GNAQ in Sturge-Weber Syndrome: what it means

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With the discovery of mutations in *GNAQ* as the cause of Sturge-Weber syndrome (SWS) and port-wine stains (PWS), we'd like to explain what we've done and what it might mean.

Background: what was known

SWS has a long history, having been described by William Sturge in the late 1800s and many others in the early 20th century. It was described based on its clinical symptoms, but the cause remained completely unknown. In the 1980s Rudolph Happle hypothesized that a set of skin disorders is caused by somatic mutations. He wrote (1987): "A genetic concept is advanced to explain the origin of several sporadic syndromes characterized by a mosaic distribution of skin defects. It is postulated that these disorders are due to the action of a lethal gene surviving by mosaicism." He suggested that a somatic mutation could cause the syndromes. What do the terms "somatic" and "mosaic" mean? Let's begin with some definitions.

What is a somatic variant? What is a chromosome? DNA? Gene?

Each of us has 23 pairs of chromosomes. They are numbered 1-22, X and Y; boys are XY and girls are XX. For the other chromosomes, including chromosome 9 that harbors the *GNAQ* gene, each of us inherits one copy from our mother and one copy from our father. A germline mutation is passed down from the father or mother to a child. A somatic muatation arises in development, even before birth.

What is a mutation? DNA is arranged in the famous double helix. It is composed of four bases (abbreviated A, C, G, T) that form pairs. Chromosome 9, an average sized chromosome, has 141,213,431 base pairs—that's 141 million base pairs! If you add up the base pairs of all the chromosomes you get the total: each of us has a genome consisting of about 3.2 billion base pairs. When we refer to a mutation, this means that one base pair changes. It may be a G changing to an A, for example. Most changes will have no effect on a person's health. Some changes cause diseases.

And what is a gene? A gene is a functional unit of DNA that typically can be turned into a protein. For example, the gene called *GNAQ* on chromosome 9 can make a protein called $G\alpha_q$ (pronounced G alpha q). Proteins carry out various functions in a cell.

There are thought to be 20,800 genes in each of our genomes. Chromosome 9 has just over 1,500 genes spread along its length. *GNAQ* is spread out over a region of 300,000 base pairs along chromosome 9; most other genes are more compact. The protein made by the *GNAQ* gene is average sized, consisting of a string of 359 amino acids.

We've defined a mutation as a change in DNA, and a somatic mutation as one that occurs during a person's life (during early development or anytime later in life). A somatic mosaic mutation is "mosaic" because it affects only parts of the body.

Testing the hypothesis 5 or 10 or 20 years ago

In the recent past—say in the 1980s, 1990s, and 2000s up to 2010—researchers had very few tools to look for the kind of somatic mutation that was expected to occur in SWS. In the case of cancer, some forms of somatic mutation were fairly easy to find. One could compare the DNA from a tumor sample to DNA from an unaffected part of the body and find large changes. (By large we mean visible under a specialized microscope, and affecting many millions of base pairs.) Finding very small mutations was not possible when we didn't know where to start looking.

Testing the hypothesis today: finding a needle in a million haystacks

A new approach to sequencing DNA, called "next-generation sequencing," burst onto the scene in 2005 and is allowing vast amounts of DNA to be sequenced at relatively low cost. The Human Genome Project, completed in 2003, succeeded in sequencing one human genome at a cost of about $1 billion. Today in 2013 it costs about $5,000 or so to sequence a complete human genome.

We began by studying just three people with SWS. (And here's a chance to say a huge THANK YOU to all who participate in research!) For each of the three people, we took small samples of regions of the body that were believed to be affected (e.g. a skin biopsy from a PWS region) or unaffected (e.g. a blood sample). In all there were six samples from three people. We sequenced these six entire genomes. (The human genome is about 3 billion base pairs but we sequenced each one many times over to make sure we were getting accurate sequences. From these 6 genomes we obtained about 700 billion bases of DNA sequence!)

