



Quadrivium: A Journal of Multidisciplinary Scholarship

Volume 5
Issue 1 Issue 5, Fall 2013


Article 7

10-28-2013

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Schmitt Lavin, Emily F. (2013) "Exploring Life and Death at the Cellular Level: An Examination of How Our Cells Can Live Without Us," *Quadrivium: A Journal of Multidisciplinary Scholarship*: Vol. 5: Iss. 1, Article 7.
Available at: <http://nsuworks.nova.edu/quadrivium/vol5/iss1/7>

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Exploring life and death at the cellular level: An examination of how our cells can live without us

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Abstract

It is possible for cells from the human body to live on long after the person they came from has died. One of the most famous of these cell lines was harvested from a tumor found in the body of Henrietta Lacks. Although Henrietta died in 1951 at the age of 31 from cervical cancer, her cells were able to grow in culture, outside of her body. The study of the more than 50 million tons of her cells, which would conservatively equal the amount of cells in one billion people, has resulted in nearly 11,000 patents, including the polio vaccine. Known simply as HeLa cells, they have transformed the face of medical research, prolonged many lives, and continue to live (62 years after Henrietta's death) in laboratories around the world. These cells have become a focus of public attention since Rebecca Skloot's book; *"The Immortal Life of Henrietta Lacks"* was published in 2010. Examples of how and why cells have lived on long after the individual they came from has died are presented. Questions such as, "Who owns our biological samples once they leave our bodies?" and, "Is cell culture a form of immortality?" will be discussed.

Introduction

During the academic year (2012-2013) students and faculty in the Farquhar College of Arts and Sciences focused on the annual theme of "Life and Death." To this end, we have been focusing all aspects of academic growth around this theme. It has guided our coursework, study groups, invited lecture series, faculty lecture series, and even our commencement speaker's address (Farquhar College of Arts and Sciences, 2013). In this paper the idea of life and death will be examined from a cellular and molecular level, and the idea of what happens to our biological pieces (cells, proteins, tissues, etc.) once they are separated from our bodies will be explored. This idea has become increasingly more intriguing as humanity has discovered ways of keeping these biological parts viable and useful while outside of the original body from which they were derived (Lodish et al., 2012). When thinking about how to approach the annual theme from a biological point of view, I began asking students and community members, "Do you really think that a person's cells can live outside the body?" Most had not given the idea much thought and were astonished to learn that the answer was a resounding, "Yes!" Although it is not that likely, even your own cells could be living in a culture dish somewhere right now without your knowledge! (Eiseman and Haga, 1999). This paper explores the history of cell culture, including cell strains and lines, their purpose in basic research and medical advances, and some of the legal and ethical underpinnings surrounding their development and use.

Historical Overview of Cell Culture

Starting in 1907, Ross Harrison, a scientist with dual degrees (M.D. and Ph.D.) and a faculty member at Johns Hopkins and then at Yale, grew nerve fibers from frogs outside the frog's body in Petri dishes (Corning Life Sciences, 2013). A few years later, Alexis Carrel (an accomplished surgeon and cellular researcher who worked at the Rockefeller Institute of Medical Research for 33 years) became very successful in tissue culture. He was able to maintain tissues outside the body in dog, cat, rat, guinea pig, and human tumor models for several months (PBS.org, 2013). In the 1930s, Charles Linbergh, of aviation fame, engineered devices to make cell culture easier. Although human cell lines became possible in the 1950s, other types of cells had been cultured previously for at least an additional 50 years. The concept of cells living outside of their host has been possible for at least 100 years. By the 1950s large scale bioreactors were in place to allow for the widespread production of cells for various research applications, including vaccine development. These bioreactors are involved in an annual multi-billion dollar industry in the U.S., with additional revenues throughout the world (Corning Life Sciences, 2013). Cell culture is an extremely valuable tool for the study of various aspects of biological science, including cell biology, virology, and cancer. It is also a major production tool for the development of cell-based vaccines, monoclonal antibodies, and cell-based drugs (Rivard, 2013).

