Harrison Gabel PhD’s talk TBRS and

*Gene Therapy Considerations in TBRS*

2025-April

As an opening statement, Dr Harrison Gabel mentioned that he saw a recent NY Times article on gene therapy. He acknowledged that gene therapy is one type of treatment that could be applied to TBRS in the future, and therefore the TBRS community felt it would be valuable to discuss gene therapy, the NY Times story and how gene therapy might be applied to TBRS in the future.

Harrison introduced himself as a PreClinical Researcher. He studies neurodevelopmental disorders of epigenetic causes. He reminded us that he is NOT a physician, despite the title of “Doctor” in front of his name, and therefore he can tell us where the research is going but he cannot provide advice on health management for individuals with TBRS.

Harrison has studied DNMT3A for 10 years. At first, he studied the neurocognitive aspects of DNMT3A, but now he is more generalized. He is working more with clinicians these days and serves as an adviser to the Community. He has experience with Rett syndrome, which is caused by disruption of a gene that works downstream from the TBRS gene, DNMT3A. This talk was occasioned by the excitement in the TBRS Community which was generated by a newspaper article about an infant with a congenital, life-threatening mutation who had been much improved with gene therapy.

First, Harrison considered, “What is Gene Therapy?” He first discussed ‘what is a gene?’, then went on to discuss this particular therapy which was performed to repair, in situ, the damaged gene in this baby. For those who do not yet know all the genetics-related vocabulary, he recommended “www/genome.gov/genetics-glossary”.

The baby known as KJ, recently had a damaged gene repaired. KJ had PCPS1 deficiency. In his case, there are enzymes in his liver that don’t work correctly in processing proteins, so one of the break-down products of proteins, ammonia, builds up in the blood. Ammonia damages the brain, and too much ammonia is eventually fatal. Unlike in TBRS, both copies of this child’s CPS1 gene were defective. He needed to be treated as early as possible. He was treated at Children’s Hospital of Pennsylvania (CHOP), arguably one of the foremost pediatric hospitals in the world. A massive undertaking brought scientists in a number of fields together for a crash, first in human, proof-of-concept gene therapy. In the meanwhile, CHOP used every known method to reduce KJ’s blood ammonia level, with some mild success. Within a few months, there was a bespoke gene therapy to fix his liver! (This therapy was delivered ONLY to his liver, but since the disease is manifest primarily in the liver, that theoretically should be enough. This is different from TBRS where the manifestations are throughout the body and brain.)

Simplistically re-stated, the group created a bespoke CRISPER gene – editing package, inserted into lipid nano-particles (little fat globules), which were picked up by the liver. Once inside the cells, the packets opened and the CRISPER tool cut the deficient gene out of the host cell and replaced it with the fixed, bespoke gene. The liver, naturally enough, digested many of the lipid packets, but enough of the lipid packets were opened by liver cells and got their genes fixed in order to reduce the ammonia in KJ’s blood. The newly inserted genes began to code for the protein which turns harmful ammonia into blood urea nitrogen (BUN). Shortly after the therapy was administered, the child had dramatically lowered ammonia levels. His other drug doses were reduced, and he started to grow.

Dr. Gabel offered caveats. This is only one child, but the case showed that it is possible to do this kind of gene editing in vivo. The speed in which it happened was amazing. Clearly, before this therapy can be in common use, the therapy needs to be made more simply and cheaply. This proof of concept was a herculean effort and cost many tens of thousands of dollars, as well as required a multi-specialty collaboration.

Dr. Gabel then went on to ask, “Does this change anything for the TBRS community? Yes and no.” Yes, because as with any successful experiment, once demonstrated that a therapy is possible, it powers more research, which will help. No, because there are still many hurdles to determine if gene therapy can be safe and effective to replace or repair the damaged DNMT3A in individuals with TBRS. In some disorders, people affected by single gene damage are too old to benefit from getting their damaged gene fixed, even were it possible. In general, the earlier such a therapy can be administered, the better.

He went on to add, “So much more needs to be understood, and the risk is as yet unknown.”

Because TBRS is caused by a large array of different mutations, it would be more feasible in TBRS to add in a complete new DNMT3A gene. TBRS, the condition, is basically caused by having one copy of the DNMT3A gene that does not do its job correctly (we call that haplo-insufficiency), and the remaining good copy of the gene doesn’t make enough of the dnmt3a protein. (A conceptus with 2 damaged DNMT3A genes doesn’t survive.)

What is DNMT3A? Every gene codes for a protein, each of which has its own job to do. DNMT3A, the gene, codes for a protein, dnmt3a, which puts a methyl group onto other genes. The methylation then tells the methylated gene to start their own jobs. It is deeply involved in development.

