Title
Preventing Food Contamination: A Need for Innovation in Food Production

Journal
UC Merced Undergraduate Research Journal, 5(1)

Author
Reynolds, Mark

Publication Date
2013

DOI
10.5070/M451020768

Undergraduate

Preventing Food Contamination

A Need for Innovation in Food Production

By Mark Reynold

Introduction

Although it is essentially impossible to prevent any entity that was previously exposed to the environment from microorganismal colonization, most complex organisms have long been accustomed to both their existence and abundance. In fact, humans and nearly all the components of the immune system evolved in response to their coexistence to microorganisms (Janeway *et. al*, 2001). The majority of the effects they have on human health are relatively mild due to the repeated exposure we have been succumbed to over the course of humanity. However, there are particular subsets of contaminants that are the culprit of an unwanted and unfair phenomenon: foodborne illness. These pathogens that are responsible pose as a serious threat to the human population and have so for many years.

The “stomach flu”, or more formally known as gastroenteritis, from various strains including members of the *Salmonella* genus, for example, kills 155,00 people worldwide while simultaneously infecting over 94 million people in any given year (Majowicz et. al, 2010). To the misfortune of the consumer population, a little over 85% of those are foodborne cases (Majowicz et. al, 2010) and can often be traced back to the mass production of soil-derived produce and other mishandled consumables.

Salmonella, E. coli, and other pathogenic bacteria aren't even the only major players in this worldwide and national scare. From the years of 1973 to 2006, 60 percent of U.S. foodborne outbreaks associated with eating leafy greens were caused by noroviruses alone along with other viral subsets, such as rotaviruses. Contrast that with *Salmonella* and E. *coli* which only accounted for 10 percent of those outbreaks (Food Safety News, 2013) and the situation becomes even more of a scary mystery.

In response to the concern of eating food and becoming ill, numerous precautionary measures have been developed in the last two centuries to ensure thorough cooking of perishables. In addition to this, there has been much advancement in food storage to further prevent microbial contamination. Due largely because of this, recent studies suggest that foodborne infections have declined in the United States since 2000. As favorable as this outcome may sound, it comes with a cost; both food-related hospitalizations and deaths are on the rise. It is strikingly apparent when looking at the statistics that further prevention measures need to be taken, for the pathogens are evolving to become more resistant and thus more of a threat to the population's health.

Throughout this assessment, I argue that the relatively innovative-absent field of food production (which will hereby include the subfields of packaging, storage, transportation, and safety) can transiently be modified to incorporate recent ground-breaking research findings through the following individual and/or dual implementation of two novel operations: Firstly, intra-ozone production in various packaged goods to act as a safe and effective disinfectant. Secondly, the exploitation of special conditions to create an unfavorable environment for microorganisms during the storage of various prone consumables. In theory, the combination of the two operations would be highly effective on common pathogen-susceptible items fated to be sold in the grocery store. Therefore, due to the simplicity of production and the commonality of being a prone to pathogen colonization, I will be further examining lettuce and other leafy greens.

(i) Background: Foodborne illness & lettuce production statistics

From a study published over a decade ago, Mead *et. al* estimated that foodborne illnesses cause approximately 14 million illnesses, 60,000 hospitalizations, and 1,800 deaths in the United States each year (Mead, 1999). In a more recent estimate, the Center of Disease Control and Prevention reported 7.8 million illnesses, 127,839 hospitalizations, and 3,037 deaths for the year of 2011 (CDC, 2013). The majority of these rates can be seen in underdeveloped countries as well as those who commonly eat food raw. Whatever the cause of the gastroenteritis, the possible outcome is similar. Symptoms range from abrupt 24 hours symptoms with a quick recovery to death attributed to uncontrollable diarrhea, a problem that affects 1.5 million children worldwide (WHO & Unicef, 2009). According to data from the Center for Science in the Public Interest, 12% of those illnesses mentioned above are linked to produce (Klonsky, 2006).

