Agilent Microarray Scanner and Feature Extraction Software
Express it your way.

Agilent’s flexible printed microarray solutions give you the freedom to drive your research where you want it to go—getting you miles ahead of the competition. You can express it your way—starting with Agilent’s high-performance microarray scanner and expand later to include other parts of Agilent’s gene expression analysis products and services.

Agilent’s gene expression analysis platform has been developed around the flexible, industry-standard 1” x 3” (25mm x 75mm) glass slide microarray format. Agilent is committed to enabling more researchers with high-quality, open and accessible microarray-based research tools. This foundation has resulted in an integrated suite of products spanning printed microarrays and reagents, microarray processing tools and sophisticated analysis software platforms. Together, they create a system that grows with your needs.
The Agilent Microarray Scanner at a glance.

- SureScan technology for higher sensitivity scanning with minimal interaction
- Tight resolution (5 micron) scanning and data acquisition
- Feature Extraction software with built-in error models
- Flexible 1" x 3" (25mm x 75mm) glass format
- 48-position slide carousel for walk-away ease
- Peace-of-mind operation backed by Agilent’s worldwide service and support
- Rugged optics and optical bench for optimum scans
- Dual-laser scanning for 2-color microarray formats
Performance. Experience it.

Agilent’s been in the microarray scanning business for years. Starting in 1997, Agilent, then Hewlett-Packard, created the GeneArray Scanner to enable Affymetrix® to read higher density probe array formats while delivering more robust scanning performance. Now Agilent takes microarray scanning to new heights with its second-generation platform that enables industry-standard 1” x 3” (25mm x 75mm) glass slide formats.

The Agilent Microarray Scanner is a product of advanced engineering and performance—the perfect combination of elegance and power. Today, top research labs have scrapped older scanning technologies for Agilent’s high performance microarray scanner. They’ve experienced greater sensitivity, more robust scans and higher confidence data that are hallmarks of Agilent’s Microarray Scanner. It’s all about data…and Agilent delivers.
Performance.
Painting the picture with data.

Comparative analysis proves that Agilent’s Microarray Scanner scans glass slides more accurately
To demonstrate the sensitivity and accuracy of Agilent’s scanner compared to 2 other commercially-available 1” x 3” laser-based single-slide scanner, Agilent performed an in-depth study that was presented at the 2003 Associated Biomolecular Research Facility (ABRF) meeting. The results of this experiment are presented here. The experimental goals sought to measure the performance parameters of the microarray scanners as independently as possible from other experimental variables. To achieve this, a method of immediate, repeated scans of the same microarray. All measures of noise/variability are then defined as the standard deviation(s) in the results for any given feature between the repeated scans. This method excludes effects of microarray fabrication, sample labeling, hybridization/washing, and biological inputs. Three nominally identical microarrays were used and 8 repeated scans of each microarray were performed on each scanner. For each microarray, the scanner order was varied, to eliminate bias due to ordering of scanners.

Replicate Variability:
This group of figures shows the effects of different scanners on the total experimental error of a microarray experiment. The 3 microarrays each had 100 probes that were replicated in 10 positions on the microarray. For a perfect experiment, all 300 replicates (10 each from 3 microarrays) should all report the same log(Ratio).

The combined plot shows the absolute value of the average log(Ratio) for each probe on each microarray minus 3 times the standard deviation. When this value is above 0, we can call the probe differentially expressed to a confidence of better than 3σ. The Agilent Microarray Scanner sees all 15 data points as significantly differentially expressed. Brand Y only 8/15. Brand Z identifies none of the 15 probes as significantly differentially expressed. It should be noted that the combined inter-array and intra-array σ(log(Ratio)) of 0.05 is achieved using the combined Agilent system (oligo microarrays, sample labeling, hybridization, scanner, feature extraction).
Performance.
Picture getting better data with Agilent.

