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RATIO

The academic journal of the next generation



**Ratio is the academic journal of the next generation, created by
students at Wheaton Academy.**

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*Ratio is published twice a year and accepts submissions from students.
Please send all submissions to lregan@wheatonacademy.org.*

What is Ratio?

Ratio is a journal dedicated to giving all students a space to publish their academic work. The word "ratio" is Latin for reason, and the journal welcomes all rational and reasonable discussion of the world around us. It is a space for physicists and philosophers, innovators and historians; all who want to speak toward what is true, beautiful, and good have a place in the conversation. Because the journal accommodates the passion and practice of diverse students, formatting styles may vary, tones may shift, and topics may contrast. But, in all cases, students will argue something about the world around them and, in doing so, will have the chance to participate in an academic conversation about what is real and relevant.

Ratio has another meaning as well. It suggests a proportion, one quantity in relation to another. Every student brings different experiences and training to the table. Some have wrestled with a topic or process for years, while others are just beginning. Generally, the more practiced thinker has more to offer. We, however, are not merely looking for the best work of experienced students. We want the best work of all students. It is not a question of quantity, but proportion. All work that approaches the capacity of the student is welcome. In the parable of the talents, Jesus describes the master's reaction to three different servants who were given three different sums of money. Regardless of what they received, it was those who doubled the deposit that received their master's praise. Only the one who kept his talent hidden was found lacking.

There are too many ways for student talents to remain hidden from those outside the classroom. We want to be a means by which students can grow what they have been given. We're excited to see them reason about the world and to engage with the tangible evidence of their efforts. We hope you enjoy being a part of it too.

Maddie Hazel

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Ellen Shales

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SELF-DIRECTED RESEARCH

Laboratory research is time-consuming, expensive, and, occasionally, maddening. Students must learn to ask the right kinds of questions, devote themselves to understanding the system they are studying, meticulously test an aspect of that system, fail, and then try again. These papers represent a semester spent in the midst of that process. From the initial question to the final analysis, these students conducted real research, and, in doing so, found a way to say something new about the world around them.

Examining circadian oscillators: effects of oxidized quinones and light exposure on growth rates in *S. Elongatus*

Ellen Shales

*Circadian rhythm is a mechanism commonly associated with daily sleep patterns in mammals. In actuality, the function of circadian rhythm encompasses a variety of cyclic patterns found within complex organisms down to simple bacteria. Regulation of downstream gene expression is controlled by a trio of Kai proteins, comprising the core oscillator. Kai proteins receive input from the redox state of the plastoquinone pool, thus revealing sensitivity to oxidized quinones. In this study, oxidized quinones were applied to *S. elongatus* cultures trained under various light environments. Results indicate the combination of oxidized quinones and specific light environments allow for visible and measurable changes in bacterial growth.*

Introduction

Circadian rhythm is a phenomenon found in thousands of creatures varying in complexity. This rhythm is most commonly understood in humans and is typically associated with sleep. While sleep is an expression of circadian rhythm particularly suited to humanity's needs, it is a narrow facet of a greater mechanism spanning many organisms.

Circadian rhythm is most often seen as a response to light and its inevitable fluctuations. In humans, sleep occurs during a dark period in which efficient work is more difficult to achieve. This sleeping period allows for recuperation and restoration in preparation for the continuation of the cycle, especially needed for optimal brain function. Beyond sleep in humans, circadian rhythm can be viewed in other organisms as a response to variations in light intensity. The majority of plant life must accommodate itself to the levels of light in their environment in addition to various other factors. These plants therefore adapt their cellular functions to optimize efficiency during the light and dark periods.

Circadian rhythm can also be viewed as the cyclic change of internal processes. Based on environmental cues, organisms format their activity around the unique nature of the surroundings. If a certain aspect of the environment is necessary for survival, this factor becomes the basis by which the circadian rhythm is established. For most organisms, this key ingredient is light. By this understanding, the

scale and depth of a circadian rhythm becomes more apparent.

While circadian rhythm can be a direct result of the environment, it also has the potential to be completely internalized, lacking external information input and triggers. Organisms lacking the structures imperative for light sensitivity show continued symptoms of a regulatory cycle. This regulatory cycle can trigger downstream processes at certain time intervals. Processes controlled by a circadian rhythm may involve alimentary related functions and low-activity versus high-activity periods.

Circadian Rhythm in Cyanobacteria

True circadian oscillator requires persistence, resetting, and temperature compensation. Thus, circadian rhythms were previously thought to be isolated to eukaryotic organisms. Prokaryotic organisms were believed to lack the complexity required to produce a circadian clock meeting the criteria. Bacteria in particular reproduce at a rate perceived to be sufficiently rapid to lack an integrated circadian oscillator (Kippert, 1987). Several rhythmic phenomena involving photosynthesis and nitrogen fixation were observed in strains of cyanobacteria. These observations, however, were attributed to cellular processes as opposed to the presence of a functioning circadian clock (Cohen and Golden, 2015). A model example of bacterial circadian oscillator emerged in 1986 in the discovery of nitrogen fixation and amino acid uptake in *Synechococcus* sp. RF-1 (Grobbelaar,

Huang, Lin, & Chow, 1986). This research reveals the presence of circadian rhythms in prokaryotes, disproving the previously held belief of sole eukaryotic possession of such mechanisms. The organism *Synechococcus elongatus* emerges as a premier model of studying prokaryotic circadian rhythm. (Cohen and Golden, 2015).

Mechanisms of the Circadian Oscillator in Cyanobacteria

The *S. elongatus* core oscillator is comprised of a three protein complex encoded by the *kaiA*, *kaiB*, and *kaiC* genes which regulate a variety of cellular processes such as global gene expression. (Ishiura, Kutsuna, Aoki, Iwasaki, Andersson, Tanabe, Golden, Johnson, and Kondo, 1998). Cell division timing and chromosome compaction have also been revealed to function under the guidance of the *kai* genes (Mori, Binder, and Johnson, 1996; Smith & Williams, 2006). The circadian oscillator remains independent from the cell cycle, and operates with a periodicity of ~24 hours despite the division rate of the cells (Mori et al., 1996). The oscillator does however regulate the timing of the cell division to define a portion of the early night in which cell division is prohibited (Dong, Yang, Wang, Kim, Wood, Osteryoung, van Oudenaarden, & Golden, 2010).

While eukaryotic organisms display circadian clocks based upon a transcription-translation feedback loop, cyanobacterial clocks function with a post-translational oscillator (PTO) involving the KaiABC proteins (Cohen & Golden, 2015). The interactions of KaiA and KaiB with the phosphorylation state of KaiC are the primary driving force of circadian rhythmicity. The KaiC protein is an autokinase and autophosphatase (Nishiwaki, Satomi, Nakajima, Lee, Kiyohara, Kageyama, Kitayama, Temamoto, Yamaguchi, Hijikata, Go, Iwasaki, Takao, & Kondo, 2004). The KaiA protein binds to KaiC during the illuminated portion of the day, promoting KaiC autokinase activity (Kim, Dong, Carruthers Jr., Golden, & LiWang, 2008). The KaiB protein counteracts KaiA's effects by drawing KaiA away from the KaiC protein, thus encouraging KaiC's autophosphatase activity (Kim et al., 2008).

The structural arrangement of the KaiB protein structure has a large effect on KaiC

phosphorylation activity, and imparts a necessary time delay for consistent 24 hour rhythm, as seen in a 2015 study conducted by Chang, Cohen, Phong, Myers, Kim, Tseng, Lin, Zhang, Boyd, Lee, Kang, Lee, Li, Britt, Rust, Golden, and LiWang. This study also showed that while KaiB typically exists as a tetramer, it was shown to switch to a monomer when binded to KaiC. This monomeric form of KaiB no longer exhibits a ground-state (gs) fold found in its common tetrameric form, but rearranges into a fold-switched (fs) state. Mutations supporting the fs-KaiB version induce rapid KaiC binding, as opposed to gs-KaiB, where it may take hours before a KaiB/KaiC interaction occurs. The transition from gs-KaiB to fs-KaiB is infrequent, thus allowing for a time delay of the entire circadian clock. Such a delay is vital to sustaining the 24 rhythm.

An interesting characteristic of a PTO like the KaiABC complex is its ability to be reconstituted *in vitro* (Nakajima, Imai, Ito, Nishiwaki, Murayama, Iwasaki, Oyama, & Kondo, 2005). Purified KaiA, KaiB, and KaiC genes combined with ATP permits oscillations in KaiC phosphorylation for days, even weeks (Nakajima et al., 2005). The total amount of Kai protein also remained constant, indicating that neither phosphorylated nor unphosphorylated KaiC proteins were degraded; thus KaiC phosphorylation is generated autonomously with the cooperation of the three proteins (Nakajima et al., 2005).

Inputs Although the cyanobacterial clock possesses a foundational oscillator that is self-sustaining, it retains a sensitivity to environmental cues, even allowing such stimuli to reset the clock (Cohen & Golden, 2015). The core oscillator proteins KaiA and KaiC are shown to directly detect metabolites of photosynthesis to synchronize the clock to the external environment (Wood, Bridwell-Rabb, Kim, Gao, Chang, LiWang, Barondeau, & Golden, 2010). Forgoing the direct use of photoreceptors as seen in eukaryotic organisms, cyanobacteria also indirectly sense the day-night cycle through the redox state of the plastoquinone pool, a result of photosynthetic activity in relation to light intensity variations (Cohen & Golden, 2015). The redox state of the plastoquinone pool changes depending on the cyanobacterial photosynthetic

activity, specifically the pool becomes oxidized with the onset of darkness (Kim, Vinyard, Ananyev, Dismukes, & Golden, 2012). A prominent redox-sensitive input pathway was found in circadian input kinase A (CikA) protein (Schmitz, Katayama, Williams, Kondo, & Golden, 2000). The fact that cikA mutants are unable to reset gene oscillation expression after a 5 hour dark pulse further solidifies CikA's status as an input pathway (Schmitz et al., 2000). CikA levels vary inversely with the intensity of light, showing high levels during the night and low levels during the day (Ivleva, Gao, LiWang, & Golden, 2006). CikA proteins bind directly to oxidized, but not reduced, quinones (Ivleva et al., 2006). The addition of oxidized quinones mimics darkness, and when applied during the day resets the circadian clock both *in vivo* and *in vitro* (Ivleva et al., 2006). Oxidized quinones bind directly to the pseudo-receiver (PsR) domains in CikA as well KaiA itself (Wood et al., 2010). Aggregation and degradation of KaiA and CikA as a result of the addition of oxidized quinones is a potential mechanism by which the clock adjusts to the day-night cycle (Wood et al., 2010). Oxidized quinones affect KaiA's ability to stimulate KaiC phosphorylation, thus impacting the entire circadian oscillator (Wood et al., 2010).

The circadian clock, in addition to receiving input from the cellular redox state, receives cues about the environmental state through the metabolites of photosynthesis (Cohen & Golden, 2015). Rust et al. showed that KaiC directly senses the ATP/ADP ratio of the cell over the course of the day. In addition they showed that cells experience a decrease in the ATP/ADP ratio over the course of the subjective night or during a dark pulse, irrespective of the time administered. This characteristic is a typical circadian response. The Kai oscillator showed heightened sensitivity to changes in ATP/ADP during the middle of the subjective day, corresponding to a point when KaiC phosphorylation was increasing. This input method combined with the redox status of the quinone pool contribute to the circadian clock adapting to the light levels and thereby environment (Kim et al., 2012).

Outputs The output pathways from the core Kai oscillator rest centrally on the histidine

kinase SasA and cognate response regulator RpaA (Iwasaki, Williams, Kitayama, Ishiura, Golden, & Kondo, 2000). The SasA and RpaA output pathways are required for global gene expression and gating cell division (Dong et al., 2010). The rhythms of chromosome compaction, however, are independent of the SasA pathway, suggesting other output functions (Smith and Williams, 2006). Loss of sasA mainly results in low-amplitude rhythms of gene expression, solidifying its role as an output pathway (Cohen and Golden, 2015). SasA interacts with KaiC through its N-terminal domain, similar to that of KaiB, which provokes SasA autophosphorylation and phototransfer to RpaA (Iwasaki et al., 2000). Once gs-KaiB changes to fs-KaiB conformation, KaiB and SasA compete for binding sites of KaiC (Tseng, Chang, Bravo, Latham, Chaudhary, Kuo, & Liwang, 2014). fs-KaiB has been shown to displace SasA from KaiC, implying its role in regulating and timing of the output functions (Chang, Cohen, Phong, Myers, Kim, Tseng, Lin, Zhang, Boyd, Lee, Kang, Lee, Li, Britt, Rust, Golden, & LiWang, 2015). fs-KaiB can also interact with the PsR domain of cikA, and this association stimulates the phosphatase activity of cikA towards RpaA (Chang et al., 2015). KaiA, through KaiB, also displaces SasA from KaiC, thus posing a role in the output pathways (Tseng et al., 2014).

While the loss of sasA produces altered gene rhythmicity, the loss of rpaA destroys the rhythm of gene expression entirely (Iwasaki et al., 2000). This fact illustrates RpaA's importance to the circadian output pathways. The protein RpaA functions as a DNA-binding response regulator, binding to ~100 targets in the *S. elongatus* genome while in its phosphorylated state (Markson, Piechura, Puszyńska, & O'Shea, 2013). The binding of RpaA to these target sites occurs in phases (Cohen and Golden, 2015). The RpaA regulon consists of over a hundred transcripts, including the expression of 8 transcription factors. These targets are likely to be more important for genome-wide rhythmicity amongst locations not directly under RpaA influence (Markson et al., 2013). The phosphorylated form of RpaA is shown to be the active state both for driving gene expression rhythms and the gating of cell division. Rhythms of KaiC phosphorylation persist in the absence of

rpaA, although gene expression is arrhythmic (Markson et al., 2013). This suggests the loss of gene expression is not due to an effect on the oscillator itself, thus further identifying RpaA as a downstream clock component (Cohen and Golden, 2015).

Circadian Effects on Glycogen Levels

The downstream effects of the circadian oscillator are most commonly studied through the cell's glycogen levels. Diamond, Jun, Rubin, and Golden provide a model (2015) of the oscillations of glycogen abundance of cells grown in a 24-h light-dark cycle. While these oscillations may be a product of environmental cycles, the circadian clock most likely is revealing its influence. Glycogen accumulation in kaiC mutant cells occurs significantly faster over a 12-h light period as opposed to the wild type cells in the same conditions. The kaiC mutant also displayed fluctuating rates of glycogen accumulation, whereas the wild type counterpart held a consistent rate over the full 12-h period. In contrast, both the kaiC mutant and wild type strain showed similar results in glycogen degradation. While the kaiC mutant showed a slightly faster rate of degradation, both strains degrade a similar fraction of glycogen within their overnight period. These data illustrate the circadian oscillator's refinement of the timing of glycogen accumulation to occur at a constant rate, while darkness is adequate to instigate glycogen degradation. In another study conducted in 2014 by Pattanayak, Phong, and Rust, it was shown that the circadian clock forces cells to accumulate and degrade glycogen during the subjective day and night, respectively, even in constant light conditions. It was also shown that the cellular energy, or ATP/ADP ratio, falls faster and reaches lower levels when a dark pulse occurs near subjective dawn, when darkness is not anticipated, as compared to subjective dusk. The results of these two studies depict the importance of the circadian oscillator's role in downstream processes, such as glycogen accumulation and degradation. These results also, therefore, remove the notion of the circadian clock as an isolated mechanism with minimal, overall cellular effects.

Summary

Through the input pathways of KaiA and CikA, the core oscillator can be targeted with the use of oxidized quinones. Oxidized quinones bind to the PsR domain of KaiA and prevent its stimulatory effect on KaiC phosphorylation. This impacts the driving force of the oscillator itself. The combined effect of oxidized quinones on KaiA and CikA mimics a dark pulse that resets the circadian clock entirely. It was also shown that the circadian clock controls glycogen levels, thereby playing a role in the metabolic cycles. The question then presents itself: can a lack of circadian rhythmicity reveal itself in completely external symptoms, such as population growth? Or does a lack of rhythmicity only express itself through minute gene expression levels, contained to the cellular level? These questions comprise the fundamental pursuit of this study.

Methods

While the majority of existing research on the Kai oscillator targets change at the molecular level, this study seeks to examine changes in overall growth rate in response to changes in light, as well as the addition of oxidized quinones during specific transitions during the circadian rhythm.

Preparation of *Synechococcus elongatus* culture

Synechococcus elongatus culture was obtained from UTEX and cultured according to the recommendations listed in the UTEX Culture Maintenance Guide. Specifically, in preparation for exposure to the oxidized quinones, a 3 mL stock culture of was created using BG-11 growth medium. Cultures were grown in ambient light levels for nine days, and on the tenth day the solutions were rocked at 120 RPMs. After one day of rocking, nine separate subcultures containing three milliliters of BG-11 growth media and 100 μ L of starter culture were established.

Preparation of Oxidized Quinones

A 1,000,000x solution was created from 0.5 grams of oxidized quinones dissolved in 1.55 mL of 100% ethanol.

Light Environment and Exposure Conditioning

The bacteria were grown in a 20 °C environment with warm white lights. These lights were chosen according to the UTEX Culture Maintenance Guide. To reduce the impact of ambient light, three separate chambers were created. Each chamber consisted of thick draft board with dimensions of 19 inches by 6 inches by 6 inches. An opening was made in the top of the chamber and fitted with a light, designed to stabilize the light levels. A 6 watt, 650 lumen, 110° radiation angle flood light was suspended in each box, 13 inches away from the bacteria. The light intensity was 2225 lux. Outlet timers were used to create different 12 hr periods of light in two of the three chambers.

To train different cultures of *S. elongatus* to correspond to different periods of light and dark, cultures were placed into each chamber and exposed to different light conditions. The first environment was constant, 24 hour light. The second was a 12 hour light cycle on a 6 am to 6 pm schedule. The third was a 12 hour light cycle on a 6 pm to 6 am schedule.

Determination of initial growth rates

The bacterial subcultures were placed into their respective light environments, 3 per environment. Before the establishment of subcultures, a hemocytometer was used to identify colony area of the initial starter culture from a 10 µL sample. A 1 square mm slide was used to determine colony size for each test. Successive hemocytometer tests were administered on days 6, 9, and 15 following the establishment of the nine subcultures, labeled D6, D9, and D15, respectively. For each of these tests, a 10 µL sample from each of the nine subcultures was measured and the results averaged by light environments yielding three values for colony area per test.

Testing

A second round of subcultures was created as outlined above, with instead 6 subcultures per environment. The subcultures were grown for 15 days total. Dosing of the oxidized quinones began on D7 and lasted until

D15. A spectrophotometer and hemocytometer test were run on each subculture on D1, D4, D7, D10, D13, D15. The spectrophotometer test was programmed to identify levels of 550 nm wavelength light. The first set of three subcultures in each environment received 3.1 µL of 100% ethanol at 6 am on D8-D15. The second set of three subcultures received 3.1 µL of the oxidized quinone solution at 6 am on D8-D15. Dosing on D7 occurred at 2 pm with 9.25 µL of solution applied. Application of the oxidized quinones and the 100% ethanol occurred every day within the D7 to D15 period. Due to evaporation, the ethanol levels in the oxidized quinone solution decreased with time. To combat this, the ethanol was refilled to the calculated amount on D11.

Data Analysis

A repeated-measures ANOVA was used to compare the spectrophotometer readings among the various treatments. Significant differences between individual treatments were identified using Tukey post-hoc analysis.