Many unfavorable outcomes become troublesome when looking at foodborne outbreak statistics and trends in recent decades. For instance, it has been unearthed that between 1995 and 2005, 19 individual outbreaks of *E. coli* foodborne illness were attributed to fresh-cut lettuce alone (Klonsky, 2006). Of particular interest was a November 2006 *E. coli* outbreak that originated in Central California that eventually spanned a total 26 states. During the monthlong outbreak, 3 individuals were killed and a hefty 200 were sickened (Klonksy, 2006).

The quick migration of the contamination in the example given above is unconventional in a sense because it was not necessarily attributed to human-to-human transmission, as are most “outbreaks”. One could argue the reason behind this is because food corporations have become so monopolized and often care only about high quantity output. Proof of this comes with knowledge that two companies, Dole and Fresh Express, control 88% of sales in the pre-washed packaged salad greens industry (Klonsky, 2006). This insinuates that if any contamination occurs within the facilities of these corporations, the general public and average consumer is likely to become sickened. The multitude of people at risk from a corporations' standpoint becomes numerical grasped when one observes that an estimated 80% of US households purchase leafy greens of some sort to serve alongside family meals (Klonsky, 2006).

(ii) Background: Present need for innovation

In addition to the glaring statistics, further reasoning behind why its important to make advancements and alterations to the current food production industry lies within the fact that most produce sold in supermarkets undergo approximately 7-10 minutes of chlorine washing during production. Many research studies have demonstrated that this semi-controversial method of treating the consumables is unsafe and ineffective (Vleugels *et. al,* 2005). For example, a study conducted in 1999 had proved that under specific chlorinated conditions only a log reduction of 1.75 in the microflora present on shredded iceberg lettuce was achieved (Vleugels *et. al,* 2005). With its mediocre effect as a microbiocide and also the health risks it poses to humans, the need for an alternative to chlorine washing is becoming highly sought after. A final proposed reason for a desperate need of additional food safety measures can be debated through the fact that consumers' eating habits are changing such that the most convenient, quick, and easy to prepare meal is the most favored. Companies succumbing to these wishes are also potentially increasing the ease of transmission through the introduction more and more ready-to-eat meals and snacks (Akishev *et. al,* 2004).

The more outbreaks and subsequent product recalls that occur, the more people become interested in finding solutions to prevent food contamination and ultimately silence the illness in the future. In 2013 alone, two major referendums to current methods were introduced. In an early July report, recent trends and issues in global food packaging were discussed. Many important matters were addressed, however, there are two particular interests that compliment my proposition: the need to "increase shelf life product" and "reduce bacteria growth" (PR Newswire, 2013). In addition to this, the FDA had released a "Lettuce Safety Initiative" with four major changes that needed to be included in current practices. All four bear great importance to my overall goal, however, two in particular can potentially be fulfilled with my soon to be elaborated on idea: that is to "stimulate advancements" in addressing all aspects of improving lettuce safety. And also, to find out what is causing contamination and hereafter re-develop strategies (FDA, 2013).

Therefore, with all that said, I propose safety guidelines for future developments and examinations in food production. One that would exponentially prevent food contamination in the industry. The most highly effective inclusion of the additional two-step operation (listed below) would be seen primarily in corporations that produce lettuce and other leafy greens as discussed below.

1). Use of a plasma generating refraction device immediately after packaging (i.e *post-harvesting)* in a process that generates ozone and disinfects a lettuce head while sustaining the consumers' safety in the process. By performing such a feat, the presence of any soil-borne microorganisms will likely be diminished.

2). Implementing knowledge stemming from microgravitational simulated experiments that confirmed increased proliferation and virulence of a pathogen due to lack of fluid dynamics under such conditions. These findings and experimental conditions can be reversed such that a high fluid shear environment will be *unfavorable* to the pathogen. Thus, the possibility of food contamination prior to being shipped to supermarkets would be drastically lowered. Such an act would benefit most during the *post-packaging* of the edible item and ideally during the storage of the goods prior to transportation.