Gene variants, or mutations, cause the protein to be malformed in some way, causing “a broken protein” if you will. This causes a paucity of functional dnmt3a proteins. Since in TBRS only one gene is broken, there is still enough of the protein to permit the baby to survive, but not quite enough to develop quite normally. Were an entire new gene to be loaded into the cells of an affected person, the extra copy of the DNMT3A gene should help add to the amount of the protein in the cell, and perhaps, thereby augmenting the amount of functional DNMT3A protein. However, as a caution, it should be mentioned that mutations of DNMT3A which cause “gain of function” – too much protein – also cause a bad syndrome.

Adding an extra DNMT3A gene would not require CRISPER, which corrects the defective gene, and is defective-gene specific. Adding an extra, complete gene is already possible using conventional viral gene therapy.

KJ’s therapy was different from adding a new gene; using CRISPER they actually REPAIRED the damaged gene. To fix one gene in place is wonderful!. But not as practical for TBRS.

The viral gene therapy option has the potential to insert TOO MUCH of the gene and therefore its product. Too much DNMT3A may contribute to a whole different and even worse syndrome, Heyn Sproul Jackson syndrome, which is sort of a reverse of TBRS. (HSJ children are small, have small heads but also suffer intellectual disability.)

Timing is, of course, important. We used to think that for a developmental disorder, fixing a gene after development has already taken place would be futile because development had already taken place. These days we think also in terms of the ongoing function of the gene, even after primary development has taken place. We don’t yet know how exact the timing needs to be in TBRS: what is the right time, the right dose, and where should we deliver the repaired genes?

We may have to make choices about where the delivery is most important. Finally, we have to make it safe. One proof of concept for a different gene therapy is not enough to make gene therapy common.

What about timing:? Does putting something back in an already born or developed person really help anything? In Rett syndrome, (caused by mutations of the MeCP2 gene) there is apparently normal development up to 6-12 months followed by loss of controlled hand movement, worsening language and communication, sleep and bowel dysfunction. The affected person has seizures, becomes wheelchair bound and often nonverbal.

Rett Syndrome is carried on the X chromosome. About 20 years ago, Dr Gabel’s group studied Rett syndrome and learned that on an anatomic level, the brain tissue tissues were still there; they just didn’t work well. Since experimenting on humans has ethical issues, they made a mouse model. The mouse is severely affected and dies at about 8 months of age. (Much of this work was done by Adrian Bird some years ago.) Dr Bird wondered if putting the gene back would make the mouse better? He developed a mouse where he could turn on and off the affected gene. (This is called a ‘Knock-Out’ mouse model.) He turned the gene back on, and LO! the mouse acted much better, actually seemed healthy. A striking finding! This “corrected” mouse had improved dramatically! Not only that, the mouse with the “corrected” gene improved even if the gene was turned back on after physiologic maturity.

But, Dr. Gabel wondered, “How far can this be extrapolated?”

We are now exploring what can be done in TBRS. We have several mouse models of TBRS, each with different mutations. Most of those mouse models are the kind where the gene can be turned on and off (so-called Knock-Out models). We are turning the gene back on at different times in the mouse’s development to see how much of the syndrome can improved at various ages. Stay Tuned!

Right after birth, some of the molecular deficits are corrected. Other investigators are working on other schedules; the answers aren’t all in yet.

Dr. Tim Ley studies DNMT3A in cancer. Genes don’t just mutate before birth or in germ cells. They can also mutate in an already grown-up body. Those are called “Somatic Mutations”. Bone marrow keeps dividing and maturing throughout one’s life and therefore tends to pick up a lot of somatic mutations. Leukemias and other blood cancers are often caused by somatic mutations and often involve mutations in DNMT3A. (This is why early on, we worried that people with TBRS would be especially prone to leukemia. So far, it appears there have been only a few with leukemia.)

Dr. Ley wondered what would happen if one put the DNMT3A function back into the blood when there is a somatic mutation of DNMT3A leading to blood cancer (a hematologic malignancy)? He transplanted blood progenitor cells with normal DNMT3A function back into the marrow of DNMT3A deficient mice. That corrected a number of the molecular damages in the blood. This suggests that at least *some* benefit may arise even when a correction is applied fairly late in life. To date, the efficacy and the dangers of doing so have not fully been understood.

In other words, timing will certainly be important, but it is likely that therapy delivered “late” (still to be defined in humans) may still offer some benefit.

Next, let’s consider the “dose” of the therapy. If TBRS is caused by having only one fully functioning DNMT3A gene, then the syndrome IS “dose-sensitive”. This is common in other neurodevelopmental disorders. And “natural experiments” – when people are born with extra copies of some of these genes, a whole different syndrome manifests. These people have Too Much of the dnmt3a protein.

