men for sexual violence (Vonderhaar & Carmody, 2015a). They are supported through the socialization of women to dress and act in certain ways to decrease the chances of victimization, and through the socialization of men to act strong and tough (Vonderhaar & Carmody, 2015a). Common rape myths include the belief that rape is often falsely reported, that rape is not a serious crime, and that some aspect of a victim’s appearance or behavior led to the rape (Smith, Wilkes, & Bouffard, 2016).

The acceptance of rape myths leads to a phenomenon known as victim blaming. Victim blaming is “where individuals find instances within the victim's behavior, such as drinking alcohol, to hold the victim at least partially responsible for the incident” (Hayes, Lorenz, & Bell, 2013, p. 203). Since rape myths are typically based around female victims, victim blaming is usually directed toward female victims which can result in a societal view that victimization is feminine (Hayes et al., 2013). When a rape is consistent with rape myths, the likelihood that the victim will be blamed for the assault increases (Hockett, Smith, Klausing, & Saucier, 2016). When a rape is inconsistent with rape myths, people are less likely to blame the victim because they view him/her as a “true” victim who was not responsible for his/her victimization (Hockett,

Smith, Klausing, & Saucier, 2016). Although people are socialized to sympathize with victims, in cases of sexual assault, people tend to accuse rather than empathize (Burt, 1980). Since women are viewed as responsible for their own sexuality, when they are sexually violated, they are considered to be at fault because they did not protect their sexuality (Burt, 1980).

In cases of sexual assault, both men and women tend to victim blame, but men significantly more so (Vonderhaar & Carmody, 2015a). Prior studies have found that men are more likely to blame the victims than the actual perpetrators for crimes (Vonderhaar & Carmody, 2015a). Existing research that tests the likelihood a victim will be blamed for sexual assault is limited mainly to analyzing people’s reactions only to female victims. Since rape myths operate on the basis that females are the victims and males are the offenders, most people only consciously hold prejudices toward female victims of rape. However, the prevalence of male rape myths- though more subtle and less studied than female rape myths- creates similar negative reactions to male rape victims.. The influence of male rape myths on people’s likelihood to victim blame has rarely been tested in empirical research, but to fully examine the effects and impacts of the phenomenon of victim blaming, people’s reactions to both male and female rape victims must be explored.

Literature Review

Lack of Rape Reporting. According to the Federal Bureau of Investigation, rape is “penetration, no matter how slight, of the vagina or anus with any body part or object, or oral penetration by a sex organ of another person, without the consent of the victim” (U.S. Department of Justice, 2015). In the 2015 Uniform Crime Report (UCR), law enforcement agencies reported 105,626 cases of rape (U.S. Department of Justice, 2015). In stark contrast, the 2015 National Crime Victimization Survey (NCVS) shows that 431,840 individuals claimed they were raped (Truman

& Morgan, 2016), nearly double that of the UCR. Rape is the only crime listed in both the UCR and the NCVS that has such a dramatic difference in the number of offenses known to police and the number of offenses victims claim actually occurred. As a result, rape is the most underreported felony in the country (U.S. Department of Justice, 2015), a phenomenon which criminologists have spent decades trying to explain.

Typically, victims of rape will not report their attacks because they are not viewed as “true” victims (Hockett, Smith, Klausing, & Saucier, 2016). Based on popular perception, a true rape victim is a young woman who is not intoxicated, who is violently raped by a stranger in an abandoned public location, and who immediately informs the police of her assault while providing direct evidence of her resistance (Hockett, Smith, Klausing, & Saucier, 2016). If a victim does not meet the criteria of a “true” victim, he/she will be less likely to be believed by police, family members, and juries (Hayes et al., 2013). While male and female victims are both typically hesitant to report, male victims are 1.5 times less likely to inform the police of their rapes (Chapleau, Oswald, & Russell, 2008). Little empirical research has been conducted to determine whether or not the sex of the rapist affects the likelihood a victim will report, but Chapleau et al. theorize that male victims will be even more reluctant to go to the police if their attacker is female (2008).

Female Rape Myths. The most common explanation for the lack of sexual assault reporting is the prevalence of rape myths in modern culture (Vonderhaar & Carmody, 2015a). The root of rape myth acceptance stems from the socialization of men and women to behave in certain ways based on their genders (Burt, 1980). Men are expected to be sexually aggressive, dominant, and powerful, whereas women are taught to be sexually conservative, submissive, and weak (Yamawaki, 2007). The strength of gender roles in society tends to create a ‘no-win’ situation for

rape victims in that when victims behave according to gender roles, their victimization is dismissed as something to be expected; when victims behave opposite from gender roles, their victimization is considered to be deserved or warranted (Burt, 1980).

Primarily, women are the subject of rape myths since sexual assault typically includes a female victim (Chapleau et al., 2008). In the Updated Illinois Rape Myth Acceptance Scale, there are four categories of common female rape myths: “she asked for it,” “he didn’t mean to,” “it wasn’t really rape,” and “she lied” (Payne, Lonsway, & Fitzgerald, 1999; McMahon & Farmer, 2011). The following statements are examples of common female rape myths: “When girls go to parties wearing slutty clothes, they are asking for trouble,” “It shouldn’t be considered rape if the guy was drunk and didn’t know what he was doing,” “If a girl doesn’t physically fight back, you can’t really say it was rape,” and “A lot of times, girls who say they were raped agreed to have sex and then regret it” (Payne et al., 1999; McMahon & Farmer, 2011).

Female rape myths typically involve beliefs of how women are expected to behave based on engendered characteristics (Doherty & Anderson, 2004). For example, based on their gender roles of being submissive and vulnerable, women are expected to dress conservatively, behave passively, and accept male aggression in all forms (Chapleau et al., 2008). When women act in a way opposite from what is expected of them as females, people tend to believe that any victimization that occurs is deserved and precipitated by the woman since she was not behaving in accordance with her assigned gender role (Daigle & Mummert, 2014). Due to the social concept that women are weak, female victims typically receive no blame for not resisting their attackers because they are not expected to be able to fight back (Davies, Pollard, & Archer, 2001); however, women are typically blamed for putting themselves in potentially dangerous

situations, i.e. walking alone at night, since women are socialized to minimize their chances of being victimized (Doherty & Anderson, 2004).

Male Rape Myths. Most rape myths focus on female victims, but many stereotypes about male rape victims are prevalent in society as well (Struckman-Johnson & Struckman-Johnson, 1992). Men who have been raped are blamed primarily for their behaviors in that people believe men, unlike women, should be able to fight off or escape from their attackers (Davies et al., 2001). If male victims do not fight their rapists, they are blamed for being passive and weak since their encumbered gender roles include being dominant, strong, and powerful (Struckman-Johnson & Struckman-Johnson, 1992). Male rape victims who are attacked by men are frequently assumed to be gay since the perception exists that socially accepted “real” men would not allow themselves to be attacked by someone who is homosexual (Chapleau et al., 2014). In cases where a female is the perpetrator, the male victim is even less likely to be believed due to the assumption that all “masculine” men enjoy sex with women, regardless of the circumstances (DeVries Lawler, 2002).

Male rape victims tend to be dismissed and ignored at a greater rate than female rape victims as a result of rape myths (Struckman-Johnson & Struckman-Johnson, 1992). Since men are considered to be unemotional, male rape victims are expected to cope with the trauma of their attack easily and quickly without the assistance of therapy or the closure of a trial (Struckman-Johnson & Struckman-Johnson, 1992). The majority of men hesitate to report their attack out of fear that they will be labelled as weak and unmasculine (Doherty & Anderson, 2004). Daigle and Mummert (2014) assert that it is not the traits of masculinity themselves which frighten male victims into silence, but rather male victims’ fear that they will be revictimized through the rejection of society and the mocking of their masculinity.

Rape Myth Acceptance. Although men have a greater tendency to accept rape myths than women (Davies & McCartney, 2015), empirical research has found that both men and women perpetuate rape culture, regardless of gender and their own personal victimization (Hockett, Smith, Klausing, & Saucier, 2016). However, the reasons why men and women adhere to rape myths are different. In most rape scenarios with a female victim, the majority of men believe in stereotypes about the victim (Vonderhaar & Carmody, 2015a), whether that be her clothes were too provocative, she was “asking for it” by getting drunk, or she did not say no or fight back (Smith et al., 2016). By accepting female rape myths, men are able to remove the responsibility from the male perpetrator which reinforces the dominance of the male status hierarchy and provides support for the gender role that women are weak (Hockett, Saucier, & Badke, 2016). Through the adherence to male rape myths, men can assert their own masculinity by claiming they themselves to be too strong to be raped and that men who are sexually assaulted are not “real” men (Davies et al., 2001).

Women are almost just as likely to give credence to rape myths as men even though female acceptance of rape myths appears contradictory (Vonderhaar & Carmody, 2015b). The reason for this inconsistency may be found in the Just World Hypothesis which states that people get what they deserve (Hayes et al., 2013). In order to distance themselves from the threat of rape, women may view female rape victims as deserving of their attacks and that they themselves are “good” women who have done nothing wrong and as a reward, are not victimized (Hockett, Smith, Klausing, & Saucier, 2016). The prevalence of female acceptance of rape myths may also speak to the dominance of the patriarchy in society. Even though women may not inherently believe in the stereotypes rape myths support, they have been socialized by the male dominant

class to think in a way that protects traditionally masculine men and demeans women and gay men (Vonderhaar & Carmody, 2015a).

Victimization and Victim Blaming. The overall acceptance of rape myths in society has led to a misconstrued perception of victimization. Victimization is typically viewed as a female problem since the general perception is that only people who are weak (and therefore, feminine) will be targeted by offenders (Hayes et al., 2013). If people are “good” and strong, they will not be victimized, or if they are, they will be able to fend off their attackers (Hayes et al., 2013). As previously mentioned, sexual victimization is used as a tool to reinforce gender roles. Daigle and Mummert (2014) found that women who are perceived as feminine have a higher chance of being victimized because they are considered weak, easy targets. For example, women who take on traditional feminine roles in the home are more likely to be assaulted by their husbands than women who assume egalitarian gender roles with their husbands. However, Daigle and Mummert (2014) also found that women who portray themselves as feminists will be sexually harassed on the street at a greater rate than women who portray themselves as traditionally feminine. So while the empirical research on the relationship between gender-role identification and victimization is slightly inconsistent, the conclusion can be made that men victimize women to capitalize on their femininity or to punish them for behaving otherwise (Daigle & Mummert, 2014).

Although women have a greater chance of being sexually victimized, men have a higher chance of being violently victimized (Daigle & Mummert, 2014). The frequent threat of being attacked causes many people to feel the need to distance themselves from the possibility that they themselves may become victims (Hayes et al., 2013; Howard, 1984). The consequence of this need is the phenomenon of victim blaming. Drawing roots from the Just World Hypothesis

(good people have good experiences and bad people have bad experiences), victim blaming allows individuals to neutralize the injustice in the world, or to create rationalizations for the evils in society so that they can live with any wrongdoing that may occur (Hayes et al., 2013). By claiming individuals who are victimized did something to deserve or cause their suffering, people are able to explain why bad things happen (Howard, 1984).

The Just World Hypothesis is the primary source of victim blaming (Howard, 1984). Analogous with rape myth acceptance, both men and women have a tendency to blame rape victims for their assaults, but males victim blame at a higher rate than females (Hayes et al., 2013). Similar to the justification for rape myths, men blame victims of sexual assault in order to remove the responsibility of sexual violence from male perpetrators, i.e. men cannot be held accountable for rape if the victim did something to cause the assault (Davies & McCartney, 2003). Also comparable to their reasoning for rape myth acceptance, women tend to victim blame to protect themselves from the knowledge that they could be raped (Vonderhaar & Carmody, 2015a). Female victim blaming relies on the notion that only women who deserve to be raped will become victims of assault (Hayes et al., 2013). The justification for victim blaming is threatened in the case of a “true” victim; that is, no circumstances surrounding the crime can be misinterpreted to claim the victim somehow wanted or caused their victimization (Hayes et al., 2013).

Despite the occurrence of victim blaming in both males and females, the amount of blame attributed to a victim differs greatly depending on the victim’s gender (Davies & McCartney, 2003). Not many studies have focused on male rape victims, largely because rape myths are focused primarily on women; however, the few studies that have examined perceptions of male victims have found a large dichotomy in the way people- specifically men-

view male and female victims of sexual assault. Heterosexual men have been found to view male rape victims- both gay and straight- very negatively, more so than how they view female victims of any sexual orientation (Davies & McCartney, 2003). The reason for this can be found in the conflict between traditional masculinity and homosexuality. If heterosexual men feel sympathy for victims of male-on-male rape, then they are condoning a homosexual act and are thereby threatening their own masculinity (Doherty & Anderson, 2004). Conversely, women do not view male victims differently than female victims; rather, they tend to victim blame both genders equally (Davies & McCartney, 2003). The study conducted by Davies and McCartney (2003) showed that while heterosexual men were predominantly “anti-victim” in a male-on-male rape scenario, homosexual men and women of both orientations were predominantly “pro-victim,” leading to the conclusion that traditional masculinity is an important factor in predicting victim blaming.

The present study aims to add to the literature surrounding perceptions of rape victims by analyzing the relationship between rape myth acceptance, victim blaming, the gender of participants, and the genders of rape victims. There are four hypotheses for the present study: a) men will be more likely to accept female rape myths than women; b) men will be more likely to accept male rape myths than women; c) men will be more likely than women to victim blame rape victims, regardless of the victims’ genders; and d) men will be more likely to victim blame male rape victims than female rape victims.

Methods

This study utilized a non-probability, availability sampling design. Data was collected from 144 participants at Cabrini University in March 2017. Surveys were distributed to students

during class. To save time for other researchers, the measures for this study were included with measures from other studies on one survey.

40% (n= 58) of the participants identified as male, and 60% (n= 86) of the participants identified as female. The sample was composed of 61% white students, 22% black students, 8% Hispanic students, 2% Asian students, 7% mixed students, and 0.7% students who identified as other. The majority of participants (61.1%, n= 88) were between the ages of 20-21, and were either sophomores or juniors (70.9%, n= 102). The most frequently reported GPA was 3.00 (10%, n= 15). The sample was representative of the university’s overall population in terms of race and gender according to numbers found in Cabrini’s factbook. Juniors were overrepresented in the sample, and freshmen were underrepresented. The values for all descriptive statistics are reported in Table 1.

The independent variable for this study was the gender of the participant which was measured by asking participants to check one of the following options: male, female, or other. The dependent variables for this study were the gender of the rape victim (the sex-role identification of the person who was raped); degree of male and female rape myth acceptance (the level to which an individual believes in stereotypes about rape victims that are mostly false, but widely accepted as true); and victim blaming (an individual’s tendency to find some fault in a victim’s behavior that shifts the blame from the perpetrator to the victim).

To measure female rape myth acceptance, participants were asked to read a series of ten statements and provide a ranking for each, ranging from *1-strongly agree* to *5-strongly disagree*. Questions were taken from the Updated Illinois Rape Myth Acceptance Scale (McMahon & Farmer, 2011; Payne, Lonsway, & Fitzgerald, 1994). The ten statements are listed in Appendix A. Statement #4 was excluded from statistical analysis due to a grammatical error on half of the

Table 1. Descriptive Statistics.

| | <i>Frequency</i> | <i>Valid Percent</i> |
|--------------------|------------------|----------------------|
| Gender | | |
| Male | 58 | 40.3 |
| Female | 86 | 59.7 |
| Race | | |
| White | 84 | 60.9 |
| Black | 30 | 21.7 |
| Hispanic/Latino | 11 | 8.0 |
| Asian | 3 | 2.2 |
| Mixed | 9 | 6.5 |
| Other | 1 | 0.7 |
| Age | | |
| 18 | 11 | 7.7 |
| 19 | 22 | 15.4 |
| 20 | 47 | 32.9 |
| 21 | 41 | 28.7 |
| 22 | 17 | 11.9 |
| 23 | 5 | 3.5 |
| Class Level | | |
| Freshman | 18 | 12.5 |
| Sophomore | 41 | 28.5 |
| Junior | 61 | 42.4 |
| Senior | 21 | 14.6 |
| Other | 3 | 2.1 |
| GPA | | |
| 1.70-2.09 | 2 | .14 |
| 2.10-3.00 | 48 | 35.3 |
| 3.01-4.00 | 85 | 63 |
| Survey Type | | |
| Survey A | 83 | 57.6 |
| Survey B | 61 | 42.4 |

surveys which could have affected the interpretation of the statement. The nine remaining questions were combined into an additive scale to create an overall female rape myth acceptance variable. Creating the additive scale was appropriate since Cronbach's Alpha was determined to be greater than 0.7 (9 items, $\alpha = .891$). The scale ranged from 10 to 45, with 10 indicating high rape myth acceptance and 45 indicating low rape myth acceptance. The most frequent ranking

was 45 (16%, $n = 23$) and just over half of respondents ranked at 40 or higher (52.8%, $n = 76$).

The mean was 38.25, showing overall relatively low levels of female rape myth acceptance.

To measure male rape myth acceptance, participants were asked to read six statements and rank their level of acceptance from *1- strongly disagree* to *6- strongly agree*. Questions were taken from Struckman-Johnson and Struckman-Johnson (1992). The six statements are provided in Appendix B. In order to create an additive scale, two statements were excluded due to their potentially confusing wording which may have led participants to indicate a response opposite from what they intended. After discarding Statements #12 and #15, Cronbach's Alpha was reported to be .771 (4 items), which indicated that creating an additive scale was appropriate. The male rape myth acceptance scale ranged from 4 to 24, with 4 representing low male rape myth acceptance and 24 indicating high male rape myth acceptance. The most common ranking was 4 (41%, $n = 59$), and the majority of respondents ranked at 9 or lower (66.8%, $n = 96$). The mean was 8.34, which demonstrated low male rape myth acceptance overall.

Next, to measure victim blaming, participants were asked to read a short vignette of approximately 200 words depicting an incident of stranger rape which was adapted from Davies, Pollard, and Archer (2001). The phrasing of the vignettes was altered from British slang terms to American slang terms for clarity. For 83 participants, the victim in the vignette was male. For 61 participants, the victim in the vignette was female. After reading the vignette, participants read 12 statements about the incident and ranked each statement from *1-pro-victim judgement* to *7-anti-victim judgement*. The vignette and questions are provided in Appendix C. After assessing Cronbach's Alpha (12 items, $\alpha = .86$), all victim blaming questions were combined into an additive scale from 12 to 84, with 12 indicating low victim blaming and 84 indicating high victim blaming. One third of respondents ranked at 12 (33.3%, $n = 48$), and over two thirds of

respondents ranked at 17 or lower (63.3%, $n=91$). The mean was 17.72 and the mode was 12, which indicated low rates of victim blaming in general without accounting for the victim's gender. Descriptive statistics for all four variables are reported in Table 2.

To analyze both forms of rape myth acceptance against gender, two independent sample t-tests were conducted. This form of analysis was most appropriate for the research question since gender is a two-way categorical variable, and the rape myth acceptance scales are ratio-level variables. To analyze the level of victim blaming based on gender, a one-way analysis of variance (ANOVA) was conducted. An ANOVA was the test most appropriate since the independent variable consisted of more than two categories, and the dependent variable was continuous. To account for both the gender of the participant and the gender of the victim, four groups were created for comparison: male participant/male victim, male participant/female victim, female participant/male victim, and female participant/female victim.

Table 2. Variable Descriptive Statistics.

| | <i>N</i> | <i>Mean</i> | <i>Median</i> | <i>Mode</i> | <i>Range</i> | <i>Standard Deviation</i> |
|------------------------------------|----------|-------------|---------------|-------------|--------------|---------------------------|
| Gender | 144 | - | - | 2 | 1 | - |
| Female Rape Myth Acceptance | 138 | 38.25 | 40 | 45 | 35 | 7.00 |
| Male Rape Myth Acceptance | 143 | 8.34 | 6 | 4 | 20 | 5.25 |
| Victim Blaming | 140 | 17.72 | 14.5 | 12 | 40 | 7.75 |

Results

Female Rape Myth Acceptance. The independent sample t-test for female rape myth acceptance was significant at the $p<.01$ level. Levene's Test was significant $F(1, 97.63) = 4.65, p<.05$, therefore, equal variances were not assumed. Men ($M=35.16, SD=7.52$) reported higher levels of female rape myth acceptance than women ($M=82, SD=5.79$), $t(1, 97.63) = -4.37$. Cohen's D was used to compute the effect size which was determined to be large, $d=-0.75$. The 95% Confidence Intervals were relatively close, ranging from -7.57 to -2.84 . The results of the t-test show that men had a higher rate of female rape myth acceptance by 5 points, supporting the hypothesis that men would be more likely to accept female rape myths. The results for female rape myth acceptance are depicted in Table 3.

Male Rape Myth Acceptance. The independent sample t-test for male rape myth acceptance was found not to be significant at any level. Levene's Test was not significant $F(1, 141) = 2.64, p=.107$, so equal variances were assumed. There was no difference in the level of male rape myth acceptance between men ($M=8.47, SD=4.60$) and women ($M=8.23, SD=5.67$) with the t-test being insignificant overall $t(1, 141) = .242, p=.809$. The results of the t-test do not support the hypothesis that men would be more likely to accept male rape myths. The results for male rape myth acceptance are depicted in Table 4.

Victim Blaming. The ANOVA for victim blaming overall was found to be significant $F(1, 136) = 4.57, p<.01$, indicating that there is a relationship between gender and victim blaming. Tukey's D revealed that there was no significant difference in the amount of blame men attributed to male victims ($M=21.36, SD=9.6$) compared to female victims ($M=19.29, SD=9.01$). Despite this,

there was a significant difference in the level of victim blaming between men overall and female participant/male victim ($M=16.13, SD=6.88$) as well as female participant/female victim ($M=15.72, SD=4.69$). Of the four groups, male participant/male victim was shown to have the highest rate of victim blaming while female participant/female victim was shown to have the lowest rate of victim blaming; however, the results show that victim gender does not factor into the amount of blame men or women place on the victim. Therefore, the hypothesis which predicted men would be more likely to victim blame regardless of the victim's gender was supported, while the hypothesis which predicted men would blame male victims more than female victims was not supported. Results for the ANOVA and the post-hoc comparisons can be found in Tables 5 and 6.

Table 3. Independent Sample t-Test for Female Rape Myth Acceptance.

| | <i>F</i> | <i>Significance (Levene's)</i> | <i>t</i> | <i>df</i> | <i>Significance</i> |
|------------------------------------|----------|--------------------------------|----------|-----------|---------------------|
| Equal Variances Not Assumed | 4.65 | .033 | -4.37 | 97.63 | .000 |

Table 4. Independent Sample t-Test for Male Rape Myth Acceptance.

| | <i>F</i> | <i>Significance (Levene's)</i> | <i>t</i> | <i>df</i> | <i>Significance</i> |
|--------------------------------|----------|--------------------------------|----------|-----------|---------------------|
| Equal Variances Assumed | 2.64 | .107 | .242 | 141 | .809 |

Table 5. ANOVA for Victim Blaming.

| | <i>Sum of Squares</i> | <i>df</i> | <i>Mean Square</i> | <i>F</i> | <i>Significance</i> |
|-----------------------|-----------------------|-----------|--------------------|----------|---------------------|
| Victim Blaming | 765.08 | 3 | 255.03 | 4.57 | .004 |
| Within | 7587.05 | 136 | 55.79 | | |
| Total | 8352.14 | 139 | | | |

Table 6. Tukey HSD for Victim Blaming.

| | <i>Participant/Victim</i> | <i>Mean Difference</i> | <i>Standard Error</i> | <i>Significance</i> |
|-------------------------|---------------------------|------------------------|-----------------------|---------------------|
| Male/Male Victim | Male/Female Victim | 2.08 | 2.08 | .752 |
| | Female/Male Victim | 5.24 | 1.70 | .013 |
| | Female/Female Victim | 5.65 | 1.77 | .009 |

Discussion

The purpose of this study was to assess the relationship between rape myth acceptance, victim blaming, and gender. Based on the findings in previous literature, four hypotheses were constructed. Of the two that were supported, the first predicted that men would be more likely to accept female rape myths than women. The results support this hypothesis in that men had a higher average rape myth acceptance score by 5 points, indicating that although there is a significant difference between men and women regarding female rape myth acceptance, the difference itself is not extremely high. The second hypothesis which was supported stated that men would be more likely to victim blame than women, regardless of the gender of the victim. This claim was supported by the results in that overall, men victim blamed an average of 5 points higher than women. Both findings are evidenced in previous literature which states men are more likely to victim blame and adhere to rape myths due to patriarchal socialization, gender role expectations, and the neutralization of male sexual aggression against women (Burt, 1980).

Two hypotheses were not supported. The first unfounded hypothesis predicted that men have higher male rape myth acceptance than women. The results did not support this hypothesis since both men and women were shown to accept male rape myths at nearly the same level. There are two possible explanations for the insignificance of this relationship. The first explanation could lie in the wording of the male rape statements and the corresponding scale.

Two statements were phrased oppositely from the other four, meaning that in order for participants' answers to agree, they could not circle the same number for each statement; however, it was evident many participants did not carefully read the statements and chose the same answer for each one. Ergo, the reliability of the male rape myth scale can be questioned.

The second explanation for the insignificance of male rape myth acceptance could be due to the ages and education levels of participants. Kassing, Beesley, and Frey (2004) found that young, well-educated individuals are less likely to adhere to rape myths, particularly those concerning male victims. Since the entire sample was composed of college students in their late adolescence and early twenties, age and education level could have impacted the degree to which respondents accepted male rape myths. Knowing that the overall rate of rape myth acceptance was low, it can be argued that education and age may be factors that were not considered.

The second unsupported hypothesis predicted that men would attribute more blame to male victims than female victims. A possible explanation for this could be found in the selected majors of participants. Although college major was not measured, the majority of surveys were distributed in criminology and sociology classes, indicating that the majority of participants were criminology or sociology majors. Myyry and Helkama (2001) found that among social science, business, and technology majors, social science majors had the highest levels of empathy; therefore, it could be concluded that social science majors in this sample also have high levels of empathy which would translate to low levels of victim blaming. Due to the liberal nature of Cabrini's criminology and sociology curricula, it can also be assumed that participants would have been taught to sympathize with, rather than blame, the victim. The overall low levels of victim blaming across both genders further supports this assumption.

This study was limited in its relatively small sample size of 144 participants. Additionally, the sample was composed entirely of college students which limits the generalizability of the findings. The generalizability is also compromised by the sampling method used. Since participants were found through availability sampling, the sample is not entirely representative in terms of class level which may have affected the results; however, the sample was representative regarding race and gender. Also, an unequal number of Survey A and Survey B were collected which may have affected the results of the victim blaming analysis since the number of people who read about a male victim was not equivalent to the number of people who read about a female victim.

Future research may wish to explore other factors which could affect a person's likelihood to victim blame, such as the sexuality of the participant as well as the sexuality of the victim. Other factors to be explored could also be the type of rape, such as stranger rape versus acquaintance rape; whether or not drugs and alcohol were involved; and the behavior of the victim before the rape occurred. The effect of race on rape myth acceptance and victim blame was not measured, but future studies may wish to explore the relationship between race, gender, and victim blaming. Further research should also be conducted on male rape myths and male victim blame in general since the existing literature surrounding both phenomena is limited.