Then a graduate student in Jonathan's lab, Joseph Baugher tried to find some DNA change in the affected but not the unaffected samples. He didn't find anything at first. Then another graduate student, Matt Shirley, noticed that a new software tool had just been developed. When he applied that tool he found just one good candidate: a mutation of one base pair occurring in a gene called *GNAQ*. Matt had found a needle in a haystack—or more like a needle in a million haystacks. Matt is expert in biology, and he is also enormously gifted at analyzing data on a computer.

The finding of a mutation in *GNAQ* was intriguing. The gene makes a protein, $G\alpha_q$, that has a key role in cell function including the regulation of blood vessels.

Confirming the finding

The three of us (DM, JP, AC) immediately pooled our efforts to see if this finding was true. In all we studied 90 samples using two very different techniques, and we kept coming up with the same answer: a single mutation in *GNAQ* occurred in almost all affected skin samples (but not often in unaffected

ones). We also obtained brain samples, and found the mutation in affected brain. A series of samples from individuals having no known disease, or having unrelated diseases, revealed no mutations in the gene. Furthermore, the mutation occurred in port-wine stain DNA—including individuals who had PWS without Sturge-Weber syndrome.

A model for what's happening, and a connection to cancer

We believe that a Sturge-Weber syndrome occurs when a somatic mutation in *GNAQ* occurs very early in development—as early as the first trimester of life. It's possible that port-wine stains (but not SWS) happen if the mutation arises a bit later in development. It may also be important which particular cell types harbor the mutation. We're working on that question now.

Surprisingly, a mutation in the same gene (*GNAQ*) at the same position can cause a form of melanoma that affects the eye (uveal melanoma). What's different? Uveal melanoma involves a somatic mutation in a different cell type (melanocytes) at a different time of life (e.g. adulthood). It matters when and where the mutation occurs.

Do mom and dad contribute to the occurrence of SWS? Not really. Lightning strikes in the form of an early somatic mutation. SWS is not inherited. Consistent with this, twins have been found in which one has SWS and the other does not (suggesting that the inherited DNA, which is the same in identical twins, does not contribute to SWS). And people with SWS who have children are not expected to pass SWS to their children—their germ cells are not affected.

How excited should we be? Keeping our feet on the ground.

Finding the gene is a starting point for figuring out how to solve SWS. For over a thousand diseases, finding the gene does not necessarily lead to a cure or even to improved treatments. So we'll be hopeful, and at the same time we'll keep our feet on the ground. In the case of SWS, our focus can now shift to *GNAQ* and figuring out how it is disturbed and what we can possibly do about it. Our research has a very specific direction to follow.

Knowing the gene we can develop an animal model, such as a mouse model of SWS. By doing this we will be able to understand the mechanism by which $G\alpha_q$ function is disrupted and what we might be able to do to correct it.

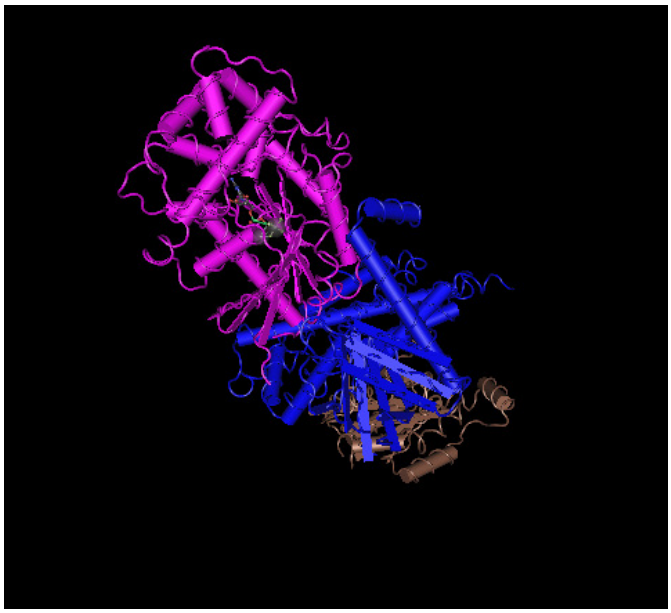
Knowing the gene we can also work with cells in a dish and study Gaq function. Led by Doug Marchuk's lab we have already begun doing this. In the paper we published today we introduced "normal" $G\alpha_q$ into cells, as well as several mutant forms of $G\alpha_q$. We identified changes in how cells process information.