(I) Why utilize plasma-generated ozone as a post-harvesting strategy?

If a gas is heated to an extremely high degree, it is possible to ionize the compositional compounds in question. I will be further specifically referring to plasma generation using the dielectric barrier discharge (DBD) method to use in lettuce production example. In this method, a gas is ionized (i.e. molecular oxygen) in between two electrodes and the result is concentrated ozone generation within a plastic encasing. DBD is used for the overall synthesis of radicals that combine to form various atmospheric gases such as carbon dioxide in addition to ozone.

Ozone is a very well known germicide as well as antimicrobial agent (Sharma & Hudson, 2008). It is important to note that ozone can be toxic to humans. However in this scenario, it naturally reverts back into oxygen quite quickly (Diver *et. al*, 2011), thus ensuring safety in the consumer. There are in fact numerous ways to create ozone, but for our purposes, the plasma generation method is by far the most efficient and safe in regards to preventing contamination of food.

(II) Why exploitation of fluid shears as a storage and/or pre-transportation strategy?

Fluid shear is the concept that a microorganism can "detect" the favorability of an environment over another based on differential fluid dynamical activity (See Figure 2 in "*Methods pertaining to fluid shear research"* section below for more). Such a phenomenon can save the microbe a vast amount of energy from doing so by proliferating only when is most convenient for them. For over a decade, researchers at Arizona State University have been investigating the threat that microorganisms may pose for astronauts during space flight. Using two culture systems that are analogous to those experienced in spaceflight, microgravity-like conditions were generated with very low fluid shear count (Nickerson et. al, 2010). Such an incredible accomplishment allowed researchers to study pathogenic microbes such as *P. aeruginosa*, *E. coli* and their subsequent gene expression in environments containing very low fluid dynamical activity (i.e. low fluid shear). Recall that even air, which may seem stationary to the naked eye, possesses fluid dynamical properties. While experiencing microgravity, such properties become virtually absent; thus creating an environment that microorganisms thrive in.

Under low fluid shear conditions, the consistent findings that were revealed during several similar research projects from those of other research teams was that external stress to at least one key sigma factor in each major strain was responsible for this unusual behavior: AlgU in *Pseudomonas aeruginosa* and RpoS in *E.coli* (Sussman, 1997). These sigma factors primarily dominate the induction and up regulation of nonconventional nucleic acid transcripts. When introduced to an external stress or an unfavorable environment, the sigma factor would act to redirect the polymerase to initiate transcription at uncommon promoter regions (Schurr *et. al,* 1996). The outcome of this is that unnecessary novel proteins are constructed that have the potential to perform unnecessary catalytic functions. This can be seen as 134 total genes related to toxic compounds were expressed in low shear microgravity vs. 9 total expressed in regular gravity (Nickerson *et. al*, 2010). In addition to this, unique histone-like proteins are also produced that are simply just not known to cause virulence here on Earth's surface, perhaps because the environmental conditions on Earth's surface rarely permit it (Klaus & Howard, 2013). Examples of other known sigma factors in microorganisms are RpoH and RpoN (which corresponds to heat shock and nitrogen deprivation, respectively).

Overall, by working with the knowledge gained from the research conducted on increased microbial virulence and proliferation in low shear microgravity environments, a contrary and very valid stance can be deduced; by similar logic, a pathogenic entity would show *decreased* proliferation and virulence in a high fluid shear and gravity accelerated environment.

(III) What are the prior research methods and conclusions reviewed prior to making such an assertion?

Methods pertaining to plasma generation research:

The prototype of interest is on that has been previously reported by Mieke Vleugles and the rest of his research group. In one experiment, their team used edible items (wrapped muffins and unwrapped bell peppers) contaminated with pathogens such as *Campylobacter* and *E.coli* The researchers have discovered the method is capable to act hundreds to thousands of times faster than chlorine in a disinfectant role (Vleugels *et. al,* 2005). The edible examples provided in the research transcript are regarding use of ozone plasma on microbial mats on the surface of bell peppers. Such use of the technique allowed for a staggering a 99.995% reduction in the bacterial colonies initially selected for along with minimal effect on the quality and nutrient content. Below, in Figure 1, a graph depicting the drastic drop in proliferation rates of microbial communities when exposed to the plasma-generated ozone is presented.