The results of study show that rape myth acceptance and victim blaming are generally low in college students which is extremely important in today's rape culture. The findings demonstrate that young people are more supportive of rape victims and are less willing to accuse victims of precipitating their attacks which holds positive implications for the treatment of rape victims in the future. Furthermore, the results indicate that social institutions, particularly colleges and universities, should readdress how they represent and treat rape victims. To

completely eliminate any degree of rape myth acceptance and victim blaming, the focus of rape-prevention programs should change from telling people how not to get raped to telling people not to rape in the first place. Then, the blame will remain solely with the perpetrator and not with the victim. Overall, the findings of this study demonstrate that hopefully rape culture is diminishing with the upcoming generations.

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Appendix A

Measures for assessing female rape myth acceptance. Adapted from Updated Illinois Rape Myth Acceptance Scale (Payne et al., 1999; McMahon & Farmer, 2011).

1. If a girl is raped while she is drunk, she is at least somewhat responsible for letting things get out of hand.
2. When girls go to parties wearing slutty clothes, they are asking for trouble.
3. When girls get raped, it's often because the way they said "no" was unclear.
4. It shouldn't be considered rape if the guy was drunk and didn't realize what he was doing.
5. If both people are drunk, it can't be rape.
6. If a girl doesn't physically resist sex—even if protesting verbally—it can't be considered rape.
7. If a girl doesn't physically fight back, you can't really say it was rape.
8. If a girl doesn't say "no" she can't claim rape.
9. A lot of times, girls who say they were raped agreed to have sex and then regret it.
10. Rape accusations are often used as a way of getting back at guys.

Appendix B

Measures for assessing male rape myth acceptance. Adapted from Struckman-Johnson and Struckman-Johnson (1992).

1. It is impossible for a man to rape a man.
2. Even a big, strong man can be raped by another man.
3. Most men who are raped by a man are somewhat to blame for not being more careful.
4. Most men who are raped by a man are somewhat to blame for not escaping/fighting off the man.
5. Most men who are raped by a man are very upset by the incident.
6. Most men who are raped by a man do not need counseling after the incident.

Appendix C

Measures for assessing victim blaming. The gender of the victim depicted in the vignette was changed from male to female for approximately half the surveys. Adapted from Davies et al., (2001).

Vignette. Steve is a 21-year-old student, in the final year of his English degree. He lives with his girlfriend, Rachel, near the university. He works several evenings a week at the local recreation center. On the weekends he enjoys going to bars and the movies. He is also an active member of the university's Drama Society and is a member of the club lacrosse team.

After finishing work, Steve made his way to his car that was parked in the nearby parking lot. The surrounding streets were quiet, but as Steve walked through the parking lot toward his car a man approached him and asked him for a light. As Steve reached into his pocket for his lighter the man suddenly grabbed him by the arm, pushing him roughly toward the bushes at the darkened rear of the parking lot. Steve, at first, resisted but the man told him threateningly that he would be very sorry if he did not do what he was told. Steve was very scared as the man forced him to the ground. Still issuing threats, the man then raped him. After the rape the man ran away, leaving Steve lying on the ground, bruised and shaken.

1. How responsible do you think [the victim] was for the attack?
2. [The victim] could have done something to prevent the attack if he/she really wanted to.
How much do you agree?
3. Do you think [the victim] can be blamed for not putting up enough of a fight?
4. How much sympathy do you feel for [the victim]?
5. Do you think [the victim] was to blame because he/she didn't try hard enough to escape?
6. How much do you think [the victim's] behavior was to blame for the attack?

7. How seriously do you think the police should take the attack?
8. How much do you think that [the victim] will be traumatized?
9. To what extent should the attacker be held responsible?
10. How severely should the attacker be punished?
11. How much do you think that [the victim's] life will be adversely affected?
12. If you were [the victim], how upset would you be?

Television Consumption and Fear of Crime

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Abstract

The research conducted looks at crime show consumption and an individual's fear of crime. The study builds upon previous literature regarding television and fear with a main focus on crime shows. A crime show is defined as a television series, fiction or non-fiction, which focuses on the criminal justice system and solving crimes. Fear of crime is conceptualized as the emotion or worry people feel over the threat of victimization (Fisher, Sloan, & Cullen, 1995). Previous literature suggests that women are overall more fearful of victimization than men (Kohm, Waid-Lindberg, Weinrath, Shelley, & Dobbs, 2012). Television, in general, is such a significant part of people's lives that the average American watches at least five hours of television a day (Hinckley, 2014). This study analyzes different crime related television shows to uncover what genre of crime show leads to the most fear of crime. The first hypothesis predicts that college students who watch more hours of non-fiction crime shows are more fearful of crime. The second one states that women are more fearful of crime compared to men. The study surveyed 154 students. Significance was found with females, individual's grades, and fear.

Introduction

The media, particularly television, plays a significant role in people's everyday lives. In 2014, the Daily News reported that the average American watches over five hours of television a day (Hinckley, 2014). This media absorption includes a slew of television genres, which makes watching television appealing to many groups of people. This exposes viewers of all ages to the news, debates, movies, and shows that are based on true events. Although not all shows on television are based on true events, the material can be relatable to everyday life. By depicting real-world crimes and events, audiences' fear of crime could increase due to these media outlets (Dowler, 2003).

There are two types of crime-related shows that are on television and expose viewers to violence and provide information on the criminal justice system (CJS): crime dramas and non-fiction crime shows. Crime dramas are fictional shows that depict crimes, but are created by writers, which means the material could be based on real-world events or formed in someone's imagination. Fictional crime shows include shows like *CSI*, *Law and Order: SVU*, and *Blue Bloods*. The other form of crime shows are non-fiction, which can consist of the news, crime documentaries, and other real-world crime events that are factual. Some examples of non-fiction crime shows are: *COPS*, *The First 48*, and the news. Fiction and non-fiction crime-related television can have an impact on viewers based on the violent and sometimes gory material.

Fear of crime can be conceptualized many different ways, but for this study, it is defined as the emotion or worry that people feel over the threat of victimization (Fisher, Sloan, & Cullen, 1995). Fear of crime is important to analyze because fear can lead to people being antisocial and avoiding one another. This in turn can lead to an overall decrease in happiness (Kohm et al., 2012). There are specific groups of people who are affected more by fear of crime than others.

Typically females are more fearful of crime and being victimized, especially in regards to sexual assault (Heath & Petraitis, 1987; Kort-Butler & Sittner-Hartshorn, 2011). This increased fear could be heightened when watching more crime-related television. In regards to the television shows, non-fiction crime shows tend to increase a viewer's fear of victimization more than fiction because the material is based on true events and it can make the situation feel more relatable (Kohm, Waid-Lindberg, Weinrath, Shelley, & Dobbs, 2012). This fear component of crime related media is an important aspect to examine since there seems to be a correlation between fear of crime and crime based television (Eschholz, 2002).

Kort-Butler and Sittner-Hartshorn (2011) found that documentary-style non-fiction crime shows are not typically represented in studies. This study will include documentary-style crime shows in the category of non-fiction to get a well-rounded analysis of this type of genre. Along with non-fiction crime shows, the current study will focus on crime dramas to determine which genre, non-fiction or fiction crime shows, causes more fear among viewers. The study will also focus on fear of crime in general to determine if a specific gender is more fearful of victimization. Crime-based programming and fear of crime is the key focus of the study and based on previous research, it is hypothesized that: women who watch more hours of non-fiction crime shows are more fearful of crime compared to men, and students who watch more hours of non-fiction crime shows are more fearful of crime than students who do not. This current study is intended to confirm previous research while identifying what types of programming affect fear of certain types of victimization. Surveys were distributed to 154 college students at Cabrini University, located in Radnor, Pennsylvania. This survey will range in questions from the types of crime shows the individual watches to how fearful he or she is of being victimized.

Literature Review

Television Perceptions

In 1994, the public's perception of crime as the most important problem (MIP) across the United States spiked from 5% in 1992 to 52% (Lowry, Nio & Leitner, 2003). Considering the crime rate only increased 7.8% from 1978 to 1994, the cause of this shift to crime as the MIP is not due to an increase in crime, but rather the way the news portrays crime (Lowry et al., 2003). In their study, Lowry et al. (2003) discovered that in New Orleans, homicide represented almost half of local news network stories, but police reports showed that homicides actually made up only 0.4% of all crimes. This false sense of reality creates a skewed view of crime in America. Gerbner, Gross, Eleey, Fox, Jackson-Beeck, and Signorielli (1976) found that the more a person is exposed to the crimes that are portrayed on television, the more likely a person is to view the world as a violent and scary place. Consuming shows and news that are not accurate to real life is beneficial to television networks because people's viewpoints are then distorted to favor the fictional world, which increases viewership and income for the companies (Heath & Petraitis, 1987). If the news is relaying skewed information and the public is believing it, then it makes sense that perceptions would favor crime shows because people are viewing the media's information as truth as opposed to looking at real-life statistics.

According to the Uniform Crime Report (UCR), violent crime has decreased in the United States by 16.5% since 2006 (U.S. Department of Justice, 2016). Despite the decline in crime, the public's perception might be the opposite due to the types of shows viewers consume and the material they are absorbing. After a perusal of the websites of the five major broadcasting networks (NBC, CBS, FOX, ABC, and The CW) in the fall of 2016, a total of 37 shows that pertained to crime were found to currently be on television. The shows were put into

the crime genre if they were fiction or nonfiction and dealt with the criminal justice system. This number does not account for crime shows on other networks or streaming services. These shows depict brutal and bloody scenes as a normal occurrence (Hust, Marett, Lei, Ren & Ran, 2015). Kort-Butler and Sittner-Hartshorn (2011) reasoned that the rise in crime shows could be occurring in order to scare viewers into concerning themselves with crimes so that support remains for a more punitive approach to crime. This may explain why *Law & Order* ran for twenty successful seasons because the drama depicted victims who were violently assaulted and their search for justice (Hust et al., 2015). Hust et al. (2015) point out that the popularity of crime drama is increasing due to the dependability and predictability of the shows' structure. Entertainment is the goal for television, but with the increasing violent media, fear is becoming a consequence.

Violent Media

In a study conducted by Eschholz (2002), crime shows were analyzed and then phone interviews were conducted to get the public's perception on crime television. The study determined that total crime television viewing is positively and significantly related to fear of crime (Eschholz, 2002). This relationship occurs due to the high amounts of violence in crime dramas and reality crime shows (Kort-Butler & Sittner-Hartshorn, 2011). Although crime dramas are fictional, some shows can affect a person's fear of being victimized because of their realistic nature (Custers & Van den Bulck, 2011). Nir and Mutz (2006) corroborate this idea by pointing out that fictional narratives have the ability to "transport" viewers into the stories. This insertion into the plotline may lead people to forget the show is fictional, therefore, they become susceptible to the political motives the show may portray, such as drugs as a problem or African-Americans as the offender (Nir & Mutz, 2006). Although someone being persuaded politically

and fearing victimization are two different concepts, they both cause the viewer to react and experience emotions even though the show is a drama. Non-fiction crime shows are more realistic and therefore, may have a larger impact on viewers (Kort-Butler & Sittner-Hartshorn, 2011). Romer, Jamieson, and Aday (2003) conclude that when men and women watch more television news, they are more likely to believe crime is a more prevalent issue. Specifically, a person who uses local news as their primary source for learning about current events is more likely to fear being victimized (Kohm, Waid-Lindberg, Weinrath, Shelley, & Dobbs, 2012). Both fiction and non-fiction crime television shows depict violence and this violence can cause someone to be fearful of crime.

Fear of Crime

Fear of crime is the emotion or worry that people feel over the threat of victimization (Fisher, Sloan, & Cullen, 1995). Fear of crime can lessen a person's satisfaction and fulfillment with life because the person may become avoidant of others due to the fact that they are fearful of trusting people (Kohm et al., 2012). Previous studies show a complex relationship between viewers and the type of television program in regards to causing fear (Kort-Butler & Sittner-Hartshorn, 2011). The varying reactions that people may have when watching crime dramas could be due to the different types of experiences a person faces. There are two types of experiences of crime that a person may encounter: direct and indirect (Tyler, 1980). Direct is when a crime occurs first-hand to someone and he/she is victimized and indirect is when the crime does not occur to that person but they hear it from another source (Tyler, 1980). Tyler (1980) defines two types of indirect experiences: informal social communication and mediated experience. The former occurs when people learn of an event from a personal interaction and the latter is information that is gained from media consumption (Tyler, 1980). Depending on certain

factors such as previous victimization, gender, and race, some people may be more susceptible to be fearful of crime than others (Eschholz, 2002; Kort-Butler & Sittner-Hartshorn, 2011). For example, if someone was victimized, they may be more fearful of crime due to that prior encounter.

Prior victimization is an important factor to consider when discussing fear of crime because victims of traumatic experiences are typically more fearful than those who have not been exploited (Heath & Petraitis, 1987). If looking at this through the cultivation hypothesis, people who encounter direct victimization may receive a "double dose" when consuming media. This means that those who already experienced a crime first-hand may be reminded of that moment when exposed to violent media (Custers & Van den Bulck, 2011). Cultivation hypothesis is a reoccurring theory when researching fear of crime and television viewing. It states that viewing large amounts of violent shows leads viewers to feel a higher level of fear of crime and mistrust (Kort-Butler & Sittner-Hartshorn, 2011). However, Tyler (1980) counteracts this idea by suggesting that if a person believes in mediated experiences, or what the media is telling him or her, then television viewing can have a higher impact on a person's level of fear even if that person has not been a victim. Although they are opposite viewpoints, both researchers agree that audiences are at risk for feeling fearful when watching crime shows. The factor of victimization is one element that should be looked into when relating fear and television consumption, and demographics, specifically gender, is another.

Gender and Fear of Crime

Gender is also associated with victimization because women have a fear of being victimized, and more specifically, being sexually assaulted (Kohm et al., 2012). Despite the fact that men have a higher chance of being a victim in all crimes except sexual assault, women are

still two to three times more fearful of victimization (Hilinski, 2008). This increased fear of all crimes can be attributed to the “shadow of sexual assault hypothesis” (Hilinski, 2008). This hypothesis states that women are most fearful of rape, but this fear is carried over into other crimes as well (Hilinski, 2008; Hilinski, Neeson, & Andrews, 2011). Two studies looked at the shadow of sexual assault hypothesis in relation to females on college campuses to see if other factors affected this increased fear. Both studies found that there are other contributing factors to fear, but being fearful of sexual assault does significantly dictate a woman’s reaction toward other crimes (Hilinski, 2008; Hilinski et al., 2011). The shadow of sexual assault hypothesis can help to explain why women are so impacted by fear.

This fear of being attacked causes women to be more fearful of crime overall, which can then transfer into being more fearful than men when consuming television (Heath & Petraitis, 1987; Kort-Butler & Sittner-Hartshorn, 2011). However, Heath and Petraitis (1987) discovered that when looking at urban fear, men have higher levels of fear than women when they are exposed to crime shows for many hours. There is a lack of gender information concerning television consumption and fear of crime, especially when it comes to who watches more television. This will be explored more in the current study because the amount of viewing time can alter a person’s perceptions (Romer, Jamieson, & Aday, 2003; Custers & Van den Bulck, 2011).

Television Viewing and Fear of Crime

The amount of television consumption plays a role in fear because television provides information that can change a person’s viewpoint and the more television is watched, the more that viewpoint can change (Romer et al., 2011). Both perceptions and fear of crime demonstrate how television can mold viewers’ opinions about the criminal justice system and potential

policies (Kort-Butler & Sittner-Hartshorn, 2011; Nir & Mutz, 2006). Using news as an example, if homicides are the most frequent news story, then people may believe more homicides are occurring more frequently and want to enforce a more punitive system. Although that is nonfiction, fiction shows have power as well based on their vivid nature (Custers & Van de Bulck, 2011).

There is a complex relationship between watching television and fear of crime because individuals interpret what they see differently than other people (Kort-Butler & Sittner-Hartshorn, 2011). Due to the complicated nature of human perceptions, there are disparities in previously conducted studies in regards to the extent that fear and television consumption is linked (Kort-Butler & Sittner-Hartshorn, 2011). However, the two most consistent pieces are that the gender of the viewer and the amount of television viewed do relate to fear of crime. Based on these factors and the information gathered, the researcher has two hypotheses. First, students who watch more hours of non-fiction crime shows are more fearful of crime than students who do not. Second, women who watch more hours of non-fiction crime shows are more fearful of crime compared to men.

Methods

This study used a non-probability, availability sampling design. The data was collected from 154 undergraduate and graduate students at Cabrini University. The surveys, which were approved by the institutional review board (IRB), were distributed in the spring semester of 2017 to participants during class. The majority of the students who took the survey were either freshmen or in a class provided by the Sociology and Criminology department.

Demographics

The sample (N=154) consisted of 98 (64.1%) participants who identified as female and 55 (35.9) who considered themselves male. Fifty-nine percent (n=89) of the sample was white students, 26.5% (n=40) black, 4.6% (n=7) mixed, 6.6% (n=10) Hispanic, 2.6% (n=4) Asian, and 0.7% (n=1) of students identified as “other.” Over half of the participants (52.6%, n=81) receive mostly B’s in their classes. The majority of the students (66%, n=101) were between the ages of 19 and 21. The descriptive statistics for the sample are available in Table 1 below.

Table 1. *Descriptive Statistics*

| <i>Variable</i> | <i>Frequency</i> | <i>Valid Percent</i> |
|-------------------|------------------|----------------------|
| Age | | |
| 18 | 20 | 13.1 |
| 19 | 33 | 21.6 |
| 20 | 36 | 23.5 |
| 21 | 32 | 20.9 |
| 22 | 18 | 11.8 |
| 23 | 2 | 1.3 |
| 24 | 4 | 2.6 |
| 25 | 4 | 2.6 |
| 36 | 1 | .7 |
| 39 | 1 | .7 |
| 43 | 1 | .7 |
| 57 | 1 | .7 |
| Gender | | |
| Female | 98 | 64.1 |
| Male | 55 | 35.9 |
| Race | | |
| White | 89 | 58.9 |
| Black | 40 | 26.5 |
| Mixed | 7 | 4.6 |
| Hispanic | 10 | 6.6 |
| Asian | 4 | 2.6 |
| Other | 1 | .7 |
| Grades | | |
| Mostly A’s | 53 | 34.6 |
| Mostly B’s | 81 | 52.9 |
| Mostly C’s | 17 | 11.1 |
| Mostly D’s or F’s | 2 | 1.3 |

Variables of Interest

This study consisted of two independent variables, one dependent variable, and five control variables. The first independent variable was gender of the participant. The variable was measured by having the student circle if they were male, female, or other. The distribution for gender (males= 35.9%, n=55; females=64.1%, n=98) are in Table 1. The second independent variable was the amount of crime shows consumed. A crime show was conceptualized as a television series, fiction or non-fiction, which focuses on the criminal justice system and solving crimes. The dependent variable was fear of crime, which was conceptualized as the emotion or worry people feel over the threat of victimization (Fisher, Sloan, & Cullen, 1995). The control variables consisted of age, prior victimization, grades, race, and gender.

In order to measure crime show consumption, participants were asked to circle how frequently they watched certain television series. There were nine questions for each fiction and non-fiction genre of crime show. These questions were adapted from Rader, May, and Goodrum’s study from 2007. Likert style questions were used and ranged from 1=*never* to 7=*very often*. The fiction crime program questions asked “how often do you watch each of the following crime drama programs?” The show options were the following: *Law & Order, How to Get Away with Murder, Criminal Minds, NCIS, Bones, CSI, Chicago PD, Blue Bloods*, and any other fiction crime shows. The nonfiction questions were the same only pertaining to this genre. The television options were: *COPS, The First 48, Snapped, Dateline, Forensic Files, I (Almost) Got Away with It, Cold Case Files, Making a Murderer*, and any other non-fiction crime shows. The questions were combined into two additive scales based on their respective genre. The creation of these additive scales were appropriate because Cronbach’s Alpha was greater than .07 for both variable sets. Cronbach’s alphas for the 9 fiction and 9 non-fiction items were .820 and

.864, respectively. The fiction scale ranged from 9 to 63, with 9 representing little crime show consumption and 63 representing high crime show viewing. The most frequent fiction ranking was 9 (15.7%, n=22), which means not many participants watch crime dramas. The non-fiction scale ranged from 9 to 62, with 9 representing little crime show viewing and 62 representing high crime show consumption. Nine was also the most frequent non-fiction ranking (24.8%, n=35), which means not many people in the study watch reality crime television. *Figure 1* and *Figure 2* provides the distributions for the fiction and nonfiction scales. The mean of the fiction and non-fiction scales were 21.28 and 16.77, respectively, which means that there were low-levels of crime show consumption.

Figure 1. Bar graph of the fiction scale distribution

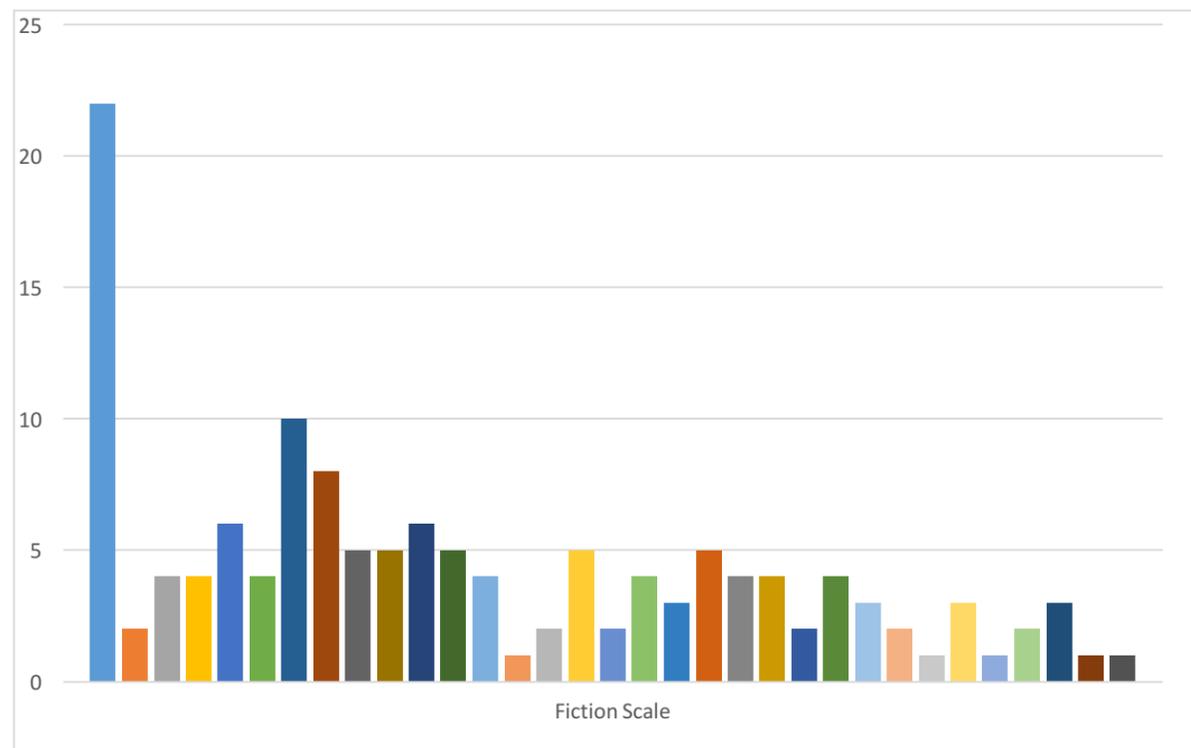
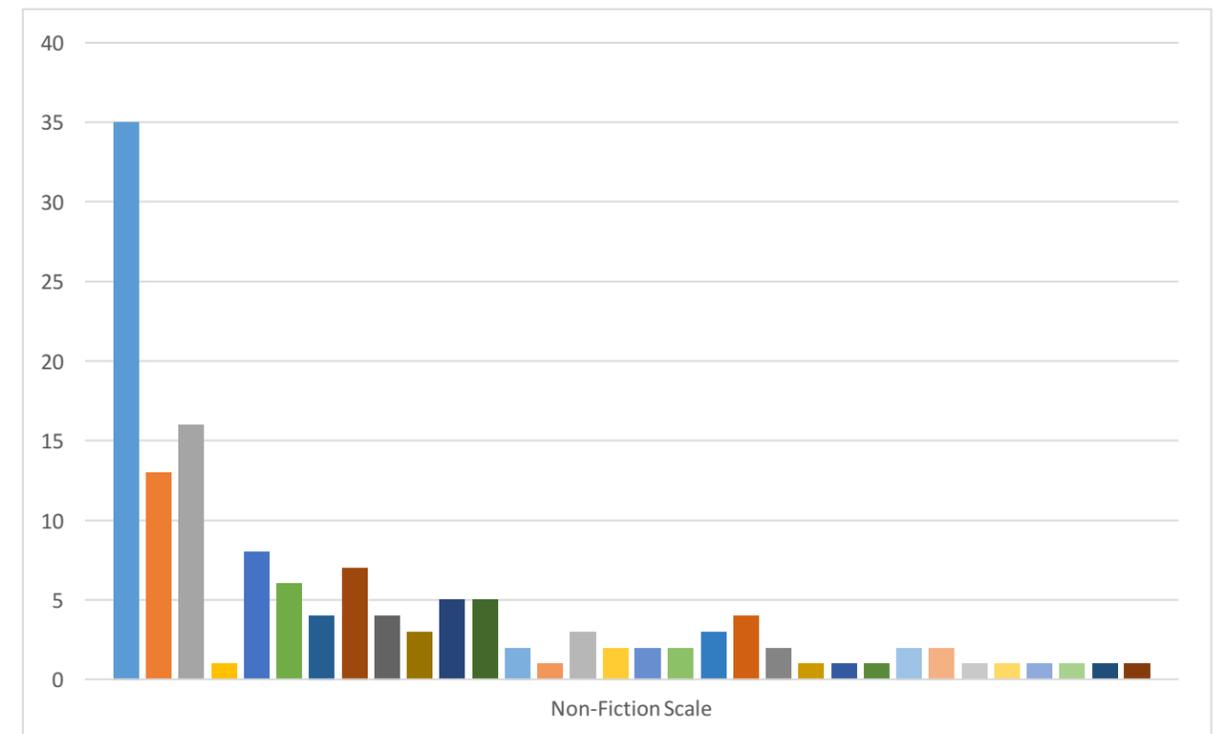
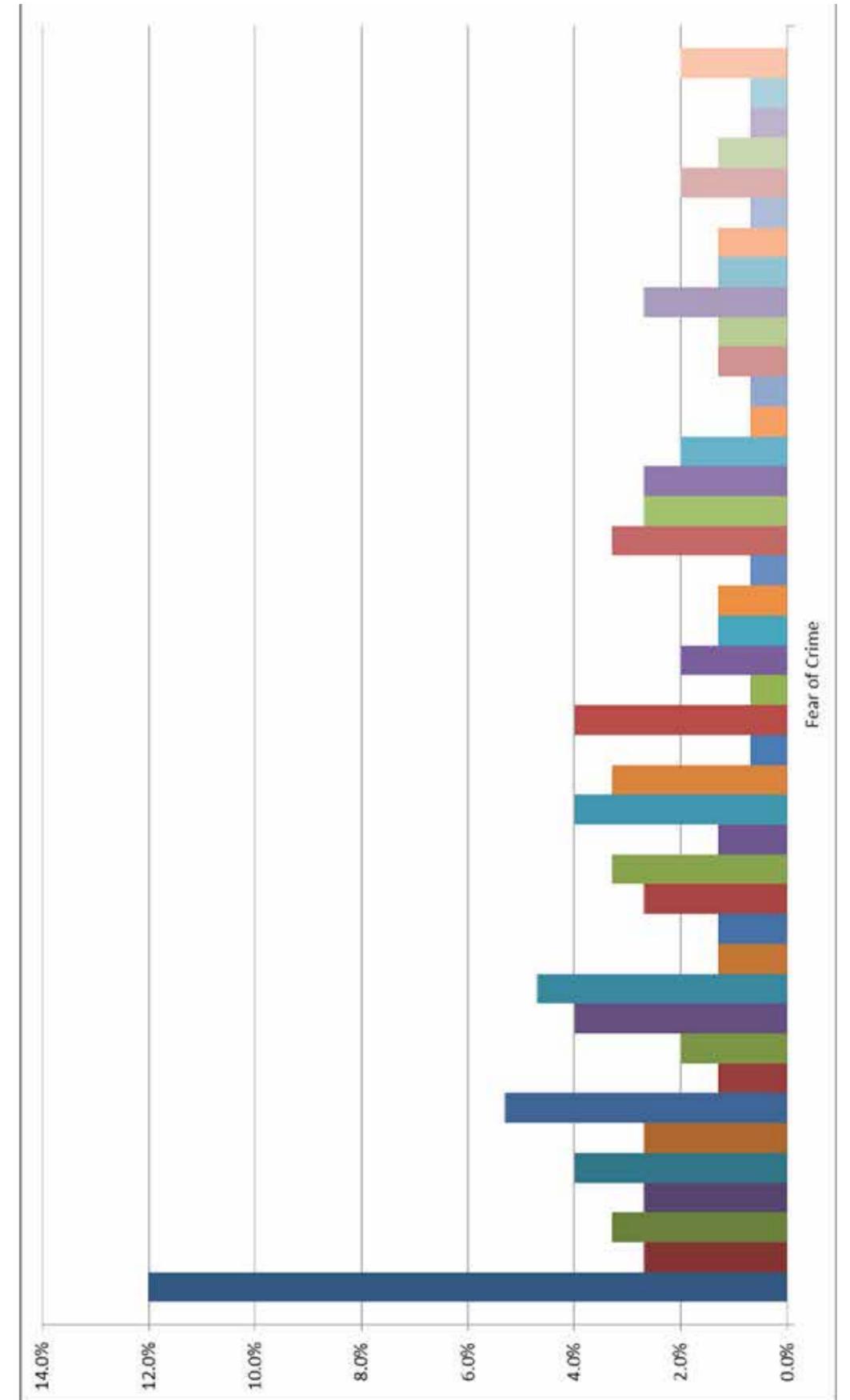


Figure 2. Bar graph of the non-fiction scale distribution



There were two questions that related to crime show consumption that had to be excluded from the analysis. The questions were: “how many days in an average week do you watch TV crime dramas?” and “How many days in an average week do you watch non-fictional programs about crime?” The response option for both of these questions was to fill in the blank. These were excluded from the data analysis for two reasons. The first reason is that there were many inconsistencies with the respondents’ answers, which the researcher would have had to interpret. Also, there was a discrepancy between these questions and the questions regarding specific show consumption. Some participants would respond that they never watch crime television, but would circle that they watched a specific show frequently. The specific crime show questions were still able to be used and they were the sole measure of crime show consumption.