Picture perfect scanner performance...only from Agilent
To test the performance characteristics of three commercial 1” x 3” (25mm x 75mm) glass slide microarray scanners (Agilent, Brand Y and Brand Z), a two color scan image of the same weak feature, within the same area of microarray #1, was scanned on each microarray scanner. The color scale for all images is logarithmic with black and brightest pixels representing those pixels outside of 1% and 99% of the distribution, respectively. Gain settings for the Brand Y and Brand Z scanners were set to be close to that of the Agilent Microarray Scanner. Brand Y scanner’s gain was set successfully within ~3% of Agilent’s scanner. Brand Z scanner’s gain was ~1.2 times that of Agilent’s Microarray Scanner.

The graphic below shows a ~3 x 3 mm section of the microarray scan with ~120 mm features, scanned on all three microarray scanners. Note the increased noise visible when comparing Agilent’s Microarray Scanner to Brand Y and Brand Z. The white outlined arrow shows the approximate location where a cut of the background is done. In the upper panel the red background counts are plotted versus pixel column along with the mean and σ of the background. Again, the difference in the noise on the background is apparent. For Brand Z, the stated σ =118 is 2.2 times larger than the measured σ because 45% of the background pixels are censored at zero counts. The same weak feature is outlined with a white box and expanded in the lower panel for each of the three scanners. Notice that the feature is clearly distinguished on the Agilent Microarray Scanner, but is barely distinguishable on the Brand Y scan, and is not detectable at all on the Brand Z scan. The signal level of the feature shown is 8 counts and 58 counts (over background) in the red and green channels, respectively, on the Agilent Microarray Scanner. Bottomline, you’re depending on your microarrays to help you discover more in your research...so discover more on the spot with Agilent’s Microarray Scanner with SureScan technology.

Discover what you’ve been missing in microarray scanning.
The Agilent Microarray Scanner is able to detect the low abundant genes or weak features, where other scanner brands (Y and Z) fail when analyzing the same weak feature from the same area of the microarray being scanned.
Minimize microarray variability while measuring more sensitively with the Agilent Microarray Scanner with SureScan technology.

This figure clearly demonstrates the robust scanning capability inherent to the Agilent Microarray Scanner. This histogram plot represents the feature standard deviation of Log(Ratio). The most common $\sigma(\text{Log(Ratio)})$ for the Agilent Microarray Scanner is 0.007. This allows for a 1.05x differentially expressed gene to be measured with a confidence interval of $3\sigma$. Brand Y has a $\sigma(\text{Log(Ratio)})$ mode = 0.03, or ~1.23x minimum differential expression. Brand Z has a $\sigma(\text{Log(Ratio)})$ mode = 0.11, or ~2.14x minimum differential expression. This figure clearly shows the impact of the scanner’s S/N on the experimental error bars. The $\sigma(\text{Log(Ratio)})$ from the scanner allows for a lower limit on the minimum detectable differential expression in your microarray experiment.

Low cross talk

Agilent’s Microarray Scanner demonstrates minimal crosstalk—less than 0.1% for each channel. In the plots below, the intensity of the last two features were assumed to be exactly 65,535, contributing to a signal that was no more than 20 counts (or 0.05% crosstalk).

This study clearly demonstrates that if a microarray feature (spot) becomes saturated in the red channel, good measurement in the green channel will result. The same holds true for a feature saturated in the green channel.
Performance + Productivity.
Walk away from scanning. Return with confidence.

- Liberate yourself from single-slide scanning
- Make more time for yourself and your research
- Gain convenience—scan Agilent 60-mer oligo microarrays with other 1" x 3" formats in the same run

Completely or partially fill the 48-slide “load ‘n’ go” carousel for scanning. Either way, you’ll save time.

It’s easy to run almost any 1” x 3” (25mm x 75mm) microarray glass slide on the Agilent Microarray Scanner. Load your microarray slide, lock it into place in the secure slide holder and place in the carousel for convenient, walk-away scanning.

