Scientists use RNA to reprogram one cell type into another

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(PhysOrg.com) -- For the past decade, researchers have tried to tweak cells at the gene and nucleus level to reprogram their identity. Now, working on the idea that the signature of a cell is defined by molecules called messenger RNAs, which contain the chemical blueprint for how to make a protein, researchers at the University of Pennsylvania School of Medicine, School of Arts and Sciences and School of Engineering have found another way to change one cell type into another.

By simply flooding one cell type, a nerve cell, with the an abundance of a specific type of messenger RNA (mRNA) from another cell type, the investigators changed a neuron into an astrocyte-like cell, a star-shaped brain cell that helps to maintain the blood-brain barrier, regulates the chemical environment around cells, responds to injury, and releases regulatory substances.

James Eberwine, PhD, Elmer Holmes Bobst Professor of Pharmacology, Junhyong Kim, PhD, Edmund J. and Louise W. Kahn Term Endowed Professor of Biology and first author Jai-Yoon Sul, PhD, Assistant Professor of Pharmacology, and colleagues report their findings online this week in the Proceedings of the National Academy of Sciences. This approach offers the possibility for a new type of cell-based therapy for neurodegenerative and other diseases.

"In some ways, this is akin to what a virus does," explains Eberwine, "When a virus infects a cell it affects the host cell genome and the RNAs that it can make." By putting the RNA of one cell type, in the correct

amounts, into another cell type, we were able to change its function."

"This research overturns the notion that all cells are permanently hardwired with little ability to change their physiology," notes Sul.

"What's new about this approach is that we didn't have to make the host cell pluripotent, that is the ability to develop into any of three major tissue types, we can directly convert from one cell type to another, without the intermediate step," explains Eberwine. The scientists put in an excess of astrocyte messenger RNAs into the neuron cell body using phototransfection, a method they created a few years ago that creates temporary pores in the cell membrane. "The RNA population was then diffused into the cell and the host cell did the rest," adds Eberwine.

"We liken the differentiated cells to ecological communities, forests and meadows," notes Kim. "Each have similar organisms but have settled on particular characteristics that we recognize as distinct. And, just as ecological communities can be nudged from one type to another, we thought we could nudge differentiated cells from one type to another through the use of the RNA population. So, we like to think of cells as an ecological community of molecules, with dynamic molecular interactions producing a larger system-level cell function similar to how organism interactions generate forests and meadows.

The approach they used, called Transcriptome induced phenotype remodeling, or TIPeR, is distinct from the induced pluripotent stem cell (iPS) approach in that host cells do not have to be dedifferentiated to a pluripotent state and then redifferentiated with growth factors to the destination cell type. This work is more similar to the prior nuclear transfer work in which the nucleus of one cell is transferred into another cell where upon the transferred nucleus then directs the cell to change phenotype based upon the RNAs that are made.

TIPeR uses RNA populations to direct the DNA in the host nucleus to change the cell's RNA populations to that of the destination cell type, which in turn changes the phenotype of the cell.

There are about 100,000 mRNA molecules in a neuron at any one time. The researchers transferred nearly double that: About 200,000 astrocyte mRNAs were transferred into the neuron, effectively dampening the neuron mRNA's ability to be translated and made into protein.

Essentially the team extracted and produced mRNA from an astrocyte, then used phototransfection to create pores in the neuron cell membrane to flood it with an excess of astrocyte mRNAs, which reside in the neuron host cell cytoplasm. Because there are now so many astrocyte mRNAs versus neuron mRNAs, they take over like a virus and the astrocyte mRNAs are translated into astrocyte proteins in the cytoplasm. These astrocyte proteins then influence gene expression in the host nucleus so that astrocyte genes are turned on and astrocyte cell-enriched proteins are made.

To track the change from a neuron to an astrocyte, the team looked at the RNA profile, shape, and physiology of the new cell. "For now, these are astrocyte-like cells," says Eberwine. "While the cells don't look like neurons any longer, they don't have the mature star-like astrocyte shape, but rather look like immature astrocytes. The new cell expresses astrocyte proteins and has an astrocyte-like physiology. We start to see changes within a week and they are stable over the life of the primary cell culture."

These studies were enabled through the collaboration of a number of investigators spanning multiple disciplines including David Meaney from Bioengineering, Vijay Kumar and David Cappelleri from Mechanical Engineering and Junhyong Kim and Miler Lee from Biology. The additional Pharmacology Department contributors include Chia-wen

Wu, Fanyi Zeng, Jeanine Jochems, Tae Kyung Kim, Tiina Peritz, Peter Buckley and Minsun Kim.

Future studies are envisioned towards the generation of distinct cell types and dissection of the core set of RNAs responsible for the generation of particular cellular phenotypes.

Source: University of Pennsylvania School of Medicine (news : web)