The dependent variable, fear of crime, was measured by having participants circle how fearful they were of being victimized in specific ways. There were five questions that related to fear of crime and they were taken from Fisher, Sloan, and Cullen (1995). The questions were Likert style and ranged from 1=*not afraid at all* to 10=*very afraid*. The questions asked “how afraid are you that you will...” and the options were: have your property stolen without the use of force; be robbed or mugged by force; have someone attack you, beat you up, or injure you without using a weapon; be attacked and injured by someone with a weapon; and be raped or sexually assaulted. These five questions were combined into an additive scale to create the fear of crime variable. The additive scale was appropriate because Cronbach’s Alpha was greater than .07 (5 items, $\alpha=.867$). The fear of crime scale ranged from 5 to 50, with 5 indicating low fear of crime and 50 representing high fear of crime levels. The most frequent ranking of fear of crime was 5 (12%, n=18). The mean for the fear of crime scale was 20.79, which means there is a relatively low level of fear of victimization among participants. For the distribution of the fear of crime scale, see *Figure 3*.



Five control variables were used to help account for the other factors that might crime show viewing and fear of crime. The first control variable was race and this variable was recoded to be a binary variable (white and non-white) due to the small percentage of respondents identifying as a race other than black (26.5%, n=40) or white (58.9%, n=89). This question asked the respondent “what race do you identify as?” The participants had an open-ended response option for this question and the researchers then coded the races based on the responses (white=1, black=2, mixed=3, Hispanic=4, other=5, and Asian=6). These variables were then recoded into white=1 and non-white=2. The next control variable was gender, which was also binary (female=1, male=2). The survey question asked the participant to circle their gender and the options were: female, male, and other. The majority of the people who took this survey were female (64.1%, n=98). The variable of grades was ordinal and respondents were asked to circle one category that best described their overall grades on their last report card. The options were the following: mostly A’s, mostly B’s, mostly C’s, and mostly D’s or F’s. Students were most likely to have mostly B’s on their report cards (52.9%, n=81). Age was the fourth control variable. This variable was ratio and the participant’s ages ranged from 18 to 57. The question was open ended and asked “how old are you today?” The most common ages were those who were 19 (21.6%, n=33), 20 (23.5%, n=36), and 21 (20.9%, n=32). The final control variable was prior victimization. This question was binary (yes=1, no=0) and asked the participant if they had ever been a victim of crime (robbery, sexual assault, physical assault, burglary, larceny-theft, etc.). The majority of the respondents answered no to prior victimization (62.9%, n=95). Descriptive statistics for the independent variables, dependent variable, and control variables are all available below in Table 2.

Table 2. *Variable Descriptive Statistics*

| <i>Variable</i> | <i>N</i> | <i>Mean</i> | <i>Median</i> | <i>Mode</i> | <i>Range</i> | <i>Standard Deviation</i> |
|----------------------------|----------|-------------|---------------|-------------|--------------|---------------------------|
| Fear of Crime Scale | 150 | 20.79 | 18.5 | 5.00 | 45 | 12.78 |
| Fiction Scale | 140 | 21.28 | 18.5 | 9.00 | 54 | 10.99 |
| Non-fiction Scale | 141 | 16.77 | 13.00 | 9.00 | 53 | 9.82 |
| Age | 153 | 20.86 | 20.00 | 20.00 | 39 | 4.29 |
| Grades | 153 | 1.79 | 2.00 | 2.00 | 3 | 0.69 |
| Race | 151 | 1.72 | 1.00 | 1.00 | 5 | 1.15 |
| Gender | 153 | 1.36 | 1.00 | 1.00 | 1 | 0.48 |
| Prior Victimization | 151 | 0.37 | 0.00 | 0.00 | 1 | 0.48 |

Plans for Analysis

In order to analyze crime show consumption and fear of crime, and also include the control variables, a multiple regression was conducted. This form of analysis was the best test to run in order to answer the research question because of the variables’ levels of measurement. In order to run a multivariate regression, the variables need to be ratio, interval, or binary, which all of these variables were.

Results

Table 3 shows the results of the multiple regression for crime show consumption and fear of crime. Neither the non-fiction scale (*Beta*=-.044) nor the fiction scale (*Beta*=-.027) are significantly related to fear of crime. This means that watching crime shows does not significantly affect a person’s fear of crime. This goes against the first hypothesis, therefore, hypothesis 1 is not supported. However, there is significant relationship between gender (*b*=-8.878, *p*<.01) and fear of crime. If an individual is a woman, they are almost 9 times more likely

to be fearful of crime than men, which supports hypothesis 2. Gender is the best predictor of fear of crime ($Beta = -.342$). Surprisingly, grades are the second best predictor of fear of crime ($Beta = -.186$). Grades ($b = -3.431, p < .05$) are significantly related to fear of crime, which means the better a person's grades, the more fearful they are of crime. The r-squared for this model explains 7.8% of the variance between the independent variables, dependent variables, and control variables ($R^2 = .078$).

Table 3. *Multiple Regression Results for Fear of Crime*

| Coefficients | <i>b</i> | SE | Beta |
|------------------------|----------|-------|-------|
| Constant | 40.267** | 7.266 | |
| Nonfiction Crime Shows | -.063 | .162 | -.044 |
| Fiction Crime Shows | -.033 | .141 | -.027 |
| Race | -1.544 | 2.376 | -.058 |
| Victim of a Crime | .946 | 2.322 | .036 |
| Gender | -8.878** | 2.531 | -.324 |
| Grades | -3.431* | 1.689 | -.186 |
| Age | .039 | .255 | .013 |
| Model Summary | | | |
| R-Squared | | .141 | |
| Adjusted R-Squared | | .078 | |

* $p < .05$, ** $p < .01$

Discussion

This study tested the relationship between crime show consumption and the viewer's fear of crime among college students. Based on previous studies and literature, it was hypothesized that crime show consumption was related to fear of victimization. However, the results from the multiple regression did not support this hypothesis. The regression did show one of the hypotheses, gender and fear of crime, to be supported. The results also had an unexpected

significance between one of the control variables and fear of crime, which could be beneficial to further studies.

The first hypothesis stated that students who watch more hours of non-fiction crime shows are more fearful of crime than those students who do not. However, the results showed no significant relationship between either non-fiction or fiction crime shows in relation to fear of crime. These results indicate that television consumption of crime shows is not related to fear of crime. This finding goes against the majority of the literature that states watching television, specifically of the violent variety, increases a person's fear of crime (Kohm et al., 2012). However, it does support Kort-Butler and Sittner-Hartshorn's (2011) statement that human perceptions are complicated, which can create disparities among research that looks at the link between television consumption and fear of crime. There are a couple reasons as to why this hypothesis was not significant.

The first reason could be because of the low amount of data collected. The majority of the participants who took the survey indicated that they did not watch any crime shows, whether they be fiction (15.7%, $n = 22$) or non-fiction (24.8%, $n = 35$). This lack of data in regards to crime show consumption could be a reason why the finding proved not to be significant. Another explanation for these results could be the idea of desensitization. Desensitization is the weakening of emotional and or physiological reactivity to certain situations after repeatedly encountering violence (Mrug, Madan, Cook, & Wright, 2014). There have been some studies that have found violent media consumption, such as video games, television, or movies, can cause people to become desensitized to violence and other situations that may lead to fear (Anderson, Bushman, Donnerstein, Hummer, & Warburton, 2015; Fanti, Vanman, Henrich, & Avraamides, 2009; Mrug et al., 2014). One of the studies looked at college students and found

exposure to televised violence did not affect a person's emotional functioning (Mrug et al., 2014). This could explain why those who watch crime shows are not fearful of crime, because their emotional functioning is not affected. The study by Fanti et al. (2009) also found that exposure to violent media made people less sympathetic to victims. A third study concurred with Fanti et al.'s (2009) findings and explained how over time, exposure to media violence can affect an adolescent's brain and make them less desensitized and sympathetic in nature (Anderson et al., 2015). These three studies explain desensitization and how exposure to media violence can affect one's sympathy and cognitive ability. This lack of sympathy can be applied to one's own life as well because if a person does not feel sympathetic to a person in a movie or television show, then they most likely will not think of the risk factors that they themselves could be exposed to in the real world. Desensitization could be a factor in why students in this study were not fearful of crime if they watched crime shows.

The second hypothesis focused on gender and fear of crime. More specifically, it was hypothesized that females would be more fearful of crime compared to men. This hypothesis was statistically significant ($b=-8.878\%$, $p<.01$). This finding adds to the literature that says women are more fearful of crime (Hilinski, 2008; Hilinski et al., 2011; Kohm et al., 2012). A reason behind this significance could be the shadow of sexual assault hypothesis (Hilinski, 2008), and future research could be conducted to see if this theory does affect women's fear of crime levels.

One of the control variables was found to be significantly related to fear of crime as well. The variable was grades and the multiple regression showed that as grades increased, so did an individual's fear of crime. This was an unexpected result because grades were not an original variable, it was solely used as a control to make sure other aspects of demographics did not affect fear of crime. However, this finding did prove to affect fear of crime. A search of literature was

conducted after the study to see if other research had similar findings. However, there was little research to support this finding and should be looked into more to see if grades consistently impact fear of crime. A possible reason could be attributed to the location that the participant grew up. If the individual grew up in an urban area, he or she may have been exposed to more crime as opposed to someone from a rural area. Also, these locations and school funding might have impacted their educational level. This is just a possible reason and further research needs to be conducted to determine the actual cause.

Limitations, Future Research, and Implications

Some possible limitations of the study include the type of sample design, the sample itself, and the discrepancy between some of the survey questions. The first limitation is the type of sample design because availability sampling was used. Although this was the most efficient test for a college campus, these findings are difficult to generalize to a larger population. An ideal sample would be bigger and include wider age ranges and locations. The school that the sample was from is located in a suburban area, which could also relate to why fear of crime was so low because not as many people were exposed to crime. The sample itself also proved to be a limitation because the majority of the sample consisted of sociology and criminology majors, which might already have a lower fear of crime based on their field of study. One more shortcoming of the study could be the questions that were not included in the statistical analysis. The two questions pertained to how many days in a week does the participant view fiction and/or non-fiction crime shows. These questions were excluded from the analysis because of the inconsistency between these answers and the ones pertaining to specific shows. Some participants would answer that they watched no crime shows during the weeks, on average. However, in some cases those who would respond "never" would then circle crime shows that

they watch. For future research, the question should probably be a close-ended response and the directions could possibly be worded more clearly.

Future research should build upon this study. It would be interesting to see if there is a relationship between grades and fear of crime in other studies. Also, finding the reason behind this significance would be beneficial as well. Other studies should focus on fear of crime and crime show consumption because the research and literature that does exist on the subject varies. It would be beneficial to determine if there is a relationship between crime show consumption and fear of crime because it could lead to political implications. For example, if a person is more fearful of crime or has a false sense of reality based on the media, then that individual might be more likely to vote for punitive politicians (Nir & Mutz, 2007). This could also lead to researchers and community members speaking out against the discrepancies between the media and reality. On the other hand, if crime show consumption is not related to fear of crime, other implications can be drawn. This insignificance could mean that people are desensitized to the material they see, which could lead to aggressive or less empathetic human beings in society (Anderson et al., 2015; Fanti et al., 2009). Future research could help initiate change in society when it comes to media and the fear of victimization.

Conclusion

Despite the mixed support of the hypotheses, this research did find a significant relationship between gender and fear of crime. A student's grades were also found to be a possible predictor for fear of crime. This study added to research that females are more fearful of victimization while also discovering a new finding with grades that should be investigated more. Although television consumption and fear of crime were not related in this study, future research

can run similar tests to determine if this is a consistent finding and what other variables may affect a person's level of fear.

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Lo fantástico en “La noche boca arriba” y “El Sur”

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Los cuentos “El Sur” de Jorge Luis Borges y “La noche boca arriba” de Julio Cortázar son parte de la llamada narrativa fantástica y representan un nuevo estilo de narrar. Estos escritores argentinos introdujeron grandes cambios en la narrativa durante el posmodernismo y su influencia ha sido perdurable. El cuento de Jorge Luis Borges se publica en 1944 en la colección *Ficciones* y “La noche boca arriba” se publica en 1956. Estos cuentos tienen muchas semejanzas pero también se pueden apreciar marcadas diferencias. El “desdoblamiento” tanto en la forma en que se narra, como en la construcción del personaje central, es una parte integral de estos cuentos. Estas características sirven para inyectar ambigüedad y sorpresa en la lectura. Los dos cuentos cuestionan la confianza que el lector debe poner en el narrador, pero lo hacen de manera diferente. Este ensayo propone demostrar cómo se configuran los personajes principales en estos cuentos y cómo, en el montaje de estas narraciones, los dos autores proponen una nueva poética de la lectura.

Si consideramos las líneas generales de sus tramas, “El Sur” y “La noche boca arriba” son semejantes. En los dos cuentos cortos, los personajes principales sufren un accidente. En “El Sur”, Juan Dahlmann se da un golpe en la cabeza cuando sube las escaleras distraído en la lectura de una versión exclusiva de “Las mil y una noches”. En el caso de “La noche boca arriba”, el personaje central sale en motocicleta por la ciudad, pero cuando evita atropellar a una mujer de edad, pierde control de su moto y se accidenta. En los dos cuentos, los accidentes resultan en una herida en la cabeza y los personajes deben ser hospitalizados. Además, en ambos cuentos, los elementos fantásticos tienen un vínculo con las heridas y el tratamiento que reciben.

Lo fantástico en “La noche boca arriba” y “El Sur”

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Es en el hospital y bajo el efecto de medicamentos que empieza a surgir la presencia de otro mundo, ya que la percepción de la realidad de los pacientes se ve alterada por los medicamentos. Para el Dahlmann de Borges, ese mundo se transforma en un viaje hacia el mundo romántico del Sur argentino, mientras que, en el cuento de Cortázar, el personaje retrocede en el tiempo hacia el mítico mundo prehispánico.

Es importante resaltar que los dos autores se valen del artificio narrativo del “doble” para armar la estructura de sus cuentos. El “doble” es un recurso literario que hace que unos elementos del cuento tengan más de una versión. Para Borges, el uso del doble como estrategia narrativa y estructural es una forma de jugar con la idea panteísta que “...todo hombre es dos hombres...” (Alazraki 44). En ambos cuentos, el lector puede apreciar el uso del doble con referencia al personaje, el lugar donde tienen lugar los hechos, y el tiempo narrativo. Esta combinación incorpora dos secuencias narrativas que desafían y cuestionan la manera en que el lector define lo que es real y lo que es onírico. El uso del “doble” ilustra la idea que el universo no es sencillo y cuestiona las maneras en que el lector interpreta la realidad y el tiempo narrativo.

Si consideramos cómo estos autores introducen un desdoblamiento en el momento en que tiene lugar la acción, podemos ver enseguida paralelos. En el cuento de Cortázar, el escenario inicial de la acción del cuento es una gran ciudad. Con la descripción de la calle Central, los lectores pueden interpretar que se trata de la ciudad es Buenos Aires. El narrador ubica al lector al describir los edificios modernos, las lámparas en la calle, el acceso a los teléfonos públicos para llamar a una ambulancia, y los bien equipados hospitales. Pero llega un momento en que la realidad onírica se apodera de la narración y el lector es transportado al mundo de los aztecas durante la guerra florida. El escenario que se presenta es el de una selva tropical con marismas y tembladerales que no huelen bien. En este espacio, el lector penetra a un templo azteca donde

los pasillos secretos recrean la tensión anterior al sacrificio humano. Por otra parte, en el cuento de Borges, el primer escenario de la narración también es una gran ciudad. El personaje principal trabaja en una biblioteca pública y termina en el hospital después de sufrir un accidente. En este cuento, la acción se traslada también al sur de la provincia de Buenos Aires, donde la familia de Dahlmann tiene una estancia. En este cuento borgeano, la imagen del Sur se arma a partir de un contexto histórico, sugiriendo un espacio que se asemeja a la Argentina mítica de los años de la independencia, completa con una tienda general y un gaucho sentado en la esquina.

Como vemos, la manera de manipular un “doble” escenario para la acción del cuento es semejante en los dos cuentos, pero hay diferencias en cómo los dos autores usan la idea del tiempo. En “La noche boca arriba” de Cortázar queda claro que hay dos épocas, el tiempo moderno y el tiempo precolombino. Al final del cuento, sin embargo, el lector descubre que la realidad precolombina representa el presente narrativo, y que ese presente prefigura a su vez el futuro del personaje. Pero, los mundos son distintos. En “El Sur”, la bifurcación del tiempo no es tan clara para el lector. Al principio, el tiempo es específico, con descripciones como “Ocho días pasaron...” (Borges 398), pero también hay descripciones se hacen más ambiguas, como “...los días noches...” (399). Acostumbrado a seguir una secuencia sintagmática, el lector supone que todos los eventos en el Sur ocurren en un presente también, pero las descripciones del ambiente y de los personajes indican que se trata de un pasado. Por eso la subida de Dahlmann a un tren en la estación Constitución sugiere el presente narrativo, aunque el narrador aclare, “Dahlmann solía repetir que eso no es una convención y quien atraviesa esa calle entra en un mundo más antiguo y más firme” (399). La ambigüedad que inyecta el narrador en la lectura es consistente con la preferencia borgeana de cuestionar la manera en que se representa el tiempo narrativo. Cuando Dahlmann entra en un café, hay un gato que parece estar fuera del tiempo.

Este se logra cuando el narrador agrega, "...porque el hombre vive en el tiempo, en la sucesión, y el mágico animal, en la actualidad, en la eternidad del instante" (399). En su manera de imaginar el tiempo narrativo, Borges apunta a que no existe el tiempo secuencial, ya que hay muchas doctrinas que favorecen la idea del tiempo circular o un momento que incorpora todos los tiempos. En contraste con esto, el cuento de Cortázar implica que es posible que haya varias dimensiones, aunque sea raro tener acceso a más de una. El impacto del accidente con la motocicleta abre un hueco, una brecha que da apertura en el tiempo y conecta dos dimensiones. Gracias a esto, la fuerza vital del personaje logra penetrar a otra dimensión en un mismo instante.

Otro aspecto del "doble" como artificio narrativo es el desdoblamiento del ente de acción o personaje. En "La noche boca arriba", hay dos personajes. Uno es un hombre moderno ordinario que sufre un accidente en su motocicleta. Es cierto que a veces parece perderse en sus pensamientos, como antes de su accidente, pero también se puede relacionar bien con otros, como cuando él bromea con los médicos de la ambulancia. Sus bromas se basan en el accidente e indican que no le tiene miedo a la muerte. El otro actante del cuento es un indio moteca, anclado en el tiempo de los aztecas, y perseguido para ser sacrificado durante la guerra florida. El personaje es un guerrero y conoce el ambiente muy bien, pero tiene miedo de la muerte y trata de escaparla. Con los dos personajes, Cortázar ilustra cómo el hombre es producto de su ambiente. En el relato, el hombre moderno no necesita las habilidades de un moteca y su actitud frente a la muerte es diferente. Pero, el autor sugiere que estos personajes tienen una conexión que trasciende el tiempo. De esta manera se puede interpretar el deseo de Cortázar de jugar con la idea de la transmigración del alma o la fuerza vital de un individuo que lo conecta a otras dimensiones en el tiempo. También, ilustra la idea de que no importa la época o la tecnología, los seres humanos no pueden evitar la muerte (Wykes 148). En "El Sur" hay dos hombres también.

El Juan Dahlmann de Borges es urbano, alto, delgado, inexpresivo, solitario, sencillo, humilde, y falto de energía. Sin embargo, el Dahlmann que se identifica con el "Sur" de sus antepasados, es más libre y tiene una renovada energía. Tiene una energía nueva porque logra su objetivo, el de reconectarse con el pasado glorioso de la Argentina de sus antepasados y reafirmar su identidad como argentino (Saona 142).

En los dos cuentos hay eventualmente una convergencia entre las dos realidades. En "La noche boca arriba", hay una convergencia en el desenlace ya que el personaje muere víctima de un sacrificio azteca. También nos damos cuenta de que la realidad del mundo azteca, que considerábamos parte de un sueño, se impone como la realidad en el cuento. En "El Sur", Borges introduce pistas en la narración que nos hacen sospechar que la realidad que se sugiere no es confiable. Dos ejemplos centrales son la descripción del gato que sólo Dahlmann puede ver, y las semejanzas entre el empleado de la tienda y uno de los enfermeros del hospital.

El artificio narrativo de hacer converger dos realidades sirve para comunicar algo diferente en cada autor, pero en ambos casos sugiere poéticas de la lectura. El cuento de Cortázar sugiere que el lector no debe sentirse atado al tiempo secuencial, ya que como ser espiritual él puede trascender esas limitaciones. El cuento comunica la idea de que la energía vital de cada ser humano puede tener apertura a otras dimensiones. En teoría, los dos hombres del cuento son parte de un alma, sin embargo, la manera en que sus identidades se enfrentan a sus respectivos mundos es única. En la estructura y superposición de las dos realidades narrativas, Cortázar ilustra cómo realizar una lectura más enriquecedora.

Un tema central en "El Sur" es también el cuestionamiento de lo que representa la identidad. Pero el enfoque aquí es que hay en cada persona conexiones con su cultura e historia, al mismo tiempo que se sugieren otras maneras de entender cómo cada uno percibe su identidad.

Para el personaje principal, Juan Dahlmann, la historia de su familia es importante. Uno de sus abuelos era un inmigrante evangélico de Europa, y otro era un soldado que había luchado en defensa de su patria. En el cuento se puede apreciar una tensión entre los dos linajes de Dahlmann. Por un lado, su identidad como empleado público en una biblioteca e identificado con su abuelo pastor. Pero por otro, el Dahlmann que quiere volver al Sur, a la zona rural vinculada a su otro abuelo que había luchado en las guerras por la independencia. Esta realidad le atrae por ser parte de una vida romántica. Al verse herido y atrapado en el hospital, Dahlmann se resiste a una muerte corriente. Su corazón está en el Sur, y quiere morir en ese lugar, con un porcentaje de honor (Vyrdova 5). Por eso la escena que recrea su lucha a cuchillo representa el deseo de Dahlmann de reclamar para sí mismo una manera romántica de morir, como su abuelo soldado. Como otros personajes de Borges, Dahlmann quiere obtener salvación, y para él, su salvación es la vida del campesino, en el Sur (McMurray 3).

Jorge Luis Borges y Julio Cortázar quieren enseñar y desafiar a sus lectores con sus cuentos. La estructura compleja y el lenguaje ambiguo de ambos cuentos, especialmente con “El Sur”, exigen una lectura cuidadosa. Es el trabajo del lector decidir lo que ocurre. Pero, para entender “El Sur” y “La noche boca arriba”, es necesario leerlos más de una vez. Los dos cuentos tienen indicios sobre lo que es real, y lo que se podría considerar fantástico porque no siguen las reglas concretas de cómo definimos lo que es real. Esto sugiere a los lectores que es imprescindible leer textos con una mente abierta. Muchas veces, lo que es significativo en una obra no es algo fácil de desentrañar.

Con todas las semejanzas que se han indicado, es comprensible que una persona infiera que Cortázar había aprendido de Borges ya que usa muchas de sus ideas y técnicas. Pero lo que es interesante es que Cortázar no está de acuerdo con muchas de las ideas de Borges. Un

entrevistador le preguntó eso en una oportunidad, y Cortázar, “...denied any real link in subject matter between himself and his elder compatriot” pero dice que, “...he and Borges coincide in their ‘search...for a style,’” (Bell-Villada, 36). Es posible que Cortázar haya hecho un comentario válido. Hay muchas semejanzas en el estilo, técnica, y construcción de los dos cuentos, pero las interpretaciones del tema de la identidad y del tiempo son diferentes. Sin embargo, a pesar de las diferencias, los cuentos tienen el mismo objetivo – desafiarnos como lectores.

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Negative School Environments for Undocumented StudentsMaryKate Sapata

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Millions of undocumented students across our country are enrolled in public schools.

That does not mean that all students receive the same quality of education. Many children, undocumented students in particular, are at a disadvantage in school due to a variety of obstacles, especially that of feeling unwelcome in the school and community. If these students are expected to succeed, the education system must be reformed in a way that will allow every student to benefit from attending its schools. This includes all students regardless of home language, legal status, income level, or any other of the many factors placing a significant impact on a student's learning environment. Undocumented students are at a loss, condemned to failure, due to disappointing practices from the education system.