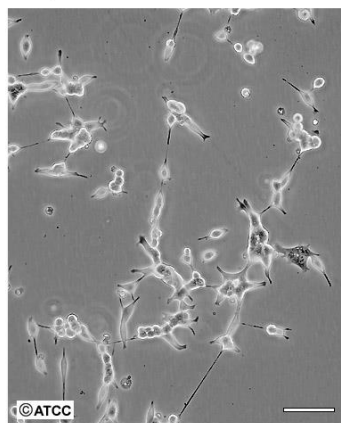
Cell Strains vs. Cell Lines

There are two main categories of cell culture (growing cells outside the original organism): primary and transformed. Primary cell culture is also referred to as the development of cell strains. The cultivation of these cells are begun from normal animal tissues (often skin, kidney, liver, or others). The cells in these tissues are specifically treated to break cell to cell and cell to matrix adhesions. Then the cells are grown on nutrient-rich media in dishes. These cells will typically divide a finite number of times (about 50) and then stop growing. However, this process can still yield a large mass of cells in culture. For example, if one starts with 10 billion cells, 50 doublings can produce 10^{20} cells, a number which is roughly equivalent to the weight of 1,000 people. The cell strain can be frozen before it goes into senescence, thus making it viable for a longer time (Lodish et al., 2012). On the other hand, transformed cell cultures have been found to be immortal. These cells can divide continually in culture for seemingly endless generations. This type of cell culture is typically derived from cancerous tissue and although they do undergo a period of senescence in culture, they emerge as a culture of cells with an indefinite life span (Lodish et al., 2012). There are entire catalogs available specifically dedicated to supplying researchers with the products involved in cell culture (American Type Culture Collection, 2013). These cells derive from a variety of sources and most of them are from human tumors. There are also some examples of normal fetal-derived human cells available for sale (Table 1; Figure 1). The ability of scientists to work with cells in culture has transformed our understanding of life at its most basic unit (Hayflick, 2012; Masters, 2002). For example, the number of scientific papers that have been published using the top 10 major cell lines alone has resulted in over 140,000 publications, with the vast majority (60,000) coming from the HeLa cell line (Biba, 2010). There are currently more than 4,000 cell lines available for sale from a variety of companies and organizations such as the American Type Culture Collection, the Coriell Institute for Medical Research (www.coriell.org), the European Collection of Cell Cultures (ECCC); (<http://www.hpacultures.org.uk/>), the German Collection of Microorganisms and Cell Cultures (DSMZ); (<http://www.dsmz.de/>), and the Bioresource and Collection Center (FIRDI-Taiwan); (<http://www.firdi.org.tw/EngWeb/2003/bcrc.htm>).

Table 1: Some of the most popularly available and used cell lines along with their biological source, cell type and reference price for a single vial to be shipped overnight to the researcher on dry ice. These cell lines were found in the American Type Culture Collection online catalog. Retrieved March, 2013 from www.atcc.org .

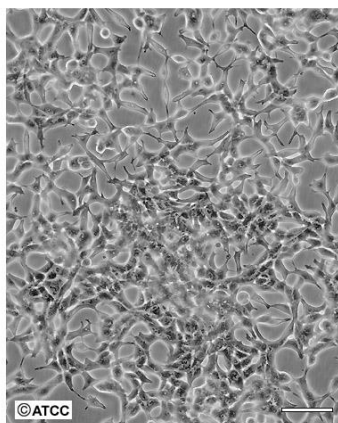
Cell Line	Biological Source	Cell Type	Reference price from www.atcc.org
MCF7	69 year; human Caucasian female	invasive breast carcinoma	\$431
JURKAT	14 year old boy	T cell leukemia; peripheral blood	\$431
HEK-293	Human fetus	Epithelial	\$431
HT-29	44 year; human Caucasian female	Epithelial; colon adenocarcinoma	\$431
LNCaP	50 year; human Caucasian male	Prostrate; carcinoma	\$431
HeLa	31 year; human Black female	Cervix; adenocarcinoma	\$431
WI-38	3 month; surgically aborted female Caucasian fetus	Normal lung fibroblast	\$431
MO	50 year, Caucasian male	T lymphocyte; hairy cell leukemia	\$551

ATCC Number: **CRL-1740**
Designation: **LNCaP clone FGC**



Low Density

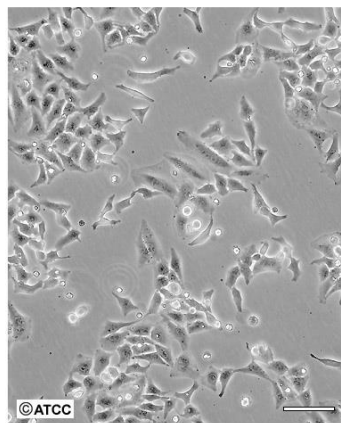
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High Density

