Eric Delpire had never given much thought to the intricate workings of the inner ear. But he learned fast when his research group made the surprising discovery that mice lacking a particular ion co-transporter are deaf.

Delpire, assistant professor of Anesthesiology and Molecular Physiology and Biophysics, generated the so-called ‘knockout’ mice to probe the biological roles of the co-transporter—a protein that moves sodium, potassium, and chloride ions across the cell membrane—in the central nervous system.

“We were hoping for significant effects related to brain development and function,” Delpire said, “but you have to go with what you find.”

But basic science research doesn’t always travel along the planned route and detours can lead to exciting discoveries. Delpire’s unexpected findings shed light on the complexities of hearing and may point to the culprit in some form of hereditary human deafness, opening the way for potential new treatments.

Do You Hear What I Hear?

We live on a noisy planet. Even a quiet place like a library is filled with the tapping of keyboards, the hum of the heating system, and the buzz of whispered conversations. All of these sounds must initiate a complex chain of events before we ‘hear’ them.

Hearing begins when sound waves, funneled in by the outer ear, bang against the eardrum. The vibrating eardrum transfers the waves through the small bones of the middle ear to the inner ear. Buried within the temporal bone, this intricate structure includes the semicircular canals, which house the equilibrium and balance machinery, and the cochlea, the snail-shaped home of the organ of hearing.

Inside the cochlea, about half the size of a split pea, a membranous tube follows the winding turns. The tube floats in one fluid, and it is filled with a special fluid called endolymph. Waves in the fluids set up shearing forces that pull on tiny hair cells, which in turn stimulate nearby neurons to send signals to the hearing centers. The endolymph and its unusual chemical composition—high potassium concentration—is key to hearing and to one thing that went wrong in Delpire’s knockout mice.

Wobbly and Deaf

When the mice that are missing a specific ion co-transporter are born, they appear normal. But within about 10 days they have a tendency to fall down, then have trouble getting back up, Delpire said. They also circle in the cage and bob their heads.
"All of the movement oddities suggested an inner ear problem," he said, "so we looked at the structure of the inner ear and tested them for hearing."

The knockout mice are deaf, and a look at the inner ear suggests why. They do not have the organized membrane compartments that characterize the inner ear.

"The endolymph is not being formed, and there is a collapse of the membranes and the cochlear duct cavity," Delpire said.

"There is also disruption of the organ of Corti—basically of the hair cells themselves."

The absence of one type of protein—the ion co-transporter—translates into dramatic changes in the structure of the inner ear.

Pass the Salt

The co-transporter protein that these knockout mice are missing is normally present on the surface of neurons and secretory epithelial cells, like those that line the lung, salivary gland, and intestines. In the inner ear, the co-transporter resides on the blood side of a layer of epithelial cells called the stria vascularis. Its likely role there, Delpire said, is to move potassium from the blood (along with sodium and chloride) into the cells. On the other side of the cells, potassium channels move the potassium into the endolymph.

"The knockout of those potassium channels also results in deafness," Delpire said. "So whichever you knock out—the molecules that move potassium across the blood side or the endolymph side of the stria vascularis cells—you get the same phenotype."

Even though Delpire wasn’t looking for hearing problems, his findings make sense. And they provide a molecular explanation for the observed ototoxic side effects of a group of drugs called loop diuretics, which includes furosemide (LASIX) and bumetanide (BUMEX).

"LASIX is probably one of the most used drugs in clinical medicine," Delpire said. "It’s used to reduce volume in a variety of conditions including hypertension and edematous states such as congestive heart failure."

Loop diuretics exert their pharmacological effect by inhibiting the activity of a kidney ion co-transporter, a sister molecule to the co-transporter that Delpire knocked out in mice. Since these drugs also inhibit the co-transporter found in the inner ear, it is now clear from Delpire’s research why one of their toxic side effects is hearing loss.
Deafness Genes

Mutation of the co-transporter in human beings might be involved in genetic hearing loss. More than 28 million Americans have hearing impairments that result from genetic or environmental factors, or a combination of the two. Although the proportion of cases with a genetic basis is not known, the list of genes linked to hereditary hearing loss is growing.

One of these genes is for the potassium channel that works in concert with the co-transporter to secrete endolymph. So a linkage between deafness and a faulty co-transporter is logical.

“There is a human deafness linked to the chromosomal region where the co-transporter gene is,” Delpire said. “This disorder was recently mapped to a different protein in the region, but that doesn’t rule out the co-transporter being involved in another form of hearing loss disorder.”

Even if co-transporter gene defects are not responsible for any forms of human deafness, Delpire’s findings offer insight to the complex biology of hearing.

“Better understanding of the physiology of hearing will potentially lead to new treatments for hearing disorders,” Delpire said.

Current treatments for hearing loss center on amplification of sound (using hearing aids) or implants that stimulate the cochlear nerve or the cochlear nucleus of the brain. New treatments might take advantage of molecular insights offered by studies like Delpire’s to introduce replacement genes that correct defective processes.

Delpire will leave further study of hearing defects in the knockout mice to other researchers while his group probes the impact of the knockout on different physiological processes. In addition, his team is currently knocking out other members of the family of ion co-transporters that are linked to diseases of the central nervous system.

Perhaps this time around, the findings will line up with the expectations. 

What’s up with the Human Genome Project?

by Leigh MacMillan

Coming this summer to a web-site near you: a “first draft” sequence of the human genome.

That is the goal of the Human Genome Project, the worldwide public consortium of academic centers that is working to decode the three billion chemical units that make up human DNA.

The Human Genome Project, funded primarily by the National Institutes of Health and the Wellcome Trust of London, began in 1990 as a 15-year project. Advances in technology and a race initiated in 1998 by the private company Celera Genomics hastened the pace of the public effort.

Regardless of which team reaches the holy grail of biology first, the unraveled genetic blueprint for a human being promises to usher in a new era of molecular medicine. Buried in the genome are each of the 60,000 to 100,000 genes—the instructions for making proteins that determine how we look, how well we fight infection, how we behave, and how we are affected by disease.

“Within five years, I believe there are going to be great strides in bringing the power of genetics and the wealth of new information from the Human Genome Project directly to the benefit of patients,” said Dr. Alfred L. George, Jr., Grant W. Liddle Professor of Medicine and director of the division of Genetic Medicine.

Those strides will likely include improved diagnosis of disease, earlier detection of genetic susceptibility to disease, rational drug design, new DNA-based drugs (gene therapy), and individually customized drug treatments.