Kunkel Laboratory Newsletter

Children's Hospital, Boston & Harvard Medical School
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microRNA May Provide New Therapeutic Targets in Muscular Dystrophy

By Iris Eisenberg-Loebl, Ph.D.

MicroRNAs (miRNAs) are a recently identified class of very small molecules that can change gene activity. The miRNAs bind to messenger RNA (mRNA) and block or inactivate the message (mRNA) that would otherwise lead to the production of proteins. It is believed that miRNA's can interfere in many basic cellular processes, including cell death, cell growth, tissue development and the immune response. When miRNAs expression is increased, they are able to interfere with messenger RNA expression acting as negative regulators, therefore, the more miRNA expression the less gene expression. Recently, studies have shown that miRNAs play important roles in healthy muscle cells; we surveyed the activity of miRNAs in several neuromuscular disorders.

We have studied the regulatory action of over 600 miRNAs that were found to associate with several neuromuscular disorders. We were able to show this association using a new type of molecular profiling, RNA expression chip technology. In collaboration with a multinational research team, we profiled 88 muscle samples taken from patients with DMD/BMD, FSHMD, LGMD 2A & 2B , Miyoshi myopathy (MM), nemaline myopathy (NM), polymyositis (PM), dermatomyosytis (DM), and inclusion body myositis (IBM). We found 185 miRNAs that had either increased or reduced activities in the diseased muscle when compared with healthy muscle. We believe that the changes in the miRNA's may provide clues to an underlying regulatory pathway in neuromuscular disease. A subgroup of 18 dysregulated miRNA molecules allowed us to accurately tell disease-affected muscle tissue from normal muscle tissue and to distinguish among the various muscle diseases.

Each of the muscle diseases studied proved to have a unique miRNA signature due presumably as a result of the underlying genetic defect for each disease. . Therefore, the miRNAs identified in this study could eventually provide targets for new drugs that mitigate many types of neuromuscular disease. Drugs that can enhance or reduce miRNA activity are still years away, but developing such drugs is a burgeoning area of research. Maybe we couldn't correct the inherited genetic defect, but maybe we could alter the course of the disease. As more miRNAs are overexpressed in dystrophic muscle, diseased muscle poses to be a very good target tissue for an antagonizing approach with anti-miRNAs. Future experiments will test whether altering the amounts of these miRNAs affect disease-related genes in ways that improve muscle function.

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HARVARD NEUROMUSCULAR DISEASE PROJECT
HARVARD MEDICAL SCHOOL • Boston Children's Hospital

Inside this issue:

Special points of interest:

- **New Kunkel Laboratory Findings**
- **Clinical Update**
- **Contact us!!!**

Meet the Kunkel Lab

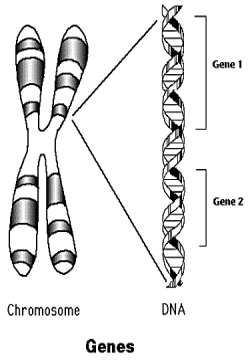
Laboratory Members: (Left-Right & Front-Back)

Hal Schneider, Marielle Thorne, Louis Kunkel, Elicia Estrella, Steven Boyden, Genri Kawahara , Hart Lidov, Jillian McCarthy, Iris Eisenberg, Mckensie Wessen, Christin Collins, Peter Kang, Juan-Carlos Casar

Missing from Photo:Dick Bennet & Alex White

Genetics 101

By Elicia Estrella, MS, CGC



Structure of DNA & Genes within a Chromosome

People, animals and many other living organisms are made of cells. All cells within an organism contain **DNA (*D*eoxyribo*n*ucleic *A*cid)**. DNA is the molecule that encodes our genetic information carrying all the instructions for making a person. DNA is made up of two twisted strands of chemical building blocks called bases. There are 4 bases that make up the language of our genes: ***A*denosine, *T*hymine, *G*uanine, and *C*ytosine**. Certain areas of our DNA contain our genes. These genes are considered our genetic blueprint or instructions. Our genes determine all of our features like height and eye color. This is why we have similarities to our parents and grandparents. Our DNA strands are coiled up and tightly packaged into structures called **chromosomes**. We have 46 chromosomes in total. They come in pairs (23 pairs), with one chromosome from each pair inherited from your mother and the other from your father. This is how our parents and grandparents have passed their genes down to the next generation. How our genes are expressed in our family is called the **inheritance pattern**.

Most MD's are inherited by the following patterns:

Autosomal Dominant
Autosomal Recessive
X-linked Recessive

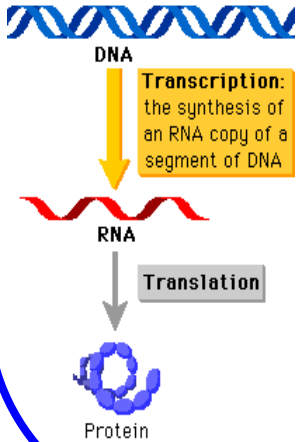
Autosome: gene is on chromosome 1-22 V.S. **X or Y**
X-Linked: gene is on X chromosome

Dominant: 1 copy of gene is changed (mutated) & trait is seen
Recessive: Both copies of a gene are changed and trait is seen

Transcription & Translation of your Genes

By Elicia Estrella, MS, CGC

"It is believed that every person has 10-15 mutations within their genome."