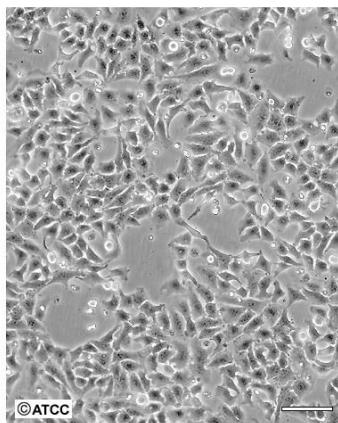
Scale Bar = 100µm

ATCC Number: **CCL-2**
Designation: **HeLa**



Low Density

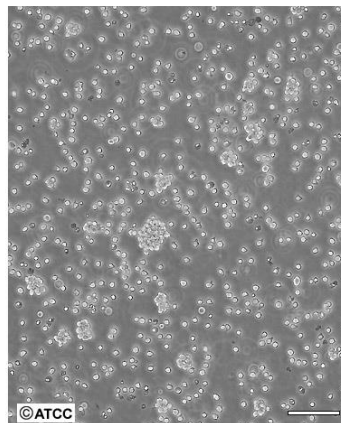
Scale Bar = 100µm



High Density

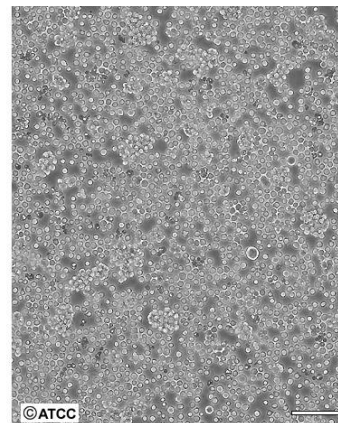
Scale Bar = 100µm

ATCC Number: **TIB-152**
Designation: **Jurkat (Clone E6-1)**



Low Density

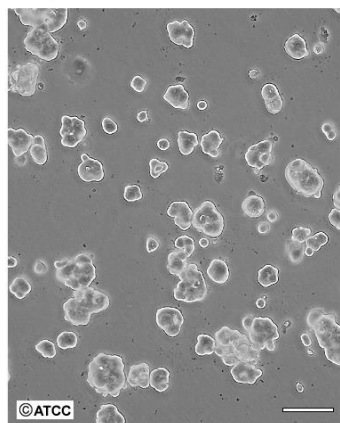
Scale Bar = 100µm



High Density

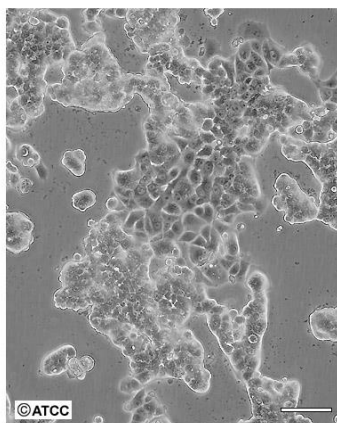
Scale Bar = 100µm

ATCC Number: **HTB-22**
Designation: **MCF-7**



Low Density

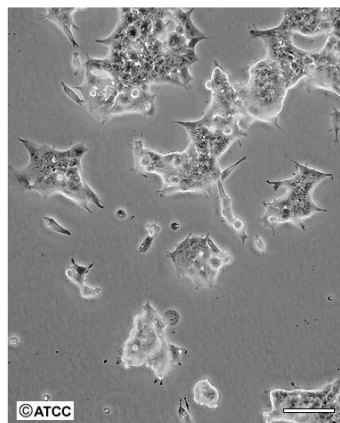
Scale Bar = 100µm



High Density

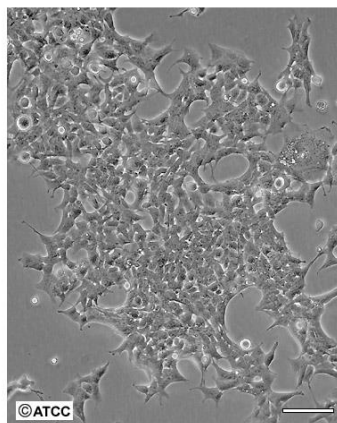
Scale Bar = 100µm

ATCC Number: **CRL-1573**
Designation: **293**



Low Density

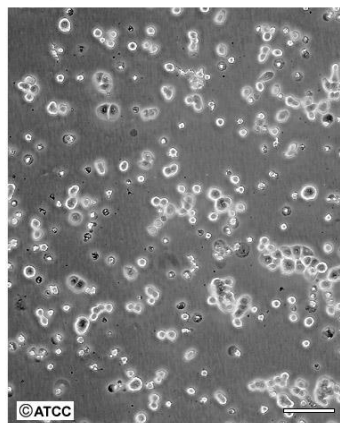
Scale Bar = 100µm



High Density

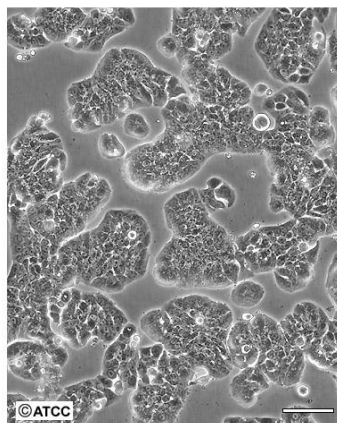
Scale Bar = 100µm

ATCC Number: **HTB-38**
Designation: **HT-29**



Low Density

Scale Bar = 100µm



High Density

Scale Bar = 100µm

Figure 1: Low density and high density microscopic views of a few cell lines available from the American Type Culture Collection (www.atcc.org).

The HeLa Cell Line

Arguably, the most well-known of all the human cell lines is currently the HeLa line. In large part this increased awareness is due to the recently published book, “*The Immortal Life of Henrietta Lacks*” (Skloot, 2010). These cells were taken while the patient, Henrietta Lacks, a 31 year old woman of low socioeconomic status and African American ancestry was being treated for a malignant tumor (carcinoma) of the uterine cervix at Johns Hopkins Hospital. These cells were found to be quite different from the normal cells from which they had originated (Grady, 2010). In fact, they were very fast replicating cells and did not have the growth requirements of normal cells. Ironically, these cells killed Henrietta very quickly in the prime of her life, leaving her husband and five young children without their wife and mother, while the cells have continued to live without her ever since (62 years and counting). Although these cells were devastating to their host body, they have provided scientists with the cellular