Agilent’s unique slide holder provides added protection against slide breakage. It accommodates Agilent 60-mer oligo microarray slides and most 1” x 3” glass slides from other sources. Load up the 48-slide “load ‘n’ go” carousel and be off to the races.

Agilent’s Microarray Scanner reads most industry standard barcode formats. An added advantage to using Agilent’s 60-mer oligo microarrays is the seamless linkage of the barcode to the scan data to the microarray design files found on the CD that comes with each Agilent microarray kit. This facilitates “hands off,” instantaneous feature information linking to the feature extraction data produced from each Agilent microarray being scanned.
Performance. Personalized.
Scan it your way.

Agilent designed its scanner to deliver added performance through a more flexible and open set of graphical user interfaces (GUIs). These value-add enhancements accommodate most 1” x 3” glass slide microarray formats—making this robust microarray scanner the smart choice for labs that desire 1” x 3” format freedom and quality scans they can count on.

Experience the ease of slot specific scanner control software which enables the input of microarray-specific scanning parameters for different microarrays to be scanned in a given run.

Customize microarray scan regions for non-Agilent 1” x 3” microarrays—making it simple to run microarrays of your choice while reducing scanning time and data storage requirements.

Webify your scanning. View scan progress remotely via the web…pick up completed scans and kick-start your data analysis.

Ensure laser longevity with Agilent’s laser saver feature. Now you can program the time when you want the lasers turned on and turned off. This increases laser life while reducing the cost of instrument maintenance.
Precision.
Agilent SureScan technology. Scan it and be sure about it.

Each Agilent Microarray Scanner comes equipped with Agilent SureScan technology—delivering precision microarray scanning for optimum results. SureScan technology is the result of Agilent’s design and engineering excellence—known for industry firsts that include “optical switches” and the “enabling technology” driving today’s optical mouse.

Agilent SureScan technology is comprised of many components that work together to deliver increased precision and ease-of-use. Core elements include 1:1 PMT Adjustment capability, automatic Laser Power Control to eliminate laser drift and the industry-first Dynamic Autofocus feature that enhances scanning sensitivity and data reliability.

![Dynamic Autofocus Comparison](image)

SureScan enabled.
See what you’ve been missing.

**Dynamic Autofocus**

Even the best glass manufacturer can’t produce a glass slide without some curvature or changes in glass thickness. These variations can cause problems with microarray scanner laser focusing. Many scanner manufacturers have addressed this problem by incorporating “single focusing” which only focuses on one area of the glass slide. This may not correct for potential glass curvature and tilt or other aberrations across the rest of the slide surface, resulting in reduced scanning efficiency and sensitivity.

Agilent combats this problem with its industry-first Dynamic Autofocus feature. This unique capability increases scanning sensitivity by continually focusing and re-focusing up to over 1 million times during the scanning process. This continual adjustment of the focal plane corrects for common glass abnormalities and gradients as well as any potential slide movement within the scanner. Bottom line, you can be sure about your scans with this differentiating dynamic autofocus feature—available only from Agilent.
1:1 PMT Control and Adjustment
Researchers will appreciate the flexibility to adjust the PMT in the Agilent Microarray Scanner knowing that reducing PMT voltage causes a commensurate reduction in background signal. This PMT Control feature correlates the PMT sensitivity level 1-to-1 with the signal level of the scanned image file. This important feature helps eliminate time-consuming pre-scanning and adjusting of PMT levels common with other commercial scanners. The scanner is also optimized for high-signal-to-noise performance in the channels for cyanine 3 (550-610 nm) and cyanine 5 (650-750 nm), with a wide dynamic range and low crosstalk. This feature allows measurement of a very broad range of gene expression levels for higher data confidence at lower signal levels.

