This Month in Genetics

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The Power of Family

Whole-exome sequencing has exploded on the scene as a way of identifying disease mutations. Roach et al. propose an even more powerful technique to zero in on causative genetic variation, and their approach also gives us some more basic information about the human genome. Whole-genome sequences from a family of four individuals, two parents and their two children were analyzed. The children are both afflicted with Miller syndrome and primary ciliary dyskinesia and had been included as two of the cases in the paper by Ng et al. (Nature Genetics *42*, 30) that used whole-exome sequencing to identify the Miller syndrome gene. Roach et al. hone right in on the causative genes for both phenotypes; the family-based approach vastly improves their ability to sort through false-positive sequence variants, significantly enhances sequence accuracy, and reduces the number of candidate genes that need to be considered in the analysis to four. This analysis also lets them precisely pinpoint sites of recombination and estimate the intergenerational mutation rate to be 1.1×10^{-8} per position per haploid genome, which is quite a bit lower than previous estimates made with the use of evolutionary data.

Roach et al. (2010) Analysis of genetic inheritance in a family quartet by whole-genome sequencing. Science Express, in press. Published online March 10, 2010. 10.1126/science.1186802.

The Mother-to-Child Transition

At the point of fertilization, gene products encoded by the mother control development. This lasts until the point of maternal-zygotic transition, at which time the zygotic genome is activated. The mechanism by which the zygotic genome is held inactive until the time of this transition is unclear, so Vastenhouw et al. studied the chromatin marks on the zygotic genome over this time period in zebrafish embryos. They mapped histone H3 trimethylation marks and found that the maternal-zygotic transition is accompanied by major changes to these modifications that may govern zygotic genome activation. The majority of genes acquire an activating H3K4Me3 mark at the point of the transition, including many inactive genes, which do not have the H3K36me3 modification that marks sites of transcriptional elongation. These genes are not stably associated with RNA polymerase II, although the mechanism by which they are maintained in a repressed state is unclear. Other inactive genes, including many developmental regulatory genes, do have a repressive H3K27Me3 modification in addition to the activating H3K4Me3, and this bivalent mark may poise them for activation while still keeping them in a repressed state. In addition to allowing us to better understand the basic biological underpinnings of this point in development, the maternal-zygotic transition coincides with the formation of pluripotent cells, so this line of research has implications for our understanding of embryonic stem cells.

Vastenhouw et al. (2010) Chromatin signature of embryonic pluripotency is established during genome activation. Nature 464, 922–926.

The Wide Reach of Cilia

Primary cilia are critical for proper development and maintenance of certain tissues, and defects in these nonmotile cilia cause a range of human diseases, from malformations in the brain to polycystic kidney disease and a variety of disorders in between. Over the past few years, several components of primary cilia have been identified, but Kim et al. decided to take one step back and look for regulators of ciliogenesis by the use of a large-scale screen. They developed a quantitative fluorescence-based assay for ciliogenesis that makes use of a green fluorescent protein–tagged ciliary protein and evaluated the impact of several thousand genes on ciliogenesis with the use of siRNAs. They found 49 positive and negative regulators of ciliogenesis, and further investigation of these regulators led them to uncover new connections between actin network formation, endocytic recycling, and the formation of cilia. They also investigated a new cellular structure, dubbed the pericentrosomal preciliary compartment, that seems as though it may provide a temporary reserve of building blocks for cilium formation.

Kim et al. (2010) Functional genomic screen for modulators of ciliogenesis and cilium length. Nature 464, 1048–1051.

Translating Genetic Variation to Gene Expression

We don't all express the same amount of every gene, and this is part of the reason that we are each unique. Gene-expression differences could be influenced by sequence variation, epigenetic regulation, or other variables completely unrelated to the DNA, but the relative importance of each of these factors is unknown. Two groups recently took a stab at figuring out how much sequence variation is likely to affect gene expression, by taking

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a genome-wide look at the relationship between genetic variation and DNase I hypersensitivity sites and/or chromatin immunoprecipitation profiles. Together, their data indicate that genetic variation can influence transcription factor binding, chromatin structure, and gene expression. McDaniell et al. show that these effects are heritable, further supporting the attribution of the expression differences to sequence variation. Although sequence variation does influence the expression of some genes, these genes are actually in the minority, so this is not the end of the story.