tool to develop 11,000 patents, more than 60,000 scientific publications, at least two Nobel prizes, and the polio vaccine. They have also assisted in developing in vitro fertilization procedures and many other medical and scientific applications. The HeLa cells have even been in space (Mullin, 2011). More than 50 million tons of these cells exist in freezers around the world and have been sold for many billions of dollars. The number of HeLa cells that exist in research laboratories around the world is enough to make up the mass of approximately 1 billion people (Zielinski, 2010). Yet, when they were first examined by Margaret Gey and Minnie (a lab technician) in the laboratory of Dr. George Gey at Johns Hopkins in 1951, the scientists were simply excited that these cells were growing and that they would not stop growing, especially since up until that point it had been exceedingly difficult to keep human cells alive in culture. Dr. George Gey sent these cells to any researcher (free of charge) who wanted to conduct research using these cells (Masters, 2002). Although these cells came from Henrietta Lacks, they are very different from her regular cells. The HeLa cell line is infected with the most virulent strain of the human papillomavirus (HPV) known to exist, and these cells ultimately killed Henrietta. Still, Henrietta Lacks’ descendents feel that when they see the HeLa cells, they are looking at their ancestor. This view is particularly poignant for Henrietta’s youngest daughter, Deborah who recounts that the first time she was able to see the HeLa cells in a lab, she felt like she was seeing a piece of her mother for the first time. Her mother died when she was only 2 years old, and just less than a year after the birth of her last child, Joseph (Skloot, 2010). Through the public awareness campaign assisted by Rebecca Skloot and many researchers that use the HeLa cells, the descendents of Henrietta Lacks have been able to see their great-grandmother’s cells and be inspired by the good that has become of them. [The great-grandchildren of Henrietta Lacks visit Johns Hopkins to see HeLa cells](#) (Video embedded link; Franzos, 2011).