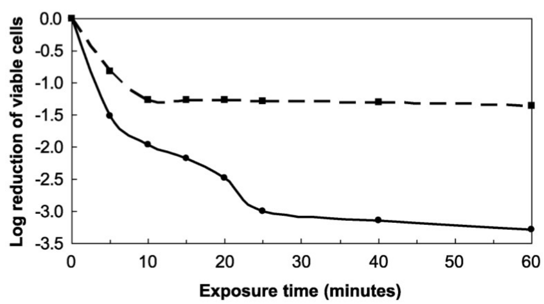

Figure 1: Viable P.agglomerans, a known gastrointestinal pathogen, and differential effects under conditions involving no treatment of plasma generation (dashed curve) as opposed to conditions involving treatment of plasma generation (solid curve).

*Image provided courtesy of Vleugels *et. al.*

Methods pertaining to fluid shear research:

Nickerson *et. al.*'s research group have been studying the fluid shear concept with a variety of different pathogenic strains of bacteria while experiencing low shear microgravitational activity for almost over a decade now. They have studied *P. aeruginosa* and also *S. typhimurium* as well as the role both's own distinct sigma factor plays in the increased virulence (AlgU and RpoS, respectively).

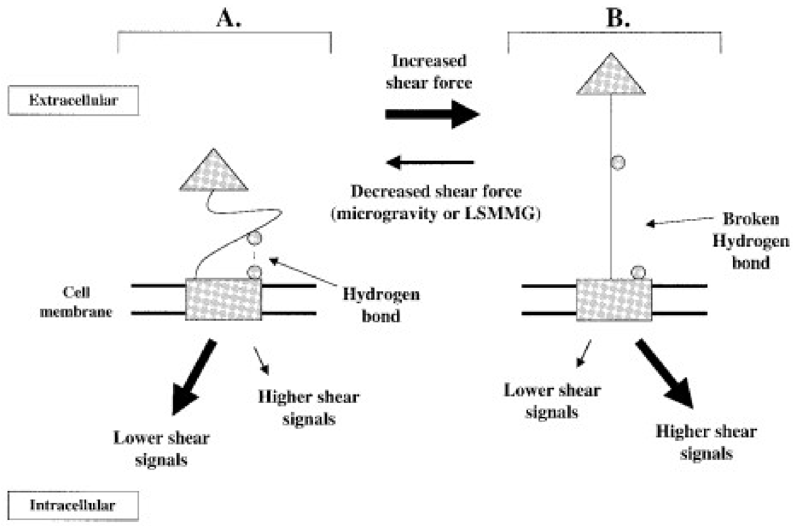

Figure 2: Schematic describing a hypothesized molecular interaction that occurs when exposed to both low fluid shear (left) and high fluid shear (right) conditions. Notice the in-tact hydrogen bond in low fluid shear depiction as opposed to the high fluid shear depiction. Perhaps this is means of signaling to the entire bacterium to safely express all the virulent proteins and also replication-promoting sigma factors.

*Image provided courtesy of Nickerson *et. al.*

They tested the virulence (concentration of a toxin, *alginate*) at regular gravity, no gravity, and low shear microgravity. Interestingly, the bacteria showed nearly the same transcriptional response in both regular gravity and zero gravity. It was only in the low shear microgravity where the unique gene expressions and subsequent increased virulence took place, proving that even the slightest stress can induce profound metabolic changes (See Figure 3 below). To further drive home the point that the conditions tested are truly relevant to the pathogens' proliferation, in their most recent publication, Nickerson *et. al.* claims that cultures exposed to regular gravity conditions experienced fluid shear levels up to a whopping 44 times the fluid shear, thus making for a much more unpleasant reproductive environment.

