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Lost in translation

A mysterious disease that causes children's brains to melt away is caused by errors in RNA translation. But biologists are realizing that this horrifying condition could shed light on more common problems. Claire Ainsworth reports.

In a consulting room in Santa Barbara, California, Michael Salsbury found himself comforting his weeping doctor. It was July 2000, and he and his wife Gabriella were worried about their baby daughter, Gracie-Sophia, who was having seizures and losing her ability to stay alert and feed. The doctor had examined her and confirmed the Salsburys' worst fears: Gracie's brain was vanishing, and he couldn't tell them why.

The doctor's tears were born of frustration and despair. Gracie was not the first of the Salsburys' children to succumb to this mysterious illness. Two other girls had perished before their first birthdays from a strange, incurable disease that robbed them of mobility and caused parts of their brains to melt away. None of their doctors had seen anything like it, and for years, the Salsburys were unable to get a full diagnosis.

Now, thanks in part to their fortitude in donating their daughters' organs for research, the Salsburys have an answer. And it seems that the inherited disease that claimed Gracie and her sisters has implications far beyond the realm of rare neurological disorders. The faulty genes involved help control a vital aspect of our biology: the translation of RNA into protein. Biologists are realizing that documenting how this control can break down should help us to understand more about common diseases such as cancer and Alzheimer's — and may one day provide new ways to treat them.

For the Salsburys, finally getting a proper diagnosis for their lost children has helped them to heal. "We went as low as any family can probably get, three times," says Michael. "Now we finally know what took the lives of our daughters."

The Salsburys' troubles began in 1993, after the birth of their second child, Stephanie. At three months, she began to have subtle epileptic seizures. Her condition soon deteriorated, while doctors ran every possible test and tried different combinations of drugs. "She was getting everything around the clock," says Gabriella, who works as an intensive-care nurse for newborn babies. "It didn't help." Stephanie died aged eight months.

No answers

Gabriella gave birth to another daughter, Jennifer, in 1995. At first, Jennifer was alert and lively. But at three months, she began to develop the same ominous symptoms. This time, knowing that nothing could be done, the Salsburys refused most medical tests. "We had been through the wringer with Stephanie," says Michael. "We wanted to keep our baby at home and comfortable, and love her for as long as we could."

Shortly afterwards, Michael, a banker, was transferred to Switzerland, where they had a healthy daughter, and then Gracie-Sophia was born. This time, Gabriella sensed that something was wrong before the symptoms began. Swiss doctors dismissed her fears, and the family planned a baptismal celebration back home in Santa Barbara. But as the plane touched down, Gabriella saw that Gracie was having subtle seizures. "And so it became a funeral again," says Michael.

After Gracie died, the Salsburys abandoned plans for a bigger family. Believing their daughters were the only known cases of the illness, they donated the girls' organs to research and hoped that someone would find an answer.

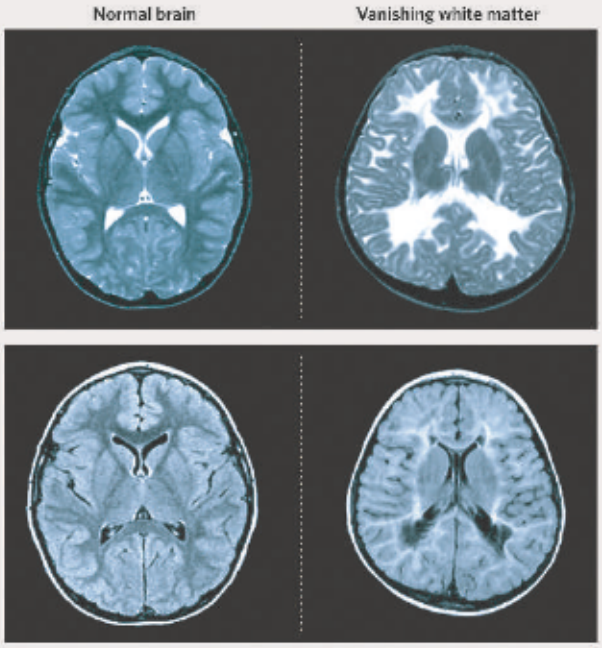
But the Salsburys were not alone. From the early 1990s, several doctors around the world had been reporting bewildering cases of ailing or comatose children who had unusual brain scans[1]. Raphael Schiffmann, a child neurologist at the National Institute of Neurological Disorders and Stroke in Bethesda, Maryland, was the first to describe and put a name to the disorder: CACH, for childhood ataxia with central hypomyelination[2].