Although it has been known for a long time that HeLa cells were far from normal, the genome of HeLa cells was recently sequenced, shedding light on just how different these cells really are from normal ones (Landry et al., 2013). HeLa cells do not have the typical number of 23 human chromosome pairs, and there are many insertions of the human papillomavirus (HPV) that have shown up within the human chromosomes (EMBL, 2013; Scudellari, 2013; Landry et al., 2013). These changes have caused many researchers to question whether or not HeLa cells are really even human, and as such whether they are the best model (or even relevant anymore) for human research (Masters, 2002). As a result of the HeLa genome being sequenced, many regions of

chromosomes were found to be re-arranged in the wrong order and there were extra or fewer copies of many genes. This chromosome shattering (the breaking and reassembling of genetic information on chromosomes) was documented and the process is estimated to exist in at least 2-3% of all human cancers (Landry et al., 2013).

Do we have a right to our cells or cell products?

Increasingly often the question of whether or not we have any property rights to our cells once they are no longer in our bodies is a topic of heated debate and even forms the basis of court cases (Skloot, 2006). Each of the following situations resulted in a different outcome regarding the patient and their samples. However, in all cases it seems to be clear that once the sample has left the body it is very difficult to claim it as one's own anymore. Laws generally protect the human subject, not their biological samples (Rivard, 2013). Many patient and tissues rights advocates such as Lori Andrews, J.D., who is the Director of the Institute for Science, Law, and Technology and a Professor of Law at the Illinois Institute of Technology champions that people should control their tissues to protect themselves from potential harm (Andrews and Nelkin, 2001). She urges us to reflect on how we can decide who gets our money after we die, shouldn't we be able to say who has a right to our biological samples as well (Andrews and Nelkin, 1998)? While there have been many cases involving property rights of one's biological samples (Skloot, 2006), consider the highlights of each of the following cases.

Henrietta Lacks

In 1951, Henrietta Lacks could barely afford her medical care and many of her descendants remain unable to afford medical insurance today (Skloot, 2010). This may seem ironic given that some genetically modified modifications of the HeLa cell line sell for up to \$10,000 (Truong, et al., 2012a and 2012b). A portion of the proceeds from the sale of *The Immortal Life of Henrietta Lacks* go to the promotion of making health care accessible, and provides other forms of financial assistance specifically for people whose biological samples have assisted scientific discoveries but whose samples were used without their knowledge, consent, or compensation (Skloot, 2010; Henrietta Lacks Foundation, 2013). The Rebecca Skloot website also promotes discussions between various readers of the Henrietta Lacks story ranging from her family members, to scientists, book club members, and the public at large (RebeccaSkloot.com; <http://rebeccaskloot.com/the-immortal-life/readers-talk/hela-forum/>). The original researcher (Dr. George Gey) from Johns Hopkins University did not personally profit from the use of Henrietta Lacks' cells as he freely shared them with interested researchers. However, the fact remains that these cells were taken and used without the precise knowledge or consent of Henrietta or her family members who remained disenfranchised from the science and medical establishments that promoted their use as major discovery tools (Skloot, 2010).

John Moore

The case of John Moore and the (Mo) cell line appears to be a bit more nefarious (McLellan, 2001). In 1976, John Moore, who was an Alaskan pipeline surveyor developed hairy-cell leukemia. He became the patient of Dr. David Golde, also a researcher at University of California-Los Angeles (UCLA). As part of treatment, John Moore had his spleen removed and

went to see Dr. Golde for follow up visits to take blood and other fluids as part of his ongoing medical care. Eventually, Dr. Golde started asking for additional and more frequent samples and started paying for John's visits. The process also began involving intricate consent forms stating that John understood his biological samples and any products derived from them did not belong to him. When John Moore refused to sign these consent forms, he was denied further medical care from Dr. Golde. He subsequently learned through his attorney that Dr. Golde was filing a patent on proteins developed from John's T cell lymphocytes which had some interesting disease-fighting properties that were keeping John's leukemia from progressing as quickly as expected. His cells naturally contained a unique protein that stimulates growth of white blood cells which fight infection (Skloot, 2006). The cell line is referred to as (Mo) and is estimated to be worth at least \$3 billion (McLellan, 2001). John Moore brought a lawsuit against Dr. Golde and UCLA in 1984. This resulted in a long court case. In 1990, the supreme court of California ruled against Moore. However, Moore did prevail on two counts; lack of informed consent and breach of fiduciary duty. John Moore died in 2001. Rebecca Skloot sums up the outcome of the Mo court case in this way, *"Any ownership you might have had in your tissues vanishes when they are removed from your body, with or without your consent. When you leave tissues in a doctor's office or a lab, you abandon them as waste. Anyone can take your garbage and sell it – the same goes for your tissue."* (Skloot, 2006). We are coming to understand that the "stuff" you leave behind in the hospital or doctor's office may not always get thrown out. It may be used in research (with or without your knowledge), or it may get stored indefinitely, or simply get thrown out. More than 307 million tissue samples from more than 178 million people are currently being stored in the United States, and this number is estimated to be increasing by more than 20 million samples each year (Eiseman and Haga, 1999).