Foremost in the discussion of undocumented students in the American school system is the fact that they are frequently singled out and left to feel rejected in their community. Nancy Gonzales states that "many Mexican-origin students do not feel supported in their schools (*qtd. in* Gibson & Benjinez, 2002), their parents do not feel welcome (*qtd. in* Valdes, 1996) and teachers do not feel efficacious in fulfilling their educational mission (*qtd. in* Jussim & Eccles, 1995)" (Gonzales, 277). The current approach to educating Latino youth is ineffective and leaves every group involved feeling dissatisfied. It is equally frustrating for the students, families, and the staff at the school. There is little communication with the parents due to linguistic, cultural, or economic barriers, and the teachers have the freedom to treat the students in any way they feel appropriate. Apathy towards the needs of the undocumented population in schools is negatively impacting the students and the overall environment of the school for these students.

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Additionally, the lives of the undocumented students become further complicated when the unwelcoming atmosphere at school makes success seem out of reach. Margaret Gibson found that, "many of the migrant students experienced a sense of marginalization and silencing in various spaces on campus, both academic and social, which limited their full participation in the life of the school (*qtd. in* Gibson & Bejinez, 2002; Koyama & Gibson, 2007)" (Gibson, 690). The life of an undocumented immigrant is a complex one. Adding extra stress through a negative school environment may be eliminating one of the student's few places where they can be distracted from the troubles of life at home. High school is where a teen can both be a student and engage in social activities, but when one or both of those factors is taken away, the experience of school is changed. The student fails to engage in academics or the joys of adolescence, and begins to view school in a much more negative light, leading to low test scores, and high dropout rates.

Not only do these undocumented students feel unwanted in school, they also feel unfit for programs that could help them as well. In mentoring programs, some students may enter with hopes that they find someone to help them cope with day-to-day struggles and to provide emotional support. However, that may not be the case. Jean Rhodes writes, "[i]n some instances, minority, lower-income, or immigrant adolescents may feel tensions between their mentors' vision of success and their family values, particularly if their mentors are middle-class and white" (Rhodes, 48). These students join such programs in an attempt to find someone who can help them improve their lives, but instead they may find someone who wants them to follow a certain path, and unknowingly advises them against their traditions. Due to a lack of understanding of the mentee's culture, the mentor may cause tensions and make the student feel uncomfortable in yet another aspect of their life. Those who do not understand minority cultures,

issues, or the struggles of poverty may do more harm than good to the student if they do not receive the proper education in dealing with these issues. It is not rare for an undocumented student to be let down by a mentor if that mentor has misconceptions about the lives of the undocumented.

Schools and teachers often set the bar low for students, giving them little room for success and placing little expectation for them to succeed. Horace Hall states that "[a]lthough we are recurrently apprised of the odds stacked against African American and Latino males, we are rarely presented with studies that highlight their ability to competently move beyond the risks that they encounter" (Hall, 218). When the student fails, it is perceived to be less of a problem due to the belief that the student simply was not capable of more than what they did. They are not pushed and, as a result, they lose confidence in their own capabilities. It is not often that minority students are recognized for their success, instead, they are pitied for their failures. Setting low standards for undocumented and disadvantaged students perpetuates the belief that they are of being incapable of doing the same work as everyone else. This gives them less motivation to beat the odds, and widens the achievement gap in schools. The students are seen as more of an inconvenience because they are a group to feel sorry for instead of an equally intelligent group with a different perspective on life.

In addition, undocumented students are unwanted in schools, and the government has not been shy in stating so. Many laws and regulations have been passed to make getting an education difficult for the undocumented. For example, Ramos explains, "an effort is underway in the House of Representatives to amend the Constitution and take U.S. citizenship away from children of undocumented immigrants. The argument was that the United States did not have enough money to pay for the... educational benefits that some of these children were going to

receive" (Ramos, 63). These children were considered to be too costly to the education departments, despite these students being lawful citizens. The revision would make the legal status of these students as undocumented, furthering their complications in life instead of propelling them forward to achieve something more. This attempted revision was an obvious act of bigotry against the undocumented population, since the cost of having these students in schools would not change considering they would be costly whether they were undocumented or otherwise. The per-student cost would not suddenly decrease if these students lost citizenship, therefore taking the time to amend the Constitution for no other reason than making American citizens' lives more difficult was a blatant act of prejudice on the part of the House of Representatives.

These standing setbacks in the education of undocumented students need not be present if schools take more initiative. It was found that "Mexican American teachers are seen as less likely to neglect Mexican students" (Jones, 154). The Mexican students felt much better appreciated and included when the teacher was of their own heritage. The teacher in this situation is much more likely to understand the students, their lives, and their unique struggles. If the districts and schools were to take this information into consideration while hiring their faculty and staff, there is a possibility that the undocumented students would feel less neglect in the classroom, excel, and be given better opportunities the reasons for which they and their families moved here. This means lower drop-out rates among Latino and undocumented students, better chances at success, and a much more pleasant school environment for all students in attendance.

In summary, the education system lacks an understanding of the various challenges in the lives of thousands of undocumented students, and it has not sufficiently catered to the needs of

these students. In contrast, they have seemingly worked against these students instead of trying to make life in the United States worthwhile. Latinos are underrepresented in the staffing, and there are few efforts made to help these students due to a lack of expectation. The effect of these actions is long-term, by virtue of the fact that these students will leave school not only undocumented, but also uneducated, and without a strong sense of self due to the constant lack of support, bias and stereotyping in their classes. In order to enhance this situation, schools and districts should be held responsible for creating more diverse school environments. By hiring a more diverse staff and adopting a curriculum that gives representation to all of its students, the entire community of the school, students and staff alike, would become educated on the unique struggles of Latino and the undocumented population, defeating stereotypes and creating a more welcoming environment for these students.

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The Impact of Anxiety and Distractors on Reading Recall

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The Impact of Anxiety and Distractors on Reading Recall

Many studies have been performed in laboratory settings to demonstrate the effect that distractors have on performance (Dolcos et al., 2008; García-Pacios, et al., 2015; García-Pacios, Del Río, Maestú, & 2014; Mueller, et al., 2015), and this research has strong implications for the classroom. For example, cell phone use in the classroom may serve as a distraction. Baker, Lusk, and Neuhauser (2012) found that about half of college students find cell phone use to be generally distracting in class and believe classroom cell phone use has negative effects on the learning process. Nonetheless, other research has shown that 80% of college students use their cell phones at least one time during each class meeting and find this to be acceptable (Berry & Westfall, 2015). Cell phones are not the only source of distraction in the classroom. Research has also shown that classroom decorations, even when educationally relevant can be a distraction for students and impact their recall on information being presented by a teacher (Fisher, Godwin, & Seltman, 2014).

In studies that have examined the impact of distractions on academic performance (Dolcos, et al., 2008; García-Pacios, et al., 2015; García-Pacios, et al., 2014; Mueller et al., 2015) results have shown that distractors do in fact inhibit memory and performance. For example, Lineweaver, et al. (2012) demonstrated that students who were distracted by visual stimuli during a comprehension test did worse than a group of non-distracted students with Attention Deficit Hyperactivity Disorder (ADHD). These results suggest that distractors do in fact detract from students' learning and that the impact is severe as it can be worse than the negative impact of psychological disorders known to deter attention (e.g., ADHD).

Distractors can impact a student's ability to perform well on tasks by inducing emotional responses and anxiety that impair an individual's working memory and in turn performance

(Chuah, et al., 2010; Dolcos, et al., 2008; Dolcos & McCarthy, 2006; García-Pacios, et al., 2015; García-Pacios, et al. 2014; Meconi, Luria, & Sessa, 2014; Mueller, et al., 2015). The may by different mechanisms may be responsible for driving this effect. Distractors alone may be responsible for shifting attention. Some distractors may evoke an emotional response that leads to anxiety and therefore lower performance as anxiety lowers the capacity of working memory. In this study we examine the role of distractors on students while reading to determine the extent to which such distractors may impact reading recall. We consider the role of both anxiety and working memory to examine their potential role in this relationship.

Emotional Responses to Stimuli

A large body of research has specifically looked at how visually distracting stimuli may also be considered emotionally distracting stimuli (Dolcos, et al., 2008; García-Pacios, et al., 2015; Mueller et al., 2015). That is, different types of visual stimuli, such as pleasant, unpleasant, or neutral stimuli (e.g., smiley faces, spiders, or a pile of bricks, respectively), may shift cognitive attention depending on how emotionally arousing the stimulus is (Chuah, et al., 2010; Dolcos, et al., 2008; García-Pacios, et al., 2015; Mueller, et al., 2015). It has been argued that individuals may be more inclined to pay attention to threatening stimuli, which may make some stimuli more important to examine if they are more detrimental to performance (García-Pacios, et al., 2015; García-Pacios, et al., 2014). Researchers have also hypothesized that we easily ignore neutral distractors compared to unpleasant or pleasant distractors because neutral distractors are less emotionally arousing (Dolcos, et al., 2008; Dolcos & McCarthy, 2006; García-Pacios, et al., 2014; Mueller et al., 2015).

In studies that have examined the effects of negative stimuli, that is unpleasant or threatening distractors, researchers have often used stimuli such as an angry face or a spider to

elicit negative emotions (Dolcos, et al., 2008; García-Pacios, et al., 2015; García-Pacios, Del Río, & Maestú, 2014; Mueller, et al., 2015). Given that threatening or fearful stimuli are presented in everyday life, such as seeing spider, or interacting with an angry or aggressive person at work, these studies have the ability to shed light on how such distractors impact our attention and performance in day to day settings. If a stimulus that elicits a negative emotional response is presented while an individual is focused on a task and it causes a distraction, this can have negative impacts on their present task performance.

One of the critical mechanisms at play that leads to lower performance when distractors are presented is working memory. Working memory is a cognitive process that allows us to hold pertinent information in mind that are necessary for completing a task, problem solving, or taking a test (Hayes, Hirsch, & Matthews, 2008). The process of working memory is essential to learning; therefore, it is important to know how working memory is negatively affected through stimuli so that the process of learning is not hindered. By knowing the factors that contribute to the inhibition of working memory, there can be greater strides to prevent distractors from interfering with the learning process. Indeed, research has found that unpleasant stimuli that are connected to a negative emotion causes individuals to lose information being held in working memory (Anticevic, Repovs, & Barch, 2010; Chuahn, et. al., 2010; Dolcos, et al., 2008; Dolcos & McCarthy, 2006; Garcia-Pacios, Del Rio, & Maestu, 2014).

Reduced working memory has also been found when non-negative stimuli are presented (Hayes, et al., 2008; Stout, Shackman, Johnson, & Larson, 2015; Vytal, Arkin, Overstreet, Lieberman, & Grillion, 2016). For example, showing test takers neutral images while taking a working memory test distracted participants and caused them to produce low working memory

test scores (Stout, et al., 2015). Given this body of research, it seems clear that visual stimuli that act as distractors have negative effects on performance by impairing working memory.

Induced Anxiety, Working Memory and Performance

The role of working memory as it pertains to distractions is important as we experience many distractions when attempting to focus. Distractors can take the form of external stimuli, such as the visual stimuli used in the aforementioned studies, however internal distractors such as anxiety can also impact working memory. Recent studies have shown that anxiety can affect working memories in a negative way (Beilock, 2008; Coy, O'Brien, Tabaczynski, Northern, & Carels, 2011; Vytal, et al., 2016). Our ability to use working memory efficiently on a task is argued to be compromised by anxiety due to the fact that working memory has a limit and anxiety, serving as a distraction, pulls our attention away from a given task (Van Dillen & Koole, 2007). For example, Coy et. al., (2011) examined how the evaluation of anxiety induced by task instructions can greatly affect a person's working memory. Participants in the study were put into two groups: an anxiety inducing group and a calming group. Those who were in the anxiety group were given anxiety inducing instructions while the calming group was given instructions that were supportive of the working memory tasks that they were completing. The results of the study showed that those who were given anxiety inducing instructions, had a greater chance of anxiety interfering with working memory than those who were given calming and supportive instructions (Coy, et. al., 2011). Research has shown that a decrease in working memory has a negative impact on academic performance. Beilock (2008) examined the effect of inducing anxiety on students taking a math test that included problems specifically designed to require working memory. Students were randomly assigned to either have anxiety induced prior to the test by telling them their name and score would be posted on a public board, having their

working memory taxed by frequently interpreting them while taking the exam, or to a control group. The results showed that when frequently distracted students made more errors on the test compared to the control group, an indication that their working memory was impacted by the interruptions. Importantly, the anxiety group made just as many errors as those in the group that was frequently interrupted. This suggests that anxiety acts in the same way as a distraction in that it inhibits one's working memory from being able to hold onto the pertinent information for a task. The results of Beilock's work are supported by research that examines how external stimuli impact specific brain activity. Research has shown that while our brains are processing emotional stimuli, the capacity for executive functioning, like working memory, is hindered (Anticevic, et al., 2010; Dolcos, et al., 2008). This may help explain how anxiety, an emotional stimulus, is impacting our working memory. Overall, anxiety has been shown to be detrimental to performance by taxing working memory. This may explain why distractors, particularly those that elicit emotional responses such as anxiety, can negatively impact performance.

Clinical Anxiety, Working Memory and Performance

While some studies have shown that induced anxiety can negatively impact working memory and performance, other studies have examined the impact of clinical anxiety. When examining the effect of distraction of high-anxiety individuals, Mueller et al. (2015) suggested that people with clinical levels of anxiety may interpret all stimuli, emotional or non-emotional eliciting, as equally threatening therefore their performance may be impacted more frequently than people with lower anxiety. Research supports this idea showing that high-anxiety individuals, such as people with a mood disorder, are distracted equally by both unpleasant and neutral distractions (García-Pacios, et al., 2014; Meconi, Luria, & Sessa, 2014). This would then suggest that high-anxiety individuals would have more deficits in keeping their attention on

cognitive tasks, for example, at work or school, due to implications from both neutral *and* unpleasant distractors.

Recent studies have shown that the neural processing in those who tend to worry more than others does suffer when placed under high stress situations (Vytal, et al., 2016). Vytal et al., (2016) examined individuals diagnosed with General Anxiety Disorder (GAD), and compared their ability to work through distractions during working memory tests with those who did not have GAD. Results demonstrated that those diagnosed with GAD found it more difficult to ignore the distractions that were emotionally disturbing or startling and therefore had lower scores on the working memory tasks than the control group.

There is a large number of school-aged youth who suffer from anxiety and its interfering symptoms, which adds a level of complexity to their lives. Recent studies have shown that GAD is present between 2.4-10.8% of children and teens in the population of average communities (Benjamin, Beidas, Comer, Puliafico, & Kendall, 2011). This implies that there are several school-aged youth impacted by GAD, and may therefore be dealing with complications such as difficulty in school processes that engage working memory.

The Current Study

Past research has shown that there is a negative impact of distractors on working memory. Fewer studies, however, have examined the practical implications of this on students' academic performance. For example, what impact do distractors have on recall of information that students read? If a student is distracted while reading, say by text messages or advertisements if reading online material, what impact do those distractors have on their recall of the information? In this study we aimed at understanding the way that distractions impact students' reading recall. We chose to distract participants during a reading recall activity rather

than a working memory test as this allowed us to examine the impact of distractors in a practical way. Therefore, we displayed distractors during the comprehension recall task to mimic the way that students may be distracted not while taking a test but while learning information through reading. As reviewed above, the emotional responses that can be derived from various stimuli can lead to different effects in people's working memory, which ultimately impacts their performance. Further, this may differ for students with different levels of anxiety. As such, we systematically tested the effect that such distractors would have on the students' recall performance of academic material and examined the potential effects that students' working memory and general anxiety may have on their recall performance.

Methods

Participants

Undergraduate students ($N=69$) enrolled in psychology courses participated in the study. The students earned credit in their psychology course for participating in the study. An alternative assignment was made available for any student wishing not to participate, however all students chose to participate in the research.

Measures & Procedure

Participants completed the research in a classroom equipped with computers. All tests and surveys were completed on a computer. A demographics survey was completed in addition to a survey that asked participants how unpleasant/pleasant 10 images were, from 1 (extremely unpleasant) to 5 (extremely pleasant), in order to validate the ability of the images to elicit positive and negative emotional responses.

The General Anxiety Disorder scale (GAD-7; Spitzer, Kurt, Williams, & Löwe, 2006; $\alpha = .92$) was included to measure students' anxiety symptoms. The GAD included seven

questions about how often the participants' anxiety symptoms occurred within the last 2 weeks rated on a scale of 0 (not at all) to 3 (nearly everyday). Scores on the GAD are categorized as low anxiety (0.00-5.50), moderate anxiety (5.51-10.00) and severe anxiety (10.10-15.00; Spitzer, et al., 2006).

An n-back working memory test was completed by all participants. Participants viewed a series of letters (e.g., A, Q, X) presented on a computer screen. Each letter was presented for 2000 milliseconds with a 500 millisecond inter-trial interval. Participants were instructed to press the "p" key when the letter on the screen matched the letter presented two screens prior, and to press the "q" key for all other instances. The n-back test has been shown to be a valid tool for assessing working memory (Kane, 2007).

A reading comprehension passage, derived from a psychology textbook, *Psychology: The Brain, the Person, the World* (Kosslyn & Roseberg, 2001), was presented for participants to read from the computer screen. After reading the passage, participants were required to answer seven multiple-choice questions about the passage. The participants were scored on their accuracy (0-100%) to the seven questions and this served as their reading comprehension recall score. The recall was completed while five positive (i.e., smiley face; positive condition), five negative (i.e., spider; negative condition), or no images (control condition) were displayed. Images flashed on the screen for 1000 milliseconds at random intervals while the participants were reading the passage. No distractors were presented while answering the questions.

Data Analysis

Given the concerns raised with null hypothesis significance testing (NHST; Cumming 2008; 2014), we interpreted the data collected in this study using effect sizes and confidence intervals. As demonstrated by Cumming (2014) *p*-values are unreliable such that they fail to

reveal what the population data look like. For example, in his simulation, Cumming (2014) conducted multiple *t*-tests between a control group and experimental group using multiple sets of sample data generated from population data with known parameters. The results showed that the *p*-values from the comparisons ranged from below .001 to above .90. This demonstrates that relying on *p*-values from one study does not necessarily tell you anything about the true underlying difference between two groups as the *p*-values can range dramatically. On the other hand, the confidence intervals generated across the multiple experiments were much more consistent in describing the true difference between the populations. While we report *p*-values in the footnotes of our tables, we did not rely on *p*-values to make conclusions about the impact of distractors and anxiety on student recall, rather we focused on effect sizes and confidence intervals.

Results

The data from students were separated into three groups based on their GAD scores (low, moderate and severe; Spitzer, et al., 2006). To ensure that students' ratings of the various stimuli were not biased based on general anxiety, we compared the rating of negative and positive stimuli between the three anxiety groups. For negative images, students in all three anxiety groups had similar average ratings between $M=1.51(.63)$ and $M=1.93(.81)$ on a scale of 1 to 5 with 1 indicating extremely unpleasant. For positive images, average ratings were also similar to each other ranging between $M=4.32(.74)$ and $M=4.72(.76)$ for all three groups. No statistically significant differences between GID groups for any stimuli were observed after, $ps=.008-.945$, using a Bonferroni adjusted $\alpha=.005$. Together, we took this as evidence of the images' ability to elicit the appropriate negative or positive emotional responses and to not differ based on the students' scores on the GAD-7.

Anxiety did not appear to have an effect on working memory scores or reading recall scores overall. The average scores for working memory were similar for all three anxiety groups ranging from $M= 0.19(0.11)$ to $M= 0.21(0.08)$ with considerable overlap in confidence intervals and small effect sizes (see Table 2). Similarly, reading recall scores overall ranged from $M= 0.30(0.27)$ to $M= 0.36(0.27)$ for all three GAD groups (see Table 2) and had overlap in confidence intervals and small effect sizes. Further there was no correlation between working memory and recall.

Table 2.

Working Memory and Recall Scores by GAD Groups

| GAD Group | Working Memory | | | Recall Scores | | |
|-----------|----------------|--------------|--------------|---------------|--------------|--------------|
| | $M(SD)$ | Lower 95% CI | Upper 95% CI | $M(SD)$ | Lower 95% CI | Upper 95% CI |
| Low | 0.19 (.11) | 0.134 | 0.251 | 0.33 (.28) | 0.18 | 0.48 |
| Moderate | 0.19 (.10) | 0.156 | 0.236 | 0.36 (.27) | 0.26 | 0.47 |
| Severe | 0.21 (.08) | 0.175 | 0.256 | 0.30 (.27) | 0.20 | 0.41 |

Note: CI=Confidence Interval; Results from a one-way ANOVA showed small effects $\eta^2_s \leq .01$ and no statistically significant differences between GID groups for both working memory and reading comprehension scores, $ps \geq .72$.

To test if distractors impacted students' performance on the reading recall differently across the three distractor conditions, we conducted a one-way ANOVA (see Table 3). Results demonstrated that distractors being present during the recall will almost always lead to a lower test score as illustrated by the confidence intervals of the difference between the control group reading scores and the two distractor groups: positive condition reading scores vs. control $M_{\text{difference}} = -.11 [-.30, .08]$; negative condition recall scores vs. control $M_{\text{difference}} = -.12 [-.33, .07]$. In this study, students had lower recall scores when they were distracted compared to the control

group, and the confidence intervals in Figure 1 show that the differences will exist for most samples of students. The type of distractor did not lead to differences in recall. As shown in Figure 1, lower recall score when experiencing negative distractors are just as likely to occur as when experiencing positive distractors. The difference between the positive and negative conditions was close to zero, $M=.01$, and had a confidence interval that ranged almost equally negative as it was positive $[-.18, .15]$. This means that both positive and negative distractors will be similar in their ability to affect students' performance.

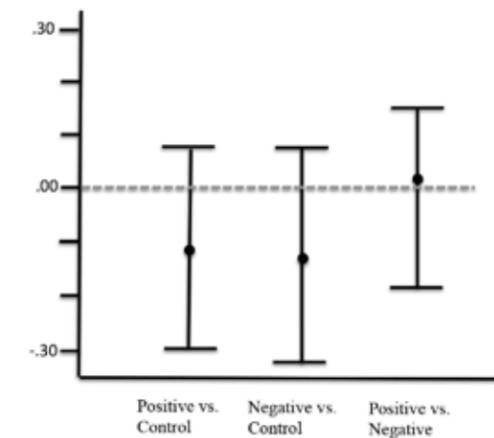


Figure 1. Error Bars for Differences in Recall Scores between Distractor Groups. Students in the positive and negative distractor groups are likely to have lower recall scores compared to the control group. However, negative and positive distractors have an average difference in scores of $M=.01$ with suggesting that positive conditions will perform better a similar amount of the time as negative conditions.

As aforementioned, the recall scores were similar across all three GAD groups. However, upon closer examination of the descriptive statistics, we found that the differences in recall scores may differ between stimuli based on GAD group. For the negative stimuli condition, all three anxiety groups had similar recall scores (see Table 3). However, for positive stimuli, follow up t -tests showed that the moderate anxiety group had a higher recall score compared to the low anxiety ($M=.53$ vs. $.37$, $d=-0.49$) and high anxiety ($M=.53$ vs. $.37$, $d=-0.48$) groups. We interpreted this to mean that positive distractions during a recall test could benefit students with

moderate levels of anxiety. However, for students with severe anxiety or no anxiety, distractors, whether they are positive or negative had consistent negative impacts as illustrated by their recall scores being lower than the control group's (Cohen's *ds* ranging from .46 - .75). These results have important implications, but should be interpreted with caution given the samples sizes here (see Table 3).

Table 3.
Descriptive Statistics for Recall Scores by Distractor and Anxiety Conditions

| | | 95% CI | | | |
|------------------|--|--------|----------|-------|-------|
| | | N | M(SD) | Lower | Upper |
| Control | | | | | |
| Total | | 18 | .53(.33) | .38 | .69 |
| Low Anxiety | | 8 | .60(.35) | -0.32 | 0.45 |
| Moderate Anxiety | | 6 | .44(.31) | 0.08 | 0.57 |
| Severe Anxiety | | 4 | .54(.33) | -0.02 | 0.36 |
| Positive | | | | | |
| Total | | 30 | .42(.30) | .31 | .54 |
| Low Anxiety | | 11 | .37(.25) | -0.14 | 0.64 |
| Moderate Anxiety | | 11 | .53(.38) | -0.15 | 0.40 |
| Severe Anxiety | | 8 | .37(.25) | 0.11 | 0.59 |
| Negative | | | | | |

| | | | | |
|------------------|----|----------|------|------|
| Total | 21 | .41(.27) | .27 | .55 |
| Low Anxiety | 8 | .42(.30) | 0.24 | 0.66 |
| Moderate Anxiety | 8 | .41(.27) | 0.07 | 0.62 |
| Severe Anxiety | 5 | .40(.27) | 0.14 | 0.49 |

Note: CI=Confidence Interval; Two-way ANOVA results showed small effects and non-significant for the interaction, $p=.56$, $\eta^2=.07$, the main effect of stimuli, $p=.17$, $\eta^2=.075$, and the main effect of GAD group, $p=.84$, $\eta^2=.005$.

Discussion

In this study we sought to understand the role of distractors on an academic recall task. While other studies have examined the role of distractors on working memory and performance in the lab, we attempted to create a more realistic experience for students by giving them an assignment based on content in their major and mimicking the study and recall process that students experience in school. Given past research, we expected that negative distractors would inhibit performance in some way but were unclear whether the distracting stimuli that elicit positive emotional responses would have similar effects given that the findings of past studies are mixed with respect to this. We also examined the role that anxiety played in the effect that distracting stimuli had on students' recall.

We found that when students are distracted while reading material they were less likely to recall the information correctly later. There was no distinction between positive and negative distractors. That is, students did just as poorly during recall when they were distracted by a positive stimuli (i.e., smiley face) as when they were distracted by negative stimuli (i.e., spider). When considering the differences in the effect of distractors on recall across groups with varying anxiety levels, we found that students with low or high anxiety were equally affected by both

positive and negative images. However, students with moderate levels of anxiety performed better when positive images were presented compared to those with low and high anxiety. This seems to suggest that presenting students who have moderate amounts of anxiety with positive stimuli are beneficial for them. This advantage was not observed for students with low or high anxiety.

The Role of General Anxiety

The fact that moderate anxiety students responded different to positive stimuli may help to explain why results in past studies have been mixed with respect to the effect of positive stimuli. It may be that the level of anxiety a student is experiencing in day-to-day life plays an integral role in how positive stimuli will affect them and this could be confounding data when groups are not separated by anxiety levels. Why would students with moderate anxiety do better on recall when positive images are presented during their reading? One way to understand this phenomenon is to compare the experiences that low, moderate and high anxiety students have when distractors are presented. For students with moderate levels of anxiety, positive distractors may relieve some of their anxiety. By doing so, they would have more working memory available to them to complete their tasks and this would allow them to perform better. In contrast to this, research has shown that students suffering from high levels of anxiety are equally likely to suffer from positive and negative distractors. Given that anxiety limits the amount of working memory available for a task (Vytal, et al., 2016) it is not surprising that distractors of any type can be detrimental as there is limited working memory available given the tax that anxiety is already having on their working memory. For students with low anxiety, because they are not suffering from anxiety in the first place, the ability of a positive stimulus to

reduce anxiety and thereby free of working memory is not an option. Instead, the positive stimuli are received simply as a distractor.