Results

Hemocytometer data indicate the total area (in mm) of bacterial colonies taken from a 10 µL sample measured across 1 square mm. While the hemocytometer measurements vary throughout the course of the experiment, observations of the cultures themselves suggested that the hemocytometer was not accurately measuring colony growth. In contrast, the spectrophotometer readings suggest growth across treatments and conditions (Table 2). The decrease in percent transmittance indicates bacterial growth in each culture, and growth continued across the 15 day testing period, for all subcultures. The 24 hour control as compared to the 6 am to 6 pm control ($p = 0.8863$) and the 6 pm to 6 am control ($p = 0.9036$) show little difference in overall growth rate. It appears that the total duration of light, whether 24 hour or 12 hour, has insubstantial effects on growth.

TABLE 1. Bacterial growth by area (mm²) from day 1 (D1) to day 15 (D15)

CONDITIONS	TREATMENT	D1		D4		D7		D10		D13		D15	
		MEAN	SD	MEAN	SD	MEAN	SD	MEAN	SD	MEAN	SD	MEAN	SD
24 Hour	0	0.004	0.006	0.001	0.001	0.002	0.002	0.0117	0.008	0.006	0.002	0.0119	0.004
	10 μ M	0.001	0.000	0.002	0.002	0.002	0.002	0.0163	0.009	0.011	0.007	0.0219	0.011
6 PM - 6 AM	0	0.000	0.000	0.001	0.001	0.001	0.001	0.0023	0.002	0.024	0.039	0.0044	0.004
	10 μ M	0.000	0.000	0.001	0.001	0.000	0.001	0.0015	0.001	0.011	0.014	0.0090	0.010
6 AM - 6 PM	0	0.000	0.000	0.000	0.000	0.004	0.003	0.0059	0.005	0.011	0.008	0.0056	0.007
	10 μ M	0.000	0.000	0.000	0.000	0.012	0.010	0.0098	0.005	0.010	0.003	0.0248	0.010

TABLE 2. Percent transmittance of light from day 1 (D1) to day 15 (D15)

CONDITIONS	TREATMENT	D1		D4		D7		D10		D13		D15	
		MEAN	SD	MEAN	SD	MEAN	SD	MEAN	SD	MEAN	SD	MEAN	SD
24 Hour	0	101.700	0.300	101.067	0.404	92.700	3.470	83.100	2.666	83.767	5.239	75.800	6.444
	10 μ M	99.800	0.917	99.867	2.268	92.867	3.017	70.533	2.710	54.633	1.656	35.300	2.615
6 PM - 6 AM	0	101.300	0.200	101.267	0.306	98.633	0.231	88.100	1.308	84.400	1.572	79.367	2.325
	10 μ M	99.467	0.462	99.700	2.488	98.967	1.779	84.167	2.926	72.767	3.362	54.600	5.745
6 AM - 6 PM	0	100.267	1.332	100.200	0.400	90.633	4.508	83.033	11.609	83.300	8.073	64.967	2.074
	10 μ M	100.500	0.624	100.967	0.950	95.100	3.900	72.533	0.929	59.000	9.822	31.733	6.824

Discussion

The hemocytometer data were too irregular for useful analysis. The spectrophotometer results showed clear and understandable correlations. The spectrophotometer for use as a bacterial growth determiner appears more accurate and precise for data measurements. All discussion of the data will concern these readings.

Overall, the subcultures dosed with oxidized quinones increased in bacterial numbers as compared to the control within all light conditions. This was unexpected, as the bacteria were predicted to be jarred by chemical signaling of darkness, thereby disrupting cellular functions such as reproduction. The oxidized quinones were expected to negatively affect conditions followed by light, and leave conditions followed by darkness relatively undisturbed. The data prove however that growth was significantly enhanced when the application of oxidized quinones directly preceded a transition to light ($p = 0.0005$) or continued light exposure ($p < 0.0001$). The condition projected to adapt the fastest, 6 pm to 6 am light, resulted in the lowest bacterial levels. The bacteria in the 6 pm to 6 am light cycle should have anticipated physical darkness and acclimated their cellular processes accordingly, however these data prove otherwise. In addition to the duration, the light condition prior to the application of oxidized quinones seemed to have little effect. The two conditions with the highest final growth were the 24 hour and 6 am to 6 pm light. These two conditions were in light and dark respectively just before the quinones were added, yet lack pronounced differences in growth ($p = 0.9970$). The crucial variable that presents itself then becomes the light condition directly after dosing. The presence of light is the factor that the 24 hour and 6 am to 6 pm conditions share. This factor was predicted to inhibit bacterial growth as the oxidized quinones induced darkness, but the data seem to suggest the contrary. The cause of this unpredictable result is unknown. For the 6 am to 6 pm condition, the bacteria may encounter the oxidized quinones' chemical darkness inducement as an extension of the true dark period and correspondingly acclimate. For both conditions, the disruption of rhythmicity may actually encourage processes tied to

replication and growth. The true reason behind this complete antithesis of the envisioned results in yet to be understood.

Limitations in the experimental setup mainly arose from the oxidized quinone solution. The quinones did not dissolve in ethanol, allowing for constant separation of liquid and solid. The solution itself was also yellow in color. This variable, however, did not affect the spectrophotometer readings as each subculture received equal dosing amounts. Preferably however, future study would eliminate this variable entirely.

Further studies may investigate the mechanisms behind the bacteria's unexpected response to the addition of oxidized quinones. By nature of the aim of this study, the simplicity in scale and equipment prevents a full understanding of such a response. The presence or absence of light after the application of oxidized quinones propounds a particularly notable avenue of research.

This study was designed to search for well understood molecular effects on a macroscopic level. The minimal equipment and lack of complexity in design further compounds the gravity of its findings. *S. elongatus* is a model organism for studying circadian rhythmicity in simplistic life forms. The bacteria's circadian processes are visible enough to research on a holistic and larger scale, as exemplified by this experiment. The possibility then arises for circadian disruption in increasingly complex organisms, and the ensuing impact on widespread health and functionality. While this study itself unearths meaningful results, the consequences it unleashes for future study of circadian rhythm are truly preeminent.

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The Testing of a New Natural Indicator Made from Lavender Lantana

Phyllis Lin

Introduction

pH indicators are commonly used to indicate the completion of chemical reactions during titrimetric analysis in research laboratories. Often, they use a color change to determine the equivalence point between the reacting species. For example, pH indicators used in conjunction with a solution of a known concentration are often used to determine concentrations of unknown solutions as the medium shifts from acid to base, or vice versa (Waghmare et al., 2013). Because of this useful property, pH indicators are used in labs throughout the world.

The commonly-used synthetic indicators, however, have some toxic and hazardous effects to the users as well as the environment which go generally unnoticed or ignored. For example, the commonly used indicator phenolphthalein ($C_{20}H_{14}O_4$) is a chemical agent, widely used in the acid-base titration. Phenolphthalein, however, has carcinogenic properties which may cause ovarian cancer (Garg et al., 2018). While it remains a useful indicator, its potential harm to humans as well as the surrounding environment is a cause for concern.

Methyl orange is also used as an indicator, which may cause local skin destruction or dermatitis. The repeated exposure of the methyl orange may cause lung damage and also may produce irritation in the eye (Garg et al., 2018). Methyl red also can produce cancer and neurological disorders and is understood to be generally toxic (Garg et al., 2018).

Taken together, these findings indicate that synthetic indicators can harm humans. Because of these unwanted and toxic effects, recent research has supported the viability of natural, non-toxic alternatives to synthetic indicators. Consequently, this focus on natural alternatives may enhance the attention paid to

natural sources of indicators that can be used in the lab (Abugri et al., 2012). To exist as a viable alternative to synthetics, these natural indicators would need to be readily available, easy to prepare, simple to extract, less toxic, inexpensive and eco-friendly. Additionally, to replace synthetic indicators, natural alternatives would need to produce comparably accurate results in the lab, have measurably fewer adverse health effects, and cost roughly the same.

Literature Review

The majority of indicators in use today are synthetic substances used to determine the pH of a material. Synthetic indicators have certain disadvantages such as high cost, availability, and chemical pollution; hence natural indicators obtained from various plant parts like flowers, fruits, and leaves will be more advantageous (Abugri et al., 2012).

Classification of Natural Indicators

In general, indicators are classified according to a few criteria. The study of classifying natural indicators is performed using analytical grade reagents, and the whole experimental work is carried out using the same set of glassware used for extraction and titration process (Garg et al., 2018).

According to Ghatage S. Trupti, Killedar G. Suresh, Hajare A. Mayuri, and Joshi M. Mrunalini (2017), the natural indicators are often classified according to the following:

1. By the Chemical Structure of the Chromophore

A chromophore is the part of a molecule responsible for its color. Classifying indicators by the chemical structures are incredibly useful because the chromophore is always involved in the color changes of an indicator. Natural indicators can be classified by taking into account the chromophore chemical structure with

conjugated systems as carotenoids, anthocyanins, betalains, caramel, synthetic indicators, and lakes, and metal-coordinated porphyrins which consist of myoglobin, chlorophyll, and their derivatives.

2. By the Structural Characteristics of the Natural indicators

Natural indicators can also be classified by their chemical structural characteristics. A chemical structure determination includes a chemist's specifying the molecular geometry and, when feasible and necessary, the electronic structure of the target molecule or other solid. The first group, called tetrapyrrole derivatives, has chlorophylls and theme colors. The second group, isoprenoid derivatives, includes carotenoids and iridoids. Additionally, the group of N-heterocyclic compounds has purines, pterins, flavins, phenazines, phenoxazines, and betalains. The next group is Benzopyran derivatives (oxygenated heterocyclic compounds), which includes anthocyanins and other flavonoid indicators. Last, the quinones contain benzoquinone, naphthoquinone, anthraquinone, and melanins.

3. By Their Origin

Indicators can be classified by origin as either natural, synthetic, or inorganic. Natural indicators are produced by living organisms such as plants, animals, fungi, and microorganisms. Synthetic indicators are obtained from synthetic chemical reactions in the lab. Natural and synthetic indicators are organic compounds. Inorganic indicators can be found in nature or reproduced by synthesis.

Preparation of Natural Indicator

Whether they are natural or synthetic, indicators are often prepared using a few established methods. The first step to prepare the natural indicator is collecting the different parts of the plants for the chemical in the natural indicator. The common plants collected this way are watermelon, fruit, red sandal bark, pomegranate seeds, hibiscus, calendula, lantana flowers, beet and carrot roots.

Take the lantana flower as an example (Cite Garg et al, 2018). To create an artificial

indicator from the lantana plant, all the flowers are collected and cleaned separately with distilled water, and petals of these flowers are kept in sunlight until they are withered entirely. The dried petals are ground into a fine powder with a mechanical blender and then ground in mortar and pestle and individually macerated in ethanol for 24 hours. The resulting solution is filtered through a muslin cloth, and the resulting extract is used as a natural indicator for acid-base titration (Garg et al., 2018). After a sufficient time interval, the filtrate is collected and the resulting extract is used as a natural indicator in various titrations (Ghatage et al., 2017). The extract of different flowers is preserved in an amber-color glass bottle with a tightly closed container and stored in room temperature or in a cool place away from sunlight.

Pre-Test of Natural Indicator

After the preparation of an indicator, but before titration, it is necessary to record the characterizations of the extracts (Journal of Advanced Research, 2017):

1) Observation of color and pH.

The extracts obtained should be observed in light, and color produced should be noted for all the extracts. Also, the pH of extracts obtained should be determined using a digital pH meter at room temp (27.50C).

2) pH sensitivity test.

All the extracts should be tested for pH sensitivity test using 0.1N HCl and NaOH solutions to understand the color changes in alkaline and acidic medium respectively which will help to know the suitability of extracts for food colorants and titrimetric analysis.

3) Effect of temperature:

All the extracts should be subjected to stability study by exposing to the different temperatures keeping in a thermostatic hot air oven at different temperatures for 1h.

Comparing the Accuracy of Natural and Synthetic Indicators.

Several studies by various investigators have reported the effectiveness of natural indicators in acid-base titrations (Ghatage et al., 2017). In general, titrimetric analysis with neutralization process follows the following procedure (Journal of Applied Pharmaceutical Science, 2014). The titrimetric analysis begins with the neutralization of a strong acid and a strong base. Typically, this involves the reaction of 10 mL of 0.1 M HCl and 50 mL of 0.1M NaOH at 25°C. The second titrimetric analysis is the neutralization between a strong acid and a weak base. This involves the reaction of 10 mL of 0.1M HCl reacts with 50 mL of 0.1M ammonia at 25°C. The third titrimetric analysis involves the neutralization of a weak acid and a strong base, which is the reaction between 10 mL of 0.1M acetic and 50 mL of 0.1M NaOH at 25°C. The last titrimetric analysis is the neutralization of a weak acid and a weak base. This involves the reaction of 10 mL of 0.1M acetic and 50 mL of 0.1M ammonia at 25°C.

Cost Comparison of Natural and Synthetic Indicators

To make natural indicators competitive, 100mL of natural indicator made in the lab should not be higher than \$10.00. The cost of the lavender lantana is \$19.99, which should be considered in the future study.

Comparing the Health Effects of Natural and Synthetic Indicators

Also, some of these synthetic indicators have toxic effects on users such as diarrhea, pulmonary edema, hypoglycemia, and pancreatitis and they can result in abdominal cramps, skin rash, eruptions, erythema, and epidermal necrosis and cause environmental pollution (Waghmare et al., 2013). Beyond the natural indicators used in high school chemistry classes, there is room for further development of natural pH indicators that serve as safe and effective alternatives to synthetic ones.

To understand deeply about the negative health effect of synthetic indicators and indicators, it is necessary to take further study for

analyzing the specific kind of molecule and their chemical structure.

Question/Hypothesis

Considering the potential for natural indicators to serve as functional, safe, and cost-effective replacements for the synthetic indicators commonly used in the lab, this comparative study of natural and synthetic indicators seeks to answer the following questions:

- Can the indicator produced from the lavender lantana's flowers accurately assess the pH of various compounds through neutralization reactions listed above?
- Does the indicator produced from the lavender lantana's flowers remain stable over an extended period of storage and regular testing?
- Can an indicator produced from the flowers of lavender lantana be produced at a cost that would make it a competitive alternative to the synthetic indicators that are currently used in the lab?

Although the chromophore structure and health impacts of the natural indicator will stay undetermined after the experiment, this preliminary analysis of a novel natural indicator seeks to provide enough information to determine whether or not the natural indicator produced is worthy future study.

Methodology

25 grams of lantana flowers were collected per the availability. Extraction was ground in mortar and pestle and macerated in 10 mL ethanol for 24 hrs, and then stored inside a brown bottle in the refrigerator. The pH of the color extracts was determined using digital pH meter and logger pro, and titrations were completed using calibrated burettes and pipettes.

The developed indicator tested for all four types of acid-base titrations: strong acid vs. strong base (HCl vs NaOH), weak acid vs. strong base (acetic acid vs NaOH), strong acid vs. weak base

(HCl vs. ammonia), and weak acid vs. weak (acetic acid vs ammonia). The sharp end point was observed for all types.

First, strong acid-strong base: 10 mL of 0.1M HCl was taken in a conical flask and 50 mL 0.1M NaOH was taken in the burette. Two to three drops of Natural indicators along synthetic indicators were added, and titration was carried out until the change in color of the solution of the flask.

Second, strong acid-weak base: 10 mL of 0.1M HCl was taken in a conical flask and 50 mL of 0.1M ammonia was taken in the burette. Two to three drops of Natural indicators along synthetic indicators were added, and titration was carried out until the change in color of the solution of the flask.

Third, weak acid-strong base: 10 mL of 0.1M acetic acid was taken in a conical flask, and 50 mL of 0.1M NaOH was taken in the burette. Two to three drops of natural indicators along synthetic indicators were added, and titration was carried out until the change in color of the solution of the flask.

Last, weak acid-weak base: 10 mL of 0.1M acetic acid was taken in a conical flask, and 50 M of 0.1 mol ammonia was taken in the burette. Two

to three drops of natural indicators along synthetic indicators were added, and titration was carried out until the change in color of the solution of the flask.

Each of the four neutralization tests was run three times at the same time. The final result for each neutralization test is the average pH value of three runs.

Results

10 mL of the natural indicator was created from lavender lantana. When combined with 10 mL of HCl, the indicator changed to a light pink color. Likewise, in the presence of 10 mL of NaOH, the indicator turned yellow (see figure 1).

Additional tests were run with different acids and bases, and, regardless of the type of acid or base used, the indicator consistently turned pink in the presence of acids and yellow in the presence of bases (see table 1).

pH analysis software determined the pH of the natural indicator to be 5.15. When the natural indicator started to change to pink in the acidic solution (HCl), the pH of the solution was 2.84. When the natural indicator started to change to yellow in the basic solution (NaOH), the pH of the solution was 10.56.

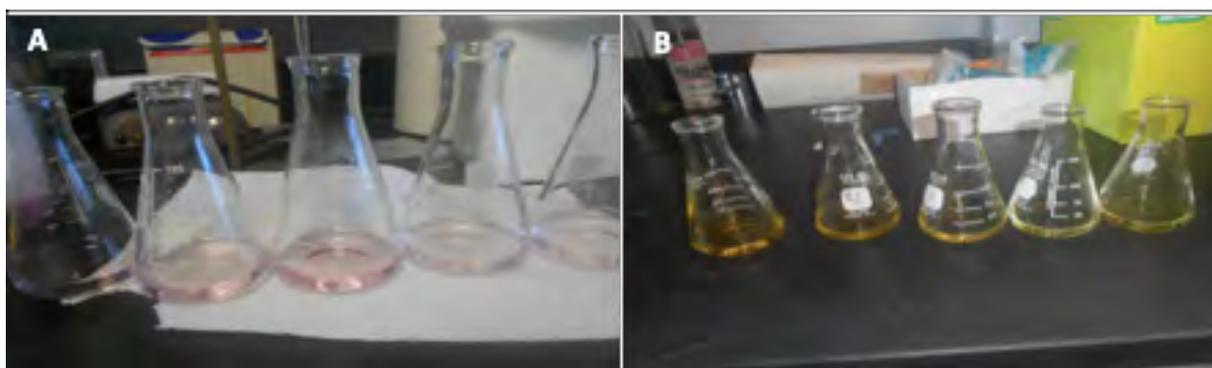


Figure 1. A) Acid (HCl or CH₃COOH) plus three drops of natural indicator results in the color changing to pink. B) Base (NaOH or ammonia) solutions plus three drops of natural indicator results in the color changing to yellow.

In order to have a better understanding of the color change of nature indicator, the UV-Vis Spectrum was run with the different pH values, resulting in absorbances at different wavelengths.

Color changes occurred at different wavelengths. Mainly at the acidic range, it showed λ value of $530 \pm 5 \text{ nm}$ while basic pH range

showed λ value of $400 \pm 5 \text{ nm}$ (Figure 2). These color changes were observed with the naked eye only, which may reveal indicator sensitivity towards pH. Over an extended period of storage and regular testing, the natural indicator produced from the lavender lantana flowers become less efficient by record the volume used for and the observation of the color change by testing indicator with 10 mL HCl/NaOH (Table 2).

Table 1. Tests of the natural indicator suggest a consistent color change in both acids and bases.

