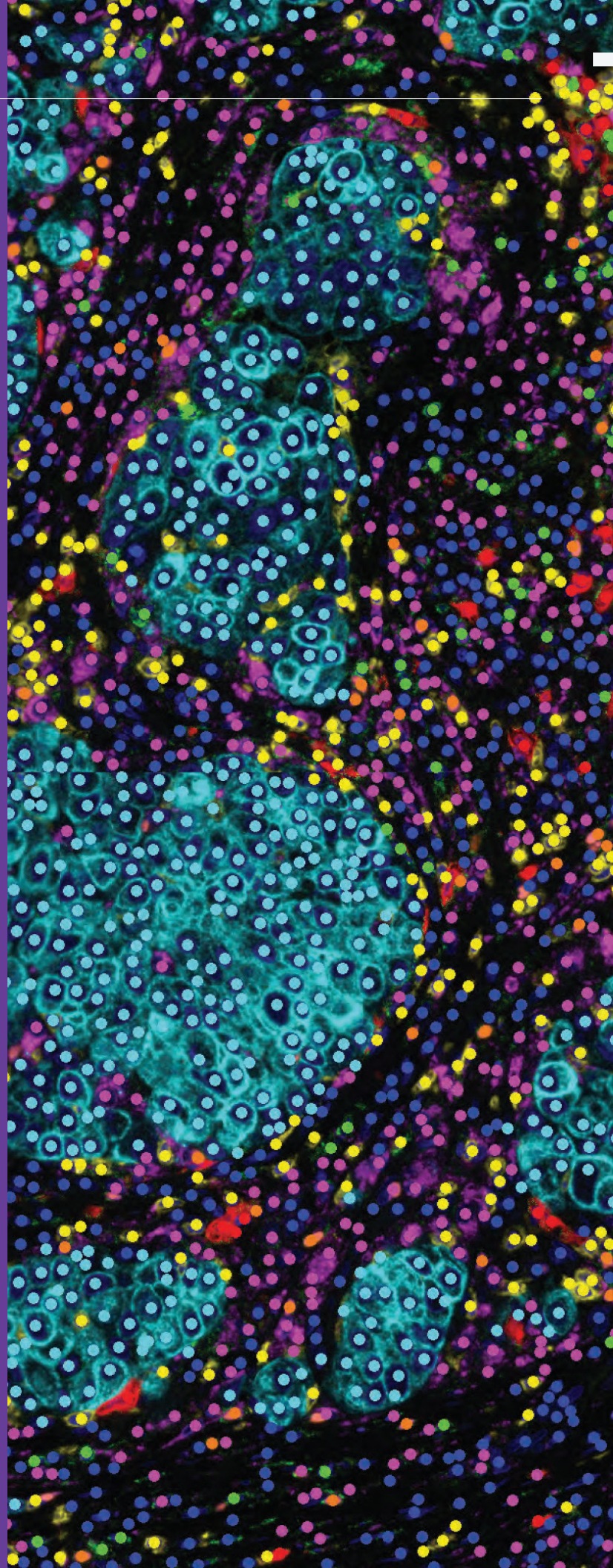


UNDERSTANDING STARTS
WHEN YOU PUT COMPLEXITY
INTO CONTEXT

THIS IS WHAT DISCOVERY LOOKS LIKE

Developing effective treatments and therapies requires an in-depth understanding of underlying disease mechanisms and biological responses, so it's critical that you see everything your tissue sample has to show you. But to get a complete picture can be challenging with so many complex biological interactions occurring simultaneously. That's what makes having a streamlined workflow a powerful solution. The Phenoptics™ workflow — multiplex immunohistochemistry staining solutions, multispectral imaging systems, and advanced image-analysis software — enables a more comprehensive and specific view and analysis of biological interactions across a digital slide, from the cellular level to the macroscopic tissue architecture, using a streamlined workflow. Better quantification of cellular interactions may reveal which disease mechanisms are in play, and help researchers discover biomarkers that may eventually lead to better subpopulation stratification methodologies. Simply put, that means a better understanding of biology that drives disease. And isn't that the goal we're all striving for?



