
# Beyond Genomics: Proteomics-Based Companion Diagnostics for Precision Medicine

The concept of precision medicine has been a goal of medicine for decades. Accordingly, oncologists have employed patient selection strategies using genetic biomarkers, a successful approach for a small subset of cancers. For the majority of patients with cancer, however, treatment success has been limited and the field is now looking beyond genomics to the proteome, aiming to accurately measure protein signaling pathways inside tumors.

Peter Blume-Jensen, MD, PhD, is the founder, president, and CEO of Acrivon Therapeutics, a clinical-stage biopharmaceutical company developing precision oncology medicines. The company uses its proprietary proteomics-based patient responder identification platform called Acrivon Predictive Precision Proteomics (AP3) to create OncoSignature® tests that are designed to match its oncology medicines to the patients whose tumors are predicted to be sensitive to a specific medicine. Dr. Blume-Jensen has extensive experience in oncology R&D and oncogenic kinase signaling and is the inventor of the AP3 platform and the OncoSignature® patient selection method.

Brian McKelligon is CEO of Akoya Biosciences, a company specializing in spatial biology platforms that enable and catalyze next-generation tissue analysis. Spatial biology combines whole-slide imaging of tissue sections at single-cell resolution with quantitative analysis of spatial parameters that are ultimately important to understand the tumor microenvironment and develop effective therapies and predictive diagnostics. It allows visualization and quantitation of the expression of multiple biomarkers at one time and reveals how cells interact and organize across the entire tissue architecture.

Acrivon is partnering with Akoya for the clinical development of an OncoSignature test as a companion diagnostic to identify cancer patients most likely to respond to treatment with Acrivon's ACR-368, an experimental DNA damage response (DDR) inhibitor therapy. ACR-368 is being studied in a Phase 2 trial of patients with ovarian, endometrial, and urothelial cancers.

Blume-Jensen and McKelligon recently spoke with *Inside Precision Medicine*'s editor in chief, Damian Doherty, about their partnership and how multiplex, spatial protein signatures have the potential to predict response to treatment for the estimated 90% of cancers not defined by single gene driver mutations. The ultimate goal, they say, is to reduce the alarming attrition rate of drug candidates in development.

**Q: Tell me about Acrivon and your approach to the development of cancer therapeutics.**

**Peter Blume-Jensen:** More than 50 cancer drugs targeting proteins encoded by single driver mutations have been approved through the accelerated pathway with the FDA, with notable examples including Zelboraf® (vemurafenib) for the treatment of BRAF-mutated metastatic melanoma and Xalkori® (crizotinib) for the treatment of EML4-ALK–driven lung cancer back in 2011. While approvals for these types of drugs tend to be fast

and there is a simple way to identify patient responders, only a very small subset of cancers, perhaps 10%, has a single genetic driver. When you look at genetic alterations in the other 90% of cancers, however, it becomes too complex to predict patient response and direct treatment using genetics-based diagnostics. This lack of ability to predict response affects the reported efficacy of experimental drugs and is a major contributor to attrition of candidates in Phase II clinical trials.

At Acrivon, we apply our AP3 platform to cancers where we believe genetics is, or has proven to be, insufficient. Instead, our approach is designed to look at what drives the disease from the perspective of proteomics and match the drug mechanism of action with the disease-driving mechanisms. Results from the AP3 platform are used to generate our proprietary OncoSignature biomarker tests, each of which are tailored to our individual drug candidates. We apply our OncoSignature test to a pretreatment tumor biopsy from each patient we intend to treat, with the goal of predicting whether the patient will benefit from our drug candidate.

We applied this approach to the clinical-stage candidate prexasertib, now designated ACR-368 and advancing in Phase II studies, which we in-licensed from Eli Lilly. It is a targeted DNA damage repair inhibitor that previously has been evaluated at the recommended Phase II dosage in more than 400 patients and demonstrated durable monotherapy activity in a proportion of patients with platinum-resistant ovarian cancer and squamous cell cancers, including head and neck and anal cancers. Despite significant efforts, Lilly was not able to identify a patient selection method for ACR-368, and without that, despite the promising clinical activity, the response rate was insufficient to advance it into subsequent clinical trials.

Peter Blume-Jensen, MD, PhD
founder, president, and CEO
Acrivon Therapeutics

We generated a three biomarker OncoSignature test that links the active tumor-driving mechanisms with the mode-of-action of ACR-368. This test was used in all of our evaluation studies, including those using pre-treatment tumor biopsies collected from past ovarian cancer trials with the drug. We have been able to demonstrate a significant enrichment of the responders, which are patients with all three biomarkers present at a minimal pre-specified level. We've also used the same OncoSignature test to identify other tumor types that are sensitive to ACR-368, which we call "predicted tumor types," and thus far, they include endometrial and bladder cancer.

Overall, the underlying principles of our AP3 platform approach are broadly applicable, and we also have a preclinical pipeline in the DDR and cell cycle pathways for which we intend to develop drug-tailored OncoSignature tests.

### Q: What is the goal of your partnership with Akoya?

**PB-J:** I've known members of the Akoya team for many years, and we consider them to be the leading provider of the spatial biology technology needed to further develop and validate our OncoSignature test into a companion diagnostic test. Together, we're pioneering an approach that hasn't been used before.