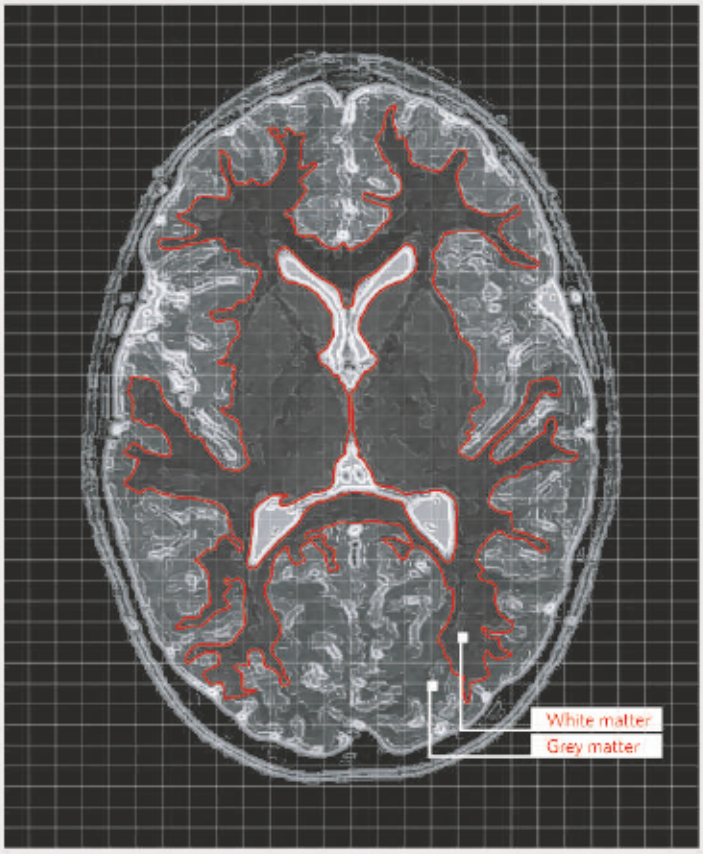
At the Free University Medical Center in Amsterdam, neurologist Marjo van der Knaap was also seeing similar cases. The children, normally less than five years old, would initially seem normal. Then, following a bang on the head, fever or even a fright, they would begin to lose muscle coordination. In some cases, the subsequent decline came in episodes and took years. But in others, it was frighteningly rapid. "On, say, Thursday, a child is unwell — starting to have a fever or a little bit of a cold," explains van der Knaap. "Then on Friday, the child becomes irritable and limp, lies on the couch and doesn't do much. By Sunday, the child is in a coma."

Magnetic resonance imaging (MRI) scans showed that the white matter in the patients' brains was abnormal. White matter acts as the main communication link for sending commands from the brain's grey matter to the rest of the body. It consists mostly of axons, the long extensions of nerve cells that form the links to different parts of the nervous system. These connections are encased in a substance called myelin, which acts like the plastic insulation around an electrical wire. With the loss

MYSTERIOUS DISAPPEARANCE

Two types of magnetic resonance image highlight the devastating effect of a genetic condition that destroys the brain's white matter (outlined in a normal brain, right). In the upper pair, the white matter (dark grey) in the normal brain is largely replaced by cerebrospinal fluid (white) in the diseased organ. Similarly, in the second images white matter (grey) is replaced by fluid (dark grey).





M. S. VAN DER KNAAP/VU UNIV. MED. CENTER

of white matter, instructions to the body would fail to get through. This fits with the symptoms: patients' cognitive function is largely unaffected, but their motor skills can be destroyed.

The MRI scans were clear: the white matter was melting away and being replaced with cerebrospinal fluid, the watery liquid that bathes the brain and spinal cord[3]. This led van der Knaap to name the condition 'vanishing white matter' or VWM disease — now known as VWM/CACH. Schiffmann has a theory about how the disease progresses. "The first thing that goes wrong is the insulation of the wire," he says. "Then at some point, the wires themselves begin to tear." In the end, the white matter disappears completely.

