Description of affyILM package

K. Myriam Kroll, Fabrice Berger, Gerard Barkema and Enrico Carlon

October 18, 2010

Contents

1 Introduction

affyILM is a preprocessing tool which estimates gene expression levels for Affymetrix Gene Expression Chips. The computation is divided into two parts:

1. Background estimation for each feature based on input from physical chemistry (nearest neighbor model) as well as on the physical location of the features and their neighboring probes.

2. Computation of gene expression levels from background-subtracted data using the Langmuir model. In contrast to other measures, this method outputs the gene expression level as concentrations measured in *pM* (picoMolar).

A linear function with 50 parameters is used to describe the background intensity for each probe. The linear least square method is used to determine these 50 parameters. In order to obtain these parameters, *affyILM* uses a standard SVD (Singular Value Decomposition) algorithm based on Golub&Reinsch (see e.g. Golub and Reinsch (1970); Golub and Loan (1996)).

18 of the 50 parameters reflect the influence of the neighboring spots on the background intensity of a particular feature. The next 16 parameters incorporate the nearest-neighbor free energies (sequence dependence) and the last 16 parameters modify the strength of the sequence dependence based upon the position of a nucleotide along the

sequence. For a more detailed description on the theoretical background of *affyILM* we refer the interested reader to Kroll et al. (2008, 2009).

affyILM allows the user to simultaneously read-in several CEL-files; it does *not* require raw data (CEL-files) to be specifically formatted like e.g. as *AffyBatch*. In case more than one CEL-file is analyzed, a simple *in-between* array normalization is done such that the median values of intensities of the CEL-files are identical.

2 Getting started

2.1 Preliminaries

To build the package from source, you have to have the following components installed on your system:

- a C compiler
- GNU Scientific Library (GSL version 1.12)
- Basic Linear Algebra Subprograms (BLAS)

affyILM makes use of a few routines of the GNU Scientific Library (GSL), a software library which is freely distributed under the GNU General Public license and can be downloaded at `http://www.gnu.org/software/gsl/`.

GSL requires a BLAS (Basic Linear Algebra Subprograms) library for basic vector and matrix operations. We recommend the user to replace the default BLAS library (supplied with GSL) by ATLAS (Automatically Tuned Linear Algebra Software), an optimized BLAS version which is freely available under `http://math-atlas.sourceforge.net`. A list of optimized BLAS libraries for a variety of computer architectures can be found here: `http://www.netlib.org/blas/faq.html#5` For instance, Mac users may use the built-in vecLib framework, while users of Intel machines may use the Math Kernel Library (MKL). A C compiler is needed to build the package as the core of the *affyILM* function is coded in C.

For the package to be installed properly you might have to type the following command before installation:

```
export LD_LIBRARY_PATH='/path/to/GSL/:/path/to/BLAS/':$LD_LIBRARY_PATH
```

which will tell **R** where your GSL and BLAS libraries (see below for more details about BLAS libraries) are. Note that this might have already been configured on your system, so you might not have to do so.
In case you need to do it, you might consider copying and pasting the line in your `.bashrc` so that you do not have to do it every time.

Now you are ready to install the package:

```
R CMD INSTALL affyILM_x.y.z.tar.gz
```

The package will look for a BLAS library on your system, and by default it will choose gslcblas, which is not optimized for your system. To use an optimized BLAS library, you can use the `--with-blas` argument which will be passed to the `configure.ac` file. For example, on a Mac with vecLib pre-installed the package may be installed via:

```
R CMD INSTALL affyILM_x.y.z.tar.gz --configure-args="--with-blas='-framework
vecLib'"
```

On a 64-bit Intel machine which has MKL as the optimized BLAS library, the command may look like:

```
R CMD INSTALL affyILM_x.y.z.tar.gz --configure-args="-
-with-blas='-L/usr/local/mkl/lib/em64t/ -lmkl -lguide -lpthread'"
```

where `/usr/local/mkl/lib/em64t/` is the path to MKL.

If you prefer to install a prebuilt binary, you need GSL for successful installation.

affyILM imports several functions from other packages. Make sure to have the following installed:
affxparser, *affy* and *gcrma*. Chip-specific probe packages which are not yet installed on your system will be automatically downloaded from the bioconductor webpage if needed.

