The stolen work of Esther Zimmer Lederberg that changed the field of Genetics

Dressed in a floor-length high school prom dress and ballet slippers that were hand-dyed to match, Esther Zimmer Lederberg stood alongside her husband, Joshua Lederberg, as he accepted the 1958 Nobel Prize in Physiology or Medicine for foundational discoveries in the mechanisms of horizontal gene transfer between bacterial cells. Normally clad in a laboratory coat and unconcerned with fashion, Esther was entirely out of place amongst the other wives. Her sincere effort to conform to the role of a Nobel Laureate's wife was surprising, in hindsight. After all, Joshua Lederberg was accepting one of the most prestigious awards a researcher can earn for work that was equally pioneered by none other than his wife, Esther, who in one evening had been demoted from co-researcher for over a decade to wife and escort.

Esther Zimmer was born in 1922 and raised in the South Bronx, New York by her parents who had immigrated from Eastern Europe. She was an incredibly bright student who had a knack for foreign languages and went on to study French with the goal of becoming a teacher at Hunter College, a women's teacher training school in New York City. In her junior year however, she made the unconventional decision to switch majors to biology despite discouragement from her male professors. Esther was persistent and enrolled in the few biology courses that were offered at Hunter College. Her lack of support from her professors galvanized her to seek a mentor outside of the campus. She found this support and mentorship in B.O. Dodge, a mycologist who studied *Neurospora* and set the stage for bacterial genetics research. Esther worked in Dodge's lab for the remainder of her undergraduate career where he imbued her with his infectious enthusiasm for laboratory research and microorganisms.

During the 1940s, one of the biggest questions in biology research was how the "gene" functions at the biochemical level. Dodge's work on *Neurospora* genetics proved valuable to geneticist George Beadle and biochemist Edward Tatum, two scientists who in 1941, formulated the One Gene-One Enzyme Hypothesis that "revealed that the properties of animals and plants are determined by proteins, and that each protein, including each enzyme, is determined by a single gene." Dodge had close professional and personal relationships with Beadle and Tatum whom Esther later worked with as she navigated the male-dominated field of genetics.

After graduating college at nineteen years old, Esther had to carve out her own career path in research. Unlikely to be granted a fellowship and unable to afford graduate school, she worked as a research technician for Alexander Holleander, a pioneering geneticist of the 1940s who took a special interest in mentoring young female scientists. Holleander taught Esther how to create mutant cell lines through UV radiation and X-rays and after 3 years of working in his laboratory, she was accepted to a Master's program at Stanford University. Esther's graduate advisor was none other than George Beadle with whom she worked with, along with Edward Tatum, for the next 4 years. Esther quickly jumped on the emerging potential of using bacteria as model organisms for genetics study. She managed to take courses with the famous microbiologist Cornelius van Niel, despite having very little funding during the summer months when his courses were offered. Esther combined her newfound knowledge, and soon love, of bacteria, with her experience in radiation biology to create the *E. coli* K-12 strain mutants that would be used to perform the work that earned her future husband and mentors a Nobel Prize.

In 1946, after Esther had earned her Master's degree from Stanford, she attended the annual Cold Spring Harbor Symposium, a conference that drew researchers from around the globe. Joshua Lederberg, a medical student at Columbia University, had just made the pivotal discovery that bacteria could transfer and recombine their genetic material and announced his findings at the symposium. Joshua had reached out to Esther prior to the conference asking to meet, as his experiments were performed using the mutant bacterial strains that Esther had prepared. After meeting at the symposium, they quickly fell in love, marrying just a few months later. Joshua later wrote, "'we had a clear contract: we would keep house with symmetrical responsibilities and acknowledge the primacy (if not supremacy) of the obligations of scientific work.'"

Joshua Lederberg was an ambitious and cerebral genius. He was a fierce debater with a view that scientific research was a competitive contest; a contest that he most certainly aimed to win. Many scientists who worked with the Lederbergs describe Esther as the experimental genius behind Joshua's work. Leonore Herzenberg of the Stanford Genetics department wrote, "'[Esther] was the ground from which Josh's brilliance grew.'" The Lederbergs' successful partnership is often described in synergistic terms, "[Esther's] 'golden hands' and deep appreciation for bacteria and their plasmids perfectly complemented Joshua's literary genius, powerful intelligence, and fierce ambition.'" However, this seemingly harmonious partnership had a tragic asymmetry that ultimately cost Esther her scientific career. She cared little about awards, fame, or accolades and was entirely devoted to the work itself. Her fascination and reverence for bacteria made her incredibly attuned to their "wonderful peculiarities." Joshua, who was effectively a dunce and far less experienced in the laboratory, relied on and exploited Esther's experimental genius and her keen knack for recognizing anomalous results that often catalyze major discoveries. After they married, the Lederbergs moved to the University of Wisconsin where Joshua was hired as an assistant professor and Esther was accepted into their PhD program. The married team was also granted funding for a new research program that would elucidate the mysteries behind the mechanism of bacterial conjugation that Joshua had originally discovered in 1946.

