ImageQualityQuantificator
Interactive Spot Quality Evaluation of Microarray Images

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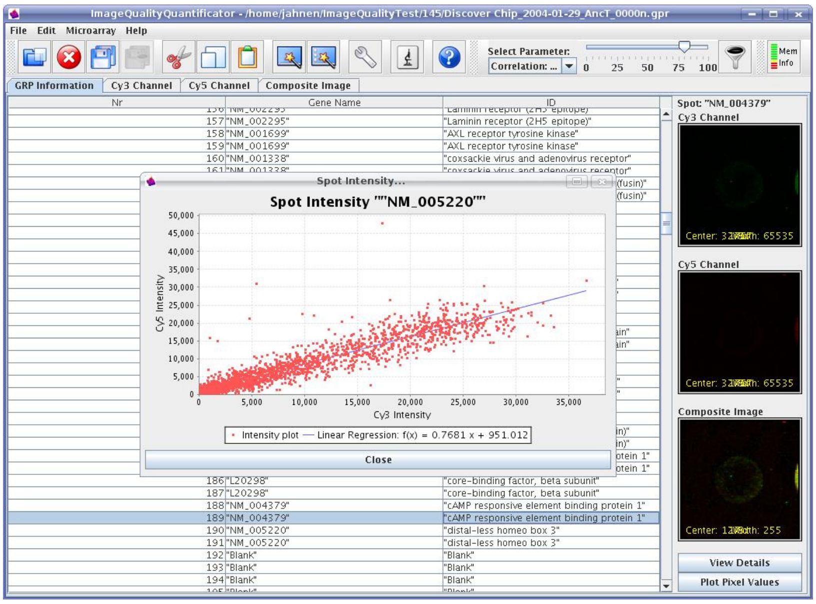
The software is available as Open Source at: http://santec.tudor.lu/bioinfo/imaging/

Abstract:

Introduction

Microarray data enable the fast and versatile exploration of genome structure, gene expression and gene function. Nevertheless the process of carrying out high quality microarray experiments is still difficult. We implemented a free and Open Source software called ImageQualityQuantificator (figure 1) that uses an interactive approach to evaluate the quality of spots from two-colour microarray experiments, based on several quality parameters.



Nr	Gene Name	ID
157	"NM_002295"	"Laminin receptor (2H5 epitope)"
158	"NM_001699"	"AXL receptor tyrosine kinase"
159	"NM_001699"	"AXL receptor tyrosine kinase"
160	"NM_001338"	"coxsackie virus and adenovirus receptor"
186	"L20298"	"core-binding factor, beta subunit"
187	"L20298"	"core-binding factor, beta subunit"
188	"NM_004379"	"cAMP responsive element binding protein 1"
189	"NM_004379"	"cAMP responsive element binding protein 1"
190	"NM_005220"	"distal-less homeo box 3"
191	"NM_005220"	"distal-less homeo box 3"
192	"Blank"	"Blank"
193	"Blank"	"Blank"
194	"Blank"	"Blank"

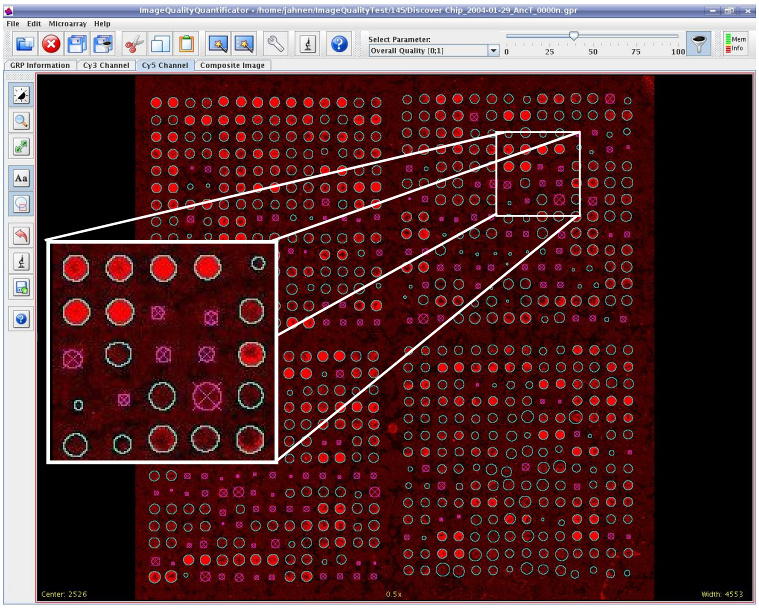
Figure 1: The software visualises the Genepix Results as well as the Images in an enhanced Graphical User Interface

Material and Methods

The software was implemented as a “stand-alone” application using Java Technology and ImageJ-based image processing [1]. As input, ImageQualityQuantificator uses conventional two-colour microarray image files in *.tiff format together with the corresponding primary analysis .gpr files generated by gridding the images using the GenePix Pro 6 software (Axon). The quality of each individual spot within the image is evaluated, and data from good quality spots are saved in an extended version of *.gpr file format. The relevance of several parameters described in the literature to quantify spot quality [2-4] was evaluated. Among them, the correlation of mean foreground signal (F), correlation of median F and linear regression (R^2) were identified as the most informative parameters and were implemented in the ImageQualityQuantificator software. In addition, an overall quality score is calculated, which is a weighted combination of the selected quality parameters. The impact of the single parameter or weighting respectively can be configured by the user, depending on his individual needs. Additional parameters are in development. Furthermore, several software applications for quality estimations or microarray image processing have been tested (e.g. MASQT, RealSpot, Spotfinder), in particular with regard to user-friendliness and an intuitive usability. The results and approaches for improvement have been considered while implementing the software.

Results

The software provides an easy-to-use graphical interface, which provides an intuitive possibility to compute and visualize each of these spot quality parameters. Using an interactive filtering procedure, for which the user can adapt the threshold by him a priori, ImageQualityQuantificator offers a comfortable possibility to select and visualize a subset of spots. The export to an extended GPR file which only contains the currently selected spots (figure 2) allows further, successive statistical analysis of the data.



Select Parameter: Overall Quality [0;1]

Figure 2: The visualisation of the filtered results allows a quick overview over the quality of the spots

Conclusion

Before any statistical analysis of data can be done, a time-consuming but essential step of initial data quality control should be carried out. The ImageQualityQuantificator supports this step by assisting the microarray spot evaluation process. The software presented allows an interactive and intuitive calculation and visualization of microarray spot quality. Quality parameters currently supported are the Correlation of Mean F, Correlation of median F and Linear Regression (R^2). Additional parameters should be incorporated in the next future.

References

[1] **M.D. Abramoff, P.J. Magelhaes,S.J. Ram**: *Image Processing with ImageJ,* Biophotonics International, volume 11, issue 7, pp. 36-42, 2004.

[2] **E. Novikov, E. Barillot:** *An algorithm for automatic evaluation of the spot quality in two-color DNA microarray experiments*, BMC Bioinformatics 2005, 6:293, doi:10.1186/1471-2105-6-293

[3] **M.R. Fielden, R.G. Halgreen, E. Dere and T.R. Zacharewski:** *GP3: GenePix post-processing programme for automated analysis of raw microarray data*, Bioinformatics vol. 18, no. 5, 2002

[4] **X. Wang, S. Ghosh and S-W Guo:** *Quantitative quality control in microarray image processing and data acquisition*, Nucleic Acid Research, 2001, vol. 29, no 15, 200