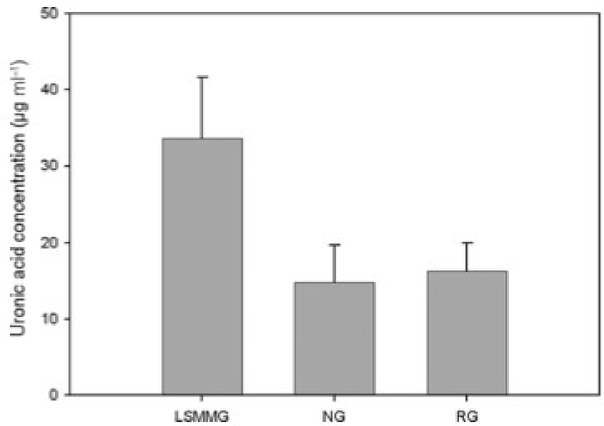

Figure 3: Portraying the differences in virulence capabilities for the *P.aeruginosa* strain under the low fluid shear, modeling microgravitational (LSMMG) conditions (left bar), conditions with zero gravitational (NG) acceleration (middle bar), and regular gravitational (RG) acceleration (right bar).
*Images provided courtesy of Nickerson *et. al. (2010)*

(IV) Future commercial use and limitations: lettuce and other leafy greens

With sufficient research, the combination of both operations described above into the conventional method of food production could be a potentially groundbreaking innovation that can be applied to packaged consumable goods of all types, both solids and maybe even liquids in the near future. However, it would hold up to extreme pressure because expectations would be high granted they ever become available on a large-scale level.

Looking ahead to this becoming a realistic possibility, it is predicted that it would not be difficult to incorporate these findings into the current method of lettuce production and commercial sale. The reasons it would be most applicable to begin the twofold operation with salad greens is because it is the leading agent that yields the most gastroenteritis cases with a staggering 23% (Painter *et. al,* 2013). Based on information provided from the University of California's Agricultural and Natural Resources department, it is shown that soil-derived lettuce is conventionally packaged naked, film-wrapped, and vacuum cooled prior to storage. To be further specific, it is stored just above freezing temperatures with a humidity of ~98% (Smith *et. al*, 2006). I believe that immediately after the wrapping (packaging) of the lettuce, measures of using the DBD refractable-plasma generator prototype can be taken to remove of any excess soil-borne microbes that persist, thus also ensuring no potentially harmful chemicals such as chlorine are involved. Figure 4 below depicts the series of events that would take place during such a cascade.

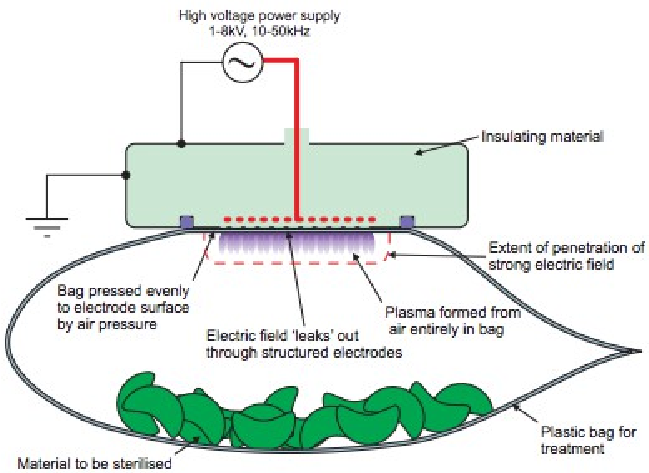

Figure 4: An illustration showing the envisioned implementation of the plasma generating device immediately after packaging of a common susceptible item, such as leafy greens for example.
*Image provided courtesy of Diver *et. al*

Hereafter, the process can resume with the storing of the lettuce. It would be during this phase that would seem most practical to begin utilizing the findings from the fluid shear experiments. Such a feat can be performed by introducing powerful, modified bladeless fans (to avoid microbial contamination even further) as a means to decrease the probability of microbial colonization on the food surfaces. In addition to this, once the theory becomes more of a possibility, modifying the gravitational vector through the alteration of Aether (or time-like vector field), which is shown mathematically to be possible in Zlosnik *et. al*'s 2008 publication (Zlosnik *et. Al*, 2008) can ultimately aid in this desired goal.