One day in 1950, Esther Lederberg entered the dingy, spare basement laboratory that she ran with her husband at the University of Wisconsin to do a routine check on her bacterial cultures. However, what she saw was most definitely not routine or regular. She noticed that some colonies looked as if they had been "nibbled" around the edges. To investigate, Esther irradiated the K-12 strain with UV light and found that she had activated the "nibbling factor." She denoted this factor, λ. She later discovered that λ was actually a bacteriophage, a latent virus that hid inside the bacterial chromosome and upon UV irradiation, lysed its host. The λ phage could even excise part of its host's chromosome and carry it to the next bacterium that it infected, a process known as generalized transduction. Shortly after this discovery, Esther was monitoring cultures when she made another unexpected discovery, "one day ZERO recombinants were recovered…I explored the notion that there was some sort of 'fertility factor,' which if absent, resulted in no recombinants. For short I named this *F.*' Esther had unearthed a process that would flip the entire field of genetics on its head. She discovered that this F-factor was highly infectious and transferred from F^+ donors to F^- recipients, thereby transmitting fertility and a segment of the donor's chromosome. While her husband had made the original discovery in 1946 that bacteria could transfer and recombine their genes, it was Esther's "golden hands" that revealed the mechanism by which this occurred. Joshua prevented Esther from pursuing further analysis and research on her discoveries of λ phage and the F-factor because "he thought finishing my thesis was a priority." Esther even noted many years later that "whenever she discovered something important, Joshua immediately took her off the project and assigned the work to someone else. She concluded that if she wanted to do scientific work it would be better for her to quietly complete the work herself, without telling Joshua."

Esther's vow to avoid telling Joshua about interesting discoveries she made in her laboratory work was not because she had a strong desire to receive recognition for her work, but rather she was invested in seeing the work through and furthering the field of genetics. Joshua's decision to delay Esther from following up on her discoveries of λ phage and the F-factor was likely partially due to his own biases about the role of women in science, often relegated to the lowly roles of assistant or technician, and his ego-driven ambition. He may have felt jealous or threatened by Esther's knack for elegant laboratory work and experimental design. However, there is another element that explains Joshua's short-sighted decisions to prevent Esther from following up on her discoveries. Despite the accumulating evidence against it, Joshua steadfastly believed that bacterial conjugation involved the mutual combination of two male and female bacterial chromosomes which formed a bacterial zygote. His insistence in this theory was based on the existing knowledge of sexual reproduction in other microbes like fungi and protists and the previous two decades of genetics research. Esther's discoveries of λ phage and the F-factor directly contradicted Joshua's bacterial zygote hypothesis and suggested instead that bacterial conjugation involved a one-way chromosome transfer. Joshua believed in his hypothesis so strongly that he even developed an alternative hypothesis to explain the unexpected results of his experiments: the post-zygotic elimination hypothesis. He insisted that "male and female cells mutually contributed their genes to each other, and then after fusion, the female genes were 'excluded.'" William Hayes, a geneticist who contributed to the one-way partial chromosome transfer hypothesis wrote, "'it was not until the hypothesis of one-way partial chromosome transfer was proven beyond reasonable doubt by the work of Wollman and Jacob four years later that Lederberg accepted it.'"

Thomas Kuhn, an historian and philosopher of science, believed that the blind devotion of scientists to a particular paradigm or theory is "[not] because they believe it is well-supported by the evidence, or because it is the 'official' way to do things…rather, they follow it because they cannot imagine doing science any other way. Were they presented with an alternative paradigm…they would find it incomprehensible." Kuhn had to reckon with the question of how science can possibly progress if scientists are incapable of considering theories and paradigms that contradict their own. He believed that "scientists' very commitment to the paradigm can push it to the point of destruction: they abide by its prescriptions, they faithfully execute its plan, yet they run into insoluble problems because the paradigm is inadequate in some way." While Joshua persistently stuck to his bacterial zygote hypothesis, rapidly accumulating evidence directly contradicted his theory, including evidence produced in his own lab by his wife Esther. Nearly every pioneering geneticist at the time continued forward with the idea that there must be some other explanation for the mechanisms underlying bacterial conjugation. Yet, Joshua's own arrogance and shortsightedness prevented him from practicing impartial and unbiased science. Kuhn's analysis of the practice of science essentially states that in order for science to progress, scientists' inability to see past their own ideas allows them to "work [the theory] to death," thus exposing its flaws. However, in the case of horizontal gene transfer and bacterial conjugation, it was the humility and hard work of Esther Lederberg, William Hayes, Lugi Cavalli, Elie Wollmann, and Francoise Jacob that led to a scientific revolution, not Joshua Lederberg's stubbornness and self-importance.