UNDERSTANDING STARTS RIGHT HERE



Combining a powerful multispectral imaging system with multiplex IHC staining, and image analysis with inForm[®] software, the Phenoptics workflow enables you to identify cellular phenotypes, assess their functional states, and measure spatial relationships. Want a workflow that enables better understanding of the complex interplay between cells and the tissue architecture? **Let's get started...**

1. IMMUNOSTAIN

THE NINE COLORS OF DISCOVERY

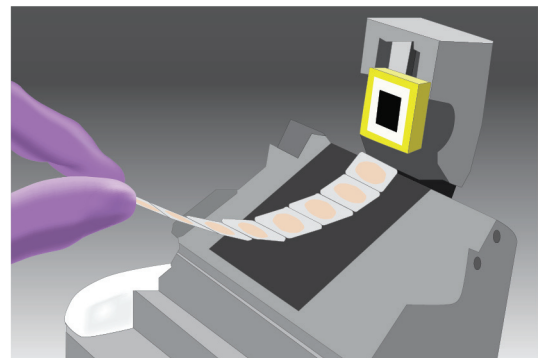
Opal Multiplex IHC kits make multiplex results accessible to anyone who works with standard immunohistochemistry, permitting enhanced visualization and understanding of complex cellular interactions. With Opal, you can select antibodies for simultaneous IHC detection based on performance rather than species. Opal kits are optimized for reliable spectral unmixing and simultaneous measurement of three to eight IHC targets, plus a nuclear stain.

Opal enables you to:

- Measure three to eight tissue biomarkers simultaneously
- Use the best primary antibodies, regardless of species — with no cross reactivity
- Identify multiple cell phenotypes while retaining spatial and morphological context that is lost with bulk measurements and flow cytometry
- Get more information from precious and scarce samples

With our **Opal Automation IHC kits** you can perform Opal multiplex staining on one of the leading research automated staining platforms — the BOND RX™ by Leica Biosystems. Automation provides you with the flexibility to support the dynamic demands of translational research.

- Quality, consistency, and reproducibility with every sample
- High-throughput protocol: perform seven-color immunofluorescence (IF) staining on 30 slides in 14 hours
- Simplified, walkaway protocol versus laborious manual process



Opal IHC works with FFPE tissue and is compatible with standard IHC workflows.



You can use the best primary antibodies together in multiplex panels, with no species-based crosstalk.



Because you retain spatial cellular context, you get more information from your precious samples.

2. IMAGE

REVEAL COMPLEX BIOLOGY IN A SINGLE TISSUE SECTION

For a deeper understanding of diseases, you need faster, better visualization and identification of disease biomarkers. And accelerating the pace of that understanding is the whole idea behind **Phenoptic™ imaging instruments**. These systems enable you to visualize, analyze, quantify, and phenotype cells in situ, in FFPE tissue sections, and TMAs. Proprietary multispectral imaging enables you to capture the multiple interactions occurring between cells because we've carefully unmixed each color from one another; while also isolating autofluorescence into its own color channel so you can easily exclude it from your digital slide analysis. That means you can have confidence in accurately quantifying the interactions that are really occurring in the biology.

Mantra Quantitative Pathology Workstation

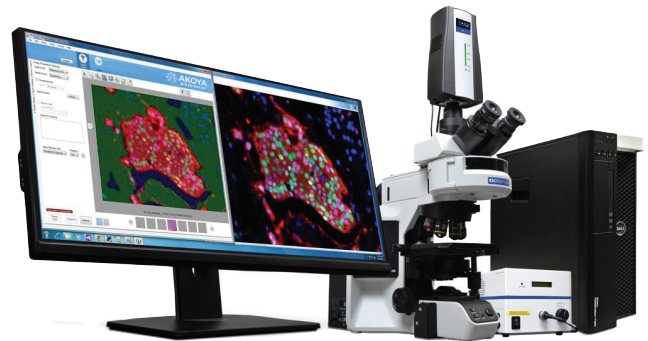
- Compact system ideal for getting started with multispectral imaging or for assay development to free up a higher throughput system such as Vectra Polaris
- Eyepiece for easy visual checking as you go
- Automated Brightfield and multichannel fluorescence (up to nine colors) capabilities
- Phenochart™ whole slide viewer for slide navigation provides context for identification of regions of interest within a digital pathology workflow

Vectra 3 Automated Quantitative Pathology Imaging System

- Detect and measure multiple weakly expressed and overlapping biomarkers within a single H&E, IHC, or IF tissue section and in TMAs
- Automatic identification of specific tissue types using integrated inForm® analysis software