Substance	Nature	Original Color	After extract addition
1 M HCl	Acidic	Colorless	Pink
1 M NaOH	Basic	Colorless	Yellow
Commercial Vinegar	Acidic	Colorless	Pink
Commercial baking soda	Basic	Colorless	Yellow
Soap	Basic	Colorless	Yellow

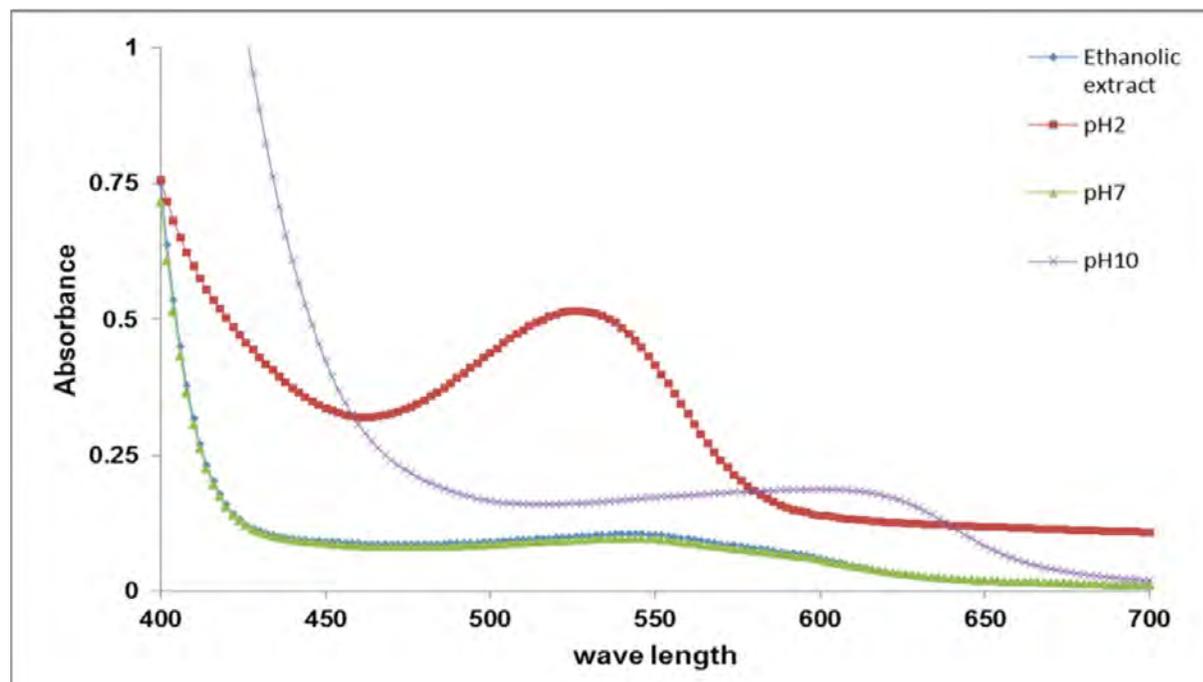
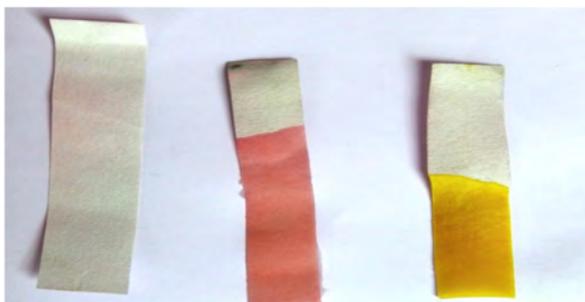


Figure 2. UV-Vis spectrum for various pH ranges, which results in different absorbances and wavelengths.

Table 2. Natural indicators stability over an extended period of storage and regular testing.

Days after the natural indicator was made	volume of the natural indicator was used to test 10 mL HCl/NaOH	color change when the HCl/NaOH with natural indicator
Day 1	1 drop	Acid: light yellow to dark pink Base: light yellow to bright yellow
Day 5	2-3 drops	Acid: light yellow to pink Base: light yellow to yellow
Day 10	5-6 drops	Acid: light yellow to light pink Base: very hard to see the color change
Day 15	No color change anymore	

In response to the decreased effectiveness of the indicator solution, indicator strips were created and tested. These strips have the potential to preserve the indicator in a more stable form. The indicator strips were then tested. In the acidic medium pH indicator strip showed pink color, while in the basic medium it showed yellow color to strip (figure 3).

**Figure 3.** pH indicator strips in acidic and basic.

Discussion

The pH of the natural indicator made from lavender lantana is 5.15. The range of the natural indicator is from 2.84 to 10.56. While the pH value of the testing solution was lower than 2.84, the color of the indicator change to pink. When the color of the indicator changed to yellow, the pH of the testing solution should be higher than 10.56. The pH value between 2.84 and 10.56 is hard to identify based on the color change of this natural indicator. There is a potential to expand the range of pH value that can be tested by the natural indicator by mixing different kinds

of natural indicators like a universal indicator. As the result of the strong acid-strong base titration, the use of natural indicator made from lavender lantana was found to be as good as a standard indicator when testing the strong acid or strong base because it gives sharp color change at the equivalence point due to the Bronsted-Lowry acid-base theory the proton from the extract received by OH ions thus causing color changes during the neutralization (see figure 1 and table 1).

The biggest problem in using the natural indicator is the natural indicator produced from the lavender lantana flowers become less efficient over an extended period of storage and regular testing (see table 2), which is because ethanol molecules in the natural indicator with 95% ethanol are volatile and have high-speed vaporization. Therefore, making the efficiency of natural indicator liquid more stable, and the exploration of the pH indicator strip can be the fields to take further study. The preparation of indicator and pH strip is accessible and student-friendly, which can be applied in the college laboratory. The color change in acidic and basic condition for extract and strip is pink and yellow, having about λ_{\max} -530 nm and λ_{\max} - 400 nm, respectively (see figure 3). The pH strip can use for various applications as initial determination. Also, if the expansion of the pH value works when the knowledge of universal indicator put into use, making a universal natural pH strip will be the step in the natural indicator study.

The synthetic indicators are toxic and have hazardous characteristics which may create problem to human health and the environment. Although it is hard to analyze the health effect on people of the natural indicator by testing them on living animals, the materials used to make natural indicator are usually flowers extract from lavender lantana, oleander, roses, which is mostly safe for people to use. As far as the economic aspect of the natural indicator is concerned, the cost of the natural indicator will be much lower when the plants which are used to prepare for the indicator is growing in the bigger scale. All the evidence shows that synthetic indicators can be replaced successfully by the natural indicators

because of economy, simplicity, and wild availability of natural indicator.

The study of the natural indicator is mostly successful by making and testing the specific natural indicator made from lavender lantana. In the future, to generalize the preparation and usage of natural indicator in the laboratory and find more potential benefits of replacing the synthetic indicator by natural indicator, it is necessary to continue to explore various plants and record the results using the similar procedure as analyzing and testing the indicator made from lavender lantana in this research.

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Arbuscular mycorrhizal fungi as a method of sustainable agriculture

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Introduction

As world populations continue to grow, modern agriculture must overcome numerous challenges to fill the increased demand for crop production. The majority of this growth is predicted to be countries showing inadequate food consumption and high levels of undernourishment (Alexandratos et al., 2012). Population increase—in addition to climate change—in food insecure countries could negatively affect global food security, especially as numerous impoverished countries must find new method to produce crops in spite of drought or flooding (Mendelsohn, R., & A. Dinar, 1999). Both favorable and these unfavourable agro-ecological environments will need to yield notably more while simultaneously avoiding further soil degradation (Alexandratos, N. & J. Bruinsma, 2012). The challenge of near-future agricultural production is compounded by the widespread dependence—and attempts to reduce dependence—of producers on agrochemicals, which are a concern in relation to the long-term health of both humans and the environment.

Arbuscular mycorrhizal fungi (AMF or AM fungus) offer a promising biofertilizer alternative through which this worldwide issue could be addressed. An oblique symbiont, AMF colonize the root cortex of plants, providing protection, water, and nutrients in exchange for photosynthetic products (Smith & Read, 2017). Plant growth is often limited by lack of nutrients, and thus an increase in accessible nutrients through AMF contribution could improve crop yield and efficiency. AMF also have potential to alleviate abiotic and biotic stress, similarly improving the health and yield of colonized plants or crops (Phillips, M., 2017).

Although a promising sustainable alternative, the use of AMF as a biofertilizer replacement of conventional fertilization practices faces a number of noteworthy

challenges, such as varied responses to inoculum and the complexity of successful large-scale inoculum production (Berruti, A., Lumini, E., Balestrini, R., & Bianciotto, V., 2016). While these primary challenges leave much room for improvement in the use of AMF as fertilizers, they do not necessarily inhibit the advancement of AMF applications.

AMF Relationship with Host Plants

AMF are soil microorganisms belonging to the phylum Glomeromycota. They are considered to be an essential part of ecosystems worldwide and form biologically diverse communities containing numerous fungal taxa (Phillips, M., 2017; Vandenkoornhuyse, P., 2002). A ubiquitous organism, the AM fungus builds a mutualistic symbiotic relationship with approximately 80% of land plant species (Parniske, M., 2005). These symbioses are unique in that they are oblique for the fungus, but generally facultative for the plant (Helgason, T., & A. H. Fitter, 2009).

Colonization of host roots The process of colonization precedes physical contact between plant and fungus. Diffusible molecule signals are exchanged between the two organisms, mediating reciprocal recognition (Lanfranco, L., Fiorilli, V., & Gutjahr, C., 2018; Bonfante, P., & Genre, A., 2015). In response to this signal, the fungus grows branching filaments called hyphae towards the root, and, upon contact with the root epidermis, develops adhesion structures. The AMF hyphae then enter the root, growing into the root cortical cells via either an intracellular or extracellular route (Gianinazzi-Pearson, V., 1996). Once inside the cortical cells, the hyphae form microscopic arbuscules composed of highly branched hyphae (Phillips, M., 2017). After the formation of arbuscules, the plant surrounds the newly formed structures with a periarbuscular membrane (PAM), which, in conjunction with the

arbuscule itself, forms the nutrient exchange interface (Lanfranco et al., 2018).

Nutrient acquisition One of the most prevalent benefits of AMF colonization—in relation to the host plant—is the increased accessibility of essential nutrients in the soil. AMF hyphae, which can grow up to 100 times the length of root hairs, have significantly more access to soil-borne nutrients than host plant roots (Javot, H., Pumplun, N., & Harrison, M. J., 2006). The increased length of hyphae allows for acquisition of resources beyond the nutrient depletion zone surrounding the root hairs and epidermis of the host plant (Javot, H. et al., 2006). To secure these nutrients, AMF secrete digestive enzymes along hyphal pathways, breaking down soil substances into elements. The resulting simple nutrients are then absorbed through pores in the hyphal cell membrane and transported to the host plant's roots for nutrient transfer (Phillips, M., 2017).

Nutrient transfer The transfer of nutrients takes place at a shared plant-fungus boundary, usually an arbuscule. The host plant releases organic carbon, which is absorbed by the fungus by means of a diffusion gradient and subsequently transported throughout the fungal organism in the form of sugars (Phillips, 2017). In exchange for the organic carbon, the AMF provides its host with a portion of the nutrients derived from the soil (Smith & Read, 2017). The transfer process is believed to be under the mutual control of host and fungi, meaning that either organism has the capability to exert control the nutrient transfer (Kiers, E. T. et al, 2011). In this manner, mutual nutrient regulation assists in maintaining a stable, often beneficial symbiosis.

Beneficial Services of AMF

One of the places plant-fungus symbiotic benefits are most noticeable is the change in total plant biomass. This change is primarily caused by increased nutrient uptake and improved stress tolerance as a result of the AMF and plant interactions.

Phosphate uptake increase Due to the aforementioned nutrient transfer between the fungus and host plant, AMF symbiosis is often highly beneficial to the growth and health of

inoculated plants. This benefit is commonly seen in the acquisition of inorganic phosphate (Pi) from the soil (Smith, S. E., Jakobsen, I., Gronlund, M., & Smith, F. A., 2011). Although phosphorus is abundant in the environment, the negative charge of its ion makes it prone to sequestering by cations (Javot, H., et al., 2007). To further complicate the issue, Pi is rather immobile in the soil, which, combined with existing cation sequestration, creates zones of Pi depletion. (Phillips, M., 2017). These properties are noteworthy in agriculture as a lack of accessible Pi significantly limits plant growth and thus has an important impact on agricultural success (Campos, P. et al., 2018). AMF ameliorate this issue by increasing the roots' access to soil and facilitating a symbiosis with phosphate-solubilizing bacteria. These bacteria produce organic acids which release phosphorus from cations in exchange for AMF carbon (Phillips, M., 2017). AMF acquire the phosphorus, delivering it to the plant via active transporters such as Pi:H⁺ symporters, many of which belong the Pht1 family (Javot, H., et al., 2007). Due to this aspect of symbiosis, phosphorus concentration has been shown to increase by up to four times in colonized host plants (Phillips, M., 2017).

Other nutrients In addition to Pi, the availability of other nutrients is often increased by AMF. AMF positively affected plant zinc concentrations in various plant tissues across distinct environmental settings (Berruti, A., 2016). Host plant nutrition involving other microminerals—specifically copper, iron, and manganese—has been shown to be benefited by the presence of the fungus (Berruti, A., 2016). Furthermore, increased amounts of nitrogen and sulfur found in colonized plants suggest that these nutrients may be transferred via AM-induced transporters as well (Chen, M., et. al., 2018).

Biotic and abiotic stress tolerance AMF have also been shown to increase tolerance of abiotic stresses caused by the environment of the host plant. Mahdavi et al. demonstrated that AMF can save vital organs of turfgrass (*Festuca arundinacea*) under severe, long, and stressful conditions, specifically drought (2018). Increased host plant drought tolerance is partially due to the increase of surface area between root and soil

and the secretion of glomalin, which assists in a better uptake of water and nutrients by the plant. Similarly, in a meta-analysis of the efficiency of AMF, Chandrasekaran et al. observed higher growth responses in inoculated plants, with *F. mosseae* being the most effective for C3 plants and *G. fasciculatum* for C4 plants (2016). Inoculated plants additionally showed an increase in K uptake and a lower Na uptake, suggesting a preferential uptake of K⁺ as opposed to the abundant Na⁺ of the saline soil. In soils prone to heavy metals, AMF also mediate heavy metal toxicity (Phillips, M., 2017). Similarly to abiotic stresses, the effects of biotic stresses have been shown to be less detrimental to the host plant with AMF inoculation. Wheat resistance to the infection *X. translucens* increased with AM symbiosis, and tomato plants exhibited enhanced disease resistance as well, decreasing the extent of infection by *Alternaria solani* (Fiorilli et al., 2018; Song et al., 2015).

Common mycorrhizal networks In addition to a relationship with a singular host plant, AMF also form poorly-understood, large, complex networks connecting numerous species of plants and fungi, referred to as common mycorrhizal networks (CMNs). These pathways allow for infochemical defense signaling across plant populations and nutrient transfer (Barto, E. K., Weidenhamer, J. D., Cipollini, D., & Rillig, M. C., 2012). CMNs have been shown to improve plant resilience to biotic stresses, as demonstrated twice by Song et al. in tomato plants (*Lycopersicon esculentum*) attacked by leaf-chewing cutworms (*Spodoptera litura*) and the pathogen *Alternaria solani*, respectively (2014; 2010). Van Der Heijden and Horton showed that CMNs generally improved seedling growth, benefiting 48% of the cases (for 21 seedling species), adversely affecting 25% and having no benefit or detriment on 27% (2009). In a similar fashion, trees can recognize offspring as genetic kin and direct nutrients to related, shaded seedlings (Phillips, M., 2017).

Challenges of AMF

Despite the benefits observed in the above studies and observations, contrasting research has demonstrated negative effects of CMNs on affected plants (Bücking, H., Mensah, J.

A., & Fellbaum, C. R., 2016). This apparent inconsistency is unsurprising as responses to AMF—including the resulting CMNs—are highly dependent on the compatibility of the plant, environment, and AMF species. Increased susceptibility to stresses and decreased nutrient uptake have been observed in a number of studies.

Dependence on nutrition supply

Although often beneficial contributors to nutrient uptake of host plants, AMF nutritional benefits are highly dependent upon the soil supply conditions. Püschel et al. (2016) determined that under a low concentration of soil nitrogen, AMF hoarded N, decreasing the host plant N uptake and lowering total and shoot N content (2016). As N concentration increased, the symbiosis became mutual. In relation to phosphorus, high availability in the soil has been associated with a decrease in mycorrhizal growth responses (MGRs) and a notable growth depression (Peng, Eissenstat, Graham, Williams, & Hodge, 1993). The cause of this depression could be attributed to the high carbon cost of AM symbiosis, which is not balanced by a gain of P (Wang, X., Zhao, S., & Bücking, H., 2016). Additionally, Wang et al. (2016) showed that plant responses differed in relation to fungal species. Due to these factors, AMF must be compatible with both the target environment and specific plant in order to benefit and protect host plants.

Biotic stress susceptibility AMF have generally been shown to have predominantly negative effects on general chewing herbivores while having predominantly positive effects on specialist chewing insects (Koricheva, J., Gange, A. C., & Jones, T., 2009). In a study of AMF colonized rice plant susceptibility to the rice water weevil (*Lissorhoptrus oryzophilus*), fall armyworm (*Spodoptera frugiperda*), and infection by sheath blight (*Rhizoctonia solani*), Bernaola, Cosme, Schneider, & Stout demonstrated that inoculation with AMF rendered plants more susceptible to pests (2018). This susceptibility resulted in increased larval growth, weights, and survival. AMF increased plant shoot biomass in two or the experiments despite the increase in larval susceptibility.

Variability in Species and Effects

The cumulative benefits of AMF on plant productivity suggest a potential role in commercial agriculture. Numerous recent and previous studies have sought answers regarding potential benefits or harms in the context of various species of plants and fungus.

Current attempts and studies
Understanding of the impact of AMF under various conditions is rather limited, but a number of studies have addressed the issue in part (Van Der Heijden, M. G. A., et al., 2015). Some suggest beneficial effects of AMF on several important agricultural crops, and others show a positive trend of host plant growth (Berruti et al., 2016; Harley, J. L., & Smith, S. E., 1983). A number of studies have also shown negative effects on colonized plants. The above mentioned study by Bernaola, Cosme, Schneider, & Stout concluded that AMF increased pest susceptibility, while Song et al. contrastingly demonstrated increased plant resilience (Bernaola, Cosme, Schneider, & Stout, 2018; Song et al., 2014). Although both testing a similar concept, the studies concluded with notably different results, ranging from beneficial to detrimental. The benefit or detriment of AMF colonization is only observable if the host plants are colonized. To determine the effectiveness of commercial inoculants in colonizing plants, Corkidi et al. determined the percent AMF colonization of corn plants and the effects of colonization on the host plant in multiple types of conditions (2004). Significant infectivity differences were observed, with minimal to no colonization in four of the ten plants and a notable colonization increase in four others. Mycorrhizal colonization was not correlated with a markable increase in host plant growth, and benefit or detriment was highly dependent upon substrate and amount of light (Corkidi et al., 2004). Faye et al. observed more successful inoculation of maize plants, with the colonization ten of the twelve commercial inoculants tested (2013). The six week testing period was too short to observe any significant shoot biomass increases. Ceballos et al. showed positive results of AMF colonization in the crop plant cassava, which is globally important for food security (2013). Inoculation with the fungus *R. irregularis* along with P fertilization increased

cassava yield by a significant amount; inoculated plant roots were 20.4% heavier than roots of non-inoculated plants (Ceballos et al., 2013). Similarly, in Cuba, inoculation with AMF and a seed coating technology was shown to increase various crop yields—including cassava, maize, and rice—by an average of approximately 46.9 percent (Rivera, R., et al., 2007).