Overall, the entire project is expected to be quite costly. For example, the idea of using the fans along with hypothetical increased gravitational conditions during the storage and possibly even transportation of the salad greens would require approximately 50 times the amount necessary to achieve a similar microorganismal deactivation that results from the presence of flowing liquid. The underlying reason behind this is because that water, as our example of a flowing liquid, possesses 50 times the viscosity levels compared to air, therefore much more would be needed (Lancaster *et. al,* 2013).

Limitations will definitely become apparent the more such an implementation becomes a realistic possibility. Therefore, the need for research becomes heavily apparent as the potential project moves forward in the future. The main concern that the plasma generation device poses is whether or not it affects the nutritional content of the food. Along with this, it is possible the use of the device may yield an undesirable physical or chemical change such as color or taste alteration in some foods. Furthermore, it was also discovered by Glasgow University researchers that the use of the plasma, using particularly the nonthermal DBD method, might lead to oxidative damage, especially in foods with high-fat content.

Examples of future areas of research lies in how the plasma generation process could work on liquids. Another manner in which further research in this field could be necessary is with other foods that are mass-produced commercially and are also at risk of microbial contamination such as nuts, eggs, imported spices, and cheese. Lastly, with a more extreme example, such precautionary measures can also be utilized in the protection of the general public against possible food terrorism, a concern that needs to be addressed as more knowledge of these pathogens becomes readily available.

(V) Conclusion/Discussion:

The salad and leafy green industry can truly benefit from firstly, the use of plasma-generated ozone in sealed packages after harvesting. And subsequently, the exploitation of low fluid shear environments during storage conditions prior to being shipped to supermarkets. Major companies and the general public would do so primarily because of the additional precautionary measures it allows for. Surely it may take some time and money to initiate the widespread and consistent use of the techniques, but the simple fact that, in theory, it would minimize food illnesses all together would allow the populations to remain healthy and ensure that the corporations would not harm their reputation due to a contamination mishap.

A common expression that is widely used in the response to a need for change is that “the only thing that is constant is change.” This is relatable to introducing novel innovations in the food production industry because although sometimes a change is not wanted to be explored, it quickly becomes a necessity that more than often benefits those who originally did not want the change. The above expression is also used in conjunction with another: “do not fix something that is not broke.” Albeit that is true in many cases (and even possibly in this example), modification, not necessarily change, is the driving force of evolution. Thus, such a fact needs to be continually remembered when advancing towards the future in regards to innovation in any field.

How this deals with my proposition of a change in the food production operation is as follows: Any revolutionary paradigm shift that has been documented in history has never been considered easy in regards to implementation. It requires an immense dedication to improve an outdated way of thinking. It becomes especially important, however, when a population’s health and wellbeing is at stake. Food enthusiast and extraordinaire, Michael Pollan, describes the dilemma best in his book *The Omnivore's Dilemma: A Natural History of Four Meals*; “But that's the challenge -- to change the system more than it changes you.” (Pollan, 2006).

References

Akishev, Yuri, Karol Hensel, and Zdenko Machala. *Plasma for Bio-Decontamination, Medicine and Food Security*. New York City: Springer, 2012. Print.

Borcia, G. & Chiper, A. "Argon Versus Helium Dielectric Barrier Discharge for Surface Modification of Polypropyleneand Poly(methyl methacrylate) Films." *Plasma Chem Plasma Process.* 33. (2013): 553-568

CDC. "Estimates of Foodborne Illness in United States (2011)." *Centers for Disease Control and Prevention*.

Diver, D.A, H.E Potts, et al. "Plasma decontamination of sealed packages." *International Conference on Phenomena in Ionized Gases*. 30. (2011):

Janeway , CA Jr. et al. Immunobiology: The Immune System in Health and Disease. 5th ed. Routledge, 2001. eBook.

