

GENORAMA[®] Genotyping Software

Genorama[™] Genotyping Software is designed for analysis of microarray-based resequencing or genotyping images.

The software can handle images from any 4-color microarray imaging system (e.g. Genorama[™] QuattroImager) and enables a kilobase readout simultaneously from both DNA strands or analyses up to thousands of mutations (SNP markers) in a single window.



MAIN FEATURES

Different color scales (grayscale and a number of pseudocolor palettes)

Grayscale images only have an 8 bit visible scale on screen. Due to this reduction of scale it is sometimes difficult to get all the visual information from microarray images having 12 to 16 bit pixel depth. The scale can be greatly enlarged by implementing different pseudocolor palettes: Plasma palette (black-red-yellow-white), Rainbow palette (black-all rainbow colors from blue to red plus white for maximum intensities) and GreenToRed palette (from green to red).

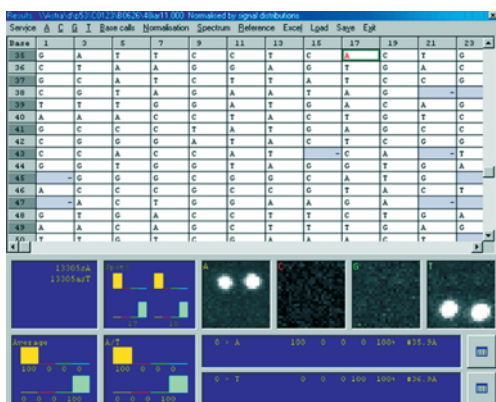
Assisted grid setup, automatic alignment according to the signals on microarray images

The software remembers the exact location of the grid from the last images analyzed and places the grid with the same size to the same location onto newly analyzed images. The grids can be also manually placed with a few mouse clicks. The sub-grid is an option to move a part of the grid independent of the rest of it. While the images are quantified, the computer will automatically perform fine alignment of the grid positions according to the location of signals.

Automatic spot identification, adaptive quantification and artifact filtering

The software identifies only round-shaped signals adapting the quantification to pixel intensities and diameter of each spot allowing each signal to be quantified independent of the size variations. Along with the automatic grid alignment feature it gives precise quantitative data. Signals with all other shapes are treated as artifacts.

The images can be analyzed by sample (one set of four images corresponding to the sample) or by SNP-s / mutations (one particular position from each set of images through all the samples to be analyzed).



Signal intensities of four images are compared and automatically translated into the presence of a nucleotide(s) on the image and analyzed sample.

FEATURES FOR ANALYSIS BY SINGLE SAMPLES

Automatic base calling

Signal intensities of four images are compared and automatically translated into the presence of a nucleotide(s) on the image and analyzed sample.

Histograms of relative signal intensities in different imaging channels

Histograms of signal values from single spots and the average value of signals are shown in the same analysis window. The highest signal will get a value of 100% and the others are compared to it.

Comparisons with wild type or reference sample

The software can compare data from analyzed images with those of wild type or reference sample and indicate all differences. This feature is useful in APEX-based resequencing assays or analysis of somatic mutations with a variable content of mutated target sequences.

Highlighting dubious positions

Genorama™ genotyping software prompts for manual analysis of all ambiguous base calls, all ambiguous positions are colored blue. This feature includes a number of variable options for recognition of the dubious base calls.

Different normalization methods of signal intensities between A, C, G and T channels

All signals are quantified in actual scale by default. However, the set of different normalization methods can be applied to minimize the signal variations due to variations in the slide processing and imaging steps.

Various formats for saving the results

The software results are presented in table format and can be saved in multiple formats: text table (Genorama base calls, *.grb), tables of signal intensities (Genorama results table, *.grs), table of notes (Genorama notes, *.grn). All the results can be exported to MS Excel in various selected formats.

FEATURES FOR ANALYSIS BY SNP-S / MUTATIONS

Image database function and comparison of multiple images

The software creates a database of microarray images corresponding to each position on the grid. This facilitates comparison of all the array positions at all samples to be analyzed in visual as well as different quantitative formats.

Absolute and relative color scales of images, grayscale and pseudocolor visualization

Absolute scale transforms the full 8, 12, 14 and 16 bit scale images to the 8bit grayscale or 24bit pseudocolor scale on the computer screen. This gives a good estimate of relative signal intensities on the database, but does not show very weak signals. A choice of relative scales will visualize all signals, including the weakest, but gives less quantitative scale on the computer screen.



Image filter

A very useful feature for images with a non-homogenous background. This option recalculates the values of each pixel taking the mean from a group of pixels for image display.

Elimination of single bright pixels

If the signal value of one pixel is several times higher than all the other signals around it (for example bright pixels of CCD chip). These bright pixels may affect the relative scale and mask possible true signals. Genorama automatically eliminates all these pixels without affecting the visible scale of the image shown.

Genotyping by groups

Scatter-plots of the signal intensities are shown in a number of graph formats allowing for manual selection of the genotype clusters and base calling by the grouped data. Different groups (homozygotes, heterozygotes) can be selected and analyzed separately.

Real time comparison of all the array positions at all samples to be analyzed in visual as well as different quantitative formats.