Too much dnmt3a protein causes problems in mice. Too much of a good thing can be a bad thing. So what else can one do? Dr. Ley wondered if it were possible to “turn up” the output of the one good copy of the DNMT3A gene that is still in the cell of a person with TBRS. Could THAT be possible without overshooting? Stay tuned for the next exciting episode in genetic research!

How might “fixing “TBRS even be possible? There are several potential types of therapies:

* One could deliver a new copy or copies of the gene into the cell using conventional gene therapy. Here we risk overshooting.
* Or perhaps to “fix” the mutation in the damaged gene. This would obviate the overshoot problem because in each cell there would be a maximum of two functioning DNMT3A genes. However, with current technology, we cannot “fix” all the cells in a body. Additionally, one would have to create a different “fix” for each different mutation of the DNMT3A gene. This is so far not yet feasible. It is not yet fast, safe and scalable.
* Potentially, a different approach would be that one could augment the amount of the RNA intermediate between the gene itself and its protein product. Functionally, this would be like turning up the volume on the DNMT3A gene that remains: augmenting its output.

Finally, a different area of scientific work is learning how to get these therapies into cells. KJ’s therapy used lipids that worked well to get his therapy into the liver, because liver cells naturally take up fat particles, but lipid nanoparticles don’t work as well in the brain. There are viruses, though, that are good at targeting the brain. We might be able to modify one of these viruses so it only delivers the therapy and does not deliver its own genome, so it doesn’t cause disease. In other words, we could potentially use one or another of these brain-targeting viruses to deliver the “fix”.

Dr. Gabel added, “It is good that there are many scientists working on different aspects of gene therapy delivery; as soon as the therapies reach maturity, we will be studying which can most benefit TBRS people.”

There is much going on in Rett Syndrome also. If anything looks promising, we will look at it to see if it could benefit people with TBRS.

Safety is paramount. Any kind of gene therapy risks changing another part of the genome and causing problems such as overshoot, cancer, or an immune response. Furthermore, a gene therapy can only be delivered ONCE; thereafter one has a dangerous, severe immune response to that viral vector. So it has to be done right the first time.

Clearly, therefore, before we start using gene therapy, we need to be sure that the benefit outweighs the risk. Currently, companies are focusing on areas where the untreated disease is *invariably fatal*. ANY therapy with potential for mitigating the disease has less downside than that disease itself. Another place where much research is being done is in eye disease, because within the eye, one can control where the virus goes and limit the risk to the rest of the body. TBRS is not in these spaces. Gene therapy has not yet advanced to where the safety outweighs the risks.

There are several hurdles we have to leap before we start administering gene therapy.

* Timing: when do you need to correct the gene for therapy for it to be effective?
* Dosing: is the gene “dose sensitive” – if we deliver too much, what happens?
* Approach: how can we replace gene function in a way that meets timing and dosing constraints?

Dr. Gabel said, “We, in our labs, are testing the latest therapies in our mouse models. This is one way to “de-risk” the therapy which is important. Once it is shown that in an animal model that the risk can be minimized, pharmaceutical companies will be more willing to develop therapies that will actually help people with specific genetic diseases such as TBRS. We are not yet there and won’t be there in the next two or three years. Yet, the research is happening very fast. “Stay tuned!”

Questions from the audience:

Is it yet possible to learn which parent the bad gene came from? Yes, but in TBRS that isn’t usually very relevant. Although there are some genes which behave differently depending on which parent it came from, so far, we haven’t found that to be the case in TBRS. Additionally, except in a few familial cases, the gene damage that happened, happened in the germ cell: either or gamete – either an egg or a sperm cell. In general, most gene mutations come from the father because of the way sperm are made. You could figure it out if you had all the money in the world, but we don’t have a reason to focus on that aspect.

Dr Laura Lavery asked about the motivation to try the therapy. Said, “My lab is looking at the RNA aspect. Do your data show that early intervention is particularly important? What are your thoughts on earlier intervention?”

Dr. Gabel answered: “The biology of the DNMT3A gene certainly shows that it does a lot of its job on the brain right after birth. That would suggest that early therapy would be most effective. But then, is later still worthwhile? We still don’t have an aged population to study. We may learn that there are very late problems that could still be corrected in the older age group. Still there is almost no scenario where earlier is not better.”

He continued, “If we can show that a given individual isn’t in severe danger now, but if not treated early, will be in severe danger later (the “N of one” experimental paradigm) it might lower the barriers to the FDA approving such therapies sooner.”