Ted Slavin

Unlike in the John Moore case, Ted Slavin's doctor advised him that his cells were interesting. Ted Slavin was a hemophiliac who had been exposed to hepatitis. It was found that he had antibodies to hepatitis in his blood, but was not sick with hepatitis. Ted contacted laboratories to see if they wanted to buy his blood for this interesting property. They did want to buy his blood. In fact, he sold his serum for \$10 per mL and \$10,000 per L. This was income that he was able to have for the rest of his life and allowed him to pay for his medical care. While being paid for his blood samples, Slavin looked for a researcher of his choice to help him find a cure for hepatitis. Ted Slavin gave his antibodies to Dr. Baruch Bloomberg (his favorite researcher), who was a Nobel prize winning hepatitis research-scientist; Ted Slavin was very hopeful for a cure. In fact, Dr. Bloomberg was able to develop the first hepatitis B vaccine (Blumberg, et al., 1985). Ted Slavin later developed a company, Essential Biologicals, which recruited and empowered others with unique and potentially lucrative biological anomalies to provide their biological components to researchers (NPR: FreshAir, 2011; Washington, 2011). Current legislation regarding patient rights in relation to their cells and tissues is mostly based on the Federal Policy for the Protection of Human Subjects (known as The Common Rule). It was passed in 1981 for the protection of the whole person, not for their excised body parts, and samples are generally considered exempt as long as they are anonymous and experts are not sure what a "good and complex consent process" to allow researchers to use biological samples would look like (Skloot, 2006; Washington, 2011).

Dr. William Catalona

In this case, Dr. William Catalona, a reputable prostate cancer researcher (Urological Research Foundation, 2013) working for Washington University collected approximately 4,000 prostate and 250,000 blood samples from at least 36,000 men. He used very detailed consent forms and the patients were happy giving samples to their beloved and trusted Dr. Catalona. However, when Dr. Catalona left Washington University his patients requested that the samples be transferred to Dr. Catalona. Washington University declined and took possession of the samples as the intellectual property of their employee (Mangan, 2008). These samples are likely to be worth more than \$15 million. This situation ended up in court, going all the way to the U.S. Supreme Court and was heard in January of 2008. It was decided that Washington University has outright ownership of the samples from the prostate cancer patients (Washington University in St. Louis, 2013). The samples do not belong to Dr. Catalona, but to his employer at the time the research was done (Mangan, 2008).

Havasupai Indians

This may be one of the only cases where the patients were able to go into the research lab and actually remove their samples from the freezers there as part of a spiritual and religious ceremony. They were one of the very few groups that have been able to get their samples back after having their trust violated. In this situation genetic samples were gathered from Havasupai Indian tribe members for use in research on diabetes, which occurs in high incidence in their population. However, these samples, collected under a very broad consent form, were actually used to study a wide variety of genetic variations, including those underpinning mental illness and related to the geographical origins of the tribe. Both of these are not culturally acceptable uses of the DNA samples for the Havasupai people (Harmon, 2010). The case of the Havasupai people against the University of Arizona was ruled on in 2010 and resulted in the assigning of individual rights to a person's DNA sample. The University of Arizona spent at least \$1.7 million fighting various lawsuits by tribal members. The University eventually settled on \$700,000 to 41 tribal members and additional assistance in the forms of scholarships and health aid to the tribal members (Harmon, 2010).

Discussion

It is a very interesting situation that now through the use of technology our cells can live separately from us, essentially without us. While cells are generally understood to be the most basic units of life, are they really life on their own (particularly in the case of multicellular organisms, like humans)? Cells from thousands of people are currently living on without them and have been useful for many purposes from medical breakthroughs to an improved basic understanding of how living systems work. Generally, the donors of this biological material are unaware and uncompensated for their contribution, while in some cases they do have knowledge and are sometimes compensated (Truog et al., 2012a; 2012b; Kominwea and Becker, 2012). United States laws remain a bit unclear and somewhat contradictory in this area (Truog et al, 2012a, 2012b; Hayflick, 2012). Biological parts are often seen as just another research tool such as a beaker, petri dish, or test tube. However, we would all be wise to remember the humanity represented by these cells and other biological parts and pieces such that we remain aware of the

life involved. By raising our sensitivity to the nature of life and its component parts, perhaps we can learn to cherish all life, all organisms, and the environment that sustains all life on this planet (D'Alessio, pers. com.).