By the late 1990s, the hunt for the genetic roots of VWM/CACH was on. We now know that it can be caused by mutations in five different genes, each on a different chromosome[4,5]. Normally, this scattering would have made it all but impossible to track the genes down. But a quirk of Dutch genealogy came to the rescue.

Local knowledge

Van der Knaap's team was collecting samples and data from families in the Netherlands, where 1 in 40,000 people are affected. Most of the families involved came from a rural region in the east of the country where the population has tended not to migrate or intermarry with people from elsewhere (see right). This gave a relatively homogeneous genetic background against which it was easier to detect disease-causing genes. And the researchers had a further stroke of luck when they found that several affected families shared a single common ancestor. Within this 'superfamily', the researchers were able to scan the genomes of affected and unaffected individuals to identify a gene involved in VWM/CACH.

The culprit, the gene for part of a protein called eIF2B, came as a surprise[6]. Far from being specific to the brain, this protein is present in almost every cell in the body. It plays a key role in stitching together amino acids to make proteins according to the recipe laid down in RNA, a process known as translation. The eIF2B protein is made of five fragments or subunits, coded for by the five different genes. It unites with ten other proteins to form a 'machine' that helps control the translation process. And when van der Knaap and her colleagues looked at other VWM/CACH patients, they found that they had mutations in the genes for these other subunits[4,5].

The discovery that faults in the control of translation can cause such a dramatic disease has thrown a fresh spotlight on the importance of the process in health and disease[7]. "It's triggered wider excitement," says Christopher Proud, now at the University of British Columbia in Vancouver, Canada, who has worked on eIF2B and related translation factors for some 15 years.

When thinking about the control of the production line from gene to protein, most

HUNTING THE GENE

In the eastern part of the Netherlands, people tend to live in small villages and do not often relocate. This allowed researchers to track down the faulty gene. They identified 252 ancestors clustered in the area of Zwolle. The numbers indicate the percentage of ancestors living in each village.

interest has focused on the earlier process of transcription. In this step, DNA is 'read' to produce messenger RNA, or mRNA. Various proteins known as transcription factors control this process, and they have come under intense scrutiny from cell biologists and researchers interested in diseases such as cancer.

The mRNA gets carefully edited and is then sent to complex molecular 'robots' called ribosomes, which read it and slot the amino acids together in the order detailed in the code. Together with its partners, eIF2B helps to position the ribosome at the correct starting point as it scans along the mRNA.

One intriguing aspect of VWM/CACH is that its severity can vary, depending on where the mutation lies in the eIF2B protein. Proud and his colleagues suggest that this is because different mutations have different effects on eIF2B's ability to function and interact with the other proteins that regulate its activity[8]. It seems that different mRNAs have different sensitivities to the level of eIF2B. This might help explain the puzzle of why the brain, and particularly the cells that make myelin, is most affected in VWM/CACH. If these cells rely especially heavily on eIF2B to make their key proteins, they will be more sensitive to drops in its level of activity.

Communication breakdown

The emerging understanding of VWM/CACH fits with a growing realization that the control of translation is more subtle than anyone thought. "It took a long time, but the wind is changing," says Arrigo De Benedetti, a molecular biologist who works on translation at Louisiana State University in Shreveport. "People are finally recognizing protein synthesis as a major player in disease in general, and in cancer in particular."

The suggestion that faulty translation could cause cancer has been around for about 15 years. In 1990, a team led by Nahum Sonenberg at McGill University in Montreal, Canada, showed that another translation factor, called eIF4E, can make cells become cancerous if present in abnormally high amounts[9]. In patients, high levels of this protein are also associated with resistance to anticancer therapy and with the disease's spread through the body. Meanwhile, other cancer researchers have noticed that some proteins are present at abnormal levels in cancer cells despite there being no obvious problem with the production of their mRNAs — hinting that translation, not transcription, is at fault.

Some mRNAs respond more to changes in the activity of eIF4E than others. Biologists now know that this is because each mRNA has a long, untranslated region at its start that affects how it responds to different translation factors, depending on its sequence or three-dimensional structure[10].