These findings could have important implications for the classroom and are important given the high numbers of children experiencing GAD. Instructors should be aware that students with moderate amounts of anxiety could be helped by presenting positive stimuli periodically throughout class. This has been supported in the literature with studies that have infused humor throughout their classes (Neumann, Hood, & Neumann, 2009). However, instructors should plan to implement methods of introducing positive stimuli to ensure that the stimuli are not presented in such a way that it will be distracting to low and high anxiety students. Most importantly, researchers should attempt to replicate the findings in this study of the interaction between anxiety and distractors as the sample size was small. The effect sizes observed ($d_s = -.48-.49$) justify future research, but until data from larger samples is available, instructors should be cautious about extrapolating these findings into practice.

Implications and Future Directions

Overall, the results of this study suggest that distractors are detrimental to students' later recall with the only exception being when students with moderate anxiety are presented with positive stimuli. The effect that stimuli had on students recall is consistent with past research. Overall, students had lower recall when they were distracted by stimuli during their reading. This is important information for teachers to share with students. Students should ensure that they are studying in locations where stimuli are limited. In the age of smartphones and electronic communication, students should take advantage of the ability to turn off notifications, or even "unplug" completely while studying. We would argue that other background stimuli, such as television and online advertisements, should be completely eliminated. At the very least

students should consider what background noises could actively distract their attention and eliminate those stimuli. Future research could look to examine the effects of background stimuli to determine how and when they can become distractors. For example, is television automatically a distractor? If streaming music online and commercials play, do they become a distractor, but not the music? Future studies are needed to tease apart the effects of these real life stimuli on students' academic performance.

In the classroom, instructors should ensure that distractors are limited. As aforementioned, studies have shown that even classroom decor can be a distraction for young children. This phenomenon has not been tested with older populations; however, our research suggests that distractions can be detrimental to a college aged population. We tested the effect of distractors on the reading-recall cycle. Do distractors have the same negative impact on other academic tasks such as active projects and homework assignments? Future studies could alter the activity students are completing while distractors are being presented to examine the extent to which the findings here generalize.

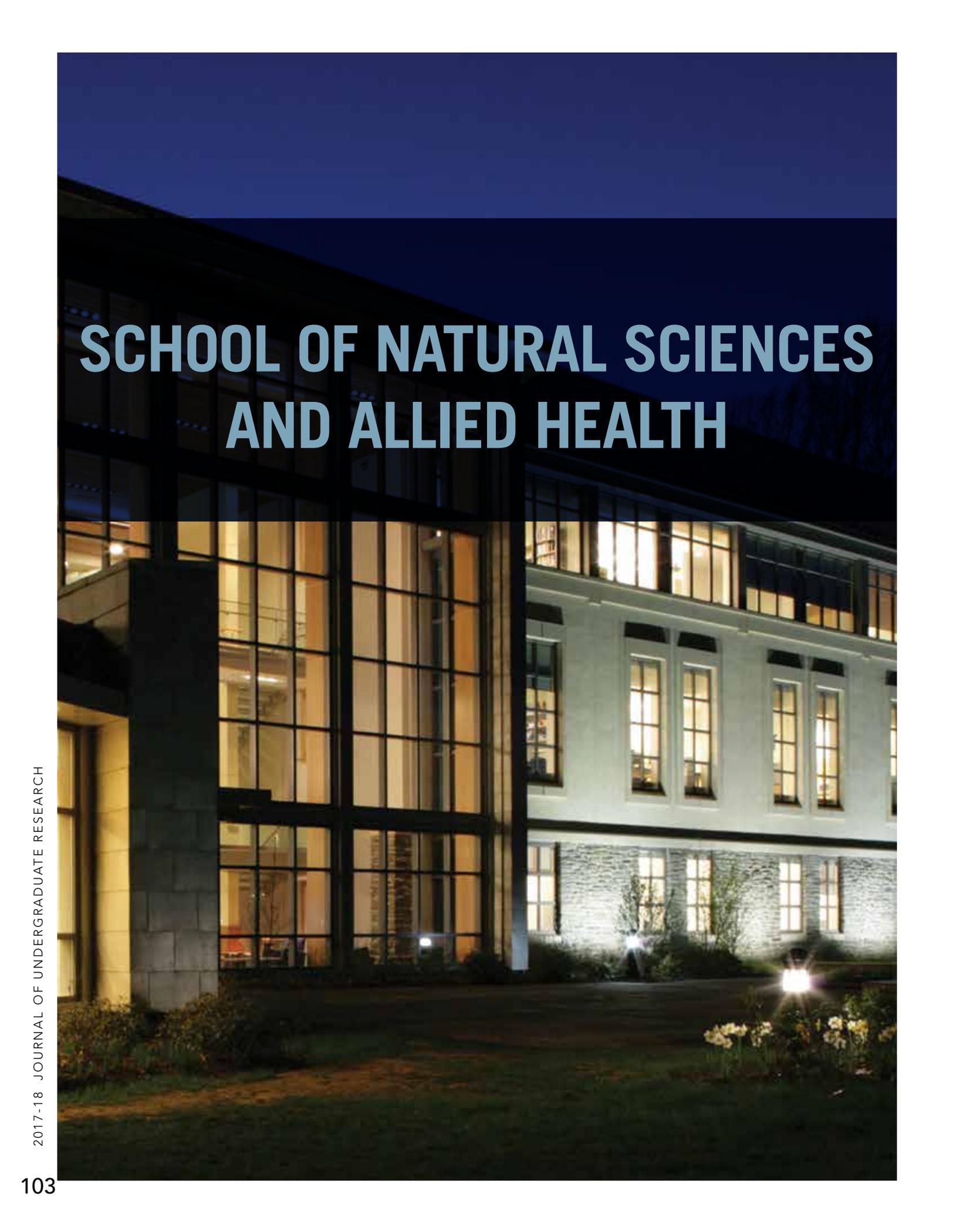
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SCHOOL OF NATURAL SCIENCES AND ALLIED HEALTH

Using Flow Cytometry for the Analysis of Macroautophagy in the Earthworm *Eisenia hortensis*
in Response to Polycyclic Aromatic Hydrocarbons for use as a Biomarker for Ecotoxicologists

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Abstract

The purpose of this research was to determine if macroautophagy in *Eisenia hortensis* could serve as a bioindicator of exposure to polycyclic aromatic hydrocarbons (PAHs) that are important pollutants that accumulate in the soil. Macroautophagy is a process in which the lysosomal machinery of the cell degrades the cellular components and is used for the removal of organelles, often induced by nutrient starvation. In our study, this cellular process was detected using a flow cytometric method permitting the measurement and analysis of individual cells in different stages of macroautophagy. Flow cytometry examines the size, granularity and fluorescence of cells as they are passed through a laser in a single cell suspension so that the laser excites one cell at a time. Macroautophagy was detected by using CYTO-ID green detection reagent which has specificity for the cytosolic double-membrane structures that form in cells undergoing macroautophagy. *In vitro* assays were conducted using earthworm coelomocytes (leukocytes) to measure the levels of macroautophagy under a range of conditions and concentrations of PAHs. Because our results did not show inter-assay reproducibility, we conclude that macroautophagy in *E. hortensis* is not a reliable bioindicator of exposure to PAHs and would therefore not be advised for use in ecotoxicological studies.

Introduction

Macroautophagy is a process where cellular components are degraded using the lysosomal machinery of the cell. In this process, an isolation membrane forms around the targeted cellular components, that closes to form a characteristic double-membrane structure known as an autophagosome. The autophagosome is then combined with the lysosomes of the

cell and the lysosomal enzymes degrade the targeted components. Once the components are broken down, the resulting macromolecules are released back into the cytosol (Kundu & Thompson, 2005).

This study examined the effects of polycyclic aromatic hydrocarbons (PAHs) on the process of macroautophagy in the coelomocytes of *Eisenia hortensis*. *E. hortensis* is a species of earthworm which extrudes its coelomocytes through its dorsal pores. These cells are leukocyte-like cells which function in the innate immune system of the earthworm and exhibit phagocytic, natural killer-like, and lytic activity (Fuller-Espie, Goodfield, Hill, Grant, & DeRogatis, 2008). In the past, *E. hortensis* has been used as a bio-indicator species to monitor ecosystem health, prevent species extinction due to pollutants, and promote ecosystem sustainability (Moore, Allen, McVeigh, & Shaw, 2006). Earthworms live in close contact with the soil and are, therefore, greatly impacted by the accumulation of contaminants in the soil. These contaminants then subsequently affect the terrestrial food chain, as earthworms represent a significant part of the diets of many vertebrate species. One type of contaminant which poses a threat to this food chain is PAHs (Hoogergrugge, Stolker, Barendregt, & Hogendoorn, 2003).

PAHs are organic compounds, containing at least two benzene rings, which are formed through the incomplete combustion and carbonization of petroleum products, and are widespread soil contaminants (Stogiannidis & Laane, 2015). These compounds are persistent due to their affinity for soil colloids, hydrophobicity and resistance to chemical, physical, and biological degradation. PAHs have also been identified as priority pollutants by the United States Environmental Protection Agency due to their neurological and toxicological health effects (Kumar, Verma, Kumar, & Sharma, 2014). In this study, the effects of two different PAHs, 7,12-dimethylbenz[a]anthracene (DMBA) and pyrene, were examined. Both DMBA and pyrene have

been used in the past to examine the use of *Eisenia fetida* as a biomarker for the presence of the PAHs (Denis et al., 1999; Fuller-Espie, Bearoff, & Minutillo, 2011). The study presented here, however, was the first to analyze the effects of PAHs on macroautophagy in *E. hortensis* using flow cytometry. It was hypothesized that coelomocytes would exhibit higher levels of macroautophagy when treated *in vitro* with PAHs, therefore indicating that macroautophagy in *E. hortensis* would hold potential as a bioindicator of PAHs in the soil.

Flow cytometry is a process, which measures a cell's size, granularity, and fluorescence using an optical-to-electronic coupling system. In this process, cells are passed through a laser in a single cell suspension. When they pass through the laser, the cells scatter the light and this scatter is detected using various photomultiplier tubes and photodiodes. The forward scatter indicates the size of the cell and the side scatter indicates its granularity, or internal complexity. The FACSCalibur flow cytometer that was used in this study is equipped with a single argon laser which detects fluorescence using one of three photomultiplier tubes, FL1, FL2, or FL3. This data is converted to electronic signals and subsequently analyzed using computer software (Becton & Dickinson, 2002). Using CYTO-ID green detection reagent, which has specificity for the cytosolic double-membrane structures (autophagosomes) that form in cells undergoing macroautophagy, flow cytometry provides a sensitive technique for measuring this cellular process.

Materials and Methods

Harvesting Coelomocytes

The night prior to beginning the extrusion process, the earthworms were taken from their habitats in groups of 6-8 and placed in Petri dishes containing paper towel pieces which had been

sprayed with fungizone (250µg/mL). Fungizone, or amphotericin B, is an antifungal agent which was used to reduce fungal contamination arising from the body surface of the animal during the *in vitro* incubation of the extruded coelomocytes. The overnight period excluded food and promoted the expulsion of gut contents, further reducing microbial contaminants during the extrusion process. After incubating in the Petri dish overnight, the earthworms were transferred into sterile plastic troughs containing 3mL of FACSFlow (BD Biosciences), which stimulated the earthworms to extrude coelomocytes (leukocytes) from the coelomic cavity through the dorsal pores. The solution containing the cells was then transferred into conical tubes and the cells were subsequently exposed to 0.5 mL of Accumax™ for 2 minutes. Accumax™ is an enzyme mixture with trypsin-like activity used to inhibit cellular aggregation. 5mL of phosphate-buffered saline (PBS) was then added to dilute the enzymes and the solution was centrifuged at 800 RPM (150g) at 4°C for 5 minutes. The pelleted cells were subsequently resuspended in 1mL of BD-BaculoGold medium (BD-BG, BD Biosciences) and enumerated using a hemocytometer. Samples were then pooled if necessary and concentrations were adjusted to 2×10^6 cells /mL.

PAH Treatment and Flow Cytometry

After extrusion, cells were loaded into a 96-well plate with their respective treatments. Cells were either treated with DMBA, pyrene, or a combination treatment of rapamycin and chloroquine. Rapamycin and chloroquine, provided by the CYTO-ID® Autophagy Detection Kit, served as positive controls. Vehicles (the solvents used to dissolve the PAHs) were included as baseline controls for comparison. Once treatments were loaded, the cells were incubated in a 5% CO₂ incubator for the appropriate time and temperature. All treatments were performed in triplicates.

After incubation, the well plates were centrifuged at 800 RPM for 5 minutes at 4°C. The pellets were resuspended in 200µL of 1X assay buffer, provided by the CYTO-ID® Autophagy Detection Kit, and the plate was centrifuged again under the same conditions. The cells were then resuspended in 100µL of 1X assay buffer, while working the wells, and then each well was transferred into flow cytometry tubes, labeled in advance. Once transferred, 100µL of CYTO-ID® 1X dye was added to each tube. The tubes were then mixed using a vortex and incubated on a shaking platform for 30 minutes at 25°C in the dark. The tubes were then centrifuged and resuspended in 200µL of 1X assay buffer twice. After being resuspended, 100µL of 7-amino actinomycin D (7-AAD) viability dye was added to any appropriate tubes, which were then incubated for 5 minutes at room temperature in the dark. Samples were then run on the FACSCalibur flow cytometer (BD Biosciences) and data was collected using Cell Quest Pro™ software. Data was analyzed in Excel and a student's t-test paired two sample for means was conducted to determine any statistical significance. Each treatment was compared to baseline values, given by vehicle-treated cells. Vehicle-treated cells received dimethyl-sulfoxide (DMSO), which was used to dissolve the PAHs, and water.

Cells were gated using a 2D dot plot which graphed cells based on their forward and side scatter properties (Figure 1A). An elliptical region was drawn around the desired cell population. Coelomocytes include three subpopulations of cells, chloragocytes, hyaline amoebocytes, and granular amoebocytes. Gating was used to exclude chloragocytes, due to their high levels of autofluorescence which would skew the data. The relative fluorescence intensity of the cells within the region, the amoebocytes, was then measured using a histogram which graphed cells based on their degree of fluorescence detected by FL1 versus cell number (Figure 1B). The voltage for FL1 was adjusted based on the levels of autofluorescence for samples, obtained by

the analysis of cells which did not receive CYTO-ID dye, so that autofluorescence fell primarily within the first decade of the histogram on a log scale. A marker region was then set beginning at the first decade and extending to the right to the end of the log scale. Any fluorescence of treated cells occurring within this marker region was considered indicative of macroautophagy. The percent of cells fluorescing in this region was recorded for each sample and the average for each triplicate was calculated. These averages were graphed and the student's t-test was used to compare the percent of fluorescence in the marker region between PAH- versus vehicle-treated samples.

Results

A total of nine separate assays were conducted. A summary of the results is shown in Table 1. With the exception of assay 6, coelomocytes were incubated at 25°C. In assay 1, DMBA was used at three concentrations starting at 100µM with a 10x serial dilution, making for three concentrations of 100, 10, and 1µM. Pyrene was used at three concentrations starting with 400µM with a 4x serial dilution, making for 400, 10, and 2.5µM. Rapamycin and chloroquine (known to induce macroautophagy in mammalian cells) were used at concentrations of 500nM and 180µM, respectively, and cells were incubated for 24 hours at 25°C. All PAH concentrations in this assay resulted in significantly elevated levels of macroautophagy. Assay 2 was an attempt to replicate these results and the serial dilutions for both DMBA and pyrene were extended to include a fourth, ultra-low concentration. The concentrations of rapamycin and chloroquine were also adjusted to 2µM and 60µM, respectively, to attempt to stimulate the cells to respond. Assay 2 revealed no significant results.

The same batch of earthworm coelomocytes were utilized for both assays 3 and 4. In assay 3, DMBA and pyrene concentrations were both increased to 250µM with 10x serial dilutions and chloroquine was increased to 180µM. Separate vehicles were used for rapamycin/chloroquine treatment and PAH treatment and cells were incubated under the same conditions as assays 1 and 2 for 24 hours. Cells in assay 4 received the same treatment, however, they were incubated for 48 hours instead of 24 hours. Significant results were only found in assay 3 at a DMBA concentration of 25µM. In addition to testing for macroautophagy, 7-AAD was used for a viability test to examine levels of cell death. This test revealed that the vehicle containing 0.4% DMSO was causing higher levels of cell death compared to media-only treated cells and that a longer incubation also resulted in higher cell death (Figure 2). For this reason, 48-hour incubation was no longer used.

Assays 5 and 6 also examined the same batch of earthworm coelomocytes. In assay 5, cells were incubated at 25°C and in assay 6 they were incubated at 29°C. The concentrations of DMBA and pyrene were lowered to 25µM and 125µM, respectively, with 5x serial dilutions. Due to a lack of significant results, rapamycin and chloroquine were not included from this point on. Significant results were found for all DMBA concentrations and for pyrene at 5µM in assay 5 and no significant results were found for assay 6. In assays 5 and 6, 7-AAD was used for another viability test which revealed higher levels of cell death in those incubated at 29°C so this temperature was no longer used. In assay 7, the concentration of pyrene was increased to 500µM with a 5x serial dilution. Significant results were only found for DMBA at 25 and 5µM.

For assays 8 and 9, coelomocytes from individual earthworms were treated instead of pooling samples from multiple earthworms due to the high levels of variation in baseline fluorescence (Figure 3). Pyrene was also no longer used due to a lack of significant results in

previous assays. In assay 8, DMBA was used at two concentrations, 25 and 5 μ M. Significant results were only found in one of the eight earthworms tested with DMBA used at 25 μ M. For the final assay, the DMBA concentration was increased to 100 μ M and it was the only concentration that was used. None of the coelomocyte samples tested in assay 9 gave significant results.

Discussion

The results in this study do not support the hypothesis that macroautophagy in *E. hortensis* is a reliable bioindicator of PAH exposure of coelomocytes in vitro. Significant results were seen in several of the assays, however, these results were not reproducible. Throughout the study, high levels of variation between assays as well as individual earthworms were observed. Based on these results, it is not recommended that the method described in this study to measure levels of macroautophagy be used by ecotoxicologists as a bioindicator of PAH contaminated soil. Future studies, however, could examine the use of alternative methods for measuring macroautophagy. The kit used in this study, including the positive controls and autophagy detection dye, was purposed for use in vertebrate organisms. Other methods of detection more suited for an invertebrate model, including *E. hortensis*, could be explored to reach a more definite conclusion.

Acknowledgements

We would like to acknowledge the Romano Scholarship Program of Cabrini University, the Pennsylvania Academy of Science, and the Cabrini Science Department for providing the funds and means to perform this research. We would also like to acknowledge Dr. Sheryl Fuller-Espie

who acted as our mentor throughout our research. Dr. Sheryl Fuller-Espie provided both her guidance and support making it possible for us to achieve our research goals.

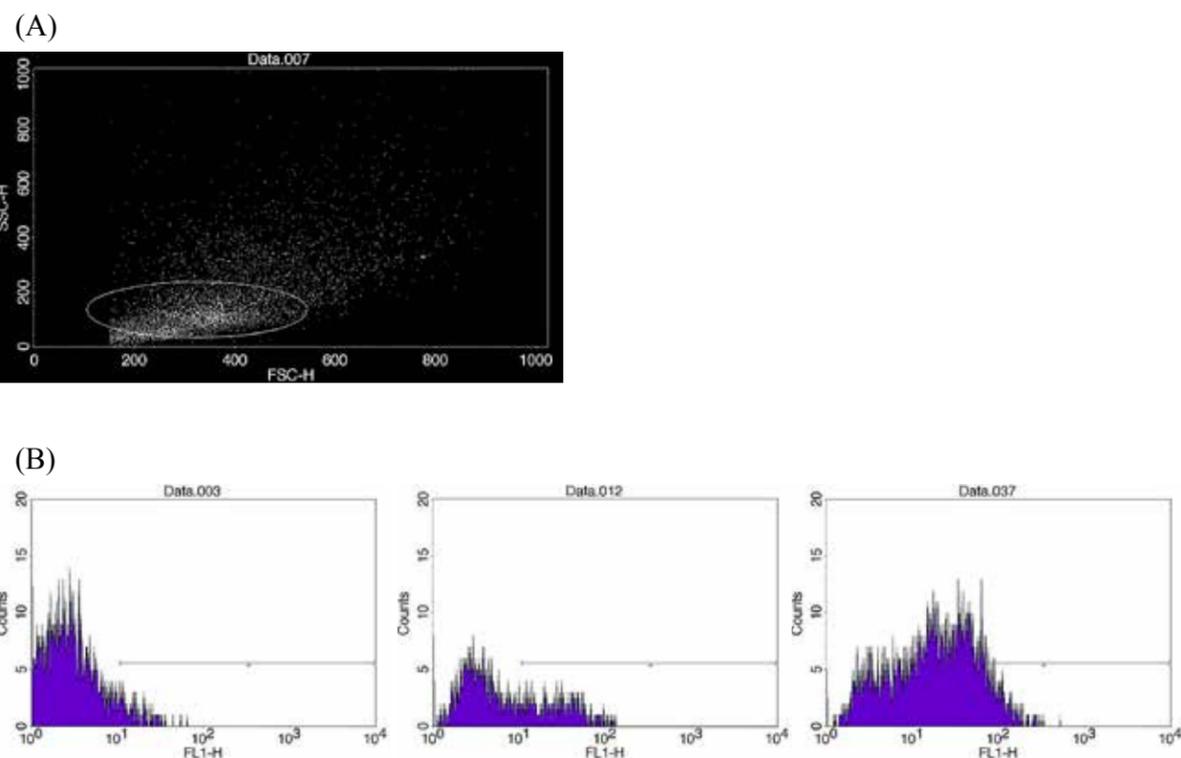


Figure 1: Gating strategy and histogram analysis. (A) A 2D dot plot was used to perform a gating strategy. Cells were graphed based on their forward (x-axis) and side (y-axis) scatter and an elliptical region (region 1) was drawn around the region corresponding to the amoebocytes. Any further analysis was performed only on cells within that region. (B) Histograms used to determine the percent of cells exhibiting macroautophagy in a sample. Cells gated on region 1 were graphed based on their degree of fluorescence detected by the FL-1 detector. A marker, indicated by the line on each histogram, was drawn based on levels of fluorescence seen in untreated cells which did not receive any CYTO-ID dye (media only cells) beginning at the first decade on a log scale. Any fluorescence by treated samples within that marker region was considered indicative of macroautophagy. The percent of cells fluorescing in this region was used for further analysis to determine statistical significance. The histograms show data from media only cells (left panel), vehicle-treated cells (middle panel), and DMBA-treated cells (right panel).

| PAH | Assay 1 | Assay 2 | Assay 3 | Assay 4 | Assay 5 | Assay 6 | Assay 7 | Assay 8 | Assay 9 |
|---------------------------|---------|---------|---------|---------|---------|---------|---------|---------|---------|
| DMBA Concentration (μM) | 100* | 100 | 250 | 250 | 25** | 25 | 25* | 25** | 100 |
| | 10* | 10 | 25** | 25 | 5* | 5 | 5* | - | - |
| | 1* | 1 | 2.5 | 2.5 | 1* | 1 | 1 | 5 | - |
| | - | 0.1 | - | - | - | - | - | - | - |
| Pyrene Concentration (μM) | 40* | 40 | 250 | 250 | 125 | 125 | 500 | - | - |
| | 10* | 10 | 25 | 25 | 25 | 25 | 100 | - | - |
| | 2.5* | 2.5 | 2.5 | 2.5 | 5 | 5 | 20 | - | - |
| | - | 0.625 | - | - | - | - | - | - | - |

Table 1: Summary of assay results. A total of 9 assays were conducted with varying concentrations of DMBA and pyrene. Significant results are indicated by an asterisk (* $p < 0.05$, ** $p < 0.005$) next to the respective PAH concentration.

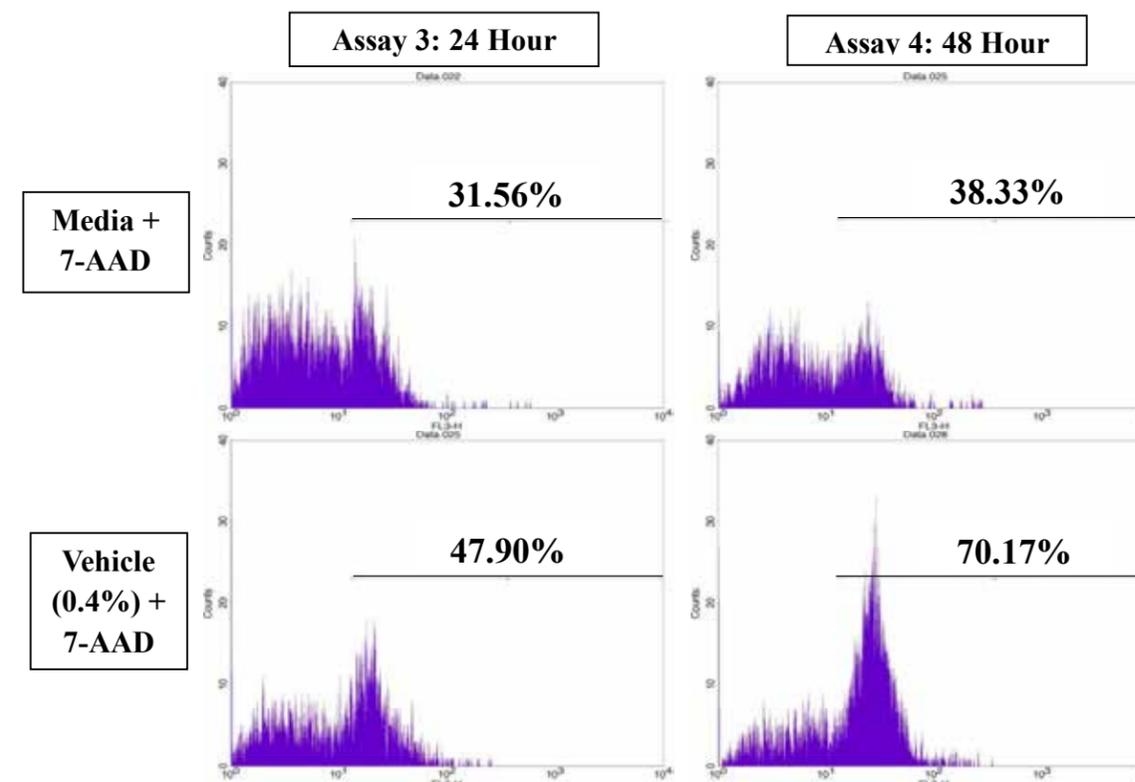


Figure 2: Histogram analyses of cell viability tests of assay 3 and 4. The results of a cell viability test using fluorescent dye 7AAD, which is detected by the FL3 detector, comparing cell death in media-only and vehicle-treated (0.4% DMSO) cells between assays 3 and 4. The x-axis represents relative fluorescence intensity detected by the FL3 detector and the y-axis represents the number of events. Listed in each graph is percent gated cells residing in the marker region, indicated by a line, which correlates with cell death.