Automatic Laser Power Control
Agilent designed its microarray scanner with a unique external laser power modulation feature on its green SH-YAG (532 nm) and red HeNe (633 nm) lasers to continuously compensate for short-term laser fluctuations. Unlike many laser power control features that can take minutes to adjust, Agilent’s laser stability mechanism adjusts instantaneously for more reliable scanning data. This laser power control feature allows Agilent to calibrate its microarray scanners relative to one another in the factory, where red and green signal intensities must agree within a +/- 20% maximum range between different scanners. Furthermore, the red and green signal intensity ratio must agree within 5% RMS on average across the same features on the slides, from scanner to scanner. This attribute allows researchers to more confidently share data between labs using Agilent’s Microarray Scanner.

The rugged, built-in optical bench adds stability during scanning, maintaining critical alignment for the most reliable scans possible.
Perfection. Agilent Feature Extraction Software.
Picture-perfect data for more confident downstream analysis.

Researchers who run microarray experiments know that image analysis and data extraction processes can be daunting—at times yielding data that can send them in the wrong direction. Until now.

Feature-rich and power-packed, Agilent’s Feature Extraction software provides more informative data so you can be sure that your research is heading in the right direction. No other feature extraction software package offers the combined benefits of sophisticated analysis algorithms plus built-in error models and more.

Extract data from an Agilent 22K microarray in less than 2 minutes

LowESS normalization algorithm to standardize your microarray analysis

Automatic feature extraction of Agilent microarrays and linking to microarray design file information facilitates data analysis

Outlier identification and local background correction for streamlined data analysis

Example images to facilitate learning and software qualification

Built-in error models—identify the best data to analyze using this powerful program

Advanced statistics to correct for biological, chemical and instrument noise
Perfection + Productivity.
Analyze and feature extract Agilent 60-mer oligo microarrays in minutes.

Agilent’s Feature Extraction software is used to qualify Agilent 60-mer oligo microarrays during the fabrication process. The data accumulated using Agilent’s sophisticated Feature Extraction software helps Agilent produce microarrays with high-quality features, for more consistent results. The design file is included on a CD that is shipped with each respective batch of microarrays. This information is linked to a unique barcode on each Agilent microarray which is seamlessly tracked during the scanning and analysis process—resulting in an instantaneous linkage to feature information generated during the feature extraction process.

Minimize false negatives and false positives while maximizing the collection of useful data
- Automatic feature finding saves time while eliminating user technique dependencies
- Smart cookie cutter algorithm that allows for statistical input as well as radius manipulation for local background manipulation
- Simple polynomial outlier flagging method lets you pick how to best flag features or local backgrounds that may lead to misleading data
- Flexible background subtraction method based on multiple user-selectable background techniques
- 2-channel background adjustment for added confidence and ease of use
- Sophisticated dye normalization with flexible options to include all significant features, list of normalized genes or rank consistent features
- Flexibility to use LOWESS or linear fitting

Utilize advanced statistical analysis with "minimal" intervention
- User-defined p-values for added data confidence
- Enhanced feature and background uniformity through outlier pixel analysis that removes pixels if their signal intensity falls outside a given statistical region
- Feature flagging to identify any feature affected by microarray imperfection such as bubbles, scratches, contaminants and abnormalities
- Pixel and background statistical errors are propagated for each feature. Statistical log ratio values enable you to evaluate the confidence of your data for every feature

Generate flexible data output options
- Tab-delimited text file (.txt)
- GEML-based (.xml) format tailored for the Rosetta Resolver® and Luminator™ Gene Expression Data Analysis Systems available only from Agilent
- “Shapes” file (.shp) which stores visual annotation of the feature extracted scan image

Automatic feature extraction ...in under 1 minute
Productivity + Performance for ANY 1” x 3” microarray
Precision feature extraction for the masses.

Now you can take advantage of a complete, integrated scanning platform that offers you the capability of walkaway scanning for any mix of 1” x 3” microarrays combined with the power-packed feature extraction software you’ve been waiting for. Agilent’s Feature Extraction software now offers the capability of feature extracting most 1” x 3” microarrays that have been scanned on the Agilent Microarray Scanner. A host of productivity-enhancing and user-friendly software features have been added to make data extraction a breeze for non-Agilent, 1” x 3” microarrays.