Klaus , David M., and Heather N. Howard. "Antibiotic efficacy and microbial virulence during space flight."*Trends in Biotechnology*. 24.3 (2006): 131-136.

Klonksy, Karen. "E. coli in Spinach, Foodborne Illnesses, and Expectations about Food Safety." *Giannini Foundation of Agricultural Economics*. 10.2 (2006): 1-4

Lancaster, Jill *et. al. Aquatic Entomology.* Oxford, U.K.: Oxford University Press, 2013. Print.

Majowicz, S.E. et. al. "The global burden of nontyphoidal Salmonella gastroenteritis." *Clinical Infectious Disease.* 50.6 (2010): 882-889.

Mead, Paul, Laurence Slutsker, et al. "Food-Related Illness and Death in the United States." *Emerging Infectious Disease*. 5.5 (1999): 607-625.

Nickerson, Cheryl, Aurelie Crabbe, et al. "Response of Pseudomonas aeruginosa PAO1 to low shear modelled microgravity involves AlgU regulation."*Environmental Microbiology*. 12.6 (2010): 1545–1564.

Nickerson, Cheryl, C. Mark Ott, et al. "Microbial Responses to Microgravity and Other Low-Shear Environments." *Microbiology and Molecular Biology Reviews*. 68.2 (2004): 345–361.

Painter, John A. *et. al*. "Attribution of Foodborne Illnesses, Hospitalizations, and Deaths to Food Commodities by using Outbreak Data, United States, 1998–2008." *Emerging Infectious Disease.* 19.3 (2013)

Sharma, M., and J.B. Hudson. "Ozone gas is an effective and practical antibacterial agent." *American Journal of Infection Control*. 36.8 (2008): 559-563.

Smith, Richard, Michael Cahn , et al. Leaf Lettuce Production in California. UC Davis Agriculture and Natural Resources: *Vegetable Production Series.* Publication 7216 (2011): 1-6.

Sussman, Max. *Escherichia coli: mechanisms of virulence*. 1. London: Cambridge University Press, 1997. Print.

Vleugels, Mieke, Gilbert Shama, et al. "Atmospheric Plasma Inactivation of Biofilm-Forming Bacteria for Food Safety Control." *IEEE TRANSACTIONS ON PLASMA SCIENCE*. 33.2 (2005): 824-828.

Zlosnik, T.G. "Modifying gravity with the Aether: an alternative to Dark Matter." *University of Oxford Astrophysics*. (2008): n. page. Web. 7 Nov. 2013.

Addy, Rod. "Plasma power fights germs in food." *Food Production Daily.* (2013).

Evelith, Rose. "In Space, Infectious Diseases Reveal Their True Nature." *Smithsonian*. (2013).

Nicholas, Sadie. "Attack of the poisoned lettuces! The dangers lurking in pre-packaged salad leaves." *Daily Mail*. (2012).

PR Newswire. "Innovations in Food Packaging, 2013 - A review of recent trends, drivers and issues in global food packaging."

U.S. Food and Drug Administration. “Lettuce Safety Initiative” (2013).

WHO. "Diarrhoea : Why children are still dying and what can be done." . World Health Organization/UNICEF. Web. 19 May 2013.

Mark Reynolds is a fourth year Biological Sciences student emphasizing in Microbiology & Immunology with a minor in Environmental Science & Sustainability. Raised in Tehachapi, CA, he had always envisioned himself attending medical school and ultimately becoming a physician. Plans changed upon enrolling at the University of California Merced where a firm interest now lies in understanding the basics of various life science sub disciplines, whether by performing research to discover or teaching and/or writing to inform those who want to learn. He plans to pursue graduate studies to obtain either a PhD in Environmental Microbiology or a Masters degree in Scientific Writing. Non-academic hobbies of Mark's include basketball, music, traveling, reading, and spending time with those close to him.