The effects of introducing AMF to an agricultural setting are highly dependent upon the host plant, fungus species, and growing conditions. Because of the specificity, commercial inoculation faces numerous challenges, such as obtaining the most effective inoculant for the specific crop, soil, and location. Even so, the prospect of AMF in sustainable agriculture is promising, and further research must be done to determine the feasibility of an effective AMF inoculum that could function as an alternative for conventional agro-chemicals or fertilizers.

In order to determine if arbuscular mycorrhizae is a feasible alternative to fertilizer or of benefit to inoculated plants, research is proposed that plants are grown in the conditions of no additives, fertilized soil, and addition of fungal spores. Consequential rate and extent of plant growth in these conditions provides a direct comparison between the variables and corresponding growth values. Three types of plants subjected to the testing represent three main food yielding types, these being fruit producing, root producing, and leaf producing. The use of three plant types gives a more comprehensive understanding of the interactions between plant and fungus.

Hypothesis The conditions of no additives, fertilized soil, and the addition of fungal spores will affect the weight and height of tested plants.

Null Hypothesis No weight or height difference will be observed between plants subjected to the different conditions above mentioned.

Methods

Inoculum must be produced to increase the concentration of AMF in the soil and to

maximize the potential benefits of the fungus. The production of inoculum faces a number of challenges, but even so has already made a noteworthy appearance in the commercial sector (Berruti et al., 2016). The main challenge of producing inoculum is the oblique symbiotic nature of AMF, meaning that a phase of cultivation with a host plant is necessary to complete the AMF life cycle (Coelho, I. R., Pedone-Bonfim, M. V., Silva, F. S., & Maia, L. C., 2014). Numerous types of production have been attempted, including substrate based, aero/aquaponic, and in vitro methods (Ijdo, M., Cranenbrouck, S., & Declerck, S., 2010). These methods are helping to make agricultural applications of AMF less expensive and more reliable (Van Der Heijden, M. G. A., Martin, F. M., Selosse, M., & Sanders, I. R., 2015). Although more reliable than previous methods, current AMF product application has not been extensively evaluated in comparison to traditional fertilizer techniques. This comparison is essential to advancement of AMF commercial applications and must be further researched.

Inoculants To research the applications of AMF, two commercial products were used, these being Mykos Pure Mycorrhizal Inoculant (Mykos Inoculant), obtained from Xtreme Gardening®, and Granular Endo Mycorrhizae (Granular Mycorrhizae), obtained from Root Naturally. The Mykos Inoculant contained only *Glomus intraradices*, and the Granular Mycorrhizae contained a *Glomus intraradices*, *Glomus mosseae*, *Glomus aggregatum*, *Glomus etunicatum* mixture. These species were easily obtained by purchasing the corresponding commercial products and were a strong representation of the current AMF species most commonly used.

Plants Due to the 28-day time frame, plant varieties with short germination times were chosen. All seed types were produced by Ferry-Morse and were container varieties. The Early Girl Tomato (*Solanum lycopersicum*) is one of the first fruiting plants, germinating in an average of 7-10 days. The Cherry Belle Radish (*Raphanus sativus*) is equally as fast to germinate, germinating in an estimated 4-7 days. The Black Seeded Simpson Lettuce (*Lactuca sativa*) also germinates in 7-10 days, similarly making it an ideal plant for the

short testing period. These three species represent the aforementioned categories of fruit producing, root producing, and leaf producing plants.

Soil The soil chosen was organic All-Purpose Potting Mix (potting mix), obtained from Garden Safe. It contained a mixture of sphagnum peat moss, composted organic matter, and perlite. The mixture was specifically formulated for container gardening. For the fertilizer trial, pre-fertilized Miracle-Gro Potting Mix (Miracle-Gro) was used.

Experimental Design

To prepare for the experiment, the potting mix was mixed with either the Granular Mycorrhizae or the Mykos Inoculant. A control with no additives was also prepared, as well as a fertilized trial using Miracle-Gro. The Mykos Inoculant was added four grams per cell, as was the Granular Mycorrhizae. Between the individual combination of each of these mixtures, both hands and the mixing bins used to combine the fungus and soil were cleansed with rubbing alcohol. Each plant type was planted in 24 cells, split into six cells for every soil variant. The tomato seeds, one per cell, were placed 0.635 cm deep into the designated tomato cells. This process was repeated with the radish seeds, but planted 1.270 cm deep and with two seeds per cell, and the lettuce seeds, 0.635 cm deep and three seeds per cell.

40 mL of water was added to each cell before planting, and after planting perlite was evenly distributed on top of all the soil to prevent further soil compaction. All of the trays were placed on a heating pad to maintain an average temperature of 26 degrees Celsius. The second day after planting, 400 mL of water was added to each tray to bottom water the seeds. Following this initial watering, regular watering was done two days a week, 30 mL the first day and 30 mL with 400 mL bottom-watered the fifth. One LED full spectrum 84-watt light was set on a 12-12 schedule, with 12 hours of light and 12 of dark per a day. Beginning at emergence from the soil, measurements of the seedlings' height were measured and recorded every three or four days.

On day 15, the number of plants was reduced to a single plant per a cell, eliminating the most underdeveloped plants, determined by relative height. Removal was performed with a wooden skewer to minimize root network damage. Skewers were switched for every tray to prevent cross contamination. Additional skewers were inserted into the soil to support the remaining sprouted plants as necessary based on instability of individuals.

Final height measurements were collected on day 21. Plants were watered with 400 mL to each tray, and the fertilizer and control trays were top watered with an additional 30 mL of water. 400 mL was bottom watered per tray, and 30 mL was top watered per cell on day day 25. On day 28 weight measurements were taken. Plants were prepared for weighing by removal of surrounding soil, both manually and with the assistance of a water bath. Significant care was taken to maintain root networks. After removal of soil, the plants were gently dried with paper towels and laid out on the counter in groups corresponding with plant type. Before weighing,

the plants' roots were once again dried by paper towel.

Results

General observations during the trials produced a number of notable developments. Fungal fruiting bodies were observed above the soil in both control and fungal cells approximately halfway through the experiment. These bodies were not disturbed or removed and thus continued to grow. Concentration of the fungus was an approximate two fungal bodies per tray, with no observed difference between presence in sprouted or unsprouted plant cells.

In addition to the presence of fungal bodies, discoloration of leaves in radish control and inoculated conditions was observed on day 25 and continued to progress through the rest of the experiment. The discoloration was most notable on the Granular Mycorrhizae (multiple fungus/multi) and Mykos Inoculant (single fungus/single), with minimal, yet noticeable, color change in the control radish plants. (Figure 1)



Figure 1: (A)Control plants, (B) Fertilizer plants, (C) Granular Mycorrhizae plants, (D) Mykos Inoculant plants

Plant growth during the tomato trials began by the 8th day with the sprouting of fertilized and multiple fungus plants (Table 1). The control and single fungus trials sprouted by the eleventh day. On the 21st day, two of the control tomato plants, one of the fertilizer tomato plants, four of the multiple fungus tomato plants, and two of the single fungus tomato plants had not sprouted. The fertilizer tomato plants displayed the highest rates of successful sprouting, and the

multiple fungus plants the lowest. All of the radish trials had at least one plant sprouted by the fifth day, and the complete 24 seedlings of radish had emerged by the eighth day (Table 2). A lettuce plant from every condition had sprouted the fifth day, and on the 21st day, all of the 24 lettuce plants had sprouted, except for one control lettuce plant and two single fungus lettuce plants (Table 3).

Table 1. Tomato plant height measured in centimeters.

Treatment	Day 5			Day 8			Day 11			Day 14			Day 18			Day 21		
	n	M	SD	n	M	SD	n	M	SD	n	M	SD	n	M	SD	n	M	SD
Control	6	0.0	0.00	6	0.0	0.00	1	3.0	*	2	3.8	0.35	3	4.0	0.50	4	5.0	0.71
Fertilizer	6	0.0	0.00	5	2.6	0.22	5	3.4	0.42	5	4.5	0.35	5	6.4	0.65	5	10.8	1.35
Multi	6	0.0	0.00	1	2.0	*	1	3.5	*	1	4.0	*	2	4.3	1.06	2	5.5	0.71
Single	6	0.0	0.00	6	0.0	0.00	1	2.5	*	3	3.2	0.58	4	3.5	1.22	4	4.3	0.29

*Standard deviation not applicable due to sample size.

Table 2 Radish plant height measured in centimeters.

Treatment	Day 5			Day 8			Day 11			Day 14			Day 18			Day 21		
	n	M	SD	n	M	SD	n	M	SD	n	M	SD	n	M	SD	n	M	SD
Control	6	1.6	0.58	6	2.6	0.49	6	3.3	0.52	6	3.4	0.49	6	3.6	0.58	6	3.9	0.58
Fertilizer	6	1.4	0.38	6	2.8	0.52	6	3.3	0.69	6	4.5	0.77	6	5.8	1.03	6	6.2	1.17
Multi	5	1.6	0.42	6	2.3	0.68	6	2.8	0.82	6	2.8	0.99	6	3.0	1.05	6	3.3	1.08
Single	6	1.7	0.9	6	2.2	0.26	6	2.7	0.26	6	2.5	0.45	6	2.6	0.49	6	2.5	0.45

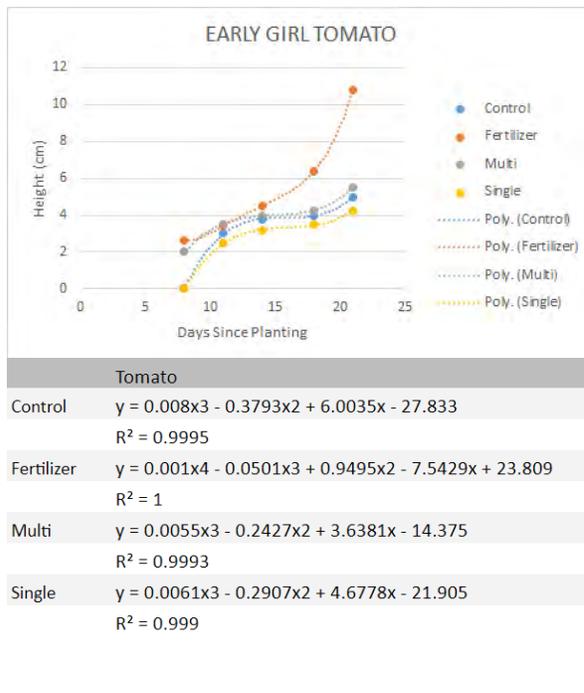
Table 3. Lettuce plant height measured in centimeters.

Treatment	Day 5			Day 8			Day 11			Day 14			Day 18			Day 21		
	n	M	SD	n	M	SD	n	M	SD	n	M	SD	n	M	SD	n	M	SD
Control	2	0.5	0	3	1.8	0.76	3	2.3	0.29	4	2.4	0.75	5	2.3	0.97	5	2.4	0.96
Fertilizer	3	1.0	0.5	6	2.4	0.38	6	3.3	0.88	6	5.4	1.59	6	5.8	1.47	6	6.2	1.03
Multi	4	0.7	0.27	6	2.3	0.52	6	3.0	0.45	6	3.3	0.88	6	3.3	0.69	6	3.4	0.86
Single	3	0.8	0.58	4	1.6	0.48	4	1.9	0.25	4	2.0	0.41	4	2.0	0	4	2.1	0.48

The control, multiple fungus, and single fungus trials follow a similar trend. Height significantly increased during the 8 day to 14 day period, plateauing from day 14 to 18, then began to rise again. The fertilizer trial height significantly exceeded the other heights, continuously increasing with the most notable change being between day 18 and 21 (Figures 2 – 4).

As observed in the tomato trials, the fertilizer plants demonstrated a more significant increase in height than the other plants. The control and multiple fungus trials showed a consistent, yet gradual increase in height, and the single fungus trial plateaued after the fifteenth day. The control trial consistently displayed higher height values than the inoculated trials. Plant height slightly decreased the fourteenth day for both inoculated conditions.

Figure 2. Growth of early girl tomato under different conditions.



Lettuce trials showed a similar trend, except with the multiple fungus plants plateauing at a notably higher value than the control or single fungus conditions. The fertilizer trial continued to achieve the highest values, reaching a more substantial height.

Figure 3. Growth of cherry belle radish under different conditions

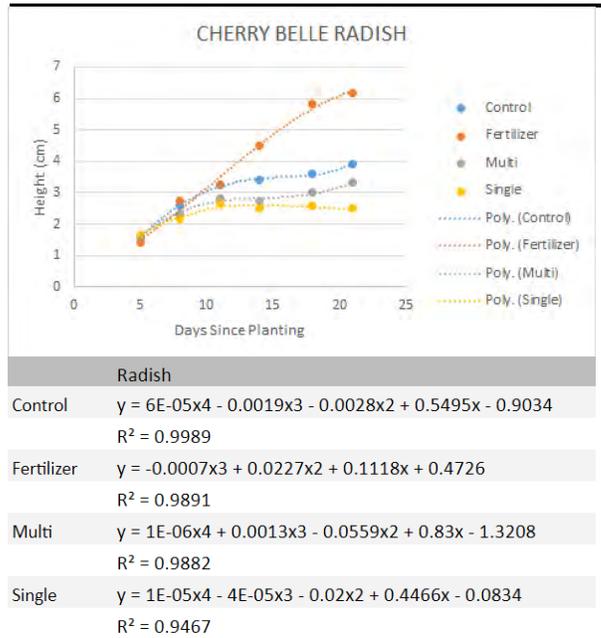
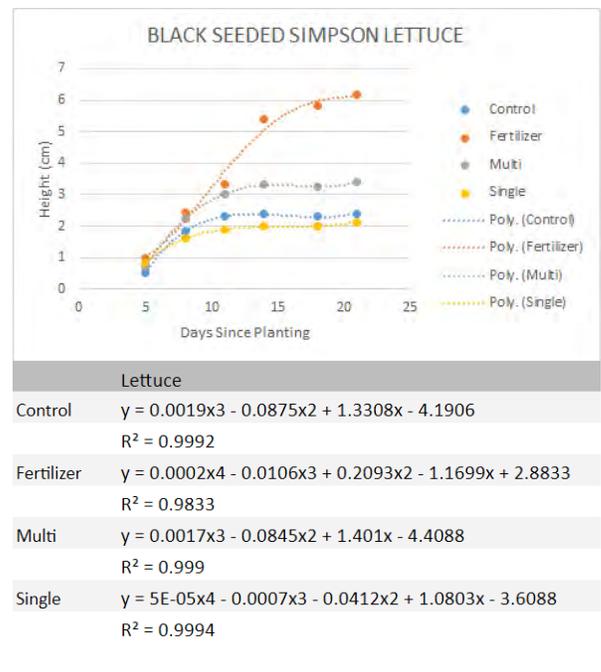


Figure 3. Growth of black seeded simpson lettuce under different conditions



At the 28th day of the tomato trial, two control tomato plants, four multiple fungus tomato plants, two single fungus tomato plants had not sprouted. All radish plants were sprouted, as aforementioned. Of the lettuce plants, all had sprouted except one control lettuce plant and two single fungus lettuce plants. Measurements of plant mass were inclusive of the root network (Table 4).

Table 4. Plants masses measured at day 28 of growth.

Treatment	Tomato		Radish		Lettuce	
	M	SD	M	SD	M	SD
Control	0.13	0.04	0.58	0.14	0.21	0.14
Fertilizer	3.97	2.58	3.70	1.66	3.13	1.03
Multi	0.18	0.06	0.69	0.18	0.51	0.21
Single	0.17	0.04	0.62	0.19	0.21	0.13

All masses measured in grams.

Mass differed significantly between the fertilizer trials and other trials (Figure 4). The tomato plants of the multiple fungus, single fungus, and control trials showed the least amount of variability in mass. The lettuce showed the most, with multiple fungus plants reaching approximately two and a half times the mass of control tomato plants. Radish total weights differed slightly, with the multiple fungus radish plants exceeding the weight of both the single fungus and control conditions.

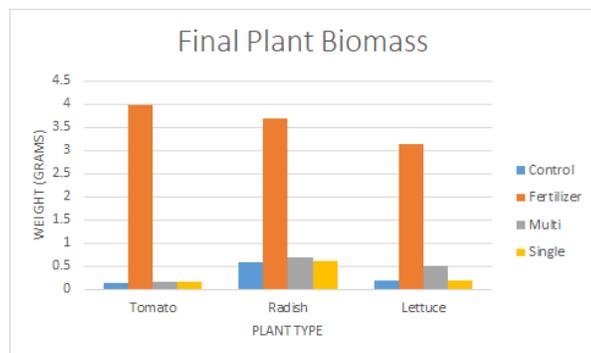


Figure 4. Final sprout and root mass measured in grams

Discussion

Observations regarding fungal bodies were made in the control cells as well as the inoculated cells. These observations imply that the fungus was a product of the potting mix used, rather than a result of the inoculants. Due to the presence in multiple conditions, this factor was not inhibitory to the data.

Plant discoloration was most overtly observed in the fungus trial, indicating that the addition of the inoculant may affect the leaf coloration. The discoloration did not affect total weight, but nevertheless suggests that the affected radish plants may not benefit from the inoculants.

For all three trials--tomato, radish, and lettuce--the height and weight values of the fertilized conditions were substantially higher than that of any other condition. The difference seen regarding these values shows a highly beneficial effect of Miracle-Gro soil on seedling growth and total mass. This benefit is primarily due to the high concentration of inorganic phosphate and micronutrients in the Miracle-Gro soil, which prevented stunting of growth caused by nutrient deficiencies.

The addition of Mykos Inoculant and Granular Mycorrhizae did not significantly improve growth of the tomato plants. Although the Granular Mycorrhizae produced higher height values than the control, the standard deviations overlap significantly. Additionally, the sample size of control--four sprouted plants--is twice the multiple fungus' two sprouted plants. Radish plants showed a similar trend, with the addition of inoculants slightly lowering the height values in relation to the control. The inoculated radish plants experienced a depression in height on day 14, which can be attributed to the addition of water. Lettuce plants showed more significant value differences, with the addition of Granular Mycorrhizae increasing weight by an approximate one hundred percent. From this increase, it can be concluded that the addition of the inoculant was beneficial to the lettuce plant growth. This benefit is likely the result of an increased nutrient uptake caused by the symbiosis.

The benefit of fertilizer for each plant type was expected as it provides the materials needed for sustained plant growth without depletion. Due to the prevalence of the nutrients in the soil, it is unsurprising that the inoculated conditions did not exhibit similarly high values. The main benefit of AMF is to acquire nutrients in nutrient depleted soil, meaning that in comparison to nutrient rich soil, any notable benefit does not compare with the benefit of plentiful nutrients.

Further research with different types of soil would show the effects of the inoculants more clearly as other organisms, such as the fungal fruiting bodies observed, would not interfere with the seedlings. Additionally, plant growth to maturity and a larger sample size would lead to more comprehensive conclusions regarding the feasibility of replacing conventional fertilizers with arbuscular mycorrhizae.

