

How to Conquer a Chromosome Abnormality—

How is the chromosome analysis that diagnosed my child different from the research study molecular analysis?



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Special Points of Interest:

- * Chromosome analysis
- * Clinical Karyotype
- * DNA structure
- * Molecular Analysis
- * Array CGH

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Chromosome Analysis

The chromosome analysis performed most commonly in clinical laboratories looks at chromosomes under a light microscope. This picture shows how the chromosomes from a white blood cell look when they have been treated with a special stain that makes them appear to have black and white bands. However, this tangled mess is hard to analyze.



Clinical Karyotype

The next step in the analysis is to use a computer program that lines up the chromosome pairs by size—with #1 being the largest. You should also notice that each chromosome has a waistband constriction somewhere along its length. This is called a centromere and it makes the chromosomes look as if they have a long and a short arm. The convention is that the chromosome is always oriented with the short arm on top.

You will also see that this particular karyotype is from a male because in the bottom right corner there are an X and a Y chromosome. If the individual were

female there would be two X chromosomes.

This picture of chromosomes lined up is called a karyotype. The analysis of these chromosomes is done by a highly skilled professional technician, who first makes sure there are no extra or missing chromosomes, and also determines if any bands are missing, added or rearranged. However, in each one of these chromosomes there are hundreds of genes. There are over 300 genes on chromosome 18. So what would appear to be a small change in the karyotype could

actually involve a large number of genes. In addition, if two different people appeared to have the same chromosome change on their karyotypes, they could in fact be very different at the gene level.



DNA

This diagram depicts the structure of a chromosome. What looks like squatty little things under a microscope are actually very long linear chemical structures that have been precisely packaged. The chemical structure called DNA is shown in red and blue. This is the DNA double helix. The pairs of blue chemical subunits are the base pairs. This chromosome is actually a long string of base pairs. This string is wound around proteins shown in green like beads on a string. This string of beads is wound in a coil. The coil is looped around and around in daisy-like loops—all together forming a very highly packaged structure.

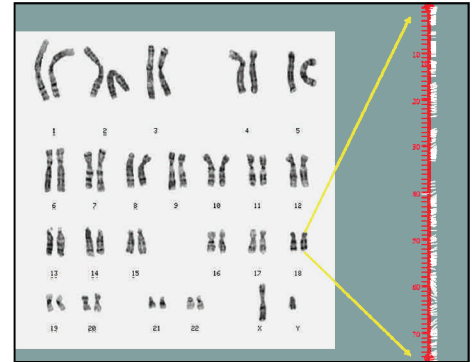


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Chromosome 18

To illustrate just how much information is actually packaged into a single chromosome, let's look at chromosome 18.

On the left is the karyotype. To the right show is red is the scale of the chemical units or base pairs of DNA. Number 1 is at the top and number 76,117,153 is at the bottom. So the numbers to the left of the red scale are 10 million base pairs, 20 million base pairs and so on. To the right of that, each of more than 300 genes is shown by the lines. You can see that they are not evenly distributed along the chromosome. There are gene deserts and gene clusters. This means that a certain distance along the chromosome cannot be equated with a certain number of genes.



Telephone Book Analogy

“Chromosome analysis is done by a highly skilled professional technician, who first makes sure there are no extra or missing chromosomes, and also determines if any bands are missing, added or rearranged.

In order to help you think about how much we really know when there is an abnormal karyotype, let's use the telephone book analogy. Here we have a pair of San Antonio telephone books. They actually do resemble chromosomes in several ways.

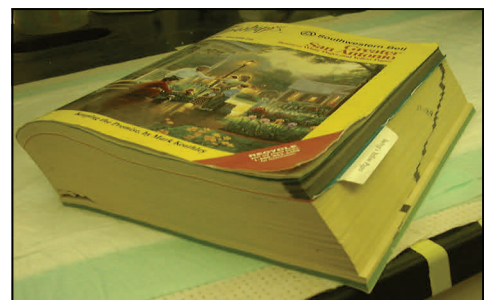
They have a p arm—the region with the black edge, a centromere—the tab, and a long arm that even has bands—region with yellow edge.