Kasowski et al. (2010 Variation in transcription factor binding among humans. Science 328, 232–235.

McDaniell et al. (2010) Heritable individual-specific and allele-specific chromatin signatures in humans. Science 328, 235–239.

Sorting through Mutations in a Cancer Genome

Now that it is feasible to sequence whole genomes, several recent papers have reported comparisons of a cancer genome relative to the constitutional genome sequence of the affected individual. Some of the cancer-specific mutations identified through these projects were undoubtedly involved in cancer development, but others are likely to have just gone along for the ride, and the task now is to figure out which is which. To give themselves a leg up in sorting through breast cancer mutations, Ding et al. compared four whole-genome sequences derived from the same patient with breast cancer: samples from her peripheral blood, samples from the primary tumor, a xenograft of the primary tumor, and a brain metastasis. This allowed them to pick out the mutations and structural changes that were specific to the tumor and then ascertain which of these were maintained or lost during metastasis and/or xenograft development. Not only are they able to find genetic changes that are gained or lost during tumor development and metastasis, but the massively parallel sequencing approach that they used to obtain their data also allowed them to estimate the relative abundance of each sequence as the tumor developed and metastasized, an analysis that supports the idea that a minority of cells from the mixed primary tumor went on to form the metastasis. Although this approach doesn't pinpoint exactly which mutations are the most important at each stage of the cancer, it does give them a global picture of the evolution of the cancer in a single individual.

Ding et al. (2010) Genome remodeling in a basal-like breast cancer metastasis and xenograft. Nature 464, 999–1005.

This Month in Our Sister Journals

We Can Make it Better, Stronger, Faster…

Although a significant fraction of individuals with idiopathic dilated cardiomyopathy (DCM) have a family history of the disorder, the genetic heterogeneity to this disorder makes it difficult to identify an underlying cause. Already, more than 30 genes have been identified for DCM, which makes genetic testing for DCM expensive and complicated. Zimmerman et al. have addressed this problem through the development of a resequencing-based array for DCM genetic testing that includes 19 genes, nine more than previous testing arrays. This approach significantly reduces the cost and turnaround time of testing and increases the likelihood that a relevant mutation will be identified. When tested on 73 DCM patients for whom no mutation had previously been identified, the approach identified seven sequence variants of likely clinical significance in the new genes included on the array.

Zimmerman et al. (2010) A novel custom resequencing array for dilated cardiomyopathy. Genetics in Medicine, in press. Published online March 11, 2010. 10.1097/GIM.0b013e3181d6f7c0.

You Can't Get There from Here

In addition to binding CD4 on target cells, HIV uses one of two different cell-surface chemokine receptors to enter cells. During primary infections, HIV almost exclusively uses CCR5 as a coreceptor, but it can switch to use of CXCR4 late in infection. Although HIV is a rapidly mutating virus and only a small number of viral mutations is needed to switch the coreceptor used by the virus, coreceptor switching does not occur as readily as might be predicted. To determine what barriers might prevent HIV coreceptor switching, da Silva et al. compared a CCR5-using parent strain of HIV and its CXCR4-using derivative. They systematically engineered mutations from the derived strain back into the parent strain singly and in combinations and measured the fitness of each in order to quantify the epistatic effect of the mutations on viral fitness. They attempted to determine the minimal mutational pathway that would be needed to get from the CCR5-using to the CXCR4-adapted daughter strain and found that some predicted steps along the way were selectively disadvantageous, a fact that could constrain viral adaptation and coreceptor switching. Because this process is associated with disease progression, an understanding of the mechanism by which HIV evolves to use a second coreceptor could help us understand how to put a stop to this wily virus.

da Silva et al. Fitness epistasis and constraints on adaptation in a human immunodeficiency virus type 1 protein region. Genetics, in press. Published online February 15, 2010. 10.1534/genetics.109.112458.