In Fetal Alcohol Study, IPA Finds the Signal in the Noise

Western University scientist Ben Laufer uses IPA from QIAGEN to take a systems approach in his study of epigenetic changes in fetal alcohol spectrum disorders.

"We didn't expect to see that broad of a change. Ingenuity not only confirmed the results but gave them a degree of power beyond just statistics by incorporating the biology into it."

At Western University, Ben Laufer is making strides in demonstrating the rampant genome-wide changes related to fetal alcohol syndrome and related disorders, a group of diseases that have been difficult to study due to heterogeneity in exposure and subsequent symptoms. By using QIAGEN's Ingenuity Pathway Analysis (IPA), he has proven that widespread epigenetic changes once written off as biological noise are in fact effects of alcohol exposure during fetal development.

Laufer says the real power of IPA is in its systems approach — the tool lets him analyze a constellation of data that would be daunting to study manually. Because that data encompasses such a broad spectrum of biological changes, even careful inspection would likely miss the common threads. But IPA can process and make sense of these large data sets, offering the narrative that explains how these changes relate to each other. In a recent experiment, Laufer fed IPA several data sets that seemed to show little biological significance. "After just 10 minutes in IPA, we had our eureka," he says. "It was great."

IPA is also an important tool for allowing biologists to perform advanced computational analyses that might otherwise be the domain of expert bioinformaticians, Laufer notes. "It helps to bridge this very wide gap that we're seeing in biology between people with a statistical or bioinformatics background and people with a biology background," he adds.

FASD FOCUS

Laufer started out with plans to attend medical school, but even during his undergraduate studies he found himself pulled toward science instead. "I realized that the people making the difference and developing the technologies that we see in medicine weren't the doctors but the scientists," he recalls. "I've always been interested in changing things rather than doing things the way they are."

He hasn't looked back since. Now a PhD candidate at Western University, his focus is on epigenetics, a field that was just taking off when he began his degree program under the guidance of Dr. Shiva Singh. "The realization that there's much more to heritable information than the four DNA bases was a big topic in the field when I started," he says. Some of his PI's lab focused on fetal alcohol spectrum disorders (FASD), so Laufer decided to apply new epigenetic technologies to that research area.

FASD offers Laufer the opportunity to do what he always wanted to: make a difference in medicine. Current diagnostics for children suspected to have FASD are rudimentary — measuring distance between the eyes, for example, to see if there's a deviation from normal. If Laufer can determine a telltale epigenetic code indicative of FASD, it would be a major step toward improved diagnostics in the field.

FASD comprises a range of conditions, with fetal alcohol syndrome the most severe. Scientists estimate there may be six babies born with FASD out of every 1,000 live births, but diagnosis is so challenging and inconsistent that some experts believe this significantly underestimates the problem. People with FASD may have symptoms from facial abnormalities and growth deficiency to cognitive impairment and other damage to the central nervous system. "Fetal alcohol really is a disease of the epigenome, with regulatory elements being most damaged," Laufer notes.

The severity of symptoms may correspond to the fetal development stage at the time of alcohol exposure, as well as the level and duration of exposure. These confounding variables make studying FASD very difficult. "A lot of past research has been bogged down by heterogeneity and the challenge of not being able to get past it statistically," Laufer says. "This area has been underexplored because of that."

THE BIG PICTURE

Much of Laufer's current work involves using microarrays to study mice modeling fetal alcohol exposure, with a specific focus on neurological changes. In a recent research project, he used gene expression arrays, DNA methylation arrays, and microRNA expression arrays. "It was an overwhelming amount of data," Laufer says. "I needed to incorporate three different technologies to get a picture of what's going on in fetal alcohol exposure in the brain."

That data storm prompted Laufer to use IPA to make sense of the data, which showed many small changes across the genome. "IPA helped us pick out these buried expression modules that we were seeing," he says. "A lot of standard analysis approaches can't tackle that type of data."

What IPA found was that the changes were not due to noise, as the team had feared since a full third of the epigenome showed alterations even to moderate alcohol exposure. "We didn't expect to see that broad of a change," Laufer says. "Ingenuity not only confirmed the results but gave them a degree of power beyond just statistics by incorporating the biology into it." IPA results indicated that the changes were not random, as they had appeared in the raw microarray data. In fact, they were associated with a number of biological functions that had previously been linked to FASD — and even some novel ones that have been implicated in schizophrenia and autism.

Laufer and his colleagues used the IPA results to determine that a paired reaction of DNA methylation and microRNAs was occurring in FASD cases. Clusters of microRNAs were highly environmentally responsive because of their reliance on epigenetic marks; alcohol exposure altered the methylation state of these regions, which in turn changed the regulation patterns of these microRNA regions. "These large clusters of microRNAs regulate a lot of complex functions that seem to be unique to mammals," Laufer notes.

"With IPA you can get a look at the big picture and see the biological system in its full context and make your inferences from there," he says, adding that these results have given the team enough confidence in being able to solve the heterogeneity dilemma that they are now moving on to human studies. They're currently collaborating with a pediatrician from a region of Canada with a high FASD prevalence.

EASE OF USE

While Laufer relies on IPA to get high-quality data analysis, he appreciates the easy-to-use interface of the tool. "If you can use Facebook and Excel, then you can use IPA," he says. "I think it would be a good fit for pretty much anyone." The application can handle everything from simple experiments with a single data set to more complicated projects integrating multiple data types and sources, he notes.

It is also essential for saving time on literature searches. "With microarrays, I can't look into every single gene that's being pulled up in every single case," Laufer says. IPA prioritizes results based on the literature, helping him avoid those tedious dead-end searches. "IPA has given me a lot of insight that I might have overlooked and really pushed my project forward."

QIAGEN Silicon Valley
1700 Seaport Blvd. Third Floor
Redwood City, CA 94063

Tel. +1 650 381 5100
Fax. +1 650 381 5190

info@ingenuity.com
www.ingenuity.com