

“Multiple buyers are needed for maximum sensitivity, specificity”

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—*Dr. Jehangir Mistry, director, R&D, Bioscience Division, Millipore Corporation, US*

Dr. Jehangir Mistry is the director, R&D, Bioscience Division, Millipore Corporation, US. He leads Millipore's Protein Research Assays Group, and also has extensive experience as director of manufacturing, director of automated systems and member of a business development team in his earlier assignment. In an interaction with BioSpectrum, Dr. Mistry speaks about the gaining popularity of protein immunoassays in protein research and drug discovery, developments and trends.

What is the importance of biomarker immunoassays in life science industry?

Biomarker immunoassays are becoming very popular for the measurement of biologically important proteins that are either involved in the etiology or could serve as markers of various diseases.

How can we address the challenges posed by these diseases?

Diseases such as diabetes, cardiovascular diseases, infectious diseases and cancer are of particular relevance for Indian researchers and drug manufacturers, given the wide footprint of these diseases among Indians. Other diseases include: autoimmune diseases, neurological diseases (e.g. Alzheimer's or Parkinson's), toxicity and others. Major pharmaceutical

companies as well as many academic clinical research centers are focused on identifying proteins that would serve as biomarkers for the purpose of either prognosis, diagnosis, monitoring drug efficacy, patient stratification, or risk assessment. These proteins either circulate in the blood or are present inside the cell.

How does Luminex xMAP technology fit in here for protein biomarkers?

The MAP stands for Multiple Analyte Profiling and x stands for any analyte of interest. The Luminex technology is based on polystyrene or magnetic beads, which are 6 micron in size. Luminex created 100 (now 500) different sets of beads, each set having a unique fluorescent signature. Each bead set can be used to conjugate an antibody to the specific analyte of interest. The second antibody is biotin-labeled and forms a 'sandwich' where the analyte binds between the two antibodies. Streptavidin-phycoerythrin (SA-PE) then binds to the biotin-labeled antibody. One can set up a multiplex immunoassay by mixing various bead sets of interest to measure specific analytes in a 96-well microtiter plate as if setting up an ELISA. At the end of the immunoreaction, the plate is inserted in the Luminex reader, which has two lasers. The red laser identifies the bead set and also the analyte. The green laser quantitates the amount of fluorescence present on the SA-PE attached to the specific biotin-labeled detection antibody, and thus quantitates the amount of analyte.

Millipore has strategically partnered with Luminex where it uses the xMAP technology to develop, manufacture and commercialize multiplex immunoassay kits for a broad range of protein biomarkers involved in various diseases or research areas.

What are the benefits and applications of protein multiplexing?

The major benefits of multiplexing include: lot more information at a much faster rate; conservation of precious biological samples; and reduced cost of analysis due to the reduction in labor, time and the cost of reagents. The xMAP technology has several advantages over other existing multiplex technologies such as planar arrays. The Luminex beads are robust, easy to handle, and are easy to couple with antibodies. The major benefit of the Luminex technology is the flexibility of measurement of only the analytes of interest by mixing only those bead sets to which antibodies to the specific analytes are conjugated. Thus, the cost per analyte per sample is usually lower with the Luminex technology compared to the planar array containing a large number of analytes.

How does Millipore support outsourcing?

Millipore not only provides the multiplex immunoassay kits to the customers who already have the Luminex reader to run the kit in-house, but also offers to run samples, and provide results for a fee to those customers who either may not have the Luminex reader or are not familiar with the technology, or simply may not want to invest in resources to run the kit in-house.

What are the significant developments and trends in protein research worldwide?

The core applications today for protein multiplexing include: studying inter-relationships among biomolecules of similar function, structure or tissue origin; for example, cytokines and hormone analysis; elucidating intra-cellular signal transduction pathways, and related biological mechanisms to understand disease pathophysiology. The next frontier for multiplexing includes biomarker analysis and validation. The fundamental concept is that multiple biomarkers are needed for maximum sensitivity and specificity. A significant amount of research is currently being done to evaluate the molecular fingerprint of multiple markers with the most predictive value.

Where does India figure in the adoption of these developments and trends?

India has been making tremendous progress in science and technology, and has attracted most foreign companies to invest in it due to its highly educated and talented workforce.

Several Indian companies and academic institutes have either already adopted the multiplexing technologies or are seriously considering it. I am glad to be a part of the Millipore India team to help our customers guide in making the decision of using the right technology that meets their research needs.

Please tell us about the portfolio of the protein assay group that you head.

My group is called the Protein Research Assays (PRA) group, and we are a part of the Life Science business unit in the Bioscience Division of Millipore. I head the R&D efforts for the PRA group, where our focus is on establishment of a vertically integrated system that develops novel recombinant proteins, antibodies and assays (RIAs, ELISAs and Luminex-based multiplex assays) for metabolic diseases (diabetes, obesity, cardiovascular, bone/cartilage metabolism), inflammation and immunology, cell signaling, epigenetics and neuroscience.

What are Millipore's future plans or focus areas in the protein assay area?

In future, we will continue to expand and introduce novel assays in the above disease/research focus areas. In addition, we have plans to develop novel assays in the oncology, toxicity and stem cell research area. Today, we have the broadest menu of panels and analytes useful for studying metabolism, inflammation and immunology, cardiovascular and bone disorders, neuroscience, toxicity and cell signaling. We offer our customers, the flexibility of selecting any analytes of their interest within a given panel. We also sell the Luminex instrument and data reduction software. We have an excellent

technical support team, and we continue to work closely with our customers to develop novel, cutting-edge assays/panels.

Are there any new therapeutic areas you plan to enter?

Yes, we plan to significantly expand our multiplex portfolio in neuroscience, cell signaling, oncology, stem cells and toxicity areas in the coming years.

Jahanara Parveen