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Effects of various minerals on remediation of soil toxicity and growth of tomato plants

Peter Sorenson

Introduction

A steady, scalable source of food is essential for population growth. Production, however, is not infinitely scalable and is limited by the amount of arable land open to development and the capacity for that land to produce food. As a population's demand for food increases with its growth, there is often increased demand for either new arable land or an increase in productivity of the existing land. Since a growing population also means an increase in the amount of land needed for infrastructure, industry, housing, and office space - all of which take up land - the amount of land open for agricultural development is often not enough to satisfy the growing demand for agricultural products. With limited land for agriculture, societies have instead focused on improving the yield of the already existing farmland. A diverse range of solutions have been tested, including fertilizers and genetic modification of plants (Delhaize & Ryan, 1995). Although there have been efforts to increase yield, there are environmental factors that can counteract these efforts including metal contamination. Metal contamination can be caused by the area surrounding a piece of land, run-off from industry, or various other processes and can severely affect the growth of plants and consequently the yield of the land. Since metal contamination is widespread, research on the reduction of metal contamination holds promise for improved agricultural results.

Heavy Metal Toxicity and Soil Acidification The toxicity of heavy metals and their impact on the environment are well understood. (Sandalio, Dalurzo, Gómez, Romero-Puertas, & Río, 2001; Rucińska, Waplak, & Gwóźdź, 1999). A significant amount of land is affected by heavy metals coming from agricultural, urban, and industrial sources (Khan, Kuek, Chaudhry, Khoo, & Hayes, 2000). Heavy metals are especially prevalent in peat and mineral soils since heavy

metals are more prone to dissolve in low pH soils. Al, for example, is soluble in soils with a pH of 5 or below (Kochian, Piñeros, & Magalhaes, 2015). Acidic soils are common as 50% of global land is also affected with low pH soils (Kochian et al, 2015). Furthermore, acidic soils can increase the toxicity of metals by reducing essential nutrients like Ca, K, Mg, and P, and the efficiency of their use in plants (Goulding, 2016; Teng, Kang, Hou, Hu, Luo, Wei, & Zhang, 2018). Soil acidification can be caused by several factors including acidic precipitation, acidifying gases and particles like sulfur dioxide (SO₂) and ammonia (NH₃), acidifying fertilizers including sulfur and urea, nutrient collection by plants, and mineralization of plant and animal matter (Goulding, 2016). Fertilizer use and the creation of acidifying gases often accompany population growth, as industry and agriculture are scaled to meet. Normally the soil pH is kept at different levels due to buffering processes, but these processes can reduce available nutrients, and can increase Al³⁺ levels in the soil (Goulding, 2016). Interestingly, heavy metals do not appear to be universally toxic to plants. In rice, tea, coffee, soybean, and maize, controlled application of heavy metals can be beneficial by priming a plant to respond to another stressor (Morkunas, Woźniak, Mai, Rucińska-Sobkowiak, & Jeandet, 2018). Also, the accumulation of metals in plants can act as a deterrent to insects and the growth of some fungi species, but they have also been shown to have a stimulating effect on other species (Boyd, 2012).

Complicating the effects of heavy metals on organisms in an ecosystem is the fact that they can also increase the concentration of metals at different levels in the food chain (Hörger, Fones, & Preston, 2013). This is concerning as trace amounts of Al can enter the bodies of tea drinkers through the gastrointestinal system, and elevated levels of aluminum could cause renal failure in humans

(Stewart, 1988). Along with being potentially dangerous to humans, excess metal cations (Cu, Fe, Zn, Mn) can become toxic to plants (Rahman, Lee, Ji, Kabir, Jones, & Lee, 2018)

Heavy Metal Tolerance Possible methods for increasing resistance to aluminum have been shown (Teng et al, 2018). Eucalyptus plants provide one example in which remediation of Al with P (phosphorus) has been tested with positive results (Teng et al, 2018). Two eucalyptus clones were subjected to Al stress with and without a P supply. Without a P supply, the concentration of P in the eucalyptus plant was significantly reduced under Al stress (Teng et al, 2018). Furthermore, it was noted that when P was applied with the Al stress, that the above ground concentration of Al decreased and the below ground concentration of Al increased (Teng et al, 2018). When P was present along with the Al stressor, the dry mass of the plant increased as compared to Al without P. When there was no Al stressor, the addition of P did nothing to increase the dry mass of the plant (Teng et al, 2018). This suggests that the addition of P to the DH32-29 Eucalyptus plant could reduce the effects of Al poisoning on that particular species of that plant by sequestering Al in the roots of the plants, even though this may seem counterintuitive as it is the roots that are generally the most affected by Al poisoning. The fact that some species have methods for regulating Al complicates the nature of the plant's interactions with the heavy metal.

There are two main ways by which plants can prevent Al poisoning: mechanisms that exclude Al from the root, and mechanisms for Al sequestration within specific cells. (Kochian et al, 2015). Many Al-resistant genotypes, including maize plants, release a variety of organic compounds, including phenolics, to bind with Al³⁺ ions, rendering them incapable of being uptaken by plants (Kochian et al, 2015). Internal Al sequestration methods are in general more complex than exclusion mechanism, and less well known, but one theory is that Al is moved from the cell wall of plant cells in other parts of the plant and into root cell vacuoles (Kochian et al, 2015). This would correlate with the higher levels of Al seen in the root of the previously mentioned eucalyptus clones. Another study done in 2006

with root tips of the Buckwheat plant confirmed that the majority of Al was located in the cell walls in the cytosol region of the root, at least with the genotype *Fagopyrum esculentum* (Wang, Raman, Zhang, Mendham, & Zhou, 2006). Although it is theoretically possible to isolate plants that are extremely efficient at exclusion and internal detoxification of Al, tolerance to aluminum is a complicated multigenic trait with multiple contributing genes, which have not been studied in a majority of plant species (Wang, et al, 2006). There are also several nutrients that have been shown to reduce Al toxicity. A study done by I. Silva in 2001 recorded the effects of magnesium and calcium on Al resistance and citrate excretion in soybeans. The study concluded that applying both Mg and Ca reduced the amount of Al in root tips and reduced damage to them while under Al stress when compared to plants just under Al stress by improving internal and external detoxification systems by the increased secretion of acids. (Silva, Smyth, Israel, Raper, & Rufty, 2001). Mg and Ca are not the only minerals known to reduce Al toxicity; phosphorus has also been shown to reduce Al toxicity, in *Lespedeza bicolor* (Sun, Shen, Zhao, Chen, & Dong, 2008), cowpea (Jemo, Abaidoo, Nolte, & Horst, 2007), and eucalyptus (Teng et al, 2018).

Measuring the Combination of Phosphorus and Aluminum on Plants Since heavy metals have shown signs of being beneficial in smaller doses, the amount of Al used in a study and the time that the Al is applied for is important. As far back as the year 1965, aluminum has been recognized as a root growth limiter in high quantities (Clarkson, 1965). Specifically, it concluded that aluminum sulfate applied in the concentration of 10⁻³ M applied over 24 hours decreased the root length in an Al-sensitive plant from 8mm before treatment to 1mm after treatment. Further research suggests that an Al solution can cause root growth suppression within 60 minutes of application, and that the most sensitive part of the root when it comes to Al toxicity is the root tip (Delhaize & Ryan, 1995). Although mineral and nutrient application is a promising area of study for the alleviation of Al toxicity in plants, the effectiveness of P on the alleviation of plant growth differs between plant

species and even some genotypes. In the *Lespedeza cuneata* species, P reduced the effects of Al poisoning, but in *Lespedeza bicolor* species, the effect was not nearly as significant (Sun et al, 2008). With this variance in response to P stimulus among genotypes of the same species of plant, it is probable that different species of plants would react differently to P stimulus. One of the plants where there is not a significant amount of literature about the application of P with Al stress is the tomato plant, even though tomatoes are an extremely common and highly cultivated food. In order to determine the effects of magnesium nitrate, calcium nitrate, and monopotassium phosphate on the growth of tomato plants under the influence of aluminum nitrate, this research seeks to test the following hypotheses: H0: The growth rates of tomato plants will not differ among the treatment groups. H1: The growth rates of tomato plants will differ among the treatment groups aluminum inhibiting growth unless applied with other nutrients.

Materials and Methods

Preparation The species of tomato selected was the beefsteak tomato, which was picked for its fast vertical growth and tall mature height. The seeds used for the experiment were purchased and stored in a cool, dry environment. To the best of our knowledge, there was no published study on the alleviation of Al poisoning in beefsteak tomatoes at the time of writing. Plants were grown in a light soil formulated for indoor potted vegetables (Miracle Grow Potting Mix). Twenty clay pots with a volume of 1.5 quarts and a rim diameter of 6.5 inches were selected. Since the plants were grown inside without the influence of solar lighting, five 300-watt equivalent full spectrum LED bulbs were wired to an analog timer. A stand was employed to raise the bulbs 0.5 meters above the top of the pots. Under the pots, a 100-watt heating mat was employed to keep the soil at 24.7C (Schwarz, Thompson, & Kläring, 2014).

Planting and Initial Growth To begin the planting process each clay pot was first filled with 1.5 quarts of soil. Then five holes were pressed into the surface of the soil, each to a depth of 6mm. Next, one seed was placed into each hole, and soil

moved to fill in the hole. Lastly, each pot was top watered with 50ml of water. Once the seeds were planted, the clay pots were placed on top of the electric heating mat. LED growth lights, set to 12 hours a day, were installed above the seedlings (Schwarz, Thompson, & Kläring, 2014). The heating mat was removed once all seeds had sprouted. During this time plants were individually watered whenever they became dry.

Replanting To reduce the difference in height between the tallest and shortest seedlings, and to address the issue of multiple seedlings per pot, all seedlings were removed from the pots and organized from tallest to shortest, after 14 days of growth (Schwarz, Thompson, & Kläring, 2014). The twenty tallest seedlings were then transplanted, along with a colored wood split identifying the plant, and a long wooden rod for the tomato plants to use as a trellis. Treatment After 25 days of growth, the plants were bottom-watered daily in preparation for treatment. Bottom watering involved adding 100ml of water per plant to each tray. At this time measurements of the seedlings were started. As the plants were watered, growth was measured using a specialized ruler to eliminate the variable of soil compaction. All plants were measured at the same time to reduce any potential error. Five groups of plants were created and organized to allow for faster treatment (Table 1). Each group was defined by color (Green, Red, Yellow, Blue, or Purple) and each seedling in a group was given a letter from "A" to "D."

Treatment Starting 26 days after planting the seeds, the application of the solutions was started. The application of the treatments was done at the same time that the growth of the plants was recorded. The green group was only treated with 20ml of water. The red group was treated with 20ml of 5mM aluminum nitrate, the same concentration that produced a significant effect in eucalyptus (Clarkson, 1965). The yellow group was treated with 10ml of 5mM magnesium nitrate along with aluminum nitrate. The blue group was treated with 10ml of 5mM calcium nitrate along with aluminum nitrate. Lastly, the purple group was treated with 10ml of 5mM monopotassium phosphate along with aluminum nitrate. The concentration of 5mM was used universally to

rule out any effects of nitrate when comparing plants since nitrate is a proven growth enhancer (Frink, Waggoner, Ausubel, 1999). Application of the treatments and measurements of the plants was stopped 35 days after initial seeding.

Results

All seedlings grew during the course of the experiment as shown by an end growth of 16.63cm for the shortest group of plants on day 35 (Table 2). Growth for all plants followed a mostly linear pattern as indicated by the R² values of .98, .956, .977, .967, and .986 (Figure 1).

Not only were all growth rates linear, but all plants also experienced very similar growth, with the difference in height between the treatments never exceeding 5cm (Figure 1). The average end growth height for the groups of plants was 16.63 to 19.53cm at day 35 (Table 2).

The growth rate for the yellow, purple, and blue groups were all within 1% of each other. The growth rates of red and green were ~4% higher than that of yellow, blue and purple (Figure 1)

Table 1. Each treatment group differed in the combination of aluminum and nutrient solutions received over the course of the experiment.

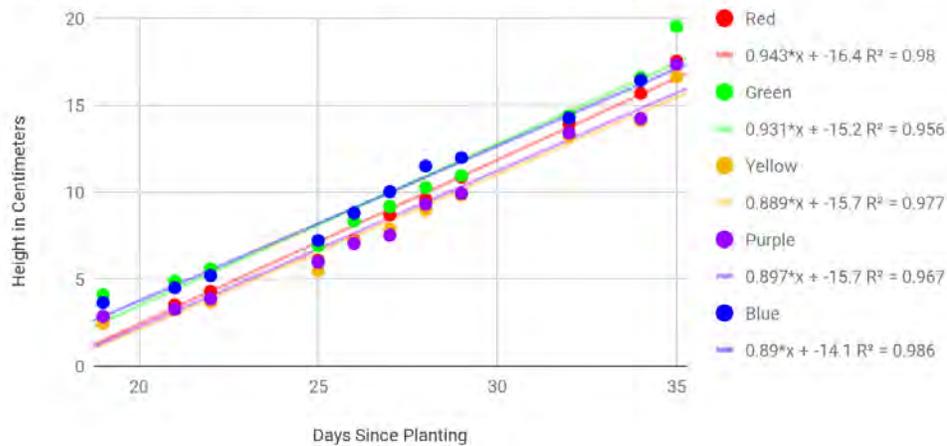
Group Number	Group Color	First Treatment	Second Treatment
1	Green	20ml of H ₂ O	Nothing
2	Red	20ml of a 5mM Al Solution	10ml of a 5mM Al(NO ₃) ₃ Solution
3	Yellow	20ml of a 5mM Al Solution	10ml of a 5mM MgNO ₃ Solution
4	Blue	20ml of a 5mM Al Solution	10ml of a 5mM Ca(NO ₃) ₂ Solution
5	Purple	20ml of a 5mM Al Solution	10 ml of a 5mM KH ₂ PO ₄ Solution

Table 2: Standard deviation (SD) and average (μ) for all groups organized by days since planting

	d19		d21		d22		d25		d26		d27		d28		d29		d32		d34		d35	
	μ	SD																				
Red	2.80	0.65	3.53	0.92	4.30	1.34	6.08	1.60	7.23	1.48	8.70	1.50	9.58	2.26	10.88	1.70	13.90	1.37	15.68	0.81	17.53	1.22
Green	4.10	1.83	4.10	2.11	5.60	2.46	6.93	2.55	8.35	2.49	9.18	2.84	10.28	2.37	10.95	2.40	14.40	1.70	16.58	1.31	19.53	1.62
Yellow	2.45	0.50	2.45	0.67	3.68	0.83	5.48	0.93	7.15	0.60	7.85	1.29	8.98	1.65	9.83	1.42	13.23	1.09	14.10	2.22	16.63	3.40
Purple	2.85	0.47	3.28	0.33	3.88	0.40	5.98	0.82	7.05	0.66	7.53	0.41	9.30	0.89	9.93	1.28	13.40	1.00	14.23	0.69	17.33	1.62
Blue	3.65	0.95	4.50	1.05	5.20	1.05	7.23	1.35	8.80	1.81	10.03	1.79	11.50	2.92	11.98	2.91	14.25	2.08	16.43	2.03	19.40	3.05

Figure 1. Plants in all treatment groups experienced similar linear growth over the course of the experiment.

Figure 1: Effects of Treatment on Plant Height over Time



To compare the effects of treatment on the growth of individual plants over the course of the experiment, a repeated-measures ANOVA was performed. The analysis suggested that there were no significant differences in growth rates among the treatment groups $F(4, 215) = 1.0753, p = 0.3696$.

Discussion

Although aluminum is generally thought to be toxic to plants, the beefsteak tomato plants tested with aluminum nitrate did not show an adverse effect in a 5mM concentration applied in 30ml quantity over six days.

There are three possible characteristics of beefsteak tomatoes that could explain why aluminum nitrate did not show a detrimental effect on the plants tested:

1. Too low of a concentration of $AlNO_3$ could have been used to show an effect. As only one concentration of aluminum was tested it was not possible to rule out the possibility that larger amounts of aluminum could have exhibited differences.
2. Beefsteak tomatoes could be naturally resistant to aluminum in any concentration. If beefsteak tomatoes are not susceptible to aluminum at all there would be no changes in tomatoes treated with $AlNO_3$ as compared to plants not treated with aluminum.

3. Aluminum delivered as a water-soluble nitrate may not be toxic to plants. If $AlNO_3$ is not absorbed by tomatoes plants, or $AlNO_3$ does not affect plants, then there would be no difference between plants, as observed.

The lack of difference between the groups treated with nutrients compared to other groups is likely due to the fact that all plants were grown in soil already treated with nutrients. Since there is a point of diminishing returns with the application of plants it is probable that the nutrients naturally occurring in the soil elevated the level of nutrients into the soil to this point.

One possible way to test whether the $AlNO_3$ was delivered in a high enough concentration to show potential effects or in a state toxic to plants would be to include a known Al-sensitive plant as a control. Since the root plant growth and plant mass were not measured it is possible that there were differences between plants below ground. A difference in root growth and mass in plants subjected to aluminum compared to the root mass and growth in plants without aluminum would be possible considering that roots are highly susceptible to Al poisoning (Delhaize & Ryan, 1995).

Further research could be done to find out if root mass, plant mass, and root length exhibit differences in beefsteak tomato plants treated with aluminum compared to tomato plants not treated with aluminum. By better

understanding how beefsteak tomato plants are affected by aluminum in different concentrations combined with nutrients, it will be possible to design treatments that more effective and efficient at remediation of the effects of AlNO_3 in the beefsteak species of tomato plant.

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PIEZO CRYSTAL GROWTH

Making crystals can be difficult. They are sensitive to slight changes in experimental conditions and demand close attention to detail. Armed only with a few materials and the desire to understand more, the following students created procedures, identified variables, collected data, and refined their technique in pursuit of the perfect crystal. None arrived at perfection, but all ended with a greater appreciation for how the scientific process moves us from ignorance to insight.

Piezoelectric Crystals: The Influence of Soda Ash and Location of Crustal Formation on Rochelle's Salt Voltage

Kathryn Kopchick

Piezoelectric crystals generate an electric charge when squeezed. One type of piezo crystals is Rochelle's salt. This crystal's ingredients include distilled water, soda ash, and cream of tartar. After combining these ingredients and leaving them to settle, Rochelle's salt is formed. Rochelle's salt forms best when the solution is left on the counter to form, and performance is best when 30 grams of soda ash are added.

Introduction

Discovered by Pierre and Paul-Jacques Curie in 1880, piezoelectric crystals are crystals that generate an electric charge when they are pressed. This function is useful in industry, providing parts for cigarette lighters, diesel engines, inkjet printers, and more.

Piezoelectric crystals come in many forms. Quartz, topaz, tourmaline, and Rochelle's salt are all examples of a piezoelectric crystal. In this experiment, Rochelle's salt was produced. The goal of this experiment was to determine the best method of creating Rochelle's salt. Using cream of tartar, distilled water, and soda ash, Rochelle's salt is easy to make, but difficult to perfect.

Materials and Methods

To create Rochelle's salt, distilled water, cream of tartar, and soda ash were used. A hot plate, funnel, coffee filters, and 2 beakers are also necessary for the experiment. 125 milliliters of distilled water were put into a 600 mL beaker, and then put on the hot plate to simmer. Once the water was steaming and slightly bubbling, 94 grams of cream of tartar were added to the water, and then were mixed in thoroughly.

One variable that was used in the experiment was the amount of soda ash that was added. After the cream of tartar was added, soda ash was added in amounts of either 26.5 grams, 30 grams, or 40 grams. To prevent spills, the soda ash was added in small increments and stirred well every time, because the solution bubbled

when it came in contact with the soda ash. After the soda ash was completely added, the hot plate was turned off. Then, the solution was drained into a small, clean beaker through a coffee filter placed in a funnel. Once the solution was completely filtered, foil was placed over the beaker to prevent possible evaporation or contamination of the solution.