All of our genes are encoded in our DNA. In order for the body to read our genes and make them into proteins for use throughout the body, our DNA is "transcribed" or re-written into RNA in a complicated process called transcription. The process of transcription is similar to a court reporter taking spoken words from one source, like a court judge, and making a copy of those words on paper.

During transcription, one gene (DNA) is "re-written" or transcribed into RNA in the nucleus. This occurs via a team of enzymes and proteins binding to the promoter, or start, of a gene. These enzymes and proteins are able to unwind the DNA double helix just at the region of the gene being transcribed. The enzyme RNA polymerase uses one of the DNA strands to make an RNA copy of that one gene. This copy, which contains the instructions to make 1 protein, is called **mRNA** or **messenger RNA**. After the mRNA is made, it is trimmed down to a final size, and shipped out of the nucleus. When the mRNA gets into the cytoplasm, it is made into protein via a process called translation.

During translation, the mRNA transported to the cytoplasm is "de-coded" or "translated" to produce the correct order of amino acids in a protein. Translation requires numerous enzymes. The enzymes help keep the mRNA is position to be decoded. The nucleic acids of the mRNA are read 3 at a time and correspond to amino acids.

By stringing together many amino acids a protein is produced. The same mRNA may be used hundreds of times during translation before it is degraded (broken down) by the cell.

All the proteins that make up YOU, your cells, your body, the foods you eat, all the living cells in the world, etc - are made this way! Every time your body needs more of a protein -

- muscle protein
- hair protein
- Enzymes
- hormones

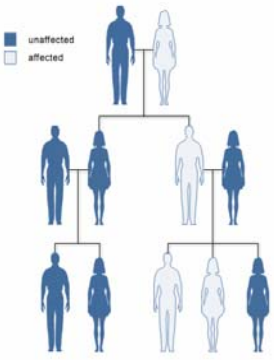
a gene carrying the information for that protein is transcribed into mRNA, and the mRNA is made into protein!

Clinical Update by Erica Sanborn, MS, CGC

The Muscular Dystrophy Association (MDA) clinic at Children's Hospital Boston is a multi-disciplinary clinic involving neurology, orthopedics, physical therapy, genetics, and social work. The clinic has about 400 outpatient visits each year, following children with a variety of neuromuscular diagnoses, Duchenne Muscular Dystrophy (DMD) and Becker Muscular Dystrophy (BMD) collectively are the biggest subset of our population. Currently we have about 100 patients with a diagnosis of either DMD or BMD.

Over the last few years research in DMD/BMD at all levels has greatly expanded. As many of you may know, a phase I trial using PTC124 was completed at a few hospitals in the USA. This medication is now being moved into a phase II study (which addresses safety and some initial efficacy). We at Childrens Hospital, Boston are hoping to be one of the trial sites for this new study. While the eligibility criteria are still being worked out, we know that boys over age 5 will likely be eligible if they have a particular genetic change known as a nonsense mutation (or premature stop codon) and are still able to walk independently. We do not have hospital approval to be a site for this study at this time and cannot field specific calls about eligibility, trial design, etc.

Another important study is trying to identify and define learning disabilities that may be associated with DMD/BMD. A study being run at Columbia University in New York, has worked with more than 250 families with DMD/BMD. Currently, the researchers are investigating early language development and reading acquisition in children with DMD/BMD. For more info about the study, please contact Abigail Batchelder, at 212-305-2394 or ab802@columbia.edu.



An example of autosomal dominant inheritance in a family tree. Lightly shaded people are affected with an "inherited condition"

Contact Us!!!

REPLY CARD

NAME: _____

(please PRINT)

Contact information Changes:

Address: _____

Phone: _____

Email: _____

Clinical updates:

_____

_____

_____ **Place a check here to be contacted about our new consent form**

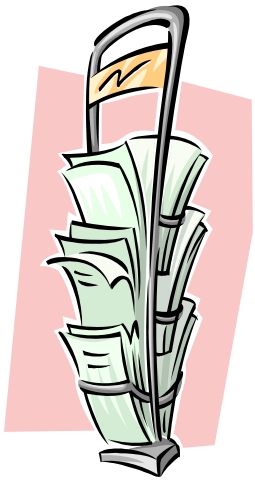
_____ **Place a check here to be taken off our mailing list**

Please sign to authenticate: _____

We would love to hear from you! Let us know:

1. Give us feedback on the newsletter
2. What would you like to see in future issues of our newsletter?
3. Update us on your contact info! (phone/email/address)
4. Take me off your mailing list!
5. Request a copy of the newsletter to be sent to someone not on our mailing list ! (family member, new study participant, your doctor, etc.)

Recent Publications:



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Guyon, J. et al. 2007. **Modeling Human Muscle Disease in Zebrafish**. *Biochim Biophys Acta*. 1772 (2):205.

Kohane, I. et al., 2007. **Medicine. Reestablishing the Researcher-Patient Compact**. *Science.* 316 (5826):836

We're on the Web!

Visit us at:

www.childrenshospital.org/cfapps/research/data_admin/Site2549/mainpageS2549P0.html

Other Websites of Interest:

- **www.clinicaltrials.gov**
- **www.MDA.org**
- **www.hhmi.org**

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