Peter Mazur (1928–2015) [1]

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Peter Mazur was a researcher in the US who developed new ways of preserving biological material by freezing it, a process called cryopreservation [5]. If done correctly, cryopreservation [5] enables scientists to store or study biological material for an extended period of time. If done incorrectly, cryopreservation [5] can easily harm or destroy biological material. Mazur worked to find the best ways to cryopreserve different cells, embryos, and organs in order to minimize the damage caused by freezing. Throughout the 1960s and 1970s, Mazur and his colleagues published a series of papers that ultimately led to the discovery of previously unexplored factors that can cause harm to cells during the cryopreservation [5] process. He called that discovery the two-factor hypothesis. That same year, Mazur also contributed to one of the first successful attempts at cryopreserving viable [6] mouse [7] embryos. Mazur's work to improve the cryopreservation [5] process helped to establish foundational knowledge that was later used in many different fields, such as reproductive health and conservation.

Mazur was born on 3 March 1928 in New York City, New York, to Adolphia Kaske and Paul Mazur. Mazur's mother stayed at the family's Manhattan home during the day, caring for Mazur and his younger sister Nancy. His father was a business writer, lecturer at Harvard University [8], in Cambridge, Massachusetts, and a partner at Lehman Brothers, a Wall Street investment firm. Mazur attended preparatory school at The Lawrenceville School, a private boarding school in Lawrenceville, New Jersey. Mazur graduated from Lawrenceville in 1946 and began attending Harvard University [8].

Mazur spent his next four years studying biology at Harvard. During his time there, he became a member of the Phi Beta Kappa Society. As of 2020, Phi Beta Kappa is one of the oldest academic honor societies in the US, and only accepts a few new members each year. Mazur graduated with his bachelor's degree in biology with honors in 1949 and stayed at Harvard until he graduated in 1953 with his doctoral degree in biology. During his time at Harvard, Mazur met his first wife, Drusilla Stevens, a native of Springfield, Massachusetts, who attended Wilson College in Chambersburg, Pennsylvania. In 1953, the two married in Springfield, Massachusetts.

After earning his doctoral degree, Mazur spent the next four years working for the US Air Force's Research and Development Command. Near the end of his service, he began carrying out research with William H. Weston [9], a Harvard faculty member whose research focused on mycology, or the study of fungi. Together, Weston and Mazur published a paper in 1956 on "The Effects of Spray Drying on the Viability of Fungous Spores," in which the researchers compared methods of preserving fungal spores by dehydrating them to identify which methods resulted in higher survival rates.

After his release from the Air Force, Mazur received a post-doctoral fellowship from the National Science Foundation, or NSF, in 1957 to continue his studies at Princeton University [10] in Princeton, New Jersey. In another publication related to his previous research on mycology, Mazur continued using fungal spores as research subjects, but instead of studying dehydration he focused on the effectiveness of freezing as a means of preserving the spores. From that point on, Mazur's research focused on freezing and the effects of low temperatures on different kinds of biological material. During his time at Princeton, Mazur also studied the survival of bacteria at low temperatures. After finishing his fellowship in 1959, Mazur was hired as a biologist at Oak Ridge National Laboratory [11], in Oak Ridge, Tennessee, where he began his career in cryobiology.

In his first decade at Oak Ridge National Laboratory [11], Mazur conducted a number of experiments that aimed to determine causes of injury to cells during freezing. For example, in his 1963 article, "Kinetics of Water Loss from Cells at Subzero Temperatures and the Likelihood of Intracellular Freezing," he noted that cells survived more often during freezing when they were cooled slowly. Later in 1965, Mazur published "The Role of Cell Membranes In the Freezing of Yeast and Other Single Cells," in which he discusses how the properties of a cell membrane impacts the likelihood for a cell to survive or die during the freezing process. Then, in 1969, Mazur was appointed as chairman for a symposium called "The Frozen Cell," in London, England. That event was part of a larger series of symposia hosted by the Ciba foundation, a Swiss nonprofit that focuses on improving collaboration among people within the medical sciences. There, Mazur led a panel composed of international scientists in discussions related to what part of the freezing process was responsible for causing damage and death to a cell.

At the time of the symposium, most scientists believed a rapid cooling rate was the best way to protect cells, which was a belief Mazur had later challenged. When cryopreserving cells, researchers freeze those cells in liquid solutions that have similar concentrations of dissolved salts as those found inside of the cells. As the solutions freeze, the water within the solutions separates from the salts as the water crystallizes into ice, leaving a higher salt concentration in the solution that has not yet become frozen. The cell's exposure to those high salt concentrations was identified as harmful to the cell by James Lovelock, a scientist who in 1953 published "The Haemolysis of Red Blood Cells during Freezing and Thawing," which detailed various freezing effects on biological material in England in the 1950s. Lovelock's results promoted the belief that a faster cooling rate was best for all cells because it would reduce the time in which the contents of a cell are exposed to high salt concentrations, therefore minimizing damage caused by that exposure. At the symposium in 1969, Mazur asked his fellow scientists to what extent those assumptions were valid, and indicated that he believed there were at least two factors responsible for cell injury when freezing cells. He then began conducting research at Oakridge National Laboratory to investigate the widespread belief that a rapid cooling rate was best.