The phone book on the left is the normal one—just as the chromosome below it is normal. The phone book to the right is missing a piece—

just as the chromosome 18 below it is missing a piece too.

When you look at the two phone books, you can see that one has a missing section. You can also see that the missing section is from the long arm of the phone book. You can even see that the missing piece is toward the end of the long arm. However, you can only make an educated guess as to the location and size of the deletion in the phone book. It looks like the deletion might be somewhere around the letter P, but that is a guess.

This is very much like the chromosome analysis. From the picture of the chromosomes, you can see that the one on the right is missing a piece—and you can see that the missing piece is from the long arm of the chromosome. You also can be reasonably sure that the missing piece is near the end or includes the end of the chromosome. This is just a guess and there is no way to know which genes are involved in this chromosome change using just this type of analysis.



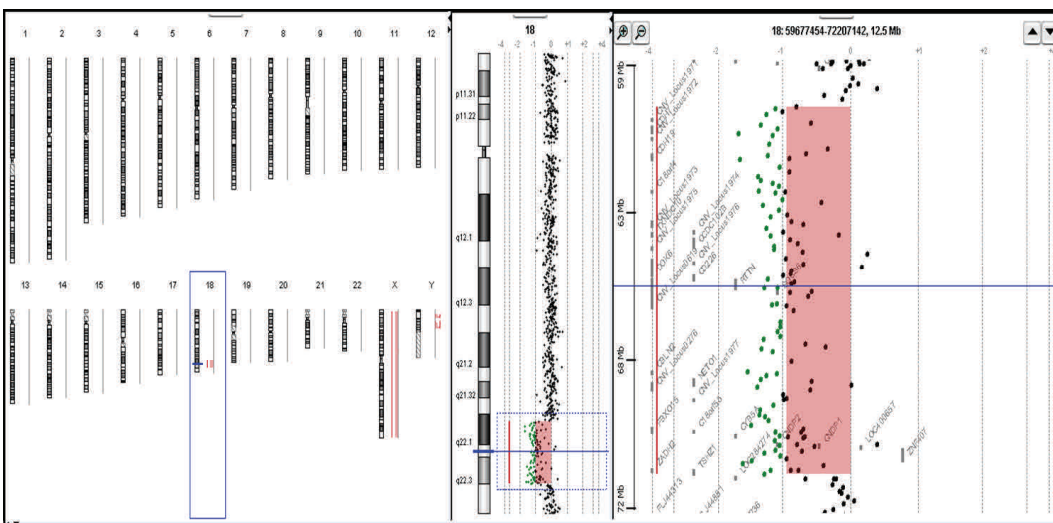
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Molecular Analysis

The molecular analysis, on the other hand, allows one to know the exact location of the deletion. This is like opening the phone book to see that the deletion goes from entries beginning with REAL to entries beginning with TREE. We can precisely know the deletion, down to the letter.



Microarray Comparative Genomic Hybridization—Chromosome Microarray Analysis



“The type of molecular analysis we use in our laboratory is called microarray comparative genomic hybridization or array CGH for short.”

Here is the molecular analysis from that same individual whose chromosome you saw with the telephone books. The type of molecular analysis we use in our laboratory is called microarray comparative genomic hybridization or array CGH for short. Some labs call it a chromosome microarray analysis.

This analysis does not show actual pictures of the chromosomes. Here are diagrams of the chromosomes with the molecular data points placed on the diagram. The panel on the left shows

every chromosome. The pink bars next to the chromosomes show which chromosomes have copy number changes. The X and Y show changes because we used DNA from a person of the opposite sex as an internal control. The only chromosome showing any aberration is chromosome 18 which has a blue box around it.

The center panel shows only chromosome 18. You see the actual data points as black dots when they are within the normal range and as green dots when they show only one copy instead of two.

The pink bar shows the region as a deletion by the statistical analysis in the program. You can see that this individual does not have a terminal deletion as you might have thought from the picture of the chromosome. But instead this person has an interstitial deletion.