One of the most important questions that arose from the Lederbergs' work was how antibiotic resistant bacterial strains arise. Is prior exposure to the antibiotic required for resistance? Or does the bacteria acquire resistance in some other way, perhaps through conjugation, prior to antibiotic exposure? In the laboratory, the only way to test this was through the mind-numbingly tedious task of transferring each bacterial colony scattered across a petri dish to a new plate containing the antibiotic in the exact same configuration. This process would take hours, requiring the inoculating instrument to be passed through a flame for sterilization between each transfer. One day, in what one could only imagine as an extemporaneous moment of playful ingenuity, Esther decided to use the pad from her makeup compact as a stamp. In just a single step, Esther pressed her makeup pad to the surface of the petri plate covered in bacterial colonies, creating a stamp that could then be pressed to another plate, thereby replicating the geometric configuration of growth from the original plate. Esther spent several months afterwards searching for the best type of fabric for the bacterial stamp and eventually refined the technique using sterilized velvet. In the original publication that shared this creative invention with the world, Joshua Lederberg was the first author and Esther Lederberg second. After Joshua's former mentor Francis J. Ryan wrote about the technique in an article on the evolution of antibiotic resistance calling it an "ingenious experiment developed and by the University of Wisconsin geneticist, Joshua Lederberg," Joshua wrote him angrily insisting that Esther was a co-author and co-inventor of the work. Similarly, in a 1956 interview with the Milwaukee Journal after he had won the Eli Lilly award in bacteriology in 1953, Joshua remarked, "Esther should have been in on that, too." Unfortunately, Joshua did not share this same sentiment when he won the Nobel Prize in Physiology or Medicine "for his discoveries concerning genetic recombination and the organization of the genetic material of bacteria."

In 1958, Esther and Joshua Lederberg traveled to Stockholm, Sweden for Joshua's acceptance of the Nobel Prize, shared with George Beadle and Edward Tatum. For Esther, this must have been widely disorienting. Once Joshua's co-researcher on the mechanisms behind bacterial conjugation for over a decade, in a single night, Esther had been demoted to wife and escort of a Nobel Laureate. In his acceptance speeches, Joshua did not even mention his wife Esther, let alone acknowledge her valuable contributions as his co-researcher. Unfortunately, Esther's situation was not at all uncommon in this era. By 1957, only one woman, Gerty Cori, had won a Nobel Prize in Physiology or Medicine which was shared with her husband, Carl Cori, and Bernardo Houssay for their research on carbohydrate metabolism. While it is true that there were significantly fewer female scientists as a result of systemic discrimination, there is an unrecognized woman behind many of the Nobel Prize-winning discoveries of the 20th century. Coined by science historian Margaret W. Rossiter in 1993, the Matilda Effect is a phenomenon where female scientists are "systematically under-recognized, either deliberately for strategic reasons or unconsciously through traditional stereotyping." The theft of Esther's Nobel recognition was likely a combination of deliberate action and cultural views and stereotypes of the 1950s woman. Married women in particular are more likely to be victims of the Matilda Effect, according to Rossiter. Reduced to "wife," a married woman's personhood is effectively stripped away as her only value becomes tied to her relationship with a man, making it all too easy to rob women of their contributions and transfer ownership to her husband.

Coupled with the theft of Esther's Nobel recognition, the obvious disparity in the Lederbergs' professional ranks led to the collapse of their marriage and collegial relationship. In 1959, the Lederbergs moved to Palo Alto, California where Joshua formed and became chair of the Genetics Department at Stanford University, employing Esther as an untenured research associate. Despite their vow to "'keep house with symmetrical responsibilities and acknowledge the primacy (if not supremacy) of the obligations of scientific work,'" Joshua hoped that Esther would resign her role as a scientist and perform the "natural" wifely and motherly duties of a mid-20th century woman. He wrote, "'Once she earned her Ph.D., our plan was to start a family, following the happy pattern we saw among our friends. That would suspend the conflicts from our intersecting careers, and enhance Esther's sense of purpose in life.'" The Lederbergs were unable to have children and their marriage ended in 1967.

The end of their marriage also meant the end of Esther's career in research. In a letter to a colleague in 1968 she wrote, "'I found out my research support has not been renewed by Josh for the coming year…the world will stop turning unless I sit in my place and play with these bacteria…It might be better to get a tan and think than streak colonies. What then I don't know.'" Esther was demoted even further when her funding was cut off by Joshua and she became a "Visitor" in Bruce Stockner's laboratory for three years. Esther was eventually appointed as a research professor, an untenured position, after she and two other women at Stanford demanded that they be appointed as professors. In 1976, she was appointed to curate the Plasmid Reference Center where she worked until her retirement in 1985. The final years of the Lederbergs' marriage marked a shift in Joshua's public defense of Esther's contributions in their discoveries. After chastising his former mentor for not giving credit to Esther for the Replica Plating Technique and insisting that she should have shared the Eli Lilly award with him in 1953, Joshua's Nobel Prize speech failed to mention Esther and in a 1989 paper on the history of genetics research, he claimed that he was the original inventor of the Replica Plating Technique.

Not only is the theft of a person's life work simply unjust, but when the loudest and most prominent voices in science only represent half of the population, there are innumerable losses in the advancement of science. Although she came to terms with her exile from scientific research, finding joy and fulfillment in music and art in her later years, it is difficult to not wonder about the advancements and discoveries Esther Zimmer Lederberg could have made if she was given the recognition and credit that she earned.

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