The second variable in the experiment was the location of crystal formation. Half of the trials (26.5 g, 30 g, 40 g) were placed in the fridge after being prepared, and half of the trials (26.5 g, 30 g, 40 g) were placed on the counter. In a few days, Rochelle's salt was formed in the beaker. The rest of the liquid solution was dumped out, and using a scoopula, the crystals were extracted from the beaker.

Results

All of the trials produced Rochelle's salt. There was one outlier in the data-- Trial 5 produced Rochelle's salt with a voltage of 194 millivolts.

Table 1. Data about Rochelle's salt produced through 6 trials -- how the location of formation and amount of soda ash impacted the voltage.

Trial Run	Location of crystal formation	Amount of Soda Ash (grams)	Voltage (millivolts)
1	Fridge	26.5	65.0
2	Fridge	30.0	41.0
3	Fridge	40.0	36.0
4	Counter	26.5	76.6
5	Counter	30.0	194.0
6	Counter	40.0	50.4

With the two variables introduced, there were two trends observed. First, it was observed that the more soda ash added to the solution, the less voltage the Rochelle's salt produced. For example, Trial 1 had 26.5 grams of soda ash added, and produced a voltage of 65 millivolts, but Trial 3 had 40 grams added, and only produced a voltage of 36 millivolts.

The second trend that was observed concerned the location of crystal formation. As seen in Figure 1, the trials left to form crystals on the counter produced a higher voltage than the trials placed in the fridge to form crystals. For example, adding 40 grams of soda ash to the solution generated crystals that produced 36 grams when placed in the fridge, but produced 50.4 grams when placed on the counter.

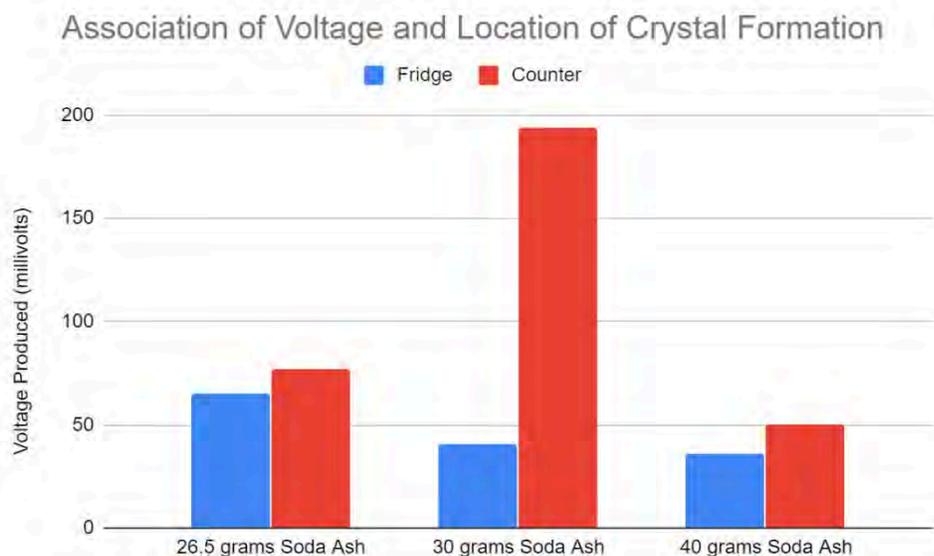


Figure 1. This figure shows the association between voltage and location of crystal formation. The trials that were located on the counter consistently produced a higher voltage compared to the trials located in the fridge.

Discussion

The crystals that were formed on the counter had a greater production of voltage overall. The reason this happened could be that the solution was able to cool slowly, which provided larger, clearer Rochelle's salt. In contrast, the solution that was put in the fridge cooled much faster because of the colder temperature of the fridge. This sudden decrease

in temperature could have disrupted the crystal forming process. Overall, the trials that had less soda ash in the solution produced crystals with a larger voltage. The solution became more saturated as the soda ash was added, and at one point, the soda ash stopped dissolving in the solution. The excess soda ash in the solution could have impaired the ability to form Rochelle's salt correctly.

Piezo Crystal Lab Report

Will Casto and Graham Shelton

Introduction

The purpose of this lab was to find the optimal method to create Piezo crystals. Piezo crystals are created by mixing Soda Ash (Na_2CO_3), water (H_2O) and ($\text{KC}_4\text{H}_5\text{O}_6$). When mechanical pressure is applied to a Piezo crystal, a measurable electric charge is created called piezoelectricity. However, in this context, optimal, the word referenced in the first sentence, means through changing certain variables creating both a sizable crystal (measured in grams) and one with a high voltage. The variables that might affect the end results are the amount of cream of tartar, amount of soda ash, and amount of distilled water used.

Materials and Methods

The materials needed to complete our experiment are as follows: at least two containers between 250 and 500 mL capacity, a coffee filter, a funnel, one hot plate, soda ash, distilled water, cream of tartar, a scale, a glass stirring rod, a small petri dish, and a multimeter. After acquiring these materials the first trial was performed. The experiment started with pouring a 150 mL of cream of tartar measured by a scale on a petri dish into a 250 mL of distilled water. Then, while stirring with a stirring rod, the container was heated by using a hot plate until simmering. Once the solution is simmering, 5 mL of soda ash were poured into the solution slowly, as to avoid bubbling over. Once the solution bubbled, the solution was stirred until there were no bubbles. This process was repeated until the solution had yellow tint and did not react when soda ash was poured in. The final amount of soda ash used was 47.5 grams. Next, the solution was poured through the coffee filter into a second container using a funnel as a guide. The solution was taken and heated until simmering and set out in a cooler environment.

A similar process was repeated for trial 2 using the same amount of cream of tartar (150g) and water (250 mL). However, the variable measured in trial 2 was the temperature the solution was kept at. In trial 1, the solution was kept at room temperature and in trial 2 the solution was kept refrigerated.

In trial 3 the variables that were changed from trial 2 were the amount of cream of tartar and water used. 200 grams of cream of tartar and 300 mL of water were used. The hypothesis was that if the amount of cream of tartar and water were increased the size and voltage of the crystal would also increase.

In trial 4 the amount of cream of tartar and water were decreased from trial 3 in hopes of maintaining the size of the crystal generated in trial 3 but with considerably more voltage. 50 grams of cream of tartar were used and 100 mL of water were used.

Results

The four trials generated very different results. A table of our variables and results are shown below. However, several trends in our data should be noted in future experiments.

Table 1. *The materials and Results of Trial 1-4.*

	<i>Cream of Tartar (g)</i>	<i>Soda Ash (g)</i>	<i>Water (mL)</i>	<i>Crystal Size (g)</i>	<i>Voltage (mV)</i>	<i>Refrigerated or Room Temperature</i>
<i>Trial 1</i>	150	47.5	250	NA	NA	Yes
<i>Trial 2</i>	150	45	250	81.6	-14.23	Room Temp
<i>Trial 3</i>	200	58.3	300	267.2	-6.56	Room Temp
<i>Trial 4</i>	50	17.3	100	11.16	-16.75	Room Temp

For instance, Trial 1 followed the same steps and measurements as Trial 2, except that

Trial 2 was left at room temperature. Trial 2 produced a sizable crystal. Additionally, trials 3 and 4 also produced crystals when left at room temperature, inferring that temperature impacts crystal growth. Another trend that can be noted is the relationship between cream of tartar and water added to the crystal size. As the amount of cream of tartar and water added increased, so too did the size of the crystal as noted by the table above. The relationship between addition of water and cream of tartar is a direct relationship. Also, the relationship between voltage and addition of water and cream of tartar should be noted. The relationship, as shown in the table is an inverse one. As the amount of cream of tartar and water increased, the voltage produced by the crystal decreased.

Figure 1. *Trial 2 produced a defined and sizable crystal.*

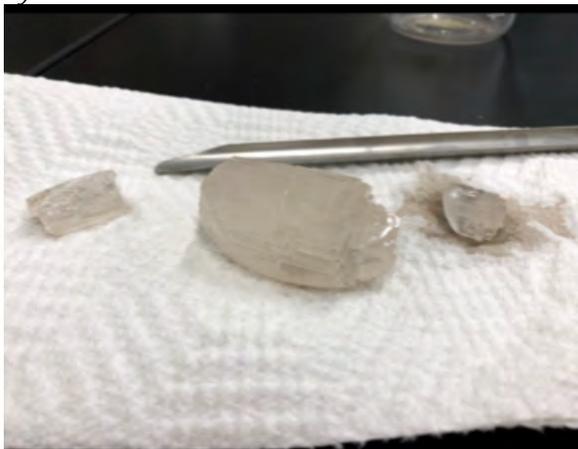
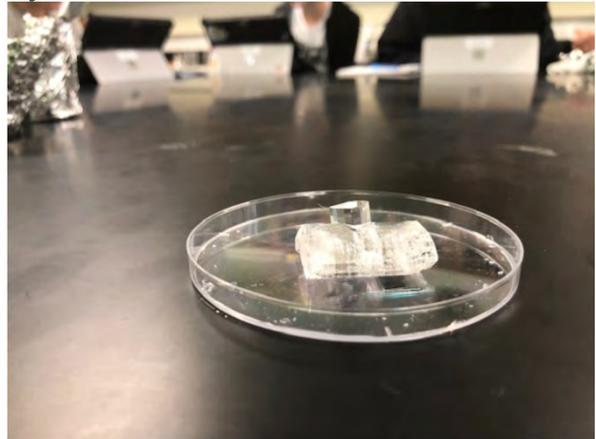


Figure 2. *Trial 3 produced a large but very grainy and not well-defined crystal.*



Figure 3. *Trial 4 produced a small but defined crystal.*



The final result that cannot be recorded on a table is the physical characteristics of each crystal. A Piezo crystal should be hard, transparent and have a defined shape. However, when 300 mL of water and 250 grams of were added, the crystals produced in trial 3 were grainy, not well defined and light brown in color. These qualities are in stark contrast to the similar looking crystals of Trials 2 and 4.

Discussion

There are many factors that go into creating the "best" crystal. The two biggest signs of a good piezo crystal are size and electricity. However size on its own does not necessarily mean the crystal is good if it does not conduct a lot of electricity. The best crystal is one that is a combination of both large size as well as being able to conduct a high amount of electricity for a piezo crystal.

In looking at the results of the sizes of the crystals, it seems pretty clear in what makes the biggest crystal. In trials 2,3, and 4, the trials that had the most grams of cream of tartar per mL of water produced the biggest crystals. Trial 3 had by far the biggest mass at 262.7g with .667 grams of cream of tartar per every mL of water, while trial 2 produced an 81.6 g crystal only having .6 grams of cream of tartar per mL of water. Lastly, the 4th trial produced the smallest crystal of 11.16g with it only having .5 grams of cream of tartar per mL of water.

However regarding creating crystals that produced the most electricity, the amount of cream of tartar had the opposite effect. The smaller crystals conducted the most electricity. The less amount of grams of cream of tartar per mL of water created the highest amount of electricity. Trial 4 produced -16.75mV while trial 2 produced -14.23mV, and with the biggest crystal, trial 3, only producing -6.56mV. Creating the best crystal would mean having a balance between having good size as well as conducting a sizeable amount of electricity. Trial 2 was the best crystal made because it had a good size of 81.6g while conducting a high amount of electricity at -14.23mV. In creating future experiments, the

crystal could be made even better if the amount of grams of cream of tartar per mL of water is the same (.6g), while increasing both the amount of water and cream of tartar. This would make a bigger crystal while still conducting a high amount of electricity for a piezo crystal.

GOVERNMENT AND FREE THOUGHT

The relationship between a government and its people is complex. Diverse stakeholders compete for access to the halls of power and expect the government to encourage a just and fair society. But what happens when the people don't agree on how the government should act? The following essays use roughly 1200 words to address how the government ought to interact with the beliefs of its citizens by examining that relationship through different lenses and in different contexts.

Each view is the author's own. While we don't endorse any of the particular positions held in the following pages, we do support the idea that rational discussion ought to grow out of comprehensive background research. We hope these essays help you come to the same conclusion as well.

Social Media's Impact on the Government and Population Relationship

Joshua Brunner

Social media has exploded since the beginning of the 21st Century. Mostly intended as a platform for posting pictures, stories, and videos, Social Media has recently taken on a political side. Chase Gunter, former FCW staff, cites a 2016 Survey that cited over 90% of U.S Representatives had adopted Facebook, Twitter, or YouTube (Chase Gunter, 2018). In addition, according to a report made by the software company Quorum, the members of the House made over one million social media posts (Kevin King, 2018). These posts range from commentary on tax reform, laws, presidents, news, opinions and more. There is not anything inherently wrong with this commentary, however, recently there has been a flood of conspiracies, half-truths, and misinformation floating around that is being posted by U.S politicians (Bolter, 2019). For example, Rep. Alexandria Ocasio-Cortez stated that the Southern Border facilities were "concentration camps" (Alexandria Ocasio-Cortez's Twitter). Alternatively, President Trump supported a conspiracy theory that the Clintons had murdered Jeffrey Epstein (Jane Timm, Liz Johnstone, 2019). Both politicians were proven wrong in their statements, however the vast majority of bought into the misinformation that the two posted. The result was mass feuds within the greater population as well as the government. Considering these online interactions are only going to increase with an ever-growing population, could the use of social media by politicians to convey information to their constituents negatively affect the relationship of the government and the population in the future?

This is not the first time that new media has caused a stir in the political scene. Television (TV) was part of a revolution in which political candidates became individuals. They did not have to go through the traditional means of media like the press; instead, they were able to propagate their agenda to every person, without fear of being censured. In a periodical during the

time, the author McLuhan, a professor at the University of Toronto who studied media theory, recognizes that it transformed political parties, how people vote, and demanded less depth than traditional post or radio (McLuhan, 1968). The reason was a movement from a regional stage to that of a global. From that stage, TV bypasses the press and the ballot box (McLuhan, 1968). McLuhan claims that TV created political representatives or individuals, who focused on image instead of any point of view, comments of the press, or interests. He even went as far as to say it ended all national and party politics-people voting along the party (Republican or Democrat) that they align with, regardless of the candidate. Some think that then end to party politics is a good thing. For example, Former Clinton Press Secretary Mike McCurry in an interview stated there had been an erosion in public confidence in political parties at that time. He claims that due to TV's infiltration into the nuclear home, a candidate could hone his agenda to fit any group that he wanted (Bigger Than TV itself, 2000). The individualistic trend or transformation stemming from the regional to the global stage is just as prevalent in today's day and age with social media.

Just like Television, Social Media is everywhere and is connecting politicians and people at a new intimate level where a candidate can interact with their base. According to Pew Research Center, 81% of all Americans own a smartphone (Demographics of Mobile Device Ownership and Adoption in the United States, 2019). To add on to that, Pew conducted a study citing that two-thirds of all American adults get their news from social media (Matsa; Shearer, 2018). To highlight the connection, take the 2020 Democratic Primary Race for example as there are over 20 candidates vying for the nomination. Contrast that to a decade ago in the 2008 Primary Race when there were only 8 candidates in the running for the primary (Burns, 2019). This increase might not be significant to some,

however, the candidates' social media following positions the massive amount of hopefuls in an interesting light. Looking at their social media reveals millions of followers on Instagram, Twitter, and Facebook (Scripps National, 2019). However, it is not just followers, it is the interactions between the candidate and their base. The Social Media Platform CrowdTangle conducted a study of the top democratic hopefuls' twitter and Facebook, finding several candidates with a massive interactive base (Lapowsky, 2019). The study demonstrates that there is a breadth of candidates for people to connect to based on who they like, instead of having only a couple to choose from. The amount of candidates paired with an interactive base shows that Social Media supports the trend first indicated with television: people are shifting their votes from along party lines to people they prefer.

Despite connecting people together, Social Media does not promote objective ideology and news. Social Media was built with society in mind. The very word social is in its name. Twitter, a highly used media platform, is a hub for discussion, especially political. The only problem is that Twitter has a character cap at 300, which is a severe limit to discussions that should be complex. In a study about political discourse on twitter in Malaysia, Khasmani, who holds a Ph.D in Political Science and studies political communication, cites a source that claims twitter and other forms of media are full of "sensation-seeking individuals" who focus on celebrities, scandals, rumor, and attacks (Can Twitter be an Effective Platform for Political Discourse in Malaysia? A Study of #PRU13, 2014). Thus, large misinformation campaigns run rampant about the latest news trying to get people to view and talk about what is being said. Michael Gurevitch, a former professor from the College of Journalism of the University of Maryland says that politicians take part in this when appealing to their base because feel like they need to appear like an ordinary person (Gurevitch, 2009). So politicians get caught up with the feuds and misinformation that dominates discourse on the platform.

An example of this trend is the tweet by Rep. Ocasio-Cortez. Her comments were brief

and simple: calling the border facilities "concentration camps". This appealed to her base while others were infuriated by it. The comments of the tweets and many others are filled with name-calling and accusations. In reality, she did not fully explain herself because the cause of her tweet was about the shortage of supplies that the CBP had. However, the truth was glossed over because it was not sensational and stimulating or in other words. On the other hand, some say social media discourse can be a good thing. Since social media allows for everybody to have a voice, democracy is strengthened since people can input their own opinion at any time (Can Twitter be an Effective Platform for Political Discourse in Malaysia? A Study of #PRU13, 2014). Combined with the character limit, dialogue can be quick and effective, offering many viewpoints instead of reading long periodicals and journals. So in another sense, politicians posting has the opportunity to generate discussion that is wholesome for democracy and possibly generate change for the better.

As time passes, we need to look not only at the present, but also to the future and the possible ramifications that social media posting has on the relationship of politicians and people. With the rise of new technological mediums such as TV and Social Media, politics takes on a more global stage. People start to resonate more with individuals instead of parties. Along with that, social media has a lasting impact on discussion, both good and bad. People fall for misinformation and get sucked into feuds. However at the same time, democracy is being elevated because more people have a voice at the table. It is tough to say that these developments have a negative effect on the relationship between the government and the people because of the trends that are happening now are not finished yet. However, one thing is for certain, Social Media has changed the way we look at parties and how we resonate with political individual (The New Media's Role in Politics, 2018). For now, with anything new, the best we can do is to watch and wait for the possible outcomes of the trends of Social Media in order to better answer the question of whether Social Media is affecting the government and people for the worse.

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The Constitutionality Behind the Gun Debate

Gabriela Salerno

In recent discussions of gun control, a controversial issue has been how to implement laws without compromising the second amendment of the United States constitution in regard to gun safety. On one hand, some argue that stricter gun control laws are necessary. From this perspective, politicians want to call for universal background checks on gun purchasers and prevent more mass shootings in the United States. On the other hand, however, others argue that stricter gun laws go against the Second Amendment. Based on the Supreme Court case *District of Columbia v. Heller*, one of this view's main proponents given by Judge Karen LeCraft Henderson was that, "the [Second] Amendment's 'core' extends to publicly carrying guns." (Sinha). According to this perspective, going against the Second Amendment of the United States Constitution is unconstitutional and an infringement on public policy in this nation. In sum, the gun control issue is whether the United States government should have regulations of gun control at all, especially since there are multiple diverse persons in the country with differing and abstract opinions.