What is GNAQ? What does Gaq look like?

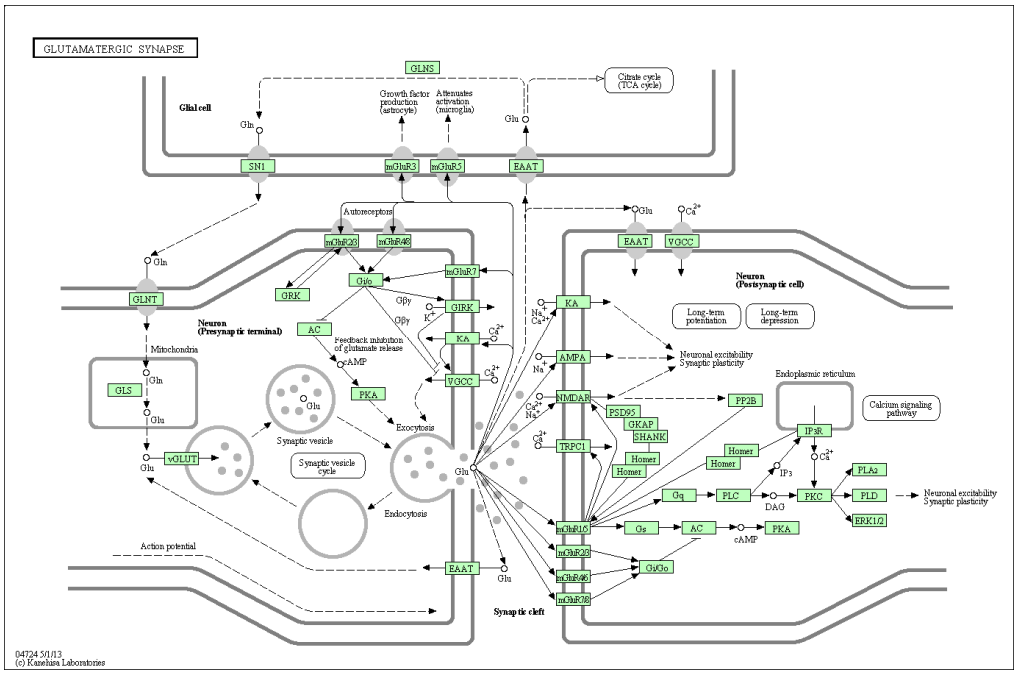
Here's one way to begin to see what is known about GNAQ and $G\alpha_q$. There is a website called the National Center for Biotechnology Information (NCBI) at http://ncbi.nlm.nih.gov. Go there, type GNAQ into the search box, and you will see some of the information we know about the gene, the protein, the pathways it participates in, and the literature.

Here's a description of $G\alpha_q$:
http://www.ncbi.nlm.nih.gov/protein/NP_002063.2

Here's what the protein looks like, touching a couple other proteins:



Here's a "circuit" diagram showing Gaq (abbreviated Gq) in two brain cells, from http://www.genome.jp/kegg-bin/show_pathway?hsa04724:



Can we fix $G\alpha_q$ pathways? We don't know yet. But that's the next problem we're working on.

What does this mean for my family?

For a person with SWS, it is technically possible to obtain a small skin sample and sequence it to confirm that there is a mutation in *GNAQ*. For most people this is probably not necessary.

For a small number of people with SWS, there could be a mutation in another region of *GNAQ*, or a different "flavor" of mutation, or perhaps a mutation in some other gene. As we do more research we will want to find the full spectrum of genetic variation.

In conclusion…

Thanks for reading about this discovery. Let us know if you have questions and we'll do our best to answer them. And thanks to everyone in the SWS community—starting with Karen Ball—for your support as we keep trying to make progress toward solving SWS.