Vectra Polaris Automated Quantitative Pathology Imaging System

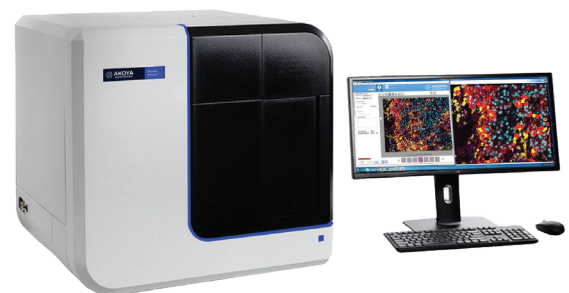
- Premier high-throughput imaging system for detecting and measuring multiple biomarkers within a single tissue section (up to nine colors)
- State-of-the-art whole slide multispectral imaging for unmixing of up to seven colors
- Fully automated digital slide scanning that can batch together brightfield with multispectral fluorescence
- Flexible data analysis, compatible with many image analysis software platforms



Mantra Quantitative Pathology Workstation



Vectra 3 Automated Quantitative Pathology Imaging System



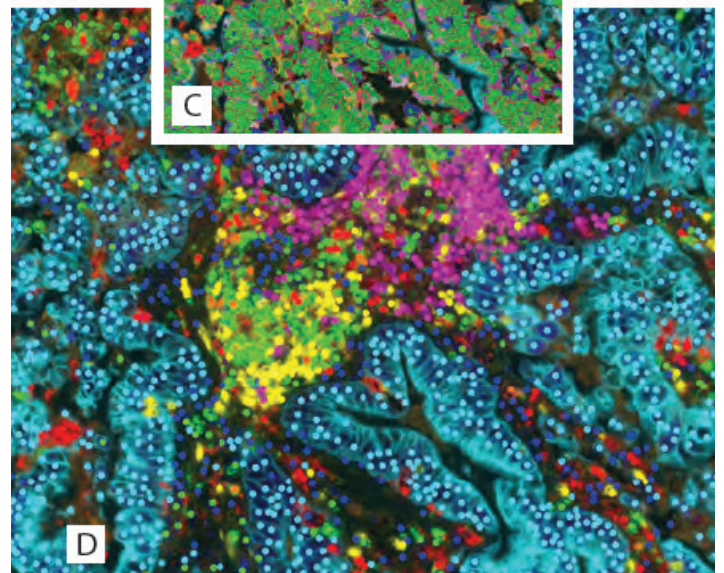
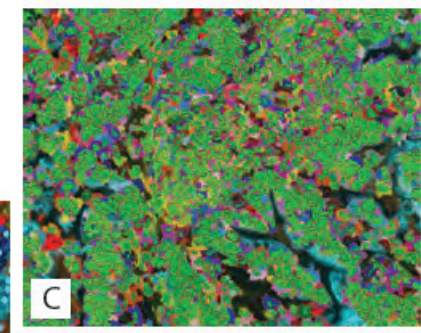
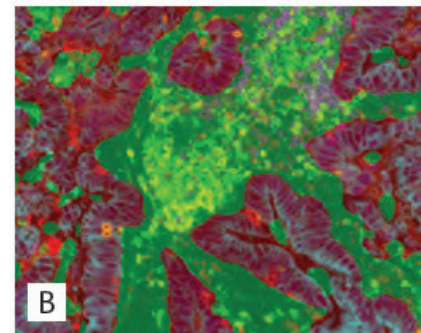
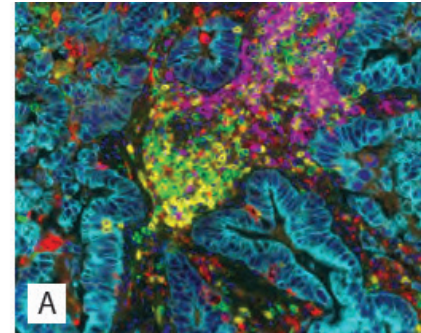
Vectra Polaris Automated Quantitative Pathology Imaging System