In order for a government to faithfully govern a group of people, it must obey its foundational principles and encompass all of its laws into its policies. When the United States became a nation, it set forth a Constitution which included a Bill of Rights which gave all citizens guaranteed personal freedoms. Within these documents lies the Second Amendment, which states that "a well-regulated militia, being necessary to the security of a free state, the right of the people to keep and bear arms, shall not be infringed." (Constitution). Throughout United States history, people have risen to contest these personal freedoms at many occasions, including Supreme Court case, *District of Columbia v. Heller*. The case began when Heller sued the District of Columbia because he sought an injunction against the enforcement of the relevant parts of D.C.'s Code and argued that they

violated the Second Amendment right to keep a functional firearm in his home without a license. (Supreme). In the case, the court held that the "Second Amendment protects an individual's right to possess a firearm unconnected with service in a militia, and to use that arm for traditionally lawful purposes, such as self-defense within the home." (United). With this being said, the United States government cannot prevent people from owning firearms, but there could be a way to prevent guns from falling into the wrong hands.

The United States stands as a supreme power in the world and is used by many countries as an example. However, there is nothing wrong with using other countries as examples too. According to an article written by an Professor of Stanford Law School with over 25 years of experience in politics and law, John J. Donohue III, in response to the controversial gun control issue, Finland issued a law stating that handgun license applicants are allowed to purchase firearms only if they can prove that they are active members of regulated shooting clubs, and before they can get a gun, they must pass an aptitude test, submit to a police interview and show they have a proper gun storage unit. And in Italy, to secure a gun license, one must establish a genuine reason to possess a firearm and pass a background check considering both criminal and mental health records. (III, John). In following either Finland or Italy, the United States would by no means be denying any citizen the opportunity to try to possess a gun, however, it would be filtering out possible mass shooters and preventing guns from landing in the wrong hands.

One would assume that implementing a similar gun control policy such as Finland or Italy would be the answer, however, those who are 100% against guns and want the strictest laws passed would say otherwise. In passing any type of law, the government must be solicitous towards the people. This includes taking into account what the people favor. According to a survey reported by Kevin Robillard, a well-

respected senior political reporter, and conducted by a CBS poll in regard to those in favor with gun control, 57% of the 620 adults who took the survey believe that gun control laws should be stricter. (Robillard). In response to similar polls, multiple Democratic candidates are trying to relate to the American people through their stance on gun control laws. For instance, in an article reported by Reid J. Epstein, who has a master's degree from the Columbia Graduate School of Journalism with over 10 years of reporting experience, presidential candidate Senator Cory Booker of New Jersey vows to reinstate a ban on assault-style weapons. (Epstein). In an age where many new voters have experienced mass shooting, candidates, such as Cory Booker, are appealing to many citizens.

Although it may seem like the right move to ban assault-style weapons and institute bans on guns, these acts would be deemed unconstitutional. Referring back to the Second Amendment, all people have the right to bear arms. If the United States were to become a nation in which all guns, or even simply assault-style guns, were to become banned, we would be implementing similar governing styles to those of the United Kingdom and Japan where handguns are illegal for private citizens (Ill, John). It is important to take into account what the people want, but to a certain extent.

Constitutional interpretation is a vital part of our government. Specifically, the United States has demonstrated, in all of its years as a nation, an "unprecedented determination" to preserve and protect its written constitution, thereby providing the American "experiment in democracy" with the oldest written Constitution still in action, according to the Supreme Court. In other words, the constitution was a carefully balanced and articulated document, which was designed to provide for a federal government sufficiently strong and flexible to meet the needs

of the republic, yet sufficiently limited and just to protect the guaranteed rights of citizens. (The Court). In its balanced, yet specificity in regard to various issues, it can be said that there is room for interpretation. This is why the Supreme Court is present. They determine if the laws are just and in accordance with the constitution.

In creating laws, it is vital for the future of the United States to be consistent, be solicitous towards the well-being of its citizens, and encompass the ideals outlined in the Constitution, especially in regard to personal liberties and freedoms. With this being said, I believe that the United States should take into account how people feel in regard to gun control. This could be done as followed:

Using either Finland or Italy's gun control policies, as exemplified prior, is a viable solution. This would create a system of background checks and limits on those purchasing guns without placing an outright ban, since a ban would be an infringement on the Second Amendment. Thus, in using a model gun control policy, we can subdue most conflicts that have risen between those who want a complete ban and those who want no limitations in purchasing or using guns.

The Constitution was written to ensure that the government would not take away the people's natural rights including those entitles to all citizens in the Bill of Rights. Therefore, when dealing with a diverse group of people in the United States, the government should implement restrictions in regard to gun control by closing the gap on those who can obtain guns but remaining in accordance with the Constitution. Only then can the United States truly implement regulations on gun control and do so in a way that would quell political debates and tension between all groups of people.

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Ethics of Surveillance

Corbin Bates

Throughout history, governments have used assorted methods to patrol and police the general population behind the scenes of society. These methods of spying and surveillance have changed and evolved through the centuries and have recently been exposed to the public through the efforts of Edward Snowden and his case against the NSA. These new events have brought to light a centuries old debate about the ethics of surveillance. In this post-Snowden era the public is outraged about the government's invasion of their privacy. Social media is ablaze with citizens claiming that their privacy has been violated. The government regularly responds through social media and public statements to these tweets by saying that this invasion of privacy is ultimately for the "safety" for the public (IV) (V). Is surveillance of US citizens by the NSA beneficial for the country? Moreover is it ethically acceptable to invade someone's privacy in order to protect the general population?

In the Texas Review of Law and Politics Clark M Neily argues that the United States government was created under the principle that ethically acceptable governmental action protects the life and liberty of its people (I) (III). This is demonstrated throughout the Declaration of Independence which reads, "We hold these truths to be self evident that all men are created equal with certain unalienable rights that are endowed to them by their creator. Among these are life, liberty, and the pursuit of happiness"(I). The declaration goes on to state that government is put in place in order to secure these rights and that their power comes from the people (I). Neiley argues that the US Constitution was created with these principals in mind and was created in order to assure these rights and freedoms (III). This infers that the violation of the people's liberty is comparable to violating the law.

The legality of the government's actions was formally challenged in the case of Jewel vs the NSA as documented by the United States Courts. The government was challenged on its

surveillance of US citizens by multiple people who raised claims against the NSA saying that their constitutional rights were violated without probable cause (II). This accusation included but was not limited to the 4th amendment. The prosecution says that under the Terrorist Surveillance Program or TSP, the NSA and other government figures violated their privacy with the help of major telecommunication companies through wire taps and internet surveillance (II). The 4th amendment protects the people's rights against search and seizure, the argument stands that the NSA illegally searched and stole personal data from people's cell phones and computers (II). The occurrence of these events is indeed verified through the revelations of Edward Snowden but whether these actions were lawful and moreover ethical remains the question. While this amendment was created in an age where digital search and seizure was not yet a reality, the founding fathers most likely never intended this amendment to be used in such a way, however the concept of one's right to their privacy without probable cause or sufficient evidence still rings true. But what qualifies as probable cause?

The United States Department of Defense or DoD argues that the prevention of a terrorist attack qualifies as probable cause (V). In 2006 before the Snowden outbreak Air Force Gen. Michael V. Hayden, principal deputy director of national intelligence made a statement on behalf of the DoD about the TSP saying, "This is focused on al Qaeda... The only justification we have to undertake this program is to detect and prevent attacks against the United States" (V). The DoD went on to argue that the TSP is specifically made to target terror attacks and that the public has nothing to fear on the subject (V). However in 2013 everything changed as Snowden leaked the NSA's surveillance as it was justified under the TSP. If this was the TSP's original state or if it was wrapped after the General's statement is unknown, but the government continues to justify this invasion of privacy by repetitively referencing the good intention of the TSP and its

war on terror. In the article *Fair Play: The Moral Dilemma of Spying* James Olsen, a 30 year Director of operations for the CIA argues that questionable actions “become honorable” when they are necessary (IV). The article argues that in a “post 9/11 world” the ends justify the means argument is more legitimate than ever (IV). However this argument is countered later in the article when the author says that ethical boundaries will prohibit the government from going too far in its spying or in any intelligence operation (IV). While they say that spying is not only necessary but also honorable, they also believe that the governments moral compass will prevent any misuse of this power (IV).

Some people however are less confident in the government's intentions and morality. Privacy International, an activist group that advocates for privacy states, “Privacy is a fundamental right, essential to autonomy and the protection of human dignity, serving as the foundation upon which many other human rights are built” (VII). Privacy International goes on to argue that privacy is a way people can protect themselves against unwanted interference in their lives from others (VII). Privacy protects against threats in power imbalances and many people think of the government as a threat (VII). For this reason many people want privacy *from* the government not privacy *through* the government.

So is the forfeit of privacy too high a price to pay for safety? It seems as if the two are equally important for privacy without life is death and life without privacy is oppression. So the question becomes when do the ends justify the means, and how far is too far? Toure-Tillery of the University of Chicago in the *Journal of Experimental Psychology* addresses this issue with the article *“The End Justifies the Means, but Only in the Middle”*. His argument states while everyone has moral standards, people are willing to compromise them in order to obtain an end goal (VI). This contradicts the CIA's statement that moral boundaries prevent people from crossing line in order to complete a task. People seem to believe that the question is less about whether the ends justify the means and more about if the people involved in the process can be trusted with the means in order to complete the end

goal. Many people entrust their privacy to friends and loved ones, however once the government gets involved they become defensive. People just do not trust the government enough to entrust them with something as valuable as their privacy.

A quote from Uncle Ben in the movie Spiderman coined the term, “with great power comes great responsibility” while this saying comes from a fictional movie its words could not ring more true today. A hero like Spiderman fighting for the people and protecting our freedom is an ideal that is very popular. However when the government is given this “great power” people are more hesitant to trust them with the responsibility it entails. The people of the United States do not fundamentally see their government as good or moral enough to possess this ideal. Is the government responsible enough and moral enough to be trusted with a person's privacy? Is it ethically acceptable to invade one's privacy even when it may serve the greater good? These are questions everyone should ask themselves in this new age. Government surveillance happens, now it's time to decide what to do about it. Is it ethical, and if so how do we make sure that it stays that way? Or is it unacceptable, and if so, is it time for people to rise up against it? What is more important to you, your freedom or your life, and whom do you entrust with them?

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The Effects of Social Media on Political Perceptions

Ellen Shales

In the 2019 election of Brazilian President Jair Bolsonaro, his supporters chanted “WhatsApp” and “Facebook,” acknowledging the role these social media platforms played in communicating his campaigns and ideology [1]. This is a shocking example of the connection between social media and a major political victory. Social media is a globalized phenomenon seated in an age dominated by technology and its accompanying ease. Social media now delves into the realm of politics and news. Modern individuals face the problem of understanding the complex relationship between political news and social media, and the question of whether or not this relationship should exist at all.

Pew Research Center conducted a study in 2018 on the viewage of news on social media platforms. This study revealed 68% of American social media users receive news information on their respective platforms, and 21% proclaim convenience as the reason behind this fact [2]. The benefit of convenience, however, is fighting a losing battle with social media’s inherent flaws. Mihaly Csikszentmihalyi, Claremont Graduate University’s Distinguished Professor of Psychology and Management, created the concept of “flow” in the 1990s to describe the optimal experience of focus and content engagement that results in a reality-shedding experience [3]. Video games and media use the concept of “flow” to suspend viewers in active states of progression and movement from challenge to challenge, post to post, video to video. In the context of social media, however, “flow” prohibits meaningful engagement with information and proper analysis of its validity [4]. Social media users feel the effects of “flow” and content gratification even while browsing through serious political content.

While social media provides convenient access to news and politics, users are often distrustful of the information they acquire. Pew

Research Center’s study discovered just over half, 57%, of consumers expect inaccuracies in the news they find on social media [2]. These consumers are more than justified in their distrust. The Knight Foundation, a non-profit organization dedicated to informed journalism, discovered a “fake news” ecosystem on the platform of Twitter before the 2016 United States Presidential election. Of the 10 million tweets analyzed, over 6.6 million tweets were linked to conspiracy news publishers a month before the election. This spread of misinformation was linked to both Democratic and Republican affiliated accounts, continuing a month after the election with a decrease to 4 million conspiracy related tweets [5].

RAND Corporation links the notion of objective truth decay to the Knight Foundation’s discoveries of “fake news” networks. In 2018, RAND published a book titled *Truth Decay*, analyzing the social trends of the 21st century and their impact on truth and the perception of truth. These trends relate to the overload of opinions in comparison to fact, and the weak distinction of factual analysis versus objective truth [6]. It is no stretch to connect these shifts to social media. RAND’s findings highlight the implications of “fake news” on society’s ability to understand objective truth.

While conspiracy theories and intentionally misleading content may cause the degradation of overall truth, they also may be feeding undemocratic behavior. Despite society’s tendencies to understand social media as a natural deterrent of authoritarianism through its global capacity and pursuit of free speech, Ronald Diebert views social media as entirely compatible with such un-democratic practices. Diebert, Professor of Political Science at the Munk School of Global Affairs and Public Policy, University of Toronto, realizes the ability of social media to overload users with misleading information to

sow confusion. He goes so far as to say that this overload of information drives authoritarian practices. "Fake news" dumps are a useful tool to the manipulative politician seeking to embed false narratives in the minds of the people. He substantiates such a claim by highlighting the emotionally charged environment social media creates [7]. A breeding ground for opinions masquerading as fact, online platforms present an efficient means of circulating false information at the behest of a political figure, whatever their intent may be. This perspective supplies a rationale behind the social media user's distrust of political information [2] and the existence of fake news ecosystems [5], allowing for a broader view on social media as an instrument of authoritarianism for political discord. In an article written by Jay David Bolter from his book *The Digital Plenitude: The Decline of Elite Culture and the Rise of New Media*, he explains the difficulty of the social media user to sort through the massive amounts of conspiracy theories, contradictory information, and wild accusations. Bolter zeroes in on Donald Trump, and his notorious presence on Twitter. The President's tweets have mentioned serious political topics such as Russian collusion, his predecessor President Obama, and his Presidential campaign opponent Hillary Clinton. During an August 2018 survey by Statista, 61% of US adults stated they believed Trump's use of Twitter was inappropriate [8]. Bolter, using Trump as his precedent, hypothesizes the continued change of political discourse as social media becomes increasingly used for convenience and subconscious gratification even on such weighty topics [4].

As the concept of political discourse changes, so does the notion of political identity. In a 2018 research study conducted by Duke University's Department of Sociology led by Professor Christopher Bail, the question of polarization was tested in social media users. Bail discovered politically antagonistic content solidifies an individual's preconceived beliefs and adherences to party values. This study focused on Twitter users, and paid them to follow a bot that retweeted posts of the opposing political views than they self-identified with. After a month of exposure to rival political views, Democrats

remained relatively unchanged in their personal opinion, while Republicans became more conservative and supportive of party views [9]. While Bail himself acknowledges the involvement of other factors that may have affected the study, the results arouse interesting notions of the social media user and their commitment to their beliefs despite exposure to antagonistic content.

While viewing opposing content solidifies political opinions, exposure to very specific forms of aligned political content further grows these opinions. In a study published in the *Journal of Experimental Psychology* by William Brady and his colleagues, the role of moral and emotive characteristics was analyzed in retweets on Twitter. He discovered a combination of moral undertones and emotion provoked the greatest diffusion of content across social networks. This implies purely emotional content lacks proper conviction for social media users to retweet. However, when the potent blend of morality and emotion does exist, the content will inevitably be spread, regardless of its veracity [10].

Zack Beauchamp, Senior Correspondent at Vox, believes the previously outlined faults in social media are slowly deteriorating democracy and jeopardizing global politics and freedom as a whole. He blames far-right politicians as misusing social media to create social divisions. Pro-democracy politicians are less likely to benefit from social media's chaotic anger as they are greater proponents of equality and freedom, says Beauchamp [11]. Disregarding the bias of Beauchamp's claims, he represents the drastically negative outlook on social media and its part in the degradation of modern globalized democracy.

Despite the realistic overlap of social media and negative concepts such as polarization and truth decay, politicians continue to use social media to bolster their campaigning. In an article co-written by Hannah Murphy, tech correspondent, and Demetri Sevastopulo, the Washington Bureau Chief at Financial Times, the connection between Instagram and 2020 election campaigning was created. Notably, Alexandria Ocasio-Cortez and Beto O'Rourke are documenting their political and lifestyle choices

through social media, forgoing traditional means of political promotion [12]. As startling relationships are established between truth decay, polarization, and authoritarianism through social media, the question of its use in current and future politics inevitably arises. The benefits to the unlimited, open discourse of social media may not be enough to hide the disconcerting shadows of its inherent shortcomings.

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Government Regulations on Gun Control

Lydia Garrett

People will never agree upon the matter of gun control regulations in the United States, but we can impinge on how people act towards the matter. In discussion of gun regulations and laws, a controversial issue is whether the United States should spread uniform regulation over a diverse group of people. While some argue that it is a part of our nation's history to allow guns with regulations, others contend that more must be done to ensure the safety of citizens.

As seen in the history of the United States, it is important for the government to balance regulations on gun ownership to ensure the safety of a diverse group of citizens. The nation's history is rooted upon the Constitution, which has been upheld for many centuries. When looking at the Second Amendment, Barlow, a J.D. candidate, argues that the framers of the Constitution allowed space for diversity. This, however, makes it difficult to have a uniform interpretation.

There are some who support regulations. In fact, gun laws and regulations have been around as early as 1791 (Congress). More recently, the Firearms Control Regulations Act of 1975 is one interpretation of the Second Amendment that has increased gun control in the United States (113th Congress). Edward D. Jones, III, a graduate from the University of Chicago and former member of the U.S. Department of Justice, writes upon the matter. In regards to the Act, he states, "The objectives were reflective of the failures of the 1968 registration law, most visible in a record number of homicides in 1974 and in the ease with which juveniles were obtaining access to handguns" (Jones). Johnathan Masters, a deputy managing editor whose works have appeared in the Atlantic, Bloomberg, and Foreign Affairs, agrees that gun control is needed because of America's history with homicides. He reported the following: "It [the United States] ranks number one in firearms per capita. The United States also has the highest homicide-by-firearm rate among the world's

most developed nations" (Masters). Having firearms readily available and accessible, they argue, comes with a price; the price of human lives every day.

The relationship between gun regulation and homicide rates is highlighted by comparing the United States with foreign nations. Countries such as Switzerland and Finland have a history of homicide rates and suicide rates that are much lower than the United States. David Hemenway, a professor at Harvard, writes, "We analyzed the relationship between homicide and gun availability using data from 26 developed countries from the early 1990s. We found that across developed countries, where guns are more available, there are more homicides" (Hemenway). If the United States decreases the amount of guns available, Hemenway implies that the homicide rate might decrease as well. From a study by University of Nevada-Reno and Harvard School of Public Health, "Homicide rates were 7.0 times higher [in the U.S.] than in other high-income countries, driven by a gun homicide rate that was 25.2 times higher." Overall, the death rate from firearms in America from all causes was 10.0 times higher (2016). This study concludes that the U.S. had a much larger and impactful gun problem than other high-income countries such as Argentina or Israel. If these conclusions are valid, it is beneficial— given our nation's income status similarity to others— to enact regulations regarding firearms in order to decrease the homicide and suicide rates in the United States.

