”The Genetic Control and Cytoplasmic Expression of 'Inducibility' in the synthesis of B-galactosidase” (1959), by Arthur B. Pardee, Francois Jacob, and Jacques Monod

Between 1957 and 1959, Arthur Pardee, François Jacob, and Jacques Monod conducted a set of experiments at the Pasteur Institute in Paris, France, that was later called the PaJaMa Experiments, a moniker derived from the researchers' last names. In these experiments, they described how genes of a species of single-celled bacteria, called Escherichia coli (E. coli), controlled the processes by which enzymes were produced in those bacteria. In 1959, the researchers published their results in a paper titled ”The Genetic Control and Cytoplasmic Expression of 'Inducibility' in the Synthesis of β-galactosidase by E. coli”. When they compared mutated strains of E. coli to a normal strain, Pardee, Jacob, and Monod identified the abnormal regulation processes and enzymes produced by the mutated genes. The results showed how enzymes break down the molecules that the bacteria ingested. The PaJaMas experiments uncovered some of the molecular mechanisms that regulate how some genes yield enzymes in many species.

In 1957, Pardee took a sabbatical year from the University of California in Berkeley, California, and he joined Monod's lab the Pasteur Institute. Monod had been studying how enzymes respond to stimuli, like sugars. Pardee had also been studying enzymes. Earlier, Jacob had worked with Elie Wollman on bacterial conjugation, the process by which individual bacteria exchange portions of DNA with each other. Monod studied how organisms, such as E. coli, created enzymes, while Jacob investigated how genes regulated the production of those enzymes. To better explain the action of enzymes in bacteria, Jacob and Monod collaborated with each other and with Pardee. As the group was familiar with the enzyme beta-galactosidase (beta-gal) system in E. coli, they chose this system for their research.

At the time, many scientists proposed the theory of enzyme adaptation, which stated that an enzyme could change shape by surrounding an external stimulus, such as a sugar, and that then the enzyme would decompose the stimulus, break down the sugar. Researchers aimed to test the enzyme adaptation theory. As researchers found beta-galactosidase in E. coli only when certain types of sugars (betagalactosides) were in E. coli's environment, researchers investigated whether or not beta-galactosidase was an enzyme that had transformed from another enzyme by changing shape.

In 1957, scientists established that the lac region of the DNA in bacteria contained genes that controlled production of the enzymes galactoside-permease and beta-galactosidase. Galactoside-permease controlled the entry of certain types of sugars, which included lactose (galactosides), into the cell. Beta-galactosidase was the enzyme that decomposed the sugar. In their 1955 paper, ”Sur le mécanisme du transfert de matériel génétique au cours de la recombinaison chez Escherichia coli” (Mechanism of the transfer of genetic material during recombination in Escherichia coli K12), Jacob and Wollman had shown that certain mutations, labeled z^-, y^-, and i^-, in a cell's lac region changed the cell's ability to decompose sugar. Jacob and Wollman observed that if the DNA was abnormal, so were the enzymatic products, but they were unable to determine if a third product mediated the production of enzymes from DNA. The z^- mutation to the lac region of the DNA left an E. coli cell unable to produce beta-galactosidase, while the y^- mutation left the cell unable to synthesize galactoside-permease. With an i^- mutation, an E. coli cell constantly made beta-galactosidase.

These results led Pardee, Jacob, and Monod to hypothesize that something must cause the production of beta-galactosidase in normal E. coli cells, or that the bacteria must always have enzymes that can re-arrange to break down sugars. As the researchers could show that a cell created the

enzyme beta-galactosidase when in the presence of sugars, but not in the absence of sugars, the researchers showed that the sugars could cause the production of enzymes. They labeled the sugars as inducers. If the sugars caused E. coli to produce enzymes, then a cell would have to make the enzymes anew whenever the cell came into contact with the sugar. But if the enzymes re-arranged their shape and decomposed sugars at any given time, then the shape re-arranging process, called constitutive expression, provided evidence for the theory of enzyme adaptation.

Jacob, Monod, and Pardee experimented with E. coli to see if, when exposed to sugars, those cells always produced new enzymes or if instead they had enzymes that rearranged themselves. The trio started with a strain of bacteria in which each bacterium in the strain had normal genes, labeled as z^+ and i^+. The researchers then developed a strain of bacteria in which the each bacterium in the strain had the same mutation or abnormal genes, labeled as z^- and i^-. Next, the researchers relied on the process of bacterial conjugation, a process in which two bacteria connect with each other and exchange genetic material, to cross bacteria from the normal strain with bacteria from the abnormal strain. In doing so, they created additional strains of bacteria with different combinations of genes. When the trio studied bacteria with z^+ and i^- genes, they noted that the cell produced beta-galactosidase constantly for a short period of time, but then stopped. When these cells were placed in the presence of the inducer, the sugar lactose, the bacteria produced beta-galactosidase. In bacteria with the z^- and i^+ genes, the cells only produced beta-galactosidase in the presence of sugar. In bacteria with the z^- and i^- genes, the cells constantly made beta-galactosidase, regardless of whether or not sugar was present with the cells.

The behavior of bacteria that had abnormal z genes indicated that something on the gene site prevented the production of beta-galactosidase. Pardee, Jacob, and François termed that something as a repressor. The behavior of bacteria that had abnormal or mutated i genes indicated that something induced cells to make the enzyme in the normal bacteria, and that cells from the mutated strains always produced the enzyme so long as they lacked repressors.

The trio's results did not support enzyme adaptation theory because E. coli made new enzymes each time sugar induced the lac operon. The researchers argued that something at the gene level regulated the production of different kinds of enzymes. There is not one enzyme that changes shape to fulfill all functions of enzymes; rather, the cell makes the type of enzyme for which it is induced. Jacob and Monod argued later in 1961 that if researchers could find similarities and differences in gene regulation between organisms within a species, as Pardee, Jacob, and Monod had done, then researchers could also identify patterns of gene regulation that were general across different species. Later experiments revealed how an inducer (sugar) attaches to a repressor and obstructs it so that the genes could be activated to make the enzyme, results published in in Monod and Jacob's 1961 paper, ”Genetic Regulatory Mechanisms of the Synthesis of Proteins”.

The PaJaMa experiment supported the hypothesis that a molecule mediated the production of proteins from DNA. While researchers had shown that lac genes helped produce the enzyme beta-gal, they couldn't explain how the DNA, which was too large to pass through the nucleus, could transfer genetic information outside of the nucleus to the ribosome, comprised of RNA. This intermediate molecule was later discovered and labeled as messenger RNA (mRNA).

After the PaJaMas experiments, Pardee left France, returning to the US in 1959. He continued to work in biochemistry while Monod and Jacob continued their collaboration in France. For the PaJaMa Experiment and the related experiments that came after, Jacob and Monod won the 1965 Nobel Prize in Physiology or Medicine. From the Federation of American Societies for Experimental Biology, Arthur Pardee later received the 3M Award in 1980 for his work on the PaJaMa experiment.

Sources

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