3. ANALYZE AND UNDERSTAND

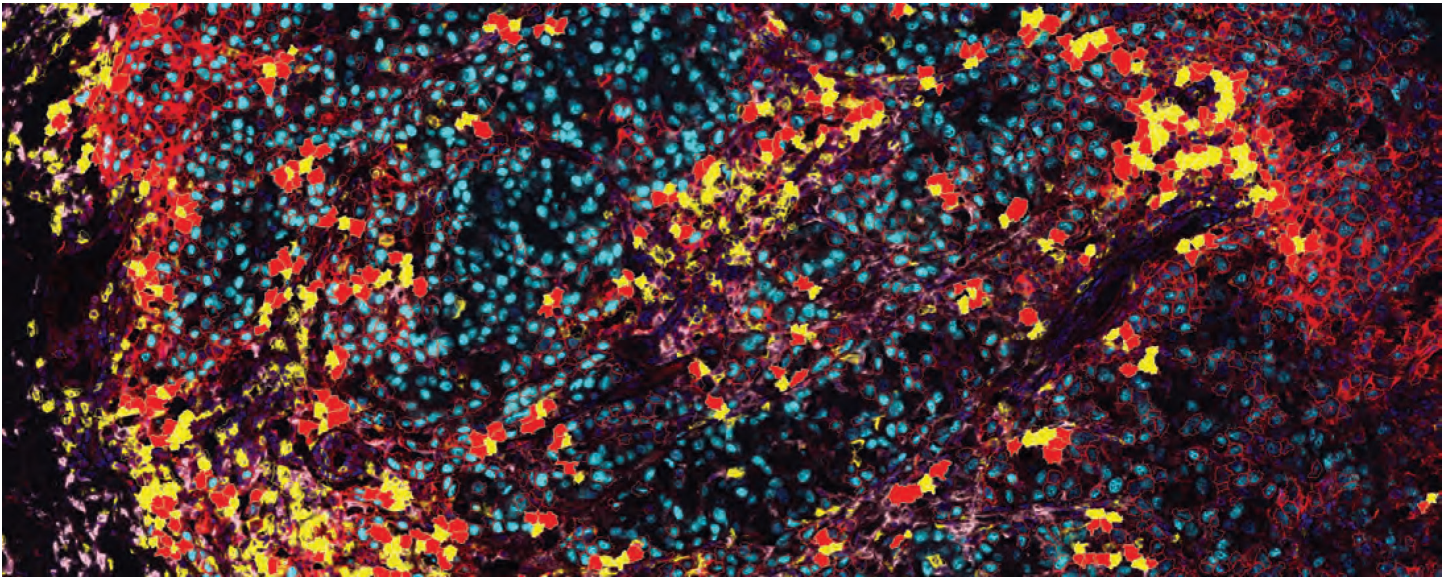
DISCOVERY COMES WITH SEEING CELL-TO-CELL INTERACTIONS

Our patented **inForm® image analysis software** allows you to accurately visualize, analyze, and quantify biomarkers in situ in solid tissue. Its powerful unmixing engine, enables the separation and quantification of weak and spectrally overlapping biomarkers within cells and cellular compartments that cannot be identified by the naked eye. Automated, trainable algorithms permit detection and segmentation of specific tissues and phenotyping of immune and other cells. Combined with powerful spatial analysis algorithms in **phenopt** and complex phenotypic analysis reporting tools in **phenoptReports**, these sensitive approaches give you the confidence to discover indicators of disease and uncover relationships between specific cell types and across the entire digital slide. Additional benefits include:

- Pathology Views™ renders immunofluorescence images as simulated H&E or DAB and hematoxylin, providing views more familiar to the pathologist
- Powerful unmixing algorithm enables identification and separation of weakly expressing and overlapping signals from background autofluorescence
- Enables per-cell analysis of H&E, IHC, IF, and RNA-ISH in FFPE tissue sections and TMAs
- Automatically classifies cell phenotypes using machinelearning algorithms
- Automated detection and segmentation of specific tissue types through patented pattern recognition algorithms
- Adaptive Cell Segmentation reliably identifies individual cell types in densely packed, complex morphologies regardless of staining heterogeneity and background levels
- Simplified whole-slide multispectral imaging workflow enables inForm analysis across the whole slide, removing selection bias



A) Spectral Unmixing; B) Tissue Segmentation; C) Cell Segmentation; D) Cell Phenotyping



Phenotyping of immune cells and cancer cells within the context of the tumor enables advanced analytics like distance mapping.

PHENOPTICS RESEARCH SERVICES

We take understanding to the next level

Want to test the Phenoptics workflow before bringing the capability in house? Let our expert research team generate the results for you. We follow a detailed staining protocol when working with your precious samples: Antibody specificity is first confirmed via monoplex with positive controls. Then the multiplex panel is tested with the same positive controls, with study samples that you provide — so you're confident the protocol works for you. Confirmation of performance levels, including multiplex staining independence and noninterference are then agreed upon. Analysis begins with multispectral imaging, providing quantitative spectral unmixing of each fluorophore signal and tissue autofluorescence, followed by tissue segmentation and cell phenotyping. This complete workflow enables new depths of understanding that cannot be achieved with standard chromogenic monoplex or duplex IHC methods.



Phenoptics Research Services Laboratory

To learn more visit [AKOYABIO.COM](https://www.akoyabio.com)
or email us at INFO@AKOYABIO.COM