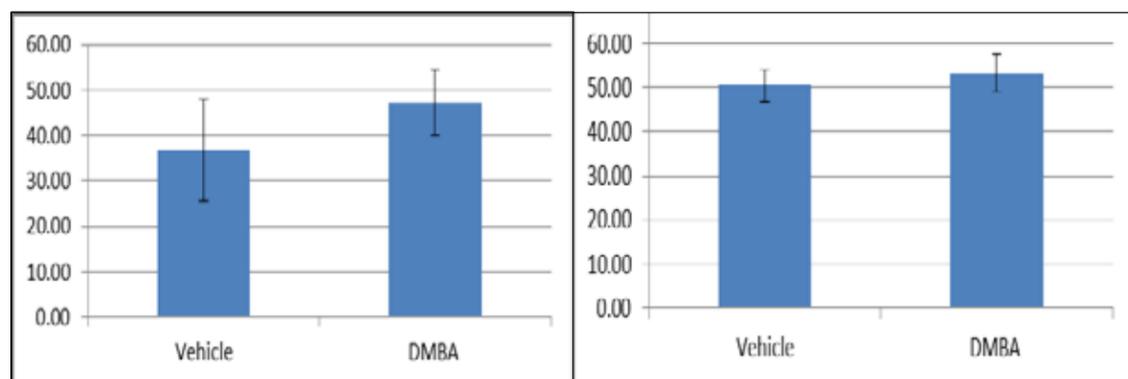


Figure 3: Comparison of individual earthworm fluorescence. Results from two earthworms, treated individually from assay 9 are shown. Each earthworm received the same treatment, however, the baseline fluorescence, given by the vehicle treatment (0.2% DMSO), was shown to have high variance between the individual earthworms. For the reason, coelomocytes from earthworms were treated individually in subsequent tests rather than using pooled coelomocyte samples from multiple earthworms in order to prevent this variance from interfering with results.

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In vitro Analysis of Cell Proliferation in Earthworm Coelomocytes in Response to Mitogenic

Agents using Click-iT EDU and Flow Cytometric Methods

Sarah Grant

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Abstract

Mitogens induce mitosis and facilitate the study of the molecular mechanisms of cell proliferation and to understand different cellular responses including the immune response to foreign components and DNA mutation. This study tested whether mitogens known to stimulate vertebrate cells also stimulate coelomocytes (leukocytes) of the invertebrate *Eisenia hortensis* (earthworm). Coelomocytes carry out innate immune defense functions to protect the coelomic cavity during infection and include chloragocytes, hyaline amoebocytes, and granular amoebocytes. Four mitogens (pokeweed mitogen, phytohemagglutinin, concanavalin A (Con A), and a mixture of phorbol-12-myristate-13-acetate and ionomycin) were used *in vitro* to stimulate coelomocytes. Newly synthesized DNA was labeled with the nucleotide analog 5-ethynyl-2'-deoxyuridine (EdU). Incorporated Edu was quantified by flow cytometric analysis using an azide dye which binds to incorporated Edu. Preliminary results confirm that Con A induces DNA synthesis in [earthworm coelomocytes](#) suggesting that growth factor-induced pathways that stimulate cell proliferation in invertebrates may use evolutionarily conserved biochemical signaling pathways used by vertebrate species. Further testing with Con A should be carried out to verify inter-assay reproducibility over a wider range of concentrations, incubation times, and temperatures with the ultimate goal of identifying the receptors and downstream effector molecules that mediate the signaling associated with DNA synthesis in this invertebrate model.

Introduction

Monitoring levels of cell proliferation can have a variety of applications such as better understanding immune responses involved in organ and stem-cell transplants (Bantly et al.,

2007). The use of mitogens, compounds which induce mitosis, is one way in which cell proliferation can be monitored. Four different mitogens known to stimulate mitosis in mammalian cells were used in this study including pokeweed mitogen, phytohemagglutinin (PHA), concanavalin A (Con A), and a mixture of phorbol-12-myristate-13-acetate (PMA) and ionomycin. This study focused primarily on Con A, which is a plant lectin isolated from the jack bean (Invitrogen, 2016). Con A is a homotetramer (Hardman & Ainsworth, 1972) which binds to two different sugar residues found on glycosylated surface proteins, α -D-mannosyl and α -D-glucosyl (Sumner, Gralén, & Eriksson-Quensel, 1938). This mitogen is known to stimulate T-cells through T-cell receptor cross-linking (Weiss, Shields, Newton, Manger, & Imboden, 1987). Con A, along with the other three mitogens, were used in an attempt to stimulate proliferation in an invertebrate model *in vitro* to determine the possibility of conserved biochemical pathways.

Traditionally, DNA synthesis, indicating active proliferation, is detected utilizing nucleotide analogs such as 5-bromo-2'-deoxyuridine (BrdU) or [3 H]-thymidine which are integrated into DNA as it is synthesized and subsequently detected. BrdU is detected using tagged anti-BrdU antibodies which is problematic because, in order for the antibodies to bind to BrdU, the DNA must first be denatured. This process requires harsh conditions which may result in DNA degradation. [3 H]-thymidine detection is also problematic as it is a process that requires a scintillation beta-counter and involves the use of radioisotopes. Click-iT EDU technology utilizes an alternative nucleotide analog, 5-ethynyl-2'-deoxyuridine (EdU), to avoid these difficulties (Base Click Product Manual).

Once incorporated into the DNA, EdU is detected using a copper-catalyzed covalent reaction between an azide and an alkyne. The azide is contained within the Click-iT[®] Alexa Fluor dye and the alkyne is contained within the EdU. After this reaction has taken place, the dye

can be detected using a flow cytometer (Invitrogen[™] Product Manual). This study examined DNA synthesis in the coelomocytes of the earthworm *Eisenia hortensis*. Coelomocytes function in the earthworm's innate immune defenses and they can be extruded through the earthworm's dorsal pores upon stimulation. They are leukocyte-like cells and exhibit phagocytic, natural killer-like, and lytic activity (Fuller-Espie, Goodfield, Hill, Grant, & DeRogatis, 2008).

Materials and Methods

Harvesting Coelomocytes

Before beginning the extrusion process, the surface of the earthworms had to be disinfected to prevent fungal contamination. To do this, the earthworms were placed in Petri dishes containing paper towel pieces which had been sprayed with fungizone (250 μ g/mL), an antifungal agent. In addition, the lack of food for the overnight period permitted defecation and elimination of gut microbes that would otherwise increase microbial contamination upon extrusion of the coelomocytes. After having incubated in the Petri dishes overnight, the earthworms were removed and transferred into sterile plastic troughs with 3mL of FACSFlow. FACSFlow is a saline solution which served as the extrusion buffer and prompted the release of the coelomocytes.

The cells were then exposed to 0.3mL of of Accumax[™], an enzyme mixture with trypsin and collagenase-like activity, to prevent cellular aggregation. After 2 minutes had passed, the enzymes were diluted with 5mL of phosphate-buffered saline (PBS) and the solution was centrifuged at 800 RPM (150g) for 5 minutes at 4°C to obtain a cell pellet. The cells were then resuspended in Dulbecco's Modified Eagle Medium (DMEM) which had been supplemented with glutamine, non-essential amino acids, penicillin, streptomycin, amphotericin B, kanamycin,

ampicillin, chloramphenicol, tetracycline, nystatin, 10mM HEPES buffer, and 10% heat-inactivated newborn calf serum according to Fuller-Espie, Harris, Daly, and Jakeman (2015). Coelomocytes were then enumerated using a hemocytometer and the concentrations were adjusted to 4×10^6 cells/mL.

EdU Flow Cytometry Assay

Once the concentration of each sample had been adjusted, cells were loaded into a 96-well plate with their respective mitogen treatment at a range of specified concentrations. In addition to the coelomocytes, K562 cells were included as positive and negative controls for the detection of DNA synthesis. Positive control cells received 5% serum while negative control cells received no serum. 20 μ M EdU, which had been pre-warmed at 25°C for 10min, was then also added to the wells and cells were incubated overnight in a 5% CO₂ at 25°C. After incubation, samples were spun at 800 RPM for 5 minutes at 4°C and pellets were re-suspended by vortexing. Cells were then incubated in 200 μ L of freshly made 4% paraformaldehyde for 15 minutes at room temperature for fixation. Fixation was followed by centrifugation and re-suspension in 200 μ L of 3% bovine serum albumin (BSA) to wash the cells. This wash step was repeated twice, however, after the final centrifugation cells were re-suspended in 200 μ L of 0.5% Triton-x-100, instead of 3% BSA, and incubated for 20 minutes at room temperature to permeabilize the cells. Fixation and permeabilization were used to allow the azide dye to easily enter the cells.

While the cells incubated, a reaction cocktail containing 1x Click-iT reaction buffer, CuSO₄, Alexa Fluor azide dye, and Click-iT reaction buffer additive was prepared. The cells then underwent three more wash steps, as previously described, however 100 μ L of the reaction cocktail was used to re-suspend cells in place of 3% BSA after the third centrifugation. The cells

were incubated with the reaction cocktail for 30 minutes at room temperature in the dark. 100 μ L of 3% BSA was then added to each well prior to three more wash steps. After the final addition of 3% BSA, cells were transferred into flow cytometry tubes and 100 μ L of FACSFlow was added to each tube. Samples were then run on a FACSCalibur flow cytometer (BD Biosciences). Data was collected and analyzed using Cell Quest Pro™ software (BD Biosciences) and statistical significance was determined using Microsoft Excel and the student's t-test paired two sample for means. Results were considered significant if they had a *p*-value less than 0.05 when compared to baseline, untreated cells providing a confidence interval of 95%. Each earthworm was treated individually and all treatments were performed in either duplicates or triplicates.

Cells were gated using a 2D dot plot graphing cells based on their forward and side scatter by drawing an elliptical region around the desired cell population. Coelomocytes include three different subpopulations, chloragocytes, hyaline amoebocytes, and granular amoebocytes. The region was drawn to include only the amoebocytes, due to the high autofluorescence of the chloragocytes which would have skewed the results (Figure 1A). The data from cells within this region were then graphed in a histogram based on their degree of fluorescence detected by the FL1 detector (Figure 1B). A marker region was set on each histogram based on the levels of baseline fluorescence seen by control cells and the percent of cells fluorescing within that marker region, indicating DNA synthesis was taking place, was recorded for each sample. The average percent of cells exhibiting DNA synthesis was then calculated for each treatment for a given worm and these averages were graphed. The student's t-test was used to determine any significant differences in the percent of cells exhibiting DNA synthesis.

Results

The K562 cells were not found to be successful negative controls throughout any of the assays conducted. Even after an extensive period of starvation, the percent of cells exhibiting DNA synthesis was not found to be significantly lower compared to samples which received 5% serum (Figure 2). Con A was found to be the only mitogen which resulted in significantly elevated levels of DNA synthesis after treatment. A representative example of these results is shown in Figure 3. In this example, cells were treated with either high (H), medium (M), or low (L) Con A concentrations of 240µg/mL (ConA H), 60µg/mL (ConA M), or 15µg/mL (ConA L). The percent cells exhibiting DNA synthesis at each concentration were compared to levels in control cells (EdU Only) and this difference was found to be statistically significant for all three concentrations. Although this increase was marginal, it was found to be both statistically significant and reliably reproducible. Con A was tested at a variety of concentrations, ranging from 15 to 240 µg/mL, and within each assay there was as high as a 67% response rate of individual earthworms at a given concentration. Table 1 shows a summary of the Con A results for all 5 experiments conducted.

Discussion

The use of K562 cells as negative controls for DNA synthesis was unsuccessful. It was hypothesized that this difficulty may have been due to the fact that K562 is a cancer cell line and, therefore, has higher than normal levels of DNA synthesis which are more difficult to decrease even with extensive starvation. Con A was the only mitogen of the four which were tested that resulted in significantly elevated levels of DNA synthesis. The fact that this increase was

marginal was postulated to be due to the low replicative capacity of earthworm coelomocytes *in vitro*. Because T-cell stimulation by Con A occurs through a T-cell receptor cross-linking pathway, there may be an analogous protein cross-linking process taking place in Con A stimulation of coelomocytes involving a membrane-associated signaling complex.

Future research aiming to better understand coelomocyte stimulation by Con A should include a control for Con A stimulation, such as EL4 thymoma cells. Viability tests should also be included using a dual staining procedure to ensure exclusion of dead cells from final analysis. The use of Accumax™ could be potentially counteracting stimulation by Con A. Accumax™ contains proteases which may cleave protein receptors needed for Con A stimulation and, therefore, may also be a cause of lower levels of DNA synthesis despite Con A treatment. An alternative to Accumax™ that would not have this effect could be used in future research. Finally, the pathways through which Con A stimulation occurs could be investigated to determine whether or not any evolutionarily conserved cytosolic signaling cascades are used in the stimulation of vertebrates and this invertebrate model.

Acknowledgements

I would like to acknowledge the Beta Beta Beta National Biology Honor Society and Cabrini Science Department for providing funding for this project. I would also like to acknowledge Dr. Sheryl Fuller-Espie for giving me the opportunity to conduct research with her and for her constant support throughout the entire process.

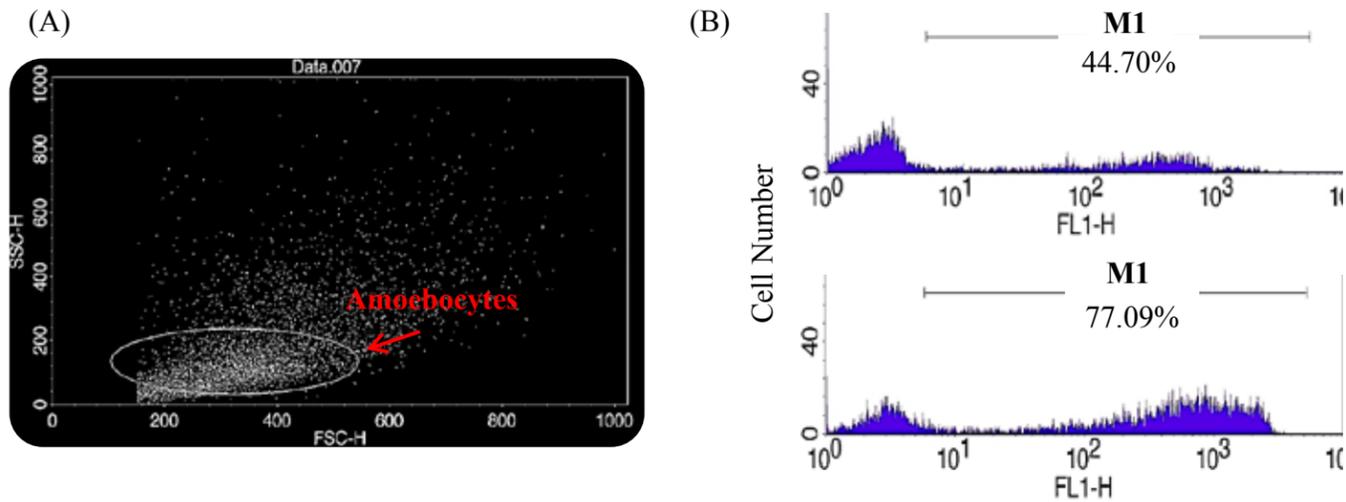


Figure 1: Gating Strategy and Histogram Analysis. (A) An example of a 2D dot plot, graphing cells based on their forward (x-axis) and side (y-axis) scatter, used to perform a gating strategy. An elliptical region was drawn around the region corresponding to the amoebocytes. All subsequent graphs and histograms only included data from the cell population within that region. (B) Two examples of histograms used to determine the percent of cells undergoing DNA synthesis. Cells were graphed based on their fluorescence detected by the FL-1 detector and a marker (M1) was drawn based on baseline fluorescence from control cells. All cells fluorescing within that region were said to be undergoing DNA synthesis. The top histogram shows K562 cells which were starved to reduce replication rates and the bottom histogram shows K562 cells which were given 5% serum. The percent of cells fluorescing in the M1 region was recorded, graphed, and used for further analysis to determine significance.

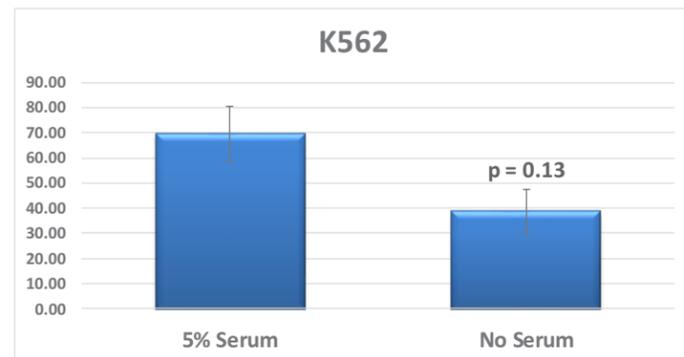


Figure 2: DNA Synthesis in K562 Cells. A representative example of levels of DNA synthesis in K562 cells after treatment with 5% serum or no serum. The y-axis shows the percent of cells exhibiting DNA synthesis. No significant decrease in DNA synthesis after starvation was seen ($p=0.13$).

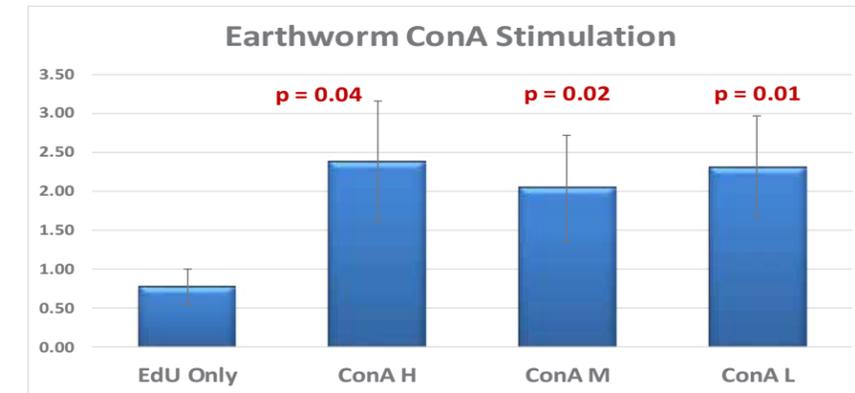


Figure 3: DNA Synthesis in Coelomocytes. A representative example of levels of DNA synthesis in coelomocytes of an individual earthworm after receiving no treatment (EdU Only) or treatment with one of three concentrations of Con A: 240 μ g/mL (ConA H), 60 μ g/mL (ConA M), or 15 μ g/mL (ConA L). All three concentrations resulted in significant increase in DNA synthesis when compared to EdU Only cells (ConA H $p=0.04$, ConA M $p=0.02$, ConA L $p=0.01$).

| <u>Experiment Number</u> | <u>ConA Concentration (μg/mL)</u> | <u>Number of Earthworms Tested</u> | <u>Percent Earthworms with Significant Results</u> |
|--------------------------|--|------------------------------------|--|
| 1 | 100 | 3 | 66.67% |
| 2 | 100 | 3 | 33.33% |
| 3 | 200 | 5 | 20% |
| 4 | 240 | 6 | 33.33% |
| - | 60 | 6 | 33.33% |
| - | 15 | 6 | 33.33% |
| 5 | 240 | 6 | 50% |
| - | 60 | 6 | 33.33% |
| - | 15 | 6 | 50% |

Table 1: Summary of Con A Results. A summary of the results seen after treatment of coelomocytes with Con A throughout the 5 assays conducted. Significance was seen in every assay and at every concentration which was tested and as high as 66.67% of earthworms in a given assay showed a statistically significant response to Con A stimulation.

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**Earthworms as Sentinel Species for the Measurement of Macroautophagic Biomarkers in
Response to Heavy Metal Environmental Pollutants, Cadmium and Copper**

Authors: Olivia Townsend, Lauren Markowitz, & Sheryl L. Fuller-Espie

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This research was funded through by a 2016 Pennsylvania Academy of Science Undergraduate Research Grant and by the Cabrini University Biology Research Fund.

ABSTRACT

Heavy metal exposure is becoming a large concern as the use of electronics has grown significantly over the years. Unknown to most people, individuals expose themselves to heavy metals, found within computers and cellphones, on a regular basis. Likewise, this becomes an increasing ecological concern as heightened toxicity exposure to the environment impacts sentinel species like the earthworm that inhabit the soil. Studies have shown the adverse effects that such exposure can have on organisms over time resulting in disease and complications. These complications begin as the cell's ability to naturally break down its old organelles, and proteins becomes dysregulated. In normal cell degradation, a process called autophagy can occur where the cell forms vesicles to digest old or damaged materials. However, this process can become irregular when the cell receives the wrong signals or carries out the process too frequently resulting in digestion of large amounts of cellular components known as macroautophagy. This ecological study sought to observe the cell's response to differing concentrations of the heavy metals copper and cadmium in order to document the levels of macroautophagy that occur within the experimental specimen *Eisenia hortensis* following exposure. In order to study the effects heavy metals have on macroautophagy, the study included both *in vivo* and *in vitro* methodologies. Analysis of macroautophagy was conducted using an

autophagy detection kit combined with flow cytometry. Preliminary assays pooled collected earthworm cells from multiple animals, but upon review, cells from individual animals were observed for macroautophagic effects specific for one earthworm at a time. Although results for three of the six treated earthworms supported the hypothesis, this experiment had low inter-assay reproducibility and further studies are needed to obtain more consistent findings.

Introduction

With the continually rising trend towards urbanization, it is no mystery as to why pollution has substantially grown over the years. Many times these pollutants can then be cycled into the soil and into the produce that the world's population consumes (Lang-Yona, Shuster-Meiseles, Mazar, Yarden, & Rudich, 2016). Improper electronic disposal contributes largely to this growing problem. As the lifespan of electronic devices shortens, the public's willingness to properly recycle used electronics decreases as well (Sepúlveda et al., 2010). This means that chemicals and heavy metals within these devices are introduced into the environment and can have adverse effects as a result. Even today, heavy metal exposure is one of the major contaminants that impedes on crop production (Sharma & Dietz, 2009). In spite of this, the environment and the organisms that live within it have adapted methods to combat these concentrations of pollutants. One of these processes carried out, on the microscopic level, is called macroautophagy.

Autophagy, is the process of organelle and protein degradation in order to maintain "cytoplasmic homeostasis" throughout the cell (Kai et al., 2016). Many times the cell will recycle nutrients digested as a result of this process (Magraoui, Reidick, Meyer, & Platta, 2015). However, the cell will not begin autophagy without stimulants to signal that it should "self eat". One cause is nutrient deprivation where the cell seeks to preserve its homeostasis and recycles its

organelles for nutrients. Another cause is associated with physical changes to the cell in which the organelles are damaged. This can result from either oxidative stress, or in the case of this study, exposure to heavy metals which induce production of reactive oxygen species (Fuller-Espie, Bearoff, & Minutillo, 2011).

The autophagic process begins as lysosomes morph into autophagosomes that form vesicles around the substances to be digested. These substances are subsequently digested into amino acids and fatty acids that the cell can use to survive (Qiankun et al., 2015). Autophagy within cells has been observed to contribute to regeneration of the cell along with aging and development. Nonetheless, irregular control of this process can lead to major diseases including that of cancer and neurological disorders (Duraes et al., 2015).

Macroautophagy refers to many of the same concepts that autophagy references. What separates macroautophagy from autophagy is the formation of a characteristic double membrane vesicle of the cytosol in which the contents can be degraded (Duraes et al., 2015). This process can also be referred to as "bulk autophagy" as large portions of cellular material are being transferred into the vesicle for digestion (Magraoui et al., 2015).

Figure 1 provides an explanation of the process of macroautophagy based upon a schematic taken from the *International Journal of Molecular Sciences* (Ziparo et al., 2013). As shown in this figure, several signals can stimulate autophagy of the cell aside from normal degradation of old organelles and proteins. When the cell does not receive enough nutrients for survival it can begin this process. This occurs from starvation of the cell. Another less common cause is exposure to toxic materials found within heavy metals (Chatterjee, Sarkar, & Bhattacharya, 2014). Heavy metals are those that have low poison concentrations with relatively high densities (Chiarelli & Roccheri, 2012). Much like any other process within the cell,

overregulation of one process can lead to complications. In the case of heavy metal exposure, the cell attempts to remove the toxic organelles, but results in degrading too much of the contents of the cell through macroautophagy that can result in the death of the entire cell (Chiarelli & Roccheri, 2012).

With over 30 different types of heavy metals considered toxic, many of which humans come in contact with daily, research into cellular exposure to these substances seems quintessential in order to understand some of their effects upon the cell itself. In regards to the human body, these heavy metals have been proven to have exceedingly adverse effects upon the cell and the resulting autophagy can lead to disease and disorder (Chiarelli & Roccheri, 2012). The metals in question for the research to follow include cadmium and copper. Both are heavy metals that can have adverse effects on the cell if overexposure becomes a factor. For example, heavy metals and metalloids have been implicated in the aggregation and misfolding of cellular proteins (Tamás, Sharma, Ibstedt, Jacobson, & Christen, 2014).

For this study, *Eisenia hortensis* was utilized as the sentinel model to observe the effects of exposure to pollutant materials within their coelomocytes, as they are sensitive to environmental changes. It is noteworthy that earthworms have also shown advantageous results when observing ecotoxicological effects (Beeby, 2001). For example, earthworms have been used to study the detrimental effects of metal-contaminated, dredged sediment on tropical ecosystems, as they are sensitive to the toxins (Cesar et al., 2014).

Coelomocytes of the earthworm are highly valuable when looking at heavy metal exposure of a cell as a flow cytometer can monitor changes in cellular processes with ease. The earthworm can be induced to excrete fluids rich in coelomocytes through its dorsal pores (Fuller-

Espie et al., 2011). Once extruded, the coelomocytes can be analyzed for induction of macroautophagic processes following exposure to environmental pollutants.

This study observed macroautophagy within the *E. hortensis* under heavy metal exposure of copper and cadmium. Samples will be observed *in vitro* and *in vivo* after being treated with five different concentrations of the metals. Coelomocytes will then have CYTO-ID® green stain solution added to them in order to monitor macroautophagy using flow cytometry (see Methods). Important ecotoxicological studies like these are quintessential when exploring potentially harmful long term effects linked to heavy metal exposure. As society's dependency on technology is exponentially growing, there is still so much more to be understood about what effect devices like cell phones and computers will have on the environment in the long-run due to improper disposal practices. Ecotoxicological studies like these are the start to understanding the major negative impact of heavy metal pollution on ecosystems that are pivotal to healthy soil and vitality of organisms that populate these habitats.

Materials and Methods

Eisenia hortensis

Our earthworms were obtained from Uncle Jim's earthworm farm (2046 Henry Ln, Spring Grove, PA 17362). We housed our earthworms in the animal auxiliary suite at Cabrini. There habitats consisted of plastic boxes containing corncob shavings, water, pine chips and shredded paper towels. They were fed with Gerber baby food and Quaker oats. The earthworm cages were changed every 2- 3 days.

Coelomocytes Extrusion

Each earthworm was placed into a sterile plastic trough containing 3 ml ice-cold extrusion buffer (FACSFlow-BD Bioscience). This buffer stimulates the coelomic cavity causing

the earthworms to expel coelomocytes from its dorsal pores. Immediately following extrusion, 0.5ml of AccumaxTM, an enzyme mixture of trypsin, collagenase and DNase was added to the coelomocyte to inhibit cellular aggregation. After 2 minutes at room temperature, 5 ml of phosphate-buffered saline (PBS) was added to dilute the enzyme. Then, the cells were centrifuged at 4 °C at 800 rpm (150g) for 5 minutes. The coelomocytes were then re-suspended in Dulbecco's Modified Eagle Medium (DMEM) supplemented with 10 % newborn calf serum, glutamine, non-essential amino acids, penicillin, streptomycin, amphotericin B, kanamycin, tetracycline, chloramphenicol, and HEPES buffer (according to Fuller-Espie, Harris, Daly, Jakeman, 2015).