When looking at countries such as Argentina and Israel, it can be tempting to use them as a model for the United States. However, the U.S. is unique in its diversity and legislative history, which is primarily seen in the Second Amendment. There have been many who interpret the Second Amendment differently, resulting in repeals of previous acts. For example, The Firearms Control Regulations Act of 1975 was repealed by the Supreme Court case *D.C. v. Heller*,

addressing the issue of the Right to Bear Arms. E. Garret Barlow wrote a Second Amendment interpretation article in the Brigham Young University Law Review. The article explains that the Heller majority argued, "The First Amendment contains the freedom-of-speech guarantees that the people ratified, which included exceptions for obscenity, libel, and disclosure of state secrets, but not for the expression of extremely unpopular and wrong-headed views. The Second Amendment is no different. Like the First, it is the very product of an interest balancing by the people" (Barlow). This Supreme Court protected the right to bear arms for self defense, in contrast to the The Firearms Control Regulations Act of 1975, even though this right creates potential conflict.

Potential conflict is inevitable, in part, because the 50 states in America all have various ideals and values. With that comes different disagreements and introduction to state laws. States with a higher homicide rate, such as Chicago, have cracked down on gun control. However, other states, such as Wyoming or Alaska, have some of the loosest gun laws in the nation. The state of California has one of the strongest gun safety laws in the nation, for they have had devastating shootings such as the Thousand Oaks shooting. Kevin Gardner, Chief of the California Department of Justice Division of Law Enforcement, signed into effect a new and amended firearm/weapons law. This includes a restraining order that, "Expands the definition of 'ammunition,' for the purposes of the Gun Violence Restraining Order (GVRO) law, to include a 'magazine'" (Gardner), along with other heightened laws. California, in the past 20 years, has cut gun deaths in half due to their strong gun laws (Jones). Certain states, like California, believe that the current national laws are not enough, so they tighten them.

These benefits, however, do not seem to show up everywhere. In a study in Applied Economics Letters by Mark Guis, in the department of economics at Quinnipiac University, "States with restrictions on the carrying of concealed weapons had higher gun-related murders... Assault weapons bans did not significantly affect murder rates at the state level.

These results suggest that restrictive concealed weapons laws may cause an increase in gun-related murders at the state level" (Guis). Criminals will not obey the law, therefore if guns are outlawed— or highly regulated to the point where it is nearly impossible to obtain a gun— only outlaws will have these weapons. It is implied that gun control laws do not decrease the rate of crime, rather the ownership of guns decrease the crime rate. What works in California, fails somewhere else, and we are left wondering if regulations have fixed the problems they sought to address.

One takeaway from the history of gun regulations in the United States and around the world, is that there are many interpretations of laws (most importantly, the Second Amendment) that lead people to believe what they do. Though gun regulations will not stop the crime rates across the entire United States, we can learn from the history of the nation on how people will react, determining where the United States should pass certain regulations of gun control over a diverse group of people.

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Does the use of social media by U.S. politicians negatively impact the relationship between the government and society?

Phyllis Lin

The use of social media in politics, including Twitter, Facebook, and YouTube has dramatically changed the way campaigns are run and how Americans interact with their elected officials. Thomas Zeitzoff, an American University associate professor, has stated that the advent of this new form of expression has enabled an unprecedented number of politicians to enter the political arena by expressing their opinions about issues in ways that can influence countless voters and officials (Zeitzoff, 2017). Social media not only reduces the costs of communication to increase the speed and dissemination of information but also gives politicians an opportunity to build trust with the public. Although it allows more engagement from the public, which is good for social and political development in the U.S, giving politicians the power to send unfiltered tweets in social media can negatively impact the relationship between the government and society.

With social media penetration deepening among both citizens and political figures, there is a pressing need to understand how political use of major platforms is electorally influential. Millions of people are using sites like Facebook and Twitter and they're using these sites for expressing their views on just about anything. The donors, board members, and local elected officials are using them too. Lily McElwee and Taha Yasseri from the University of Oxford say that few have considered implications for campaign fundraising despite its recognized importance in American politics. Drawing on candidate-level data from Facebook and Twitter, Google Trends, Wikipedia page views, and Federal Election Commission donation records, they analyze the relationship between the topic and volume of social media content and campaign funds received by all 108 candidates in

the 2016 US Senate general elections. By applying an unsupervised learning approach to identify themes in candidate content across the platforms, they find that more frequent posting overall and of issue-related content are associated with higher donation income when controlling for incumbency, state population, and information-seeking about a candidate (McElwee & Yasseri, 2017). As candidates and fundraisers, one of the primary responsibilities is to engage donors and prospective donors in conversations because "An engaged audience is a happy audience."

It is also undeniable that social media helps to build trust between politicians and the public. As Tony Tran, an Assistant Professor in the Communication Department at Boston College, mentions, more than half of voters want to see more authenticity and transparency in their government (Tran, 2019). A Columbia University and Harvard University graduate Zachary Sniderman says that almost every social media company has worked with local, state, and federal governments all over the world. All of them know the unique requirements for social communications at all levels of government to adapt to a rapidly advancing world but also to leverage the changes to their benefit (Sniderman, 2015). One great example of this is Alexandria Ocasio-Cortez, who leverages Instagram to give her followers a behind-the-curtain look at how Congress operates. As Tom Murse, a former political reporter and current Managing Editor of daily paper "LNP," says, the prevalence of social media in politics has made elected officials and candidates more accountable and accessible to voters (Murse, 2019). The social media companies' job is to look at some of the challenges governments face in communicating with the public and offer solutions on how to mitigate risk.

When politicians are more transparent and keep the public engaged, they can keep citizens informed about the policy and issues that matter most to them because as they open themselves up, the public will trust them more. Tony Tran says that a video from the National Park Service's Instagram account was designed by local governments to organize town halls, educate citizens on safety issues, and engage community groups. Even though it is merely 30 seconds long, the video was an engagement hit garnering more than 25,000 views (Tran, 2019). Social media tools have allowed Americans to quickly join together to petition the government and their elected officials, leveraging their numbers against the influence of powerful lobbyists and monied special interests (Tran, 2019). This is a way politicians can engage in conversations with the public without additional cost, which allows politicians to humanize their brand and show audiences that they are human too. Just like a business, the ability to remain authentic and honest with the audience for the government is crucial to building a good relationship with the public. Social media provides a perfect avenue to remain as transparent and clear with all the constituents as possible.

Social media can reduce the government's spending. Eric Katz, a Senior Correspondent from Government Executive states that the U.S. Social Security Administration spends about \$275 million per year on call centers to answer questions and solve issues. Social media can dramatically cut those costs. Instead of spending on advertising, government entities can now leverage more cost-efficient social platforms to raise awareness about issues the public needs to know about. After all, these are platforms their audience already uses (Katz, 2019).

It is obvious that direct access to voters also has its downside. Handlers and public-relations professionals often manage a candidate's image, and for good reason: Allowing a politician to send out unfiltered tweets or Facebook posts has landed many a candidate in hot water or embarrassing situations. A good

example is Anthony Weiner, who lost his seat in Congress after exchanging sexually explicit messages and photos with women on his Twitter and Facebook accounts. Weiner lost the New York mayor's race following a second scandal and ended up serving prison time when one of his "sexting" partners turned out to be underage (Murse, 2019).

Using social media allows politicians to engage in conversations with the public without additional cost, which allows politicians to humanize their brand and show audiences that they are human too. Just like a business, the ability to remain authentic and honest with the audience for the government is crucial to building a good relationship with the public. Social media provides a perfect avenue to remain as transparent and clear with all the constituents as possible.

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Ethics of Uniform Gun Control

Caleb Rathbun

Introduction

In the wake of continuous devastating mass shootings, Americans tend to draw heavy partisan lines, further separating the country in times of much-needed national unity. The growing Everytown for Gun Safety movement points condemning fingers at the gun industry, claiming that their business practices “directly threaten public safety” (Everytown). In return, businesses such as the NRA fire back, referencing the Second Amendment right to bear arms and diverting blame instead to the mental health of the shooters. Through all the debate, Americans take opposing sides, holding firmly to their views, believing they have the complete, perfect solution. Americans turn their attention to the government, believing it may have some clear and cohesive solution. This, however, is obviously not the case as shootings continue to abound. The government, like the people, find themselves similarly divided and therefore unable to enact any one successful solution.

Washington DC talks endlessly of gun control. The idea is thrown around with various interpretations, as nearly all would agree the consequences of gun violence is a looming phenomenon. Staggering statistics of nearly 100 gun-related deaths per day claws at the ethical values of this free and secure nation (Everytown). But, if taken too far gun control could quickly eliminate long-standing rights of the American people. Specifically, what are the ethical implications of a government-spread uniform ideology of gun control across a diverse American people? Here exists a controversial crossway of ethical ideologies, not in the right and wrongs of gun violence, but rather the prioritization of upholding law versus bending it for a greater good. There is no question whether the action and allowance of gun violence is ethical. However, the ethics of a uniform gun control become controversial upon contact with preexisting rights, specifically of the Second Amendment,

creating argument between utilitarianists and deontologists, reformers and law-followers..

Heller vs District of Columbia

For proponents of stricter gun control laws, the Second Amendment poses as a difficult obstacle. Nearly always summarized as the “right to bear arms” the interpretation of the Founders’ intentions of this right had not been debated until the recent Supreme Court case of *Heller vs District of Columbia*. The official reporting of the case by the Supreme Court interpreted the right to bear arms as a right not limited simply to military purposes. They conclude that the 18th century meaning of this amendment was “unambiguously used to refer to the carrying of weapons outside of an organized militia” (United). In regards to this ruling, a government-instated uniform gun control would likely contradict the interpretation of the Second Amendment. Despite this ruling, however, many question the fundamental ethics of this right and its interpretation amidst gun violence.

Utilitarian Ethics

These skeptics of Scalia’s ruling base their ethics through a utilitarian lens, believing that in order to create a safer nation and limit gun violence, the obstacle of the Second Amendment and its interpretation may need be revised. Lauren Hirsh, the Senior Legal Officer at Australia’s Attorney General Department, argues for the United States’ adaptation of gun laws similar to those of her nation of Australia. Hirsh recognizes the obstacle of the Second Amendment, nonexistent in Australia. She explains the implications of *Heller* instating an individualist view of the amendment – meaning that the “right to bear arms” refers to rights of an individual for self-defense rather than militia-related – creating more difficulties in gun control. However, as *Heller* also determined that the Second Amendment is not absolute, Hirsh argues that in the fight for gun control, governmental

gun control laws reflecting those of Australia may infringe on this right, but “it is nevertheless constitutional because the infringement is justified.”

Hirsh is not alone in her criticism of the Heller interpretation of the Second Amendment. Executive Director of the Coalition to Stop Gun Violence, Joshua Horwitz combats the Heller ruling, claiming the Founders’ purpose was not to “safeguard an individual right of insurrection” and that democracies are inherently safe from governmental genocide, thus the right to bear arms only increases violence and national insurrection. He believes the individual right to guns unethical and detrimental – rather than empowering – to a democratic nation.

Yet another activist challenges the individualist ruling of the Second Amendment. President of the Brady Center to Prevent Gun Violence, Dennis Henigan directly combats Justice Scalia’s ruling in the Heller case, claiming that Scalia viewed the right to bear arms outside of the historical context and misunderstood the meaning of the original text. However, in the case report, Scalia weighed in on the historical context of the linguistics of the amendment, contrary to Henigan’s criticism.

Although each activist paints their argument in a different way, Hirsh, Horwitz and Henigan all disagree with the individualist ruling of the Second Amendment and believe that, when it comes to the proposition of uniform gun control, the “right to bear arms” may be challenged and might possibly need be infringed upon, for the betterment of American safety. Although none argue to completely repeal the amendment, their utilitarian ethics prioritize gun reform above the instated Constitutional Bill of Rights and find infringement of the amendment a plausible option, or even requirement.

Deontological Ethics

Believers in the supremacy of the Second Amendment and its presiding individualist interpretation do not necessarily all agree with the right itself, but rather look through a lens of deontological ethics. More simply put, in regards to the establishment of governmental uniform gun control, they believe the Second

Amendment immovable and not to be infringed upon. Again, supporters of deontology are not inherently lesser activists in gun reform and the ethics of gun violence but look to establish laws within the bounds of the preexisting Constitution and its interpretation.

Renowned public health lawyer, as well as previous executive director of the American Society for Law, Medicine and Bioethics, Larry Gostin insists that the government must do something about the gun issue. However, he suggests that resolution can be attained without infringing upon the peoples’ right to bear arms, stating that the current reading makes gun reform “difficult, but not impossible.” He suggests various commerce-related control of guns that Congress has the power to instate, within the Constitution, but has not. To Gostin, there is no need to override preexisting law for the greater good as similar solutions may be implemented under the current law, contrary to the previous utilitarian ideologies.

Hirsh’s argument for the adaptation of laws without regard to the Second Amendment butts heads with the ethical priorities of Jonathan Weg, an upstart young lawyer of New York. Weg weighs in, rehashing the fact that Australia is without a “right to bear arms, as seen in America. Weg concludes that America cannot look to Australia for gun reform protocol as theirs are nowhere near “within the parameters set by the Constitution and Supreme Court”. America should therefore look to other countries – such as Canada – for inspiration but ultimately create regulations necessary for better safety yet still within the scope of the law. Proponents of this deontological view of ethics believe the Constitution to be the ethical priority, creating laws for gun reform within those bounds. In their eyes, if indeed uniform gun control is in contradiction with the Second Amendment, an alternate solution must be sought out. Although not always the easiest, they believe the necessity of the path to gun violence reform to be within the parameters of the current law.

Conclusion

Across the board, the majority calls for governmental action to reduce the horrific mass shootings of America. The unethical reality of continued violence is accepted. The debate instead arises in the conflict between reform within and outside the bounds of the law and its interpretation. It is this crossroads at which Americans decide their ethical priorities in the path for reform – the efficient revision of the law for a greater good or the methodical, lawful practice – both with the ending goal of increased safety and ethical justice.

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A Historical Analysis and Critique of the Spectrum of Uses for Government Surveillance

Jacob Williams

The issue of government surveillance only is increasing in priority as governments, and the technology they use, evolve through the ages. Such surveillance has been used by governments for the protection of their population, or the control of its populus. An empire such as the Soviet Union monitored its citizens for the protection of the government, while the United States has claimed its surveillance is for the protection of the citizens of the country. This historical analysis proposes a significant question: Is the surveillance over US citizens by their government beneficial to the country as a whole? This question can be answered in many ways, with focuses highlighting the economic, ethical, and social (and more) issues of this topic. However, an analysis of the historical aspects to government surveillance provides an essential insight into the crucial issues that affect the people of the world to this day.

Overall, as surveillance and its influence have increased, reactions have ranged from resounding praise to bitter protest. Kevin D. Haggerty and Richard V. Ericson, professors of sociology working at the Universities of Alberta and British Columbia, discuss how the minds of people have changed since "the proliferation of social visibility" (Haggerty, 2007). Haggerty and Ericson contrast examples of traditional power in Renaissance Europe and the present day in order to advance the idea that the increase of surveillance has caused a greater unease in citizens aware of such monitoring, causing reactions to surveillance to become more polarized. The two argue that centuries ago, in Renaissance Europe, since surveillance was conducted on a much smaller scale, less citizens being monitored neither knew of nor opposed such surveillance. Currently however, mass use of government surveillance has created great

awareness and controversy surrounding such a topic.

Prior to analyzing the different methods of government surveillance as well as the purposes such surveillance serves, an examination of the emergence of government surveillance and the effects of its spread is essential. Specifically considering the United States, dramatic events, specifically during the first and second world wars, led to an exponential increase in mass surveillance across the country. Krystal Conniry, a graduate of Portland State University with a Masters of Science in Conflict Resolution, draws a correlation between an increase in radio use for military activity and the development of specific departments in the US government, such as the Cipher Bureau in the Military Intelligence Department. With both radio developments and an increase of radio activity leading into WWII, specifically after the attack on Pearl Harbor, surveillance was trained on every Japanese in the United States; all of this caused the fear of the United States to lead to the internment of thousands of Japanese citizens. Conniry, to put it succinctly, debates how the efforts to intercept and decode enemy messages and tactics initiated the mass surveillance of US citizens (2000). These events and the distrust they carried with them, Conniry argues, are key historical catalysts that led to increased government surveillance through events such as the Patriot Act, which greatly increased the amount of surveillance the US government could use on its citizens. It should be noted however, that Conniry acknowledges how these mentioned events led to heightened government surveillance around the world.

While the benefits of increased safety and protection that government surveillance facilitates for the citizens of a country resolutely

defend the use of such a system, it is a necessity for governments to carefully weigh benefits against possible downfalls that could have drastic consequences on the country as a whole. Many argue that government surveillance has been used throughout history for the same purposes that America used it to imprison Japanese citizens: to further the government's control. An example of this use for surveillance is demonstrated in the Soviet Union. Amir Weiner, a Stanford professor with a PhD in Soviet history, argues *Getting to Know You: Soviet Surveillance and its uses*, an article in the journal *Kritika*, that Soviet surveillance, to the extent of its usage, was used to further the degree of control that gave the Soviet dictatorship power (2012). Soviet surveillance, Wiener argues, was used to further the complete control of a totalitarian regime hell-bent on staying in power. Similarly, Lora A. Esau identifies how these Soviet roots still exist in modern-day Russia, arguing in her essay for the *ILSA Journal of International and Comparative Law*, that while Russia does have parameters enacted for legal surveillance, these parameters are not sufficiently rigid, allowing for a circumnavigation around these laws, bordering on illegal activity by organizations like Russia's Federal Security Service (2017). In sum then, both authors identify significant flaws in the surveillance used by the Soviet Union and Russia, respectively, that facilitates tyrannical practices in the government.

Arguments against government surveillance based on events from the specific eras, such as Soviet Russia, are warranted, yet need to be weighed against other examples. The United States, as argued by Tamara Dinev, writing for *The Journal of Strategic Information Systems* with a PhD in theoretical physics, has heightened its surveillance not only for increased knowledge on terrorist activity, but to protect its citizens more (2008). This surveillance, Dinev argues, has benefitted US citizens greatly, which is why they are willing to be monitored if it keeps them safe from potential terrorism. Furthermore, David Lyon, a professor of sociology writing for *International Sociology*, argues that as a result of heightened surveillance since 9/11, terrorist attacks, as well as the overall effects of terrorism on the United States, have decreased significantly

(2004). Not only do these journalists agree that the United States' increase of surveillance has had positive effects for the country, but the director of the CIA, Keith B. Alexander, concurs as well. While testifying before the Senate, Alexander argues that US government agencies, such as the CIA and NSA, have managed to tighten their grip around terrorism while respecting the rights of citizens by increasing the power and extent of government surveillance in a lawful, constitutional manner (2006). In sum, Dinev, Lyon, and Alexander all agree and argue that the constitutional use and increase in government surveillance has had overall positive effects on the US as a whole.

Overall, government surveillance is a highly powerful asset to governments across the globe. It can either be used to further a totalitarian state or it can be used to serve the people by protecting their day-to-day lives. As demonstrated by the history of mass surveillance in the US, examples of surveillance in the Soviet Union, Russia, and America, as well as the effects of surveillance on citizens before and after surveillance was introduced, the US's use of government surveillance has both favorable examples and negative displays of how surveillance can be used. However, overall, as demonstrated by the arguments made by Dinev, Alexander, and others, government surveillance in the US is beneficial to the country overall due to the amount of terror it has prevented as well as the limit to which US government has used this power, avoiding moving towards a totalitarian state. As long as the government of the United States of America continues to use mass surveillance to serve the citizens of the US and not further their own agendas, then government surveillance in the US will continue to benefit the nation as a whole.

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