The region of chromosome 18 in the blue box in the central panel is shown in the panel on the right. Let's zoom in on this panel so you can see everything.

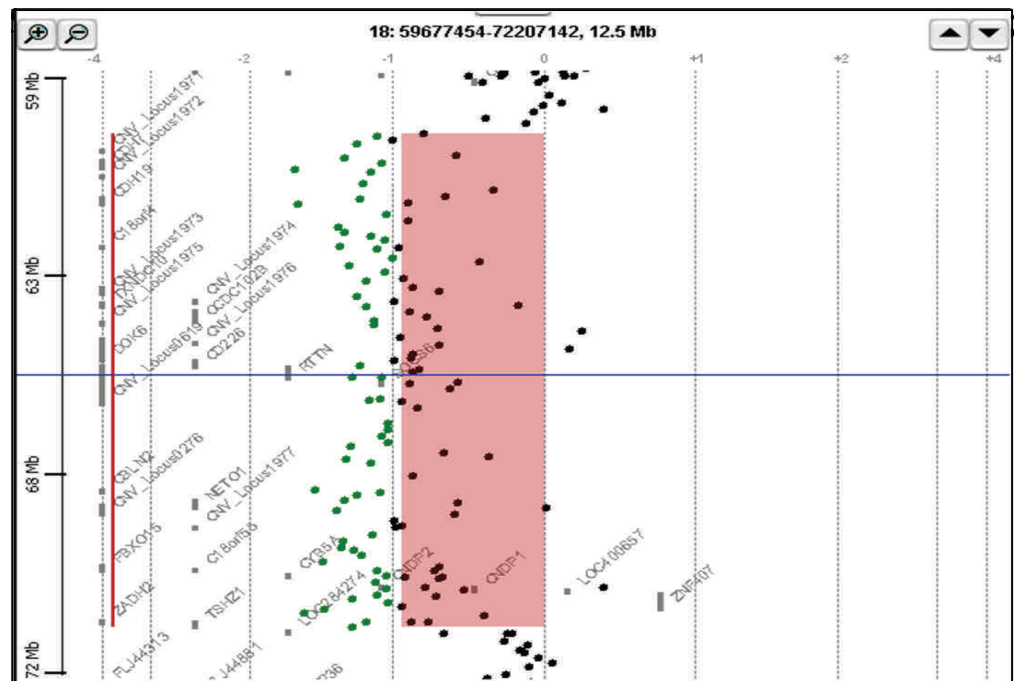
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Microarray Explanation continued

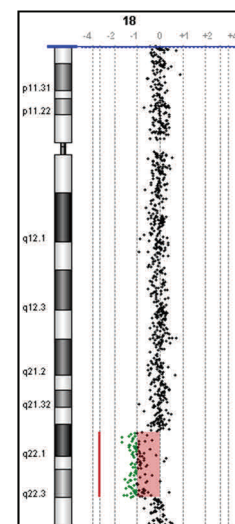
The scale on the left is the base pair scale. It goes from a position at 59 million base pairs of DNA to 72 million base pairs of DNA. The grey bars of varying lengths with letters next to them show the positions of particular genes. They are spread across the diagram for readability purposes only. This is because at this resolution if they were all in a row from top to bottom, the scale would be unreadable. The black and green dots are the

data points. The black dots are within the normal range. You can see that this technique is very “noisy” and the range of normal is scattered. The green dots are those data points that are present at half the normal amount. The red line and the pink bar indicate the region that the program has detected as present in one copy instead of two copies. From this data you can easily determine exactly which genes are present in one copy versus the normal two copies.

"From this data you can easily determine exactly which genes are present in one copy versus the normal two copies"



From this comparison, you can see how important the molecular analysis is when we are trying to tie specific genes to specific outcomes.





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Our Motto:

To provide individuals and families affected by chromosome 18 abnormalities with comprehensive medical and educational information with a focus on treatment options.

We are on the web!

<http://pediatrics.uthscsa.edu/centers/chromosome18/>

Information provided by The Chromosome 18 Clinical Research Center to:



<http://www.chromosome18.org/>
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