Making grid alignment a snap.
Agilent’s user-friendly grid alignment GUI and tools make grid alignment simple. Another industry-first is the unique, dual-view zones that allow you to easily toggle across the microarray and view the alignment at a more granular level in the windows below. This removes the hassle of toggling between windows and saves time while reducing frustration for the user.
**Productivity right to the end**
Enhanced data flow tools get you to the next step faster.

Microarrays produce vast amounts of data which can be cumbersome to load into downstream gene expression data analysis systems. Agilent has removed many of the hurdles in getting data into such systems by offering time-saving data export tools such as FTP-capabilities and MAGE-ML support functionality.

![Rosetta gene expression data analysis system users will appreciate the FTP function and capability to choose the data format for export (GEML or MAGE, JPEG or TIFF).](image)
Agilent Feature Extraction Software.
Perfection. Plus a whole lot more.

No other commercial microarray analysis software package can match the power of Agilent’s Feature Extraction software. Here’s just a glimpse of our graphical user interfaces (GUIs) designed to make feature extraction a more enabling tool for our users.

Agilent provides built-in error models
Error models come standard with Agilent Feature Extraction software—providing more confident results while saving time and money.

Choose from:

Propagated error model—good estimation of instrumentation noise and error from printing or hybridization processes

Universal error model—a good estimator of error in high intensity features, representing the expected error of the difference between the red channel and the green channel.

Hybrid error model—evaluates the propagated error model and the universal error model and reports the error based on the higher p-value of the two models.
Agilent’s algorithm offers unparalleled outlier rejection and background pixel correction.
Incorporate better outlier rejection and background pixel correction resulting in more robust data for downstream analysis.

Background unadjusted.

Automatically adjusted background.
Agilent’s background adjustment algorithm first finds the central tendency of the background-subtracted data (features shown as blue circles in the features). Using this subset of features, the algorithm then estimates the best adjustment in both red and the green channels to remove the bias. After using Agilent’s background adjustment and dye normalization, the bias is removed and the plot is linear and symmetrical (self vs. self analysis.)
Agilent Feature Extraction Software.
Perfection. Personalized.

Use advanced statistics to account for biological and instrument noise. Polynomial outlier flagger option allows users to select and reject certain microarray data (such as non-uniform features) on a statistical basis.

Get the most advanced normalization algorithm and Agilent’s exclusive rank consistency probe selection. Normalize data using the well-respected LOWESS algorithm—made available with Agilent’s Feature Extraction software.

Agilent provides visual gene and quality annotations. Simple “mouse-over” visualization of gene signal, log ratio and quality annotations that link to feature information found on the CD shipped with each Agilent 60-mer oligo microarray.

View the data the way you want it. Agilent provides flexible data viewing options.
Performance. Precision.
Perfection in Microarray Scanning and Analysis.
Backed by Agilent’s world class service and support organization.

Pharmaceutical companies worldwide have come to rely on Agilent’s service and support to keep their development and QA/OC efforts on track. Agilent brings its know-how and service record to bear on the gene expression market as well by delivering world class training, service and support. You can trust Agilent to be there every step of the way, from installation and set-up to on-site and remote access repair as well as friendly and accessible telephone support.

Get the right start today.

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Discover more on the web
Be sure to visit Agilent’s gene expression website for up-to-date product, application and support information.
www.agilent.com/chem/dna

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www.agilent.com/chem/GEnotes

Need a lab to run your microarrays?
If you are interested in accessing 60-mer oligo microarrays but need a lab to run them for you, be sure to check out one of Agilent’s Certified Service Provider Laboratories.
www.agilent.com/chem/csp

Special promotions
From time to time, Agilent will offer special promotions or incentives for some of its gene expression products and solutions. By signing up for e-notes, you’ll receive monthly updates on these and other Agilent promotions per your application of interest. View the most up-to-date list of promotions at:
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