Clinical Chemistry

Integrative Bioinformatics Analysis Reveals New Prognostic Biomarkers of Clear Cell Renal Cell Carcinoma

Article:
H. Butz et al.
Integrative Bioinformatics Analysis Reveals New Prognostic Biomarkers of Clear Cell Renal Cell Carcinoma.
Clin Chem 2014;60:1314-1326.
http://www.clinchem.org/content/60/10/1314.abstract

Guest:
Dr. George Yousef is an anatomical pathologist and a scientist at St. Michael's Hospital in Toronto, and an Associate Professor at the Department of Laboratory Medicine at the University of Toronto.

Bob Barrett: This is a podcast from *Clinical Chemistry,* sponsored by the Department of Laboratory Medicine at Boston Children's Hospital. I am Bob Barrett.

Kidney cancer is among the ten most frequently occurring cancers in the Western world and its incidence has been steadily rising each year. In the absence of symptoms, about 30% of patients with renal cell carcinomas are diagnosed with disease already in the metastatic stage.

In the October 2014 issue of *Clinical Chemistry,* researchers using an integrative system biology approach identified three novel factors as potential biomarkers involved in renal cell carcinoma pathogenesis which have not been linked to kidney cancer before.

In this podcast we are joined by the senior author of that paper, Dr. George Yousef. He is an anatomical pathologist and a scientist at St. Michael's Hospital in Toronto, and an Associate Professor in the Department of Laboratory Medicine, School of Graduate Studies and the Institute of Medical Sciences at the University of Toronto.

Dr. Yousef, in your recent publication in *Clinical Chemistry,* you describe the construction of a network of interaction and renal cell carcinoma through combined analysis of multiple publicly available databases. What's new in this paper and how is this different from a regular meta-analysis?

Dr. George Yousef: A unique feature of our study is the use of integrative analysis. We combine different types of molecular changes from genes, proteins, and microRNAs from almost 600 cases of clear cell renal cell carcinoma and 400 normal kidney controls.

And then we used an unbiased approach to identify the most significant molecular interactions that are specific to renal cell carcinoma.

So in our model the basic elements, whether these are genes, microRNAs or proteins, are called nodes; and the interactions or the relationships between them are called edges.

The relationship between the nodes is also directional, i.e., it's based on the type of interaction, whether activation or inhibition; this way our network is hierarchical, with three levels of nodes.

Nodes of the top layer are considered to be the master regulators, because they control all other lower layers in the network, but they are not affected by other nodes; and as such, this can be considered as ideal drug targets.

We also identified a number of hubs. A hub is a node that has the highest number of connections or interactions. And as such, they are considered critical molecules in the pathogenesis of clear cell renal cell carcinoma.

These molecules can be potentially useful as prognostic markers. A prognostic marker is the one that's used to assess disease aggressiveness.

We finally validated the prognostic significance of three of these hubs in patient tissues.

Bob Barrett: Recently with extensive use of sophisticated informatics analysis, many have expressed concerns about the accuracy of these in silico approaches, and if they can accurately reflect the real biology rather than just being computer artifacts.

Dr. George Yousef: This is actually a very important point, and I am glad that you brought it up. I totally agree with you that bioinformatic prediction has to be interpreted in a biological context, and that there is always a need for experimental validation of any bioinformatics-driven findings, because relying solely on informatics can be very misleading.

In our paper, for example, we have two independent validation sets; one from the Cancer Genome Atlas, which is a large database of over 450 cases in addition to our database of about 400 cases of primary renal cell carcinoma.

So in my opinion, the major advantage of bioinformatics in biomedical research is to put you in the right direction by identifying potential disease biomarkers and druggable targets, but this should not replace the need for experimental validation.

Bioinformatics should be also backed up by functional analysis. In our study, for example, we validated the potential impact of three critical molecules on disease aggressiveness using saline models. And we showed that they affect cell migration and the invasive ability of tumor cells.

Bob Barrett: Well, this brings us to the next point, which is the noticeable trend of the increasing use of integrated genomics, which has been observed in your paper and a number of studies by the Cancer Genome Atlas Group and others. What's your take on the impact of the integrated genomics in the future of cancer research?

Dr. George Yousef: Actually, this is a very interesting issue. I strongly believe that integrated genomics will revolutionize our understanding of the pathobiology of cancer.

For decades researchers were looking at cancer pathogenesis from one single narrow dimension. Some looked at mutations in cancer; others were more focused on the epigenetics, etcetera, but in reality the cancerous phenotype is the product of interaction between all these classes of molecular changes and we can't understand it by focusing on one level in isolation.

That's why incorporating different types of molecular changes will enable a much more thorough understanding for cancer pathogenesis.

Let's for example look at kidney cancer. Data from the Cancer Genome Atlas and from our group showed the presence of what we call mutually exclusive events. These are kind of different mechanisms or tools that are used by the tumor to achieve the same outcome.

For example, a tumor can activate certain oncogene through chromosomal amplifications in a subset of patients, or by using hypomyelination or microRNAs in another subset and so on.

Also, in some situations the final gene dysregulation battle can be a net outcome of a number of opposing forces that act on the target and that's the power of integrated genomics.

Bob Barrett: This is extremely interesting stuff, but let me ask you, aren't there any limitations to this integrative genomic approach?

Dr. George Yousef: Well, I share with you your concern, and I do agree that we need to understand the limitation of integrated genomics. One such drawback is our inability to transform information

from one platform to another and the lack of standardization between the different platforms.

In addition, it's important to realize that when integrating thousands of data points from different classes of molecular changes, we can run into the risk of what we call incidental findings. These are statistically significant but they are not biologically valid.

Bob Barrett: Well, finally Dr. Yousef, you also presented evidence on the role of microRNAs as master regulators of the network of kidney cancer pathogenesis. How do you envision the potential clinical utility of these microRNAs in the clinic?

Dr. George Yousef: Well, I strongly believe that microRNAs are great potential clinical tools. They can serve both as biomarkers and as therapeutic targets. These microRNAs are small, single-stranded RNA molecules; they do not code for protein, but rather function by controlling the expression of their target genes.

Interesting facts about microRNAs that make them great biomarkers are the fact that they are secreted in blood and urine and many other body fluids. They can be also extracted with high quality from formalin-fixed and paraffin-embedded tissues, in which they are stable for almost ten years.

Other exciting characteristics are that these microRNAs are very small, and as such they are very easily transected with minimum toxicity and side effects, and that a single microRNA has the ability to target multiple genes simultaneously. And that's why they represent very attractive potential therapeutic targets.

Bob Barrett: Dr. George Yousef is an anatomical pathologist and a scientist at St. Michael's Hospital in Toronto, and an Associate Professor at the Department of Laboratory Medicine at the University of Toronto. He has been our in this podcast from *Clinical Chemistry* on biomarkers of renal cell carcinoma.

I am Bob Barrett. Thanks for listening.