In 1972, along with scientists Stanley Leibo and Ernest Chu, Mazur published the paper, “A Two-Factor Hypothesis of Freezing Injury: Evidence from Chinese Hamster Tissue-culture Cells,” in which they determined two factors responsible for cell injury and death during the freezing process. They demonstrated those two factors are when a cell is exposed to high salt concentrations and when ice forms within the cell. They note scientists should account for both of those factors when determining the rate of cooling. Recognizing the harm intracellular freezing causes, Mazur explained that there was an optimum cooling rate for each different kind of cell that is slow enough to prevent intracellular freezing, while being rapid enough to minimize exposure to high salt concentrations. Mazur’s identification of the risk of intracellular ice formation helped scientists in the field of cryopreservation [5] to adjust cooling rates and increase cell survival.

In the same year that he published “A Two-Factor Hypothesis of Freezing Injury: Evidence from Chinese Hamster Tissue-culture Cells,” Mazur also contributed to one of the first successful attempts at cryopreserving viable [6] mouse [7] embryos that could still develop into offspring. In that study, Mazur recruited his Oak Ridge National Laboratory [11] colleague, Leibo, and embryologist David G. Whittingham from London, England, to conduct the experiments. Whittingham had first published results on the preservation of mouse [7] embryos the year before in 1971 in the journal*Nature*. Mazur and Leibo invited Whittingham toOak Ridge National Laboratory [11] to work with them to replicate Whittingham’s result, but could not do so successfully. It was only when they used Mazur and Leibo’s method of using liquid nitrogen to cool the cells more slowly that the team was able to create a replicable and reliable method of cryopreserving the embryos so that, when thawed, the embryos could still lead to successful births. Mazur and his team stated that they believed their success would help further the ability to freeze and store more complex mammalian organisms for medical use in the future. The team published their results in the journal *Science* in 1972. That publication provided a model for future researchers to cryopreserve other mammalian embryos.

After those two projects, Mazur’s wife Drusilla died in 1982. Two years after her death, Mazur remarried to Sara Jo Bolling, whose previous husband had died four years earlier. Bolling and her late husband had been friends and neighbors to the Mazurs up until her husband’s death. Mazur continued working in cryobiology throughout their marriage and after his second wife’s death nearly twenty years later.

After Mazur created a reliable method of preservingmouse [7] embryos, he began to determine if he could uncover a reliable procedure for the cryopreservation [5] of *Drosophilia* embryos. *Drosophilia* is the name of a species of fly that scientists commonly use for genetic research because of its ability to mutate quickly and produce many generations in a short amount of time. Though it took many trials, Mazur’s team found a reliable method several years later in 1992. That enabled scientists to not have to sustain large populations of *Drosophilia* in their laboratory, which was often costly and tedious.

Following Mazur’s experience with *Drosophilia*, the National Institutes of Health [12] provided Mazur with funding to investigate the process of cryopreserving mosquito embryos. Many labs in the 1990s were attempting to genetically-modify mosquitoes so that they would be unable to carry malaria, a disease caused by a parasite that mosquitoes can transmit to humans [13]. In humans [13], malaria can cause severe flu-like symptoms that can often be fatal. They tasked Mazur with the matter of establishing a reliable means of cryopreserving many different strains of mosquitos, with the notion that the technology could enable scientists to release mutant mosquitoes to the wildtype population of mosquitoes to spread a gene resulting in the inability to carry malaria. In order to do that, scientists would need to compare hundreds of mosquito strains to locate the ideal genes [14] to modify. If the embryos could be cryopreserved, then labs could maintain many more cryopreserved stores of mosquitos from different strains at once, which could help researchers compare between strains. However, Mazur was unable to use the same methods for preserving *Drosophilia* embryos to preserve mosquito embryos. After those endeavors, most of the rest of Mazur’s work focused on the fundamentals of broadly freezing any kind of cell rather than specific cell lines.

In 1998, Oak Ridge National Laboratory [11] closed its biology division, and Mazur moved to Knoxville, Tennessee, to work at the University of Tennessee. Mazur’s research over the next decade focused on technology surrounding cryopreservation [5], specifically to help speed up the rewarming process of frozen cells. In the late 2000s, he studied the effects of using lasers when rewarming cells. Lasers allow for a quicker rewarming process without harming the cells, which reduce the risk of them refreezing as they thaw. Mazur worked on using lasers to aid the preservation of different cell lines up until his death.

Mazur’s career established the foundations of cryobiology and helpedcryopreservation [5] become a more practical research tool. He received numerous awards throughout his career, including a Distinguished Service Award from the American Association of Tissue Banks, and a Jackson National Laboratory Award for his contribution in producing one of the first mammals born from a frozen embryo [15]. Four of Mazur’s papers out of his over 170 publications have been named Citation Classics, or highly-cited publications as identified by the Institute for Scientific Information. Additionally, he received the Society for Cryobiology’s Fellowship Medal in 2005, after having served as a member of the Society’s Board of Governors, including as president of the society in the early 1970s, as well as a member of the Editorial Board of the Society’s Journal, *Cryobiology*, from 1967 up until his death.

Mazur was diagnosed with lung cancer in 2013. Despite his illness, Mazur continued his work in cryobiology. Mazur died at his home in Oak Ridge, Tennessee, on 15 December 2015.

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