As if this weren't complicated enough, there is another kind of untranslated region that also affects how mRNAs respond to translation factors. These regions are long — up to a third of the length of the entire mRNA. They fold into elaborate structures, called internal ribosome entry sites, or IRESs, that together with control proteins summon ribosomes to read the mRNA[11]. Mutations in IRESs have been linked to a number of diseases. They have been found in cancer patients in genes that control cell division, for example.

Other translation-control mechanisms are probably waiting to be discovered. "There could be lots of other things out there," says Anne Willis, a molecular biologist at the University of Nottingham, UK, who works on IRESs and their role in cancer. "They've just not really been looked for."

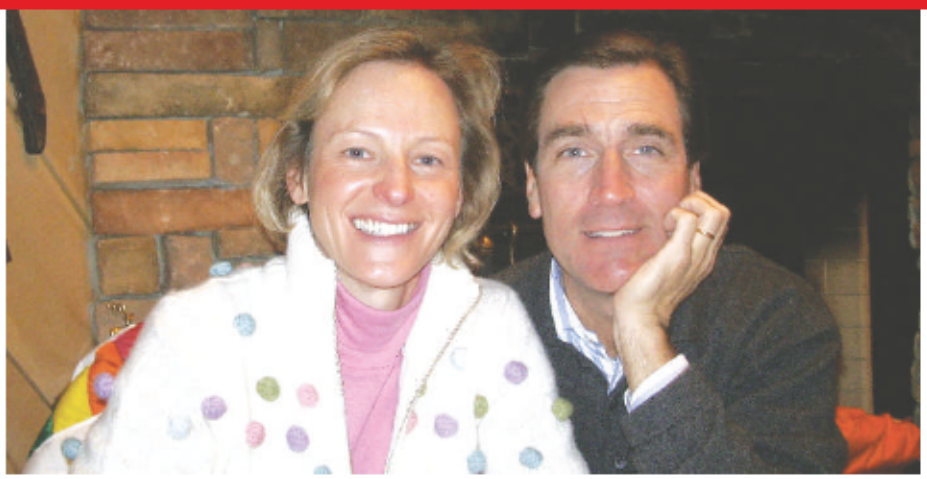
Unravelling the complexities of translational control may look daunting, but its very subtlety may be to clinicians' advantage. In theory, it should be possible to selectively target the production of particular proteins implicated in disease without causing wide-ranging side effects. "There are probably dozens, if not hundreds, of compounds that can inhibit steps in translation and have not been exploited yet to their full extent," says De Benedetti.

Targeted effect

In fact, one such drug is currently in clinical trials to treat cancer. Rapamycin, originally developed as an immunosuppressant, targets one of the systems that regulates translation[12]. The drug shuts down a protein called mTOR, which senses aspects of the cell's environment, such as the availability of nutrients, and allows the cell to grow and divide if all is well. One of the ways it does this is by controlling the activity of translation factors. In many cancers, normal mTOR signalling seems to break down.

It should also be possible to target specific translation factors directly. And in the long run, Schiffmann and others hope that it may be possible to help VWM/CACH patients by targeting eIF2B or the affected mRNAs. "The problem is that it is really too early to say how one would do that," says Proud. "We need to know a lot more about how the condition works."

Researchers working on VWM/CACH are now concentrating on developing mouse models of the disease and studying the biology of affected brain cells. They are also identifying other variants of VWM/CACH caused by different eIF2B mutations[13–15]. For example, van der Knaap has identified mutations that cause severe, early-onset disease that affects multiple organs and causes babies to die before birth or in the first few months of life[16]. It was while working on that project that she happened on organ samples and MRI data from a family that had lost three baby girls to an undiagnosed white-matter disease.



Gabriella and Michael Salsbury lost three of their baby daughters to a rare brain disease.

So it was in 2003 that the Salsburys got the phone call they had long waited for. It was van der Knaap, who had discovered eIF2B mutations in their daughters' brain tissue. As well as helping the Salsburys recover from their loss, the knowledge offers the family's surviving children the opportunity to be tested for the mutations, so they won't have to endure the same ordeal when they come to have families of their own.

The Salsburys have now set up a charity, the Three Little Angels Foundation, to raise money for research and to improve awareness of rare neurological diseases. They are passionate advocates of donating organs for research. "We see science and medicine as an evolving effort," says Michael. "Each single daughter gave science a new piece of knowledge." ■

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