“Furthermore, if even with mouse studies, if we can show that the earlier the therapy, the more effective it is, Insurance Companies will be forced to do the testing earlier. They may even be more willing to approve that therapy because it will lower their costs in the long run. That, of course, presupposes that Insurance companies think in terms of in the long run.”

Shannon wondered about the cost of such therapy. The good news is that some insurance companies are paying for gene therapy if something expensive now prevents LOTS of expenses later on.

Marcal Pastor-Anglada, Professor and father of a 33-year-old affected daughter, wondered “If later therapy might be too late for the brain but might still be efficacious for the blood?”

Dr. Vicken Totten added, “or for the bones?”

Dr. Gabel replied that theoretically, it is possible that later therapy that would not improve brain function, might still be worth pursuing to treat the blood, the immune system, reduce cancer chances, or even bone therapy. Whole body gene therapy is not currently possible.

To be feasible in clinical practice, we must be able to show improvement in clinically testable endpoints.

Dr. Shaimaa Helal, a resident physician and researcher, asked what if there were a subgroup of variants which did not just produce loss of function, but acted as negative dominants, what then? Like R882?

Dr. Gabel replied that Dr. Tim Ley has shown that adding DNMT3A back can still fix things even though the loss of DNMT3A in those mutations acts as a dominant negative. There is still functional DNMT3A protein around, perhaps at 50% to 75% loss of function. There is actually some evidence to suggest that even though the R882 mutation acts as a dominant negative, one can overpower its effect. In the case of the R882 variant, which is the most common variant, and given its special biology, a CRISPER targeted approach type of gene repair might be the preferred technology. To keep our work generalizable, Dr. Gabel added, “we work with mice both with a loss of function variant as well as one with a negative dominant effect.”

2 more questioners wanted to know if the KJ news might lead to more funding? And if the current barriers were more on the funding aspect or on the lack of specific research aspect?

Dr. Gabel answered that of course funding was always critical and pointed out that the NY Times article spent half their column space discussing the research which had lead up to this capability that treated KJ, making the case that basic science research is critical to moving towards treating humans.

Dr. Gabel recommended following the news about Angelman Syndrome and Rett Syndrome to learn what treatments might be coming down the pike. He mentioned a company that is fixing RNAs in place, perhaps called Taysha and Neurogene. Another company, called Ultragenics, is looking at delivering antisense nucleotides to cells.

Dr. Rosanne Wecksberg asked if in multisystem disorders like TBRS, one might get different impacts in different tissues? That’s something people need to look at. She wanted to layer this issue on top of everything Dr. Gabel said. She went on, “Biomarker development is critical for outcome measures. It has been shown that in clinical trials, there is a much better outcome of the entire trial if you have a biomarker that actually predicts what your clinical outcomes will be. But even more importantly, we are finding that different variants have different effects. There are a small number of other variants that fall into the milder category. It might be worthwhile talking more about the milder cases?

A biomarker is a measurable item that will show what the disease state actually is. Think about blood sugar in diabetes, or PSA in prostate cancer. You can monitor treatment to show how well or badly the disease is being treated, even though the disease may affect any number of organ systems.

Harrison Gabel praised the work that Dr. Weksberg had done on methylation in the blood of TBRS persons. This tool makes a very quantitative biomarker and would let us see if our therapy is doing its job. To further this tool, we need to know if changes in the blood also take place in other organs. Rett Syndrome doesn’t have a good biomarker, so researchers are using Cognitive Testing, which is both cumbersome and not as precise as Dr. Weksberg’s molecular biomarker would be.

Community members may be familiar with the Clinical Trial Readiness campaign that Jill Kiernan and Kit Church have been mapping out. Good biomarkers are an important part of clinical trial readiness.

Dr. Weksberg pointed out that Dr. Shaimaa Helal has been working on blood methylation levels and found that the biomarker correlates well with cognitive function, at least in the mouse equivalent of R882 TBRS.

Another listener queried how much international research information exchange was going on?

Jill answered that we, the TBRS Community, definitely have an international presence. We have come so very far in only 11 years. There are researchers from all over the world who are collaborating. As much as possible we are pulling in researchers in Great Britain and in Spain. The health care systems are so different that it makes it clinical information harder to exchange, but we still can share the biochemical data.

Cindy wondered who would determine which patients would be eligible for the treatment?

Dr. Gabel replied that it might depend on the genetic variant, the patient’s condition, and a number of considerations that we don’t even know about yet. Geography may also play a part. In the early days, clinical trials are very much a two-way street between researchers and caregivers.

Our TBRS Community is one of the more collaborative research communities that Dr Harrison Gabel knows about. He wanted to emphasize that we are keen to collaborate with researchers from any country.