In vitro Procedure

The day before extrusion, ten earthworms were placed into a Petri dish containing paper towels moistened with fungisome. The earthworms were removed from the Petri dish the next day and their coelomocytes were extruded, as indicated above. Cells from 6 individual earthworms were analyzed. The number of coelomocytes were measured using a hemocytometer and adjusted for assay set-up. 100µl of the cells (2×10^5) were added to appropriate wells of a 96- well plate. Then, vehicle, heavy metals and controls were added to designated wells to treat the cells. All treatments were done in triplicates. The heavy metals were added to specified wells in the amount of 100 µl. Cells were treated with copper and cadmium at concentrations of 5µM and 40µM in PBS vehicle control. Controls had 100µl of media added to each well with the same concentration of PBS compared to treatments. The treated cells were incubated in 5% CO₂ incubator at 25 °C for 16-18 hours. The 96- well plates were removed from the incubator and examined under a microscope to detect any contaminants. Then, the plates were centrifuged at 4°C at 800 rpm for 5 minutes. The plates were then flicked in the sink to remove supernatant and

vortexed. 200 µl of 1x assay buffer supplemented with 5% newborn calf serum was then added to each well containing cells. The plates were centrifuged again and flicked in the sink. Next, we resuspended the cells in each well in 100 µl of 1x assay buffer and transferred the mixture to labeled flow cytometry tubes.

Next, 100 µl of 1x Cyto-ID dye was added to appropriate tubes. 1x Cyto- ID was made using 1000x Cyto-ID stock and assay buffer that did not contain newborn calf serum. Therefore, the Cyto-ID staining incubation period was done in 2.5% newborn calf serum. After Cyto-ID was added to each sample it was mixed using a vortex. The tubes were covered with aluminum foil to decrease photobleaching and placed on an orbital shaker for 45 minutes at 25 °C at 200 rpm. After 45 minutes the tubes were centrifuged at 800 rpm at 4°C for 5 minutes and the supernatant was discarded. Then, 200µl of 1x assay buffer was added to each tube and the samples were centrifuged again under the same conditions. The supernatant was discarded and the cells were resuspended in 300 µl of assay buffer and placed on ice. The samples were then analyzed one by one on the flow cytometer.

Results

Figure 2 shows the results of an assay that provided favorable findings. In this experiment cells from individual earthworms were analyzed. Three worms exhibited significant results. Earthworm #2, shown in **figure 2**, had an average vesicle formation of 4.19% for vehicle #1. Vehicle #1 was a baseline comparison for cadmium and copper treatments at 40µM which had an average of 6.38% and 4.37%. PBS vehicle #2 had an average of 4.47%. Vehicle #2 was a baseline comparison for cadmium and copper treatment 5µM which had values of 4.72% and 3.65%. Earthworm #3, shown in **figure 3**, had an average vesicle formation of 4.27% for vehicle

#1. Cadmium and copper 40 μ M concentrations had values of 52.72% and 6.60%. Vehicle #2 had an average of 3.85%. This value was compared to cadmium and copper 5 μ M concentration values of 41.57% and 29.51%. Earthworm #4 is shown in **figure 4**. Vehicle #1, 40 μ M cadmium and 40 μ M copper had average values of 5.11%, 52.35% and 52.74% respectively. Vehicle #2, 5 μ M cadmium and 5 μ M copper had values of 7.42%, 54.60%, and 55.33% respectively. Flow cytometer images shown in **figure 5**, represent the shift in macroautophagy in earthworms 2, 3, and 4.

Conclusion

Many of our assays produced significant results for one or more of the heavy metal concentrations. The data shown in this report had significant results for three of the six worms analyzed. Earthworm 2 had significant results for cadmium medium. Earthworm 3 had significant results for cadmium medium and cadmium low. Earthworm 4 had significant results for copper medium, copper low, cadmium medium and cadmium low concentrations. In this experiment we used individual earthworms and we found that each earthworm had a high degree of variability between the control values. We concluded that it is important to use individual earthworms in subsequent assays to ensure the control was representative of the cells being treated. Unfortunately, throughout our study, results were highly varied yielding low inter-assay reproducibility. For this reason and because Cyto-ID is only known to detect macroautophagy in mammalian cell lines, we concluded that Cyto-ID is not a reliable source to detect macroautophagy in earthworm (invertebrate) cells.

Alongside this experimental design, we also conducted *in vivo* studies using intact earthworms and analyzing macroautophagy following 19 days exposure (data not shown). Significant results were obtained with cadmium, however, the coelomocytes were pooled from

all treatment groups, a practice found to provide non-optimal conditions because of wide variation in baseline readings between individual earthworms.

In the future, another method could be used to measure macroautophagy in earthworms. One method is analyzing the autophagy protein LC3 to monitor autophagy. LC3 is a microtubule-associated protein initially synthesized in its unprocessed form (pro LC3). Upon autophagy stimulation, the C terminus of LC3 is cleaved by the Atg4 proteases to form LC3-I (Kimura, Noda, & Yoshimori, 2007). LC3-II then forms, which remains associated to the autophagosomal membrane until this vesicle fuses with the lysosome to form the autophagolysosome where LC3-II is digested together with the rest of the autophagosomal content (Kimura et al., 2007). Conversion of LC3-I into LC3-II and changes in the subcellular distribution of LC3 are thus two hallmarks of autophagy that can be monitored to determine whether this cellular process becomes activated. Cells can be manipulated to express a recombinant form of LC3 fused with GFP (green fluorescent protein). This recombinant form of LC3 is processed in the same way as endogenous LC3, and therefore when these cells are subjected to an autophagic stimulus they exhibit a characteristic pattern of GFP-LC3 puncta (Kimura et al., 2007).

Appendix

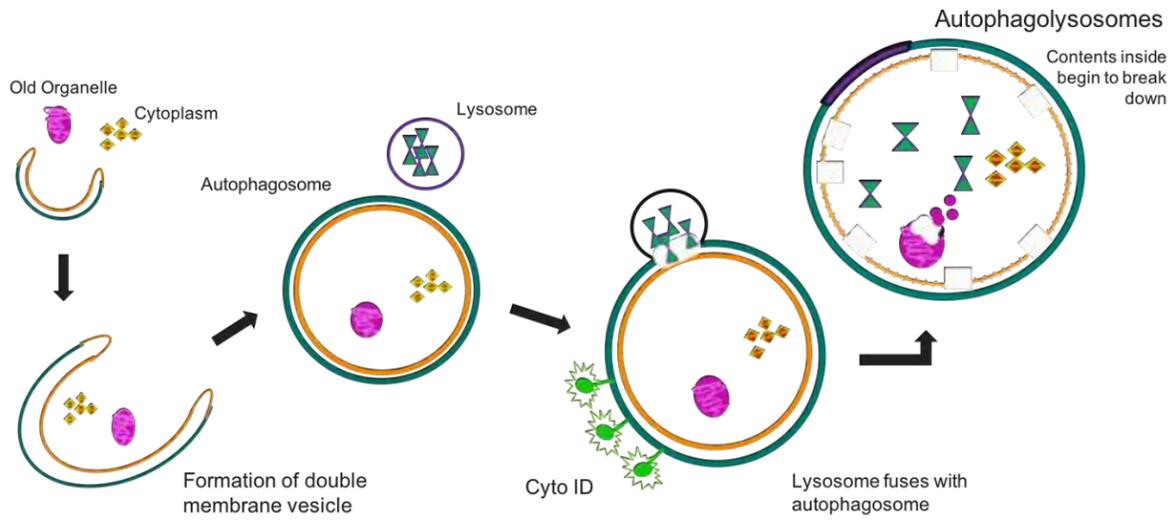


Figure 1. Autophagy in Prostate Cancer and Androgen Suppression Therapy. This figure depicts the autophagic process within a cell induced by starvation or some other stress like heavy metal exposure. The characteristic double vesicle is present within this figure as the cell begins to digest its own contents. Incorporation of Cyto ID in the double-membrane structure is illustrated to represent the process for quantifying macroautophagy in experimental trials of this study. This figure was modified from a schematic designed by Ziparo, Petrunaro, Marini, Starace, Conti, Facchiano, & Giampietri (2013).

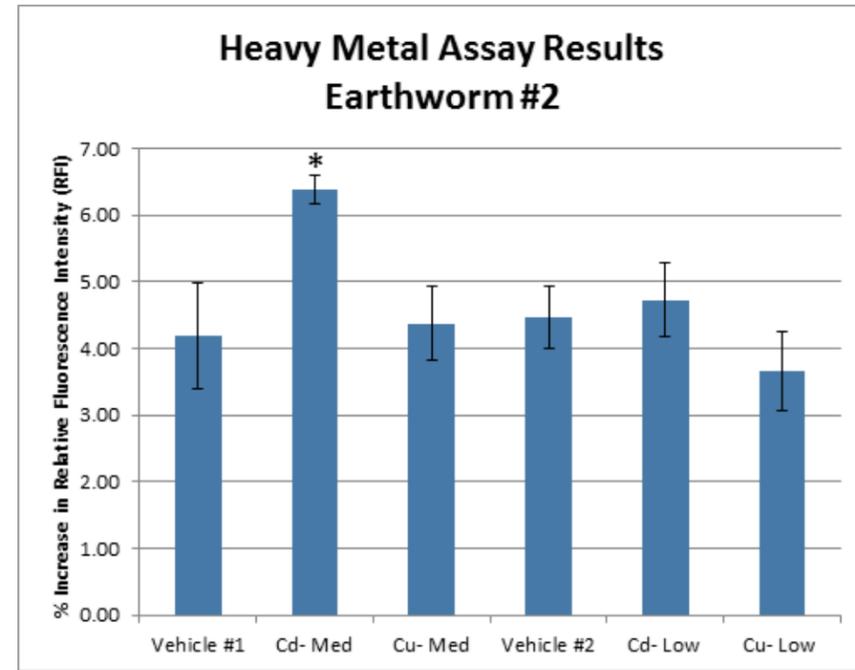


Figure 2. *In vitro* Assay. The average percentage of vesicle formation in cells obtained from earthworm #2 after treatments measured as percent increase in relative fluorescence intensity(RFI), detected by the FL-1 photomultiplier tube. The error bars represent the standard deviation, while an asterisk represents a significant result.

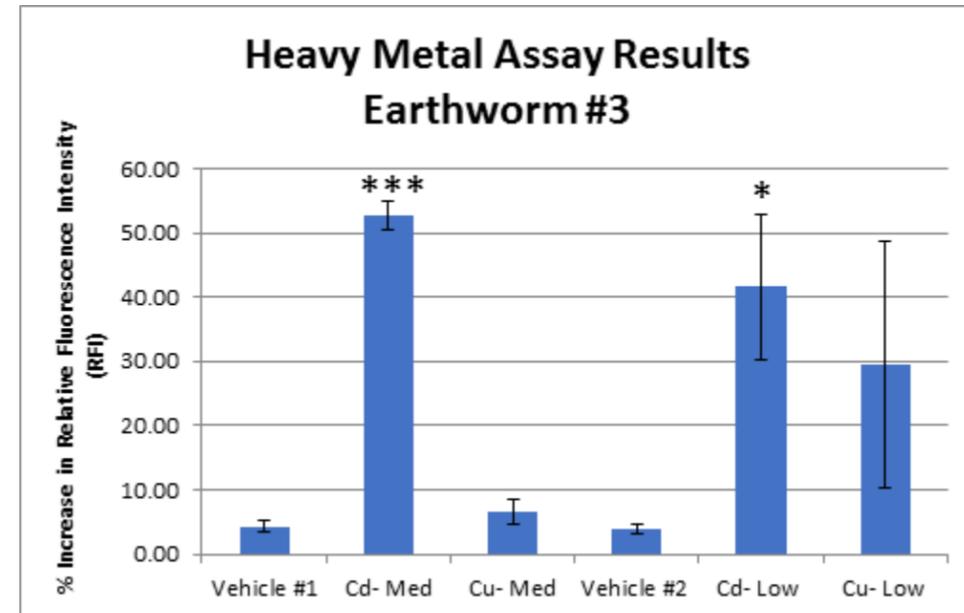


Figure 3. *In vitro* Assay. The average percentage of vesicle formation in cells obtained from earthworm #3 after treatments measured as percent increase in RFI, detected by the FL-1 photomultiplier tube. The error bars represent the standard deviation, while one asterisk represents a significant results and three asterisks represents a highly significant result.

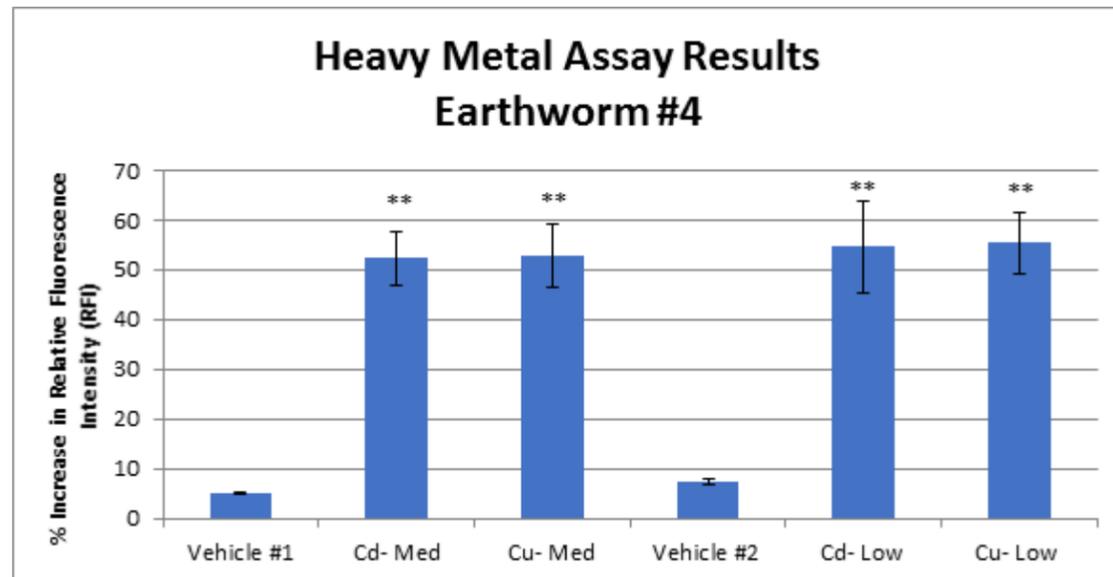


Figure 4. *In vitro* Assay. The average percentage of vesicle formation in cells obtained from earthworm #4 after treatments measured as percent increase in RFI, detected by the FL-1 photomultiplier tube. The error bars represent the standard deviation, while two asterisks represents a highly significant result.

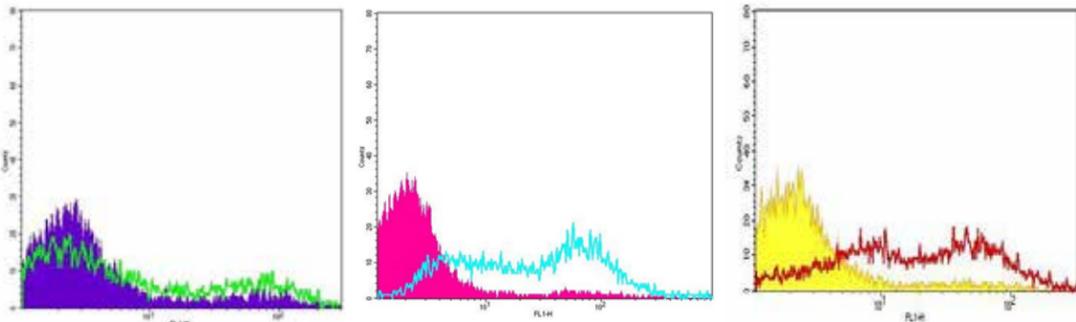


Figure 5. *In vitro* Assay. Shifts in macroautophagy levels can be seen in the histograms, from left to right, in earthworm #2, #3 and #4. The solid color represents the untreated cells and the line represents cells treated with cadmium concentration of 40µM. The x-axis shows RFI and the y-axis shows the cell count.

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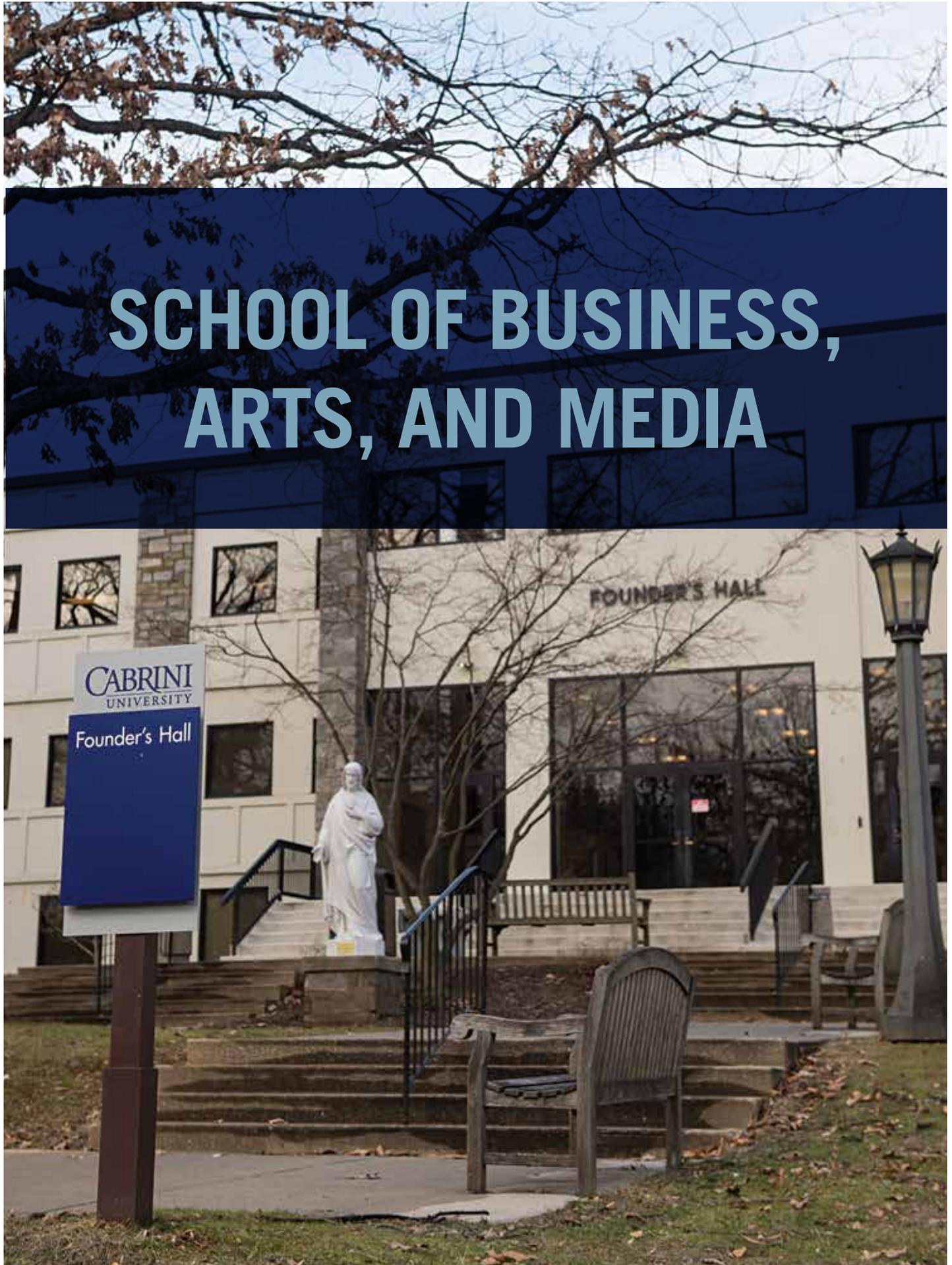
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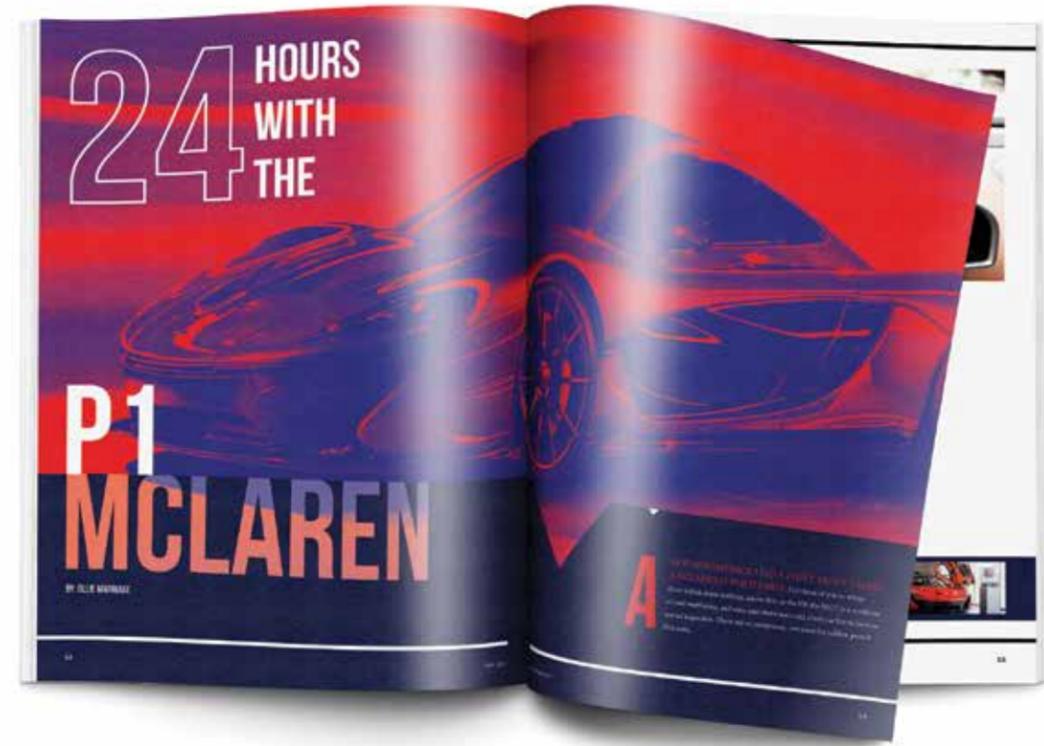
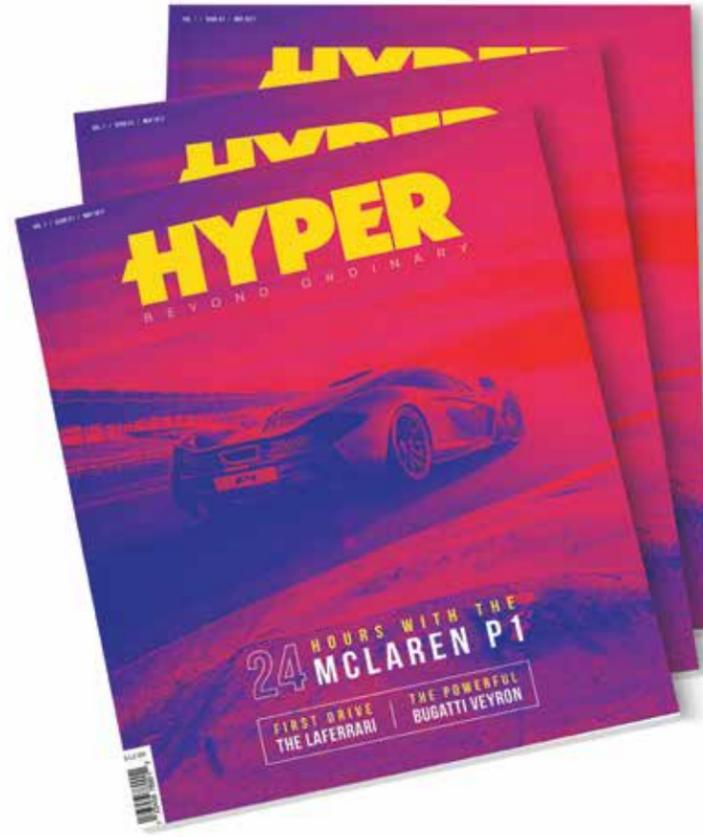
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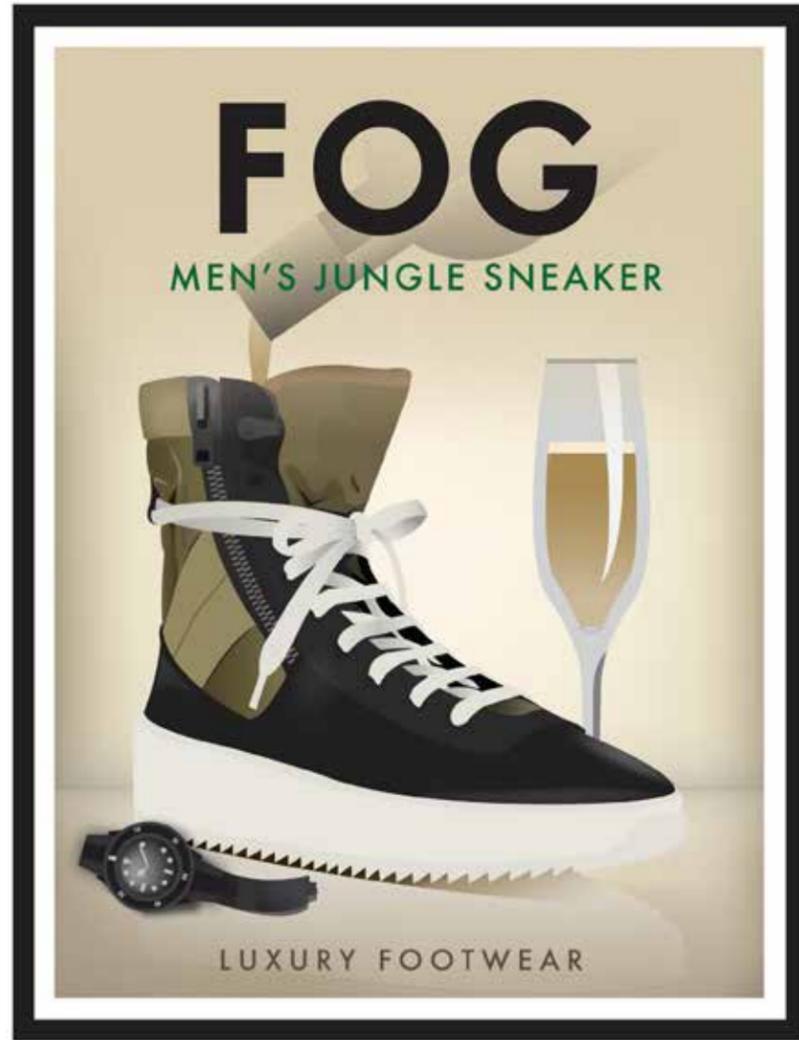
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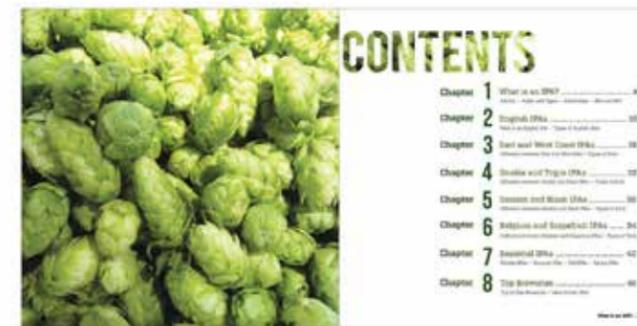
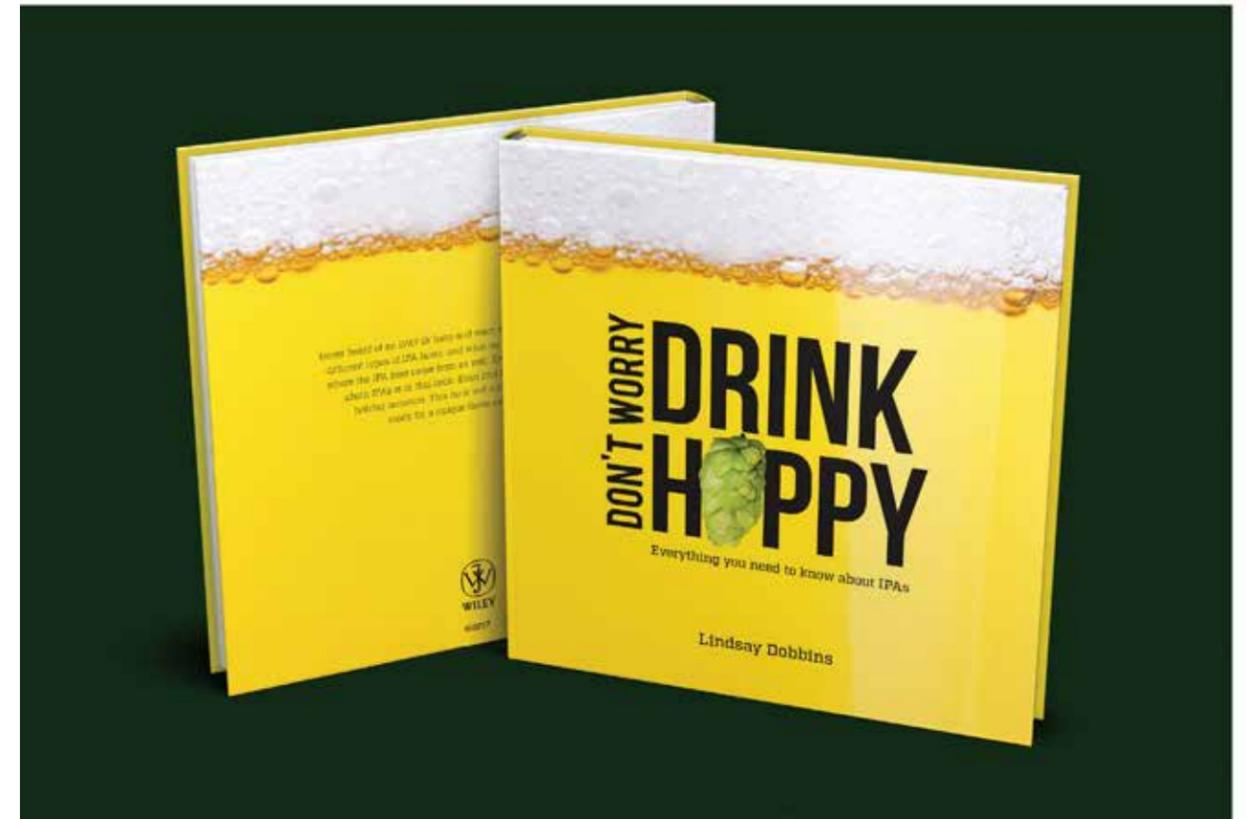




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Drink Worry, Drink Hoppy Book Design
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The Guide to All Things Rock 'N' Roll in Philly

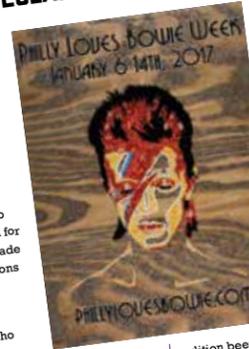
MAYOR JIM KENNEY DECLARES PHILLY LOVES DAVID BOWIE WEEK

On Thursday, January 5th, Philadelphia Mayor Jim Kenney issued the following proclamation officially declaring January 6-14 Philly Loves David Bowie Week!