> "There are many potentially efficacious drugs, but if they can't be matched to the right patients, they'll never gain approval."

The goal of our partnership with Akoya is to develop, clinically validate, seek regulatory approval for, and, pending ACR-368 approval, commercialize the multiplex OncoSignature test. Akoya would then be the exclusive provider of the companion diagnostic test required for prescribing ACR-368. The test will leverage the spatial phenotyping capabilities of their PhenoImager® platform to localize and quantify expression of the three clinically relevant protein biomarkers in the relevant biological regions within the tumor. This approach is similar to how our quantitative multiplex OncoSignature test measures biomarkers only in biologically relevant tumor regions, namely the tumor epithelium.

### Q: What is spatial biology and how is it being used in discovery and for development of diagnostics?

Brian McKelligon
CEO of Akoya Biosciences

**Brian McKelligon:** Despite the phrase "spatial biology" being relatively new, we've seen a massive acceleration of multi-marker tissue-based approaches and publications.

In discovery research studies, hundreds of markers are being simultaneously measured across tens of tissue samples with the goal of understanding the immense complexity of the tumor microenvironment and the spatial architecture of immune cells relative to each other and to cancer cells.

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When you move farther downstream towards translational and clinical applications, however, I would more aptly describe the approach as multiplex immunohistochemistry or multiplex immunofluorescence with protein being the dominant analyte measured in clinical trials. Here, relatively fewer markers will typically suffice, with each one potentially adding significantly more information. Typical studies focus on four to seven biomarkers across several hundred samples. The needs and platform requirements of a discovery researcher are quite different, therefore, from what Acrivon (and, more generally, other pharmaceutical companies) require for translational and then, ultimately, clinical applications. This is why Akoya has created a portfolio of solutions purpose built for these distinct customer segments. Discovery researchers leverage our PhenoCycler®-Fusion for high-plex and high-throughput discovery, and those interested in higher scale translational and clinical research applications use our PhenoImager HT system.

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Regardless of whether it's a 100-plex RNA and protein study or a three- to four-plex companion diagnostic, every step of the multiplex workflow, from the assay methods to image acquisition, must be done in a consistent manner. By creating this continuum, the biomarker journey from discovery to clinical use is enabled.

### Q: What excites you most about the collaboration with Acrivon?

**BM:** The Acrivon team is directly addressing the Phase II drug attrition problem where drugs that are well tolerated are not advanced since they fail to meet clinical endpoints in unselected patient populations. These drug candidates would potentially benefit from a biomarker strategy to enrich for responders in a subset of patients, but this is not always done in large pharmaceutical companies. Acrivon's biomarker discovery approach represents a convergence of expertise in multiplex spatial phenotyping from Akoya and Acrivon's expertise in biomarker discovery, validation, and clinical trial design. We see this collaboration as the tip of the iceberg for advancing companion diagnostics and the field of oncology as a whole. There are many potentially efficacious drugs, but if they can't be matched to the right patients, they'll never gain approval. That's the problem Acrivon seeks to solve, and what our platform enables.

We're excited about the possibility of additional partnerships in which our platforms and expertise can be used to define and advance emerging spatial biology–based approaches to biomarker discovery and validation in oncology and even non-malignant disease. In the field of immuno-oncology specifically, this strategy will support combination trials and enable the elucidation of predictive signatures in patient tumors where multiple drugs are directed at multiple targets, and those trials in which a single therapeutic, such as a bispecific, engages more than one target.

### Q: What do you see as the future for companion diagnostics in the cancer space?

**PB-J:** As I see it, pharma and biotech will continue to move towards proteomics-based methods to identify responders. This contrasts with genetics-based methods where the patient either has the genetic alteration or not; with only that information, you must infer and hypothesize which protein signaling pathways are driving the disease and also how the drug acts on these. With protein-based methods, you can basically measure the activity states of the pathways that are driving the disease, and you can directly measure and observe how the drug acts on these pathways. The industry is also recognizing the need to look at more than one biomarker at a time and hence be able to measure what the tumor depends on. You need functional orthogonality of biomarkers to be able to do that, as well as multi-marker tissue-based approaches.

This is the premise of what Acrivon is doing in collaboration with Akoya; the way we predict patient response is designed to be independent of underlying genetic alterations and prior therapies. The studies we have completed at Acrivon are with tumor biopsies from heavily pre-treated patients who therefore have all kinds of genetic alterations, but we simply look at the protein-based signature. Ten years from now, I believe proteomics-based technologies will have advanced sufficiently to become the gold standard for patient responder stratification for cancers in which genetics-based methods are insufficient. ■

**Damian Doherty** has been in media and publishing for nearly 30 years, beginning in the early nineties at News Corporation. Damian has managed, edited, and launched life science titles in drug discovery and precision medicine. He was features editor of *Drug Discovery World* for fourteen years and founded, established, and edited the *Journal of Precision Medicine* in 2014. In parallel, Damian founded and organized the Precision Medicine Leaders' Summit, a global, immersive 3-day senior leadership conference that still runs today. He edited *AIMed* magazine in 2019 before launching Photo51Media, a platform for illuminating untold, compelling stories in precision healthcare. Damian joined Mary Ann Liebert in 2021 to help steer the new rebrand and relaunch of *Clinical OMICS* to *Inside Precision Medicine*.