As biotechnology becomes more advanced, we are coming to realize that all of our biological parts do not stay with us for our entire lives. We are constantly shedding proteins and DNA in the form of our hair, bodily fluids, trace material left in chewing gum, tissues, and even in our excretory waste. In the last decade several artists have begun to bring this situation into the public eye. For example, information artist Heather Dewey-Hagborg creates facial 3D portraits using DNA she extracts from material she finds throughout New York City in public places (Squires, 2013). She has taken the DNA from discarded chewing gum, cigarette butts and even hair in public restrooms and analyzed it for a variety of sequences. Then she puts the genetic-marker results into a sophisticated computer program that assigns facial structure (for a person at age 25) to the outcome of those genetic markers. With that knowledge, she uses a 3D printer to create a facial portrait of the sample's source at age 25 (Gambino, 2013; Additional Information Links). In a similar vein, artists Ionat Zurr and Oron Catts have developed the Tissue and Culture Art Project (<http://tcaproject.org/>) in which they use living human cells and tissues to create works of art that have been on display in various galleries (Andrews, 2007). They are perhaps most well known for their piece, "Semi-Living Worry Dolls" (2000), in which they grew and displayed human cells on doll-shaped polymer scaffolding inside culture tubes (Andrews, 2007; Additional Information Links).

In the end, many questions remain. Questions exist such as, Do you have any rights to your biological parts? How much of you have you left behind for others to potentially examine in various ways? How would you know if someone ever did use your discarded biological parts, or parts that you unknowingly left behind? Do these ideas disturb you or really not bother you at all? Are your cells possibly living separately from you? We stand on a biological frontier; biological information that was once considered personal and was definitely inaccessible is now becoming increasingly accessible for careful study. These molecules, cells, and tissues are the results of millions of years of evolution and they are in our bodies, giving us our life, and possibly contributing to our eventual death, but do they even belong to us (Reader Survey Poll: <https://www.surveymonkey.com/s/CELLS>)?

Links to Additional Information:

1. An editorial illustration for Lisa Marginalia's review of Rebecca Skloot's book, *The Immortal life of Henrietta Lacks*. By the artist, Chiarara Andriole. October 2011. <http://www.chiararaandriole.com/The-Immortal-Life-of-Henrietta-Lacks>
2. Official book trailer with Rebecca Skloot, *The immortal life of Henrietta Lacks*. <http://www.youtube.com/watch?v=1vow1ePzuqo>
3. Images of HeLa cells and an interview with Rebecca Skloot on NPR's Fresh Air radio program. February 2, 2010 <http://www.npr.org/2010/02/02/123232331/henrietta-lacks-a-donors-immortal-legacy>

4. Documentary regarding the DNA from Havasupai Indians. Blood Journey April 2010, by Kassie Bracken and Amy Harmon
<http://www.nytimes.com/video/2010/04/21/us/1247467672743/blood-journey.html>

Links to Art Using Biological Components:

5. The Tissue Culture and Art Project. The Semi-Living Worry Dolls. By Oron Catts and Ionat Zurr, Australia. <http://sciencegallery.com/visceral/semi-living-worry-dolls/>
6. Studio 360: Kurt Andersen's DNA Mask Revealed by Kurt Andersen and Heather Dewey-Hagborg. February 7, 2013. <http://www.youtube.com/watch?v=u1jgkykLBVI>
7. Excerpt from Hands-On Ideas: Genetics with Artist Heather Dewey-Hagborg
<http://www.youtube.com/watch?v=j2SjNSIRbvM>
8. Lifelike Masks created from DNA. May 7, 2013 by GeoBeatsNews
http://www.youtube.com/watch?v=dN_STkxr91g

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http://www.wired.com/magazine/2010/01/st_henrietta/
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<http://www.firdi.org.tw/EngWeb/2003/bcrc.htm>. Last accessed. May 28, 2013.
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http://www.corning.com/lifesciences/latin_america/en/about_us/cell_culture_history.aspx

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