The City of Philadelphia is a music hub and source of professional inspiration for remarkable entertainers who have made a lasting impact on multiple generations worldwide.

The City was an integral part in the legendary career of David Bowie who performed many sold out shows that delighted Philadelphians over the past few decades. He recorded his album *Young Americans* at the iconic Sigma Sound Studios on 12th Street in August 1974. He also recorded three LPs – *David Live*, *Young Americans*, and *Stage* – in Philadelphia as well as filmed the music video for the 1983 hit single “Modern Love.” In total, his works include 27 studio albums, 9 live albums, 49 compilation albums, 6 extended plays, and 121 singles. He is estimated to have sold over 140 million records during his unparalleled lifetime career.

In 1973, Bowie published the song “My Death” in which he wrote about his supposedly impending demise – But whatever lies behind the door/There is nothing much to do/Angel or devil, I don't care/For in front of that door there is you. His fans felt that same level of connection to him. Bowie's



intense passion was about sharing love with others and encouraging them to spread their generosity. His untimely death from cancer on January 10, 2016 stunned his many fans around the world who were devastated by the tragic news. But as the musician wrote in the 1972 song “Rock'n'Roll Suicide,” just turn on with me and you're not alone.

Philadelphians will not be alone when they honor his legacy with “Philly Loves David Bowie Week” that will comprise more than a dozen unique events between January 6 and 14. The festivities are coordinated by those who were inspired by the musician and expressed their attachment to him in a special way. It includes the unveiling of a special

edition beer, a celebration on the anniversary of his birthday on January 8, a spoken word event that includes readings of his work, a special tribute in his memory, an art show, ice skating, and multiple musical homages by local artists. Proceeds of the events will benefit cancer research at the Children's Hospital of Philadelphia (CHOP).

Therefore, I, James F. Kenney, Mayor of the City of Philadelphia, do hereby proclaim the week of Friday, January 6 through Saturday, January 14, 2017 to be **Philly Loves David Bowie Week** in Philadelphia, and urge all citizens to recognize the extraordinary contributions the organizers are making to celebrate this iconic musician's roots in Philadelphia, and also help fund an important City institution to cure and prevent cancer.

MUST-SEE UPCOMING CONCERTS:

| | | | | | |
|---|---|---|--|--|--|
| February 22: Dave Hause Boot & Saddle | February 24-25: Jaspandroids Union Transfer | April 20: Drive-By Truckers World Cafe Live Wilmington | May 12: Metallica Lincoln Financial Field | June 19: Jason Isbell The Fillmore | August 31: Green Day BB&T Pavilion |
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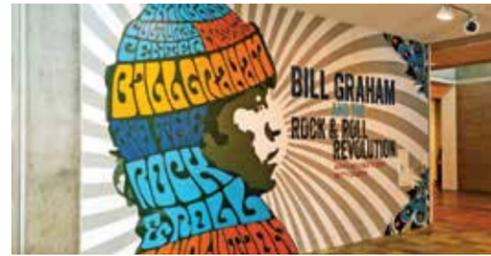
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THE SETLIST:

Philly Loves David Bowie Week
Bill Graham Honored in Exhibit
Bruce Springsteen Book Tour
Dave Hause New LP

Reviews:

Purling Hiss 'High Bias'
Drive-By Truckers 'American Band'



LATE ROCK & ROLL LEGEND BILL GRAHAM TO BE HONORED IN AN EXHIBIT AT PHILLY'S MUSEUM OF JEWISH HISTORY

Calling all Rock 'n' Roll enthusiasts, historians, museum hoppers of Philadelphia, and more. It doesn't matter who you are or where you come from, everybody will be able to identify with the traveling exhibit that is currently being hosted by The National Museum of American Jewish History.

Bill Graham and the Rock & Roll Revolution, which opened in Philadelphia on September 15, is an exhibit devoted to honoring the life and career of legendary concert promoter, the late Bill Graham. NMAJH is one of three venues who are hosting this retrospective of Graham's life which is

scheduled to remain in Philly through January 16, 2017. Graham played key roles in the careers of Rock legends like the Grateful Dead, Janis Joplin, Jimi Hendrix, Santana, Fleetwood Mac, the Who, the Doors, and the Rolling Stones just to name a few. For Graham, Rock & Roll was so much more than just music, it was an outlet through which people could promote humanitarian causes on a large scale. Of his many accomplishments, his role in organizing benefit concerts such as Live Aid in 1985 and Human Rights Now! in 1988, are some of those that are most globally noted.

The exhibit will feature more than two-hundred items of rock memorabilia, personal photos/items, vintage posters, preparatory show artwork, and much more. All of these are designed to shed light on the pivotal role that Rock played in the 60's, 70's, and 80's and to display Graham's part in these pivotal eras. The NMAJH is adjusting their hours on certain days to ensure that Philadelphia residents are able to easily access this unique exhibit despite busy schedules.

BRUCE SPRINGSTEEN STOPS IN PHILADELPHIA ON BOOK TOUR

As the misty rain fell, the line of pilgrims stretched down Vine Street, up 19th, wrapped around Callowhill, and started back down 20th. It was still several hours prior to Bruce Springsteen's scheduled arrival at the Philadelphia Free Library. Theirs were faces of a white upper-middle working class, in their 40s, 50s, 60s, and north. Most were from Philly, or the suburbs or Jersey, some had flown in from Florida or the West Coast, and a few from Europe just for the afternoon. Maybe a 60-40



women-to-men split, dressed in vintage concert t-shirts, jean jackets and leather, smiling under their umbrellas. They all seemed ready to burst from their skin.

On this third stop of Bruce Springsteen's short book tour for his new autobiography *Born to Run*, the 1200 allotted tickets were gone even quicker than to one of his concerts. There would be no book reading or talk, this was purely Springsteen pressing the flesh. For their \$33, each was guaranteed a moment with their hero, a quick word, a treasured selfie, a signed book.

The mythos of Springsteen has been well established from his stage stories: the early battles with his difficult father and his hard-won emergence from the “swamps of Jersey” into an international rock phenomenon. So you might expect more variance, more distant coolness in the chronological unfolding. But as a whole, it's a good bit like his famous marathon live shows. And it's surprising how often you get those soaring peaks, how readily it reads like the emotional experience of his music.

WINTER 2017 | AMPLIFIED

HOW DAVE HAUSE MARRIED PUNK AND AMERICANA ON MARVELOUS NEW ALBUM

Philadelphia singer-songwriter puts lyrics first on new LP

On his third solo album *Bury Me in Philly*, out now, Hause puts the emphasis squarely on emotional, vulnerable lyrics, proving that — like the Clash's Joe Strummer, Social D's Mike Ness and X's John Doe before him — the most punk thing you can sometimes do is bare your soul.

“The amount of time a guy like Gaslight's Brian Fallon or Hot Water Music's Chuck Ragan invest in a lyric, when you break down their songs to the core, they're every bit as sturdy as any Jason Isbell or Ryan Adams song,” says Hause, 38. “With Mohawks and leather jackets and the speed at which you're playing, the craft of songwriting can be lost in punk music. But today, punk is a bunch of guys in beards and flannels.”

outfit the Loved Ones, sprints off with the baton. It's an exhilarating album, a collection of 11 songs that challenge the listener before providing cathartic, hard-won release. “Helluva Home” is a harmonica-driven folk-rock about being away from Philadelphia; “The Ride,” with its Lou Reed-like sing-speak delivery, is about rushing into romance; and “Wild Love” recalls the balladry of Cyndi Lauper. Hause cites Lauper and Bryan Adams' Reckless as key influences and sees their impact today in a range of artists.

Throughout *Bury Me in Philly*, Hause sees his lyrics-first approach through to the end, reinforcing the Americana bent of the project. But even though he gushes over Patty Griffin (“She's the Holy Grail of lyricists”) and Jason Isbell



It's a tongue-in-cheek generalization, but one that becomes more apt as punk-leaning rock artists gravitate toward the Americana genre. Way back in 1999, Social Distortion's Ness explored country on *Cheating at Solitaire*; in 2002, New York hardcore fixture Jesse Malin released his introspective singer-songwriter debut *The Fine Art of Self Destruction*, produced by Ryan Adams; and last year Gaslight Anthem's Fallon explored heartland rock on his solo LP *Painkillers*.

Now, with *Bury Me in Philly*, Hause, the leader of on-again/off-again punk

(“He's got the gift”), and counts rootsy singer-songwriter Cory Branan as a close friend and collaborator, he maintains that he'll always be more punk than Americana — or country.

“There is a perceived credibility that comes with being in Americana. But for me to write an acoustic song and add fiddle and banjo is just absurd,” he says. “It would be more appropriate to put hip-hop beats under my lyrics than to put a country tinge on it. I'm not country — I'm from Philly.”

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REVIEW: PURLING HISS, 'HIGH BIAS'

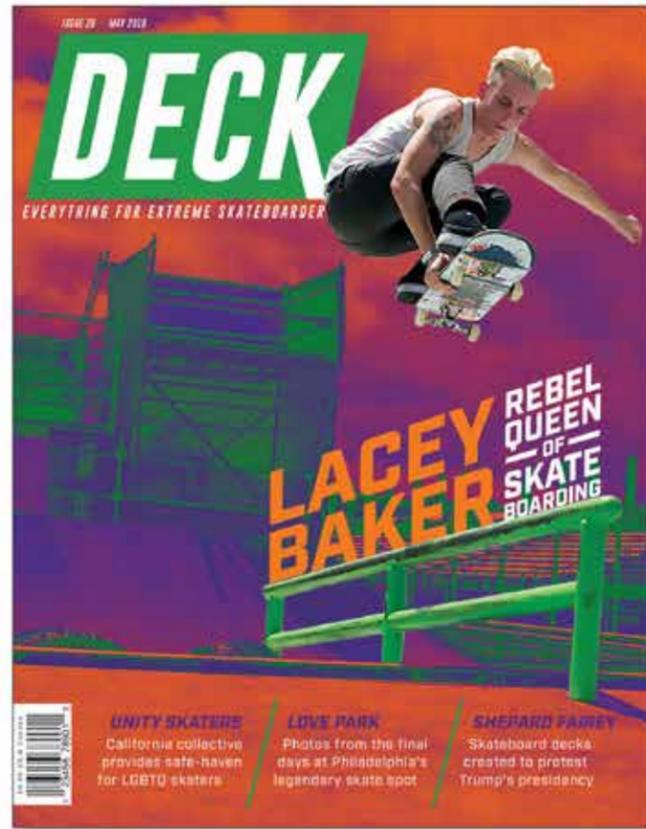
Philadelphia's Purling Hiss is now eight idiosyncratic albums into a remarkable career, and the new *High Bias* is the group's definitive work.

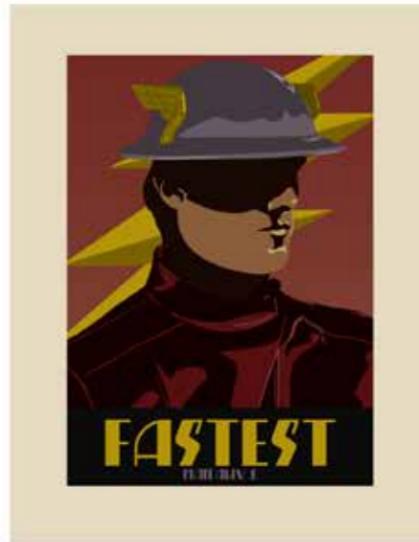
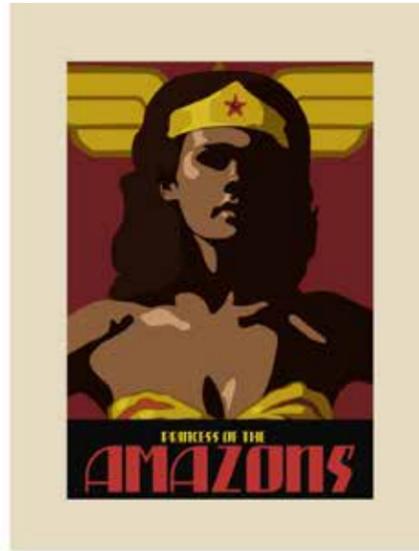
This is an unquestionably huge guitar record. In “3000 AD,” four steadily growing chords and a king-sized mattress of bass help form the kind of song FM radio once thrived upon. Polizze sings much of the record in a tuneful, upper-register bark that recalls John Lydon and Pete Shelley, in songs like “Pulsations” and “Notion Sickness” that spike *High Bias* with actual, no-joke punk speed and energy.

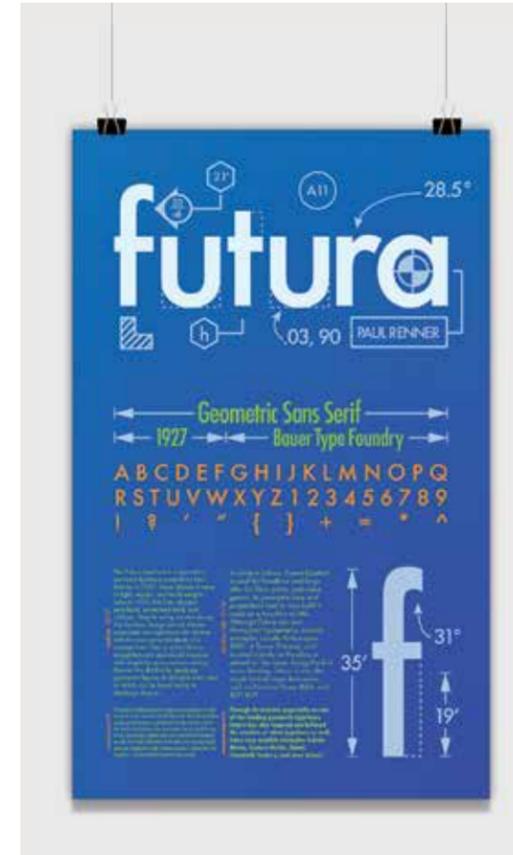
But for all its hints of Anglophilia, America is on Purling Hiss' mind. The slow-burning “Get Your Way” cannily cops the opening bars of “The Star-Spangled Banner,” while *High Bias* closes with 11-plus minutes of “Everybody In The USA,” in which Polizze exclaims, “Save me, I'm afraid of everybody in the USA.” As guitars race by in a generous chunk of frenetic soloing and hammer-ons, the song closes out an album that masterfully abstracts commonly held fears about today's world.

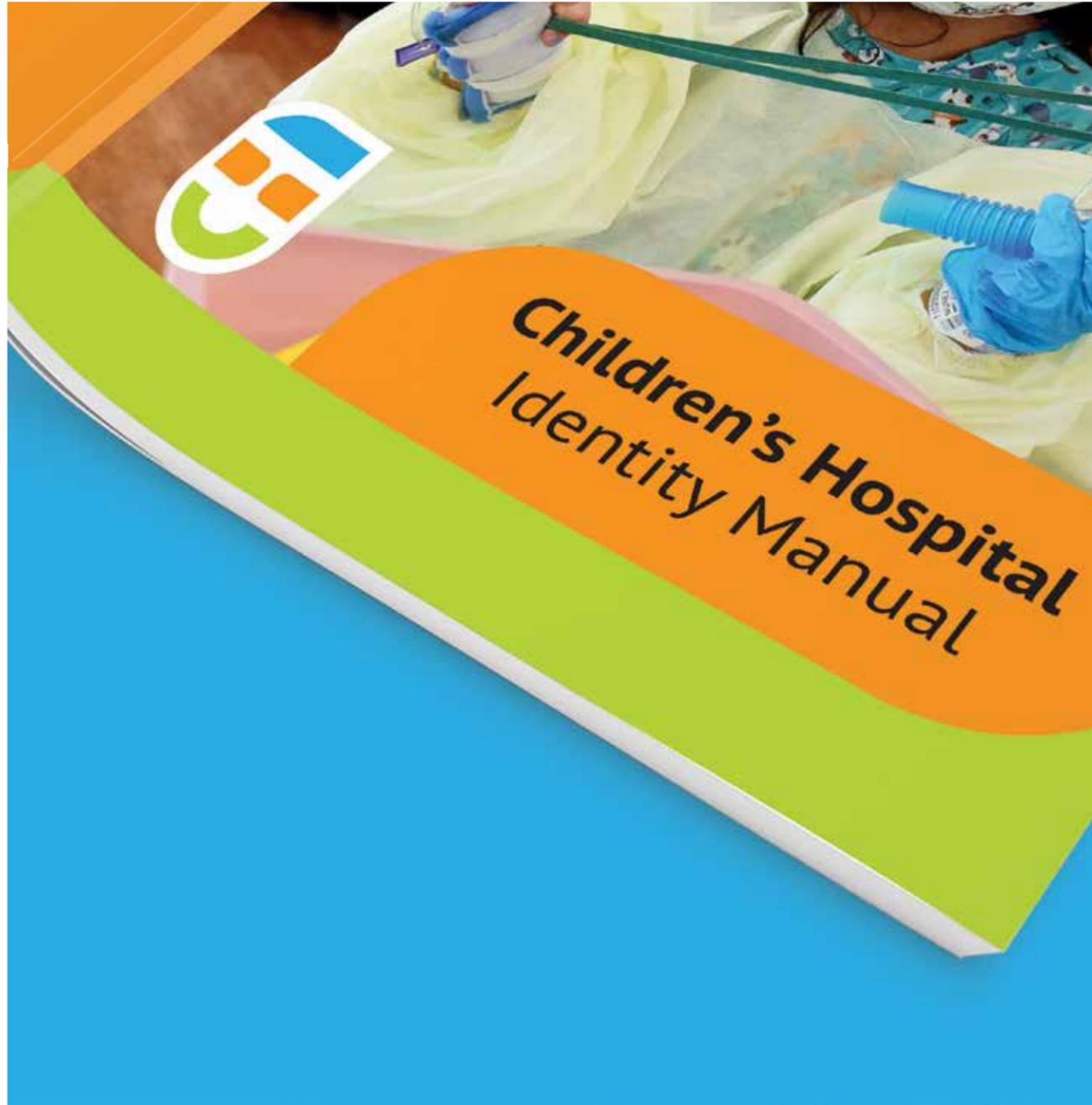
If rock music is your salve in hard times, it helps to find something that actually rocks. After casting a wide net over many styles in many years, Purling Hiss spends *High Bias* doing just that.

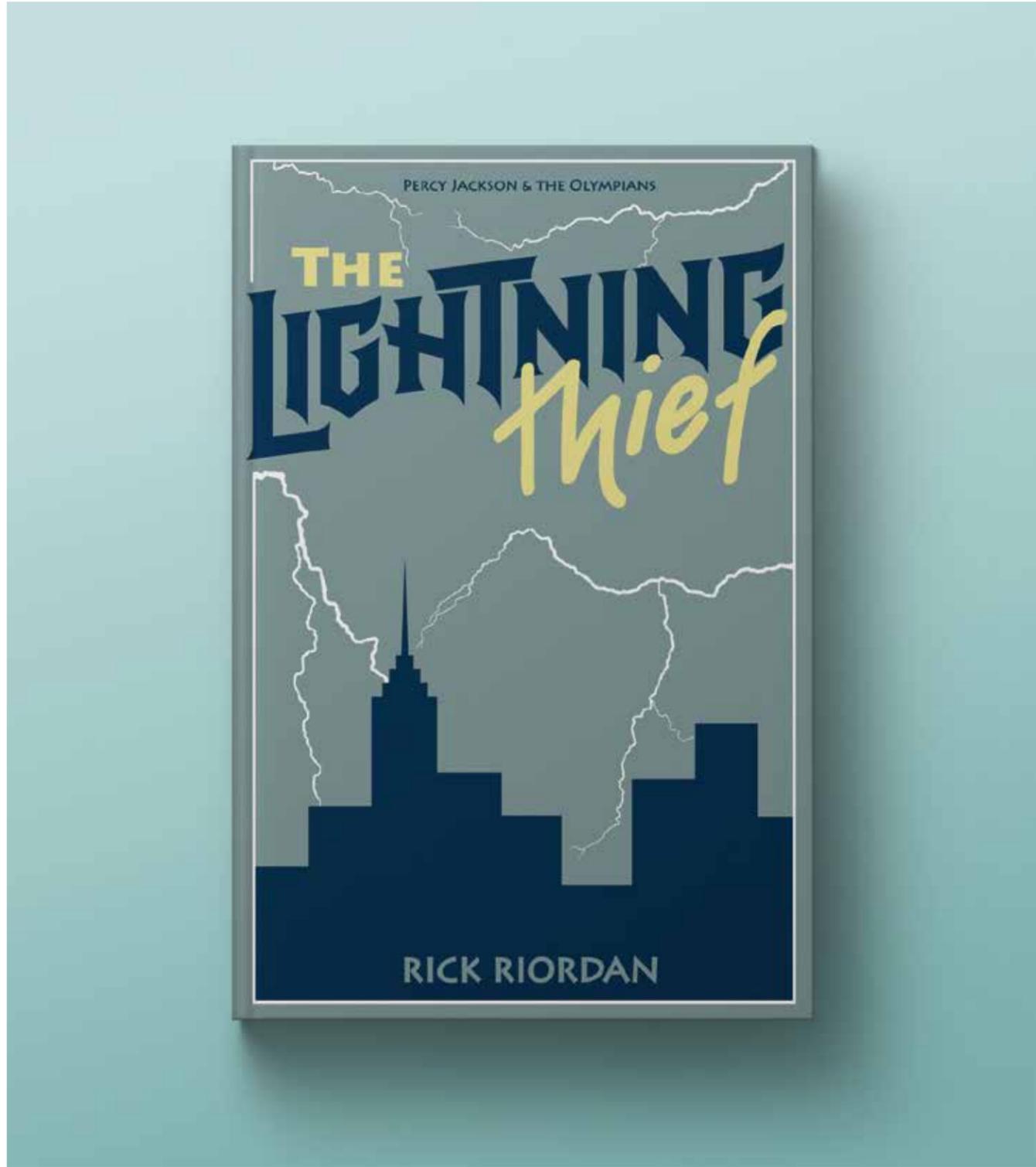
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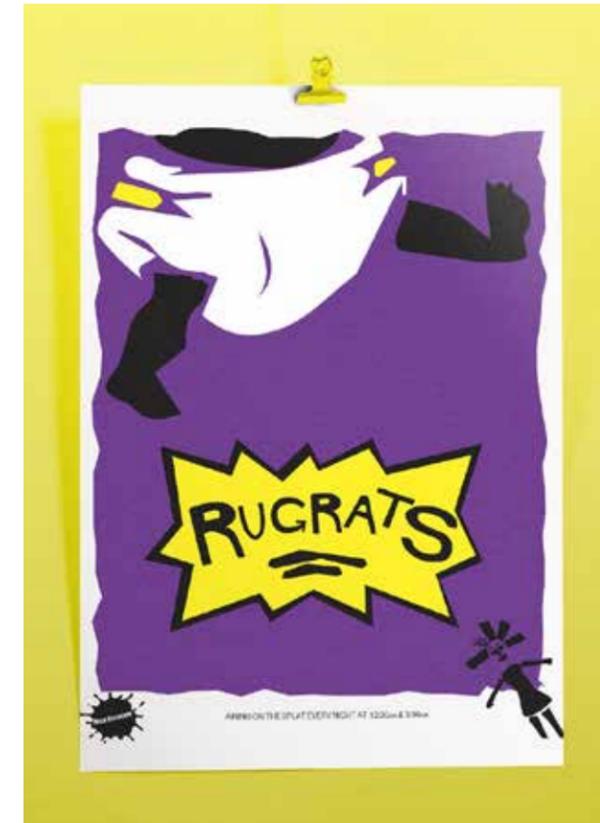






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