2.2 First Steps

For demonstration purposes we use a test CEL-file supplied by *AffymetrixDataTestFiles*.

```
> require(AffymetrixDataTestFiles)
```

Load the library

```
> library(affyILM)
```

and locate the test CEL-file

```
> file1 <- system.file("rawData", "FusionSDK_HG-Focus", "HG-Focus",
+     "2.Calvin", "HG-Focus-1-121502.CEL", package = "AffymetrixDataTestFiles")
```

Calculation of the background intensity as well as of the concentrations:

```
> result <- ilm(file1)
```

```
Chip dimension 448  x  448
[1] "Checking to see if your internet connection works..."
package 'hgfocusprobe' successfully unpacked and MD5 sums checked
```

```
The downloaded packages are in
        E:\biocbld\bbs-2.7-bioc\tmpdir\Rtmp9YXcPw\downloaded_packages
Probepackage hgfocusprobe loaded
Reading intensities...done
72482 features used for parameter estimation
Residual value is 0.000000
Background intensities calculated
Concentrations calculated
Done
------------------
```

Now let's have a look at the output printed on the screen:

- Chip dimension
- probe package downloaded if missing
- the number of features used for background estimation depends on the threshold intensity according to which features to be used for the calculation are selected. The default value is 350.
- Residual value of the linear least square problem

Take a look at the calculated background intensities as well as the experimental PM's

```
> getIntens(result, "AFFX-r2-Ec-bioD-5_at")
```

	Intensities	
Probeset	IPM-HG-Focus-1-121502.CEL	IOPM-HG-Focus-1-121502.CEL
AFFX-r2-Ec-bioD-5_at	1384.0	162.38
AFFX-r2-Ec-bioD-5_at	1410.8	136.65
AFFX-r2-Ec-bioD-5_at	2616.3	127.28
AFFX-r2-Ec-bioD-5_at	1404.0	96.57
AFFX-r2-Ec-bioD-5_at	602.5	213.75
AFFX-r2-Ec-bioD-5_at	694.3	190.60
AFFX-r2-Ec-bioD-5_at	1329.8	96.07
AFFX-r2-Ec-bioD-5_at	644.5	120.89
AFFX-r2-Ec-bioD-5_at	628.3	114.54
AFFX-r2-Ec-bioD-5_at	3265.8	172.70
AFFX-r2-Ec-bioD-5_at	1045.5	137.40

`Note:` The order in which the probes are displayed do not correspond to the original order!

Plot the result:

```
> plotIntens(result, "AFFX-r2-Ec-bioD-5_at", "HG-Focus-1-121502.CEL")
```

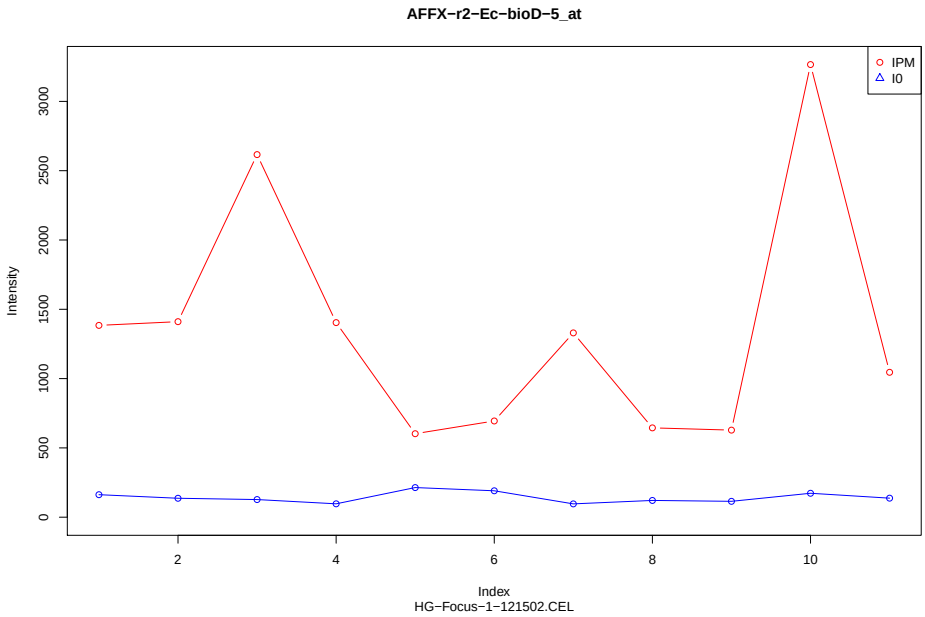

Figure 1: Comparison between PM intensity and the background values

3 More Examples with options

Analyze two or more CEL-files

```
> file2 <- system.file("rawData", "FusionSDK_HG-Focus", "HG-Focus",
+     "2.Calvin", "HG-Focus-2-121502.CEL", package = "AffymetrixDataTestFiles")
> result2files <- ilm(c(file1, file2), 400, 12000)

Chip dimension 448  x  448
Probepackage hgfocusprobe loaded
Reading intensities...done
File analyzed:  HG-Focus-1-121502.CEL
75612 features used for parameter estimation
Residual value is 0.000000
Background intensities calculated
Concentrations calculated
Done
------------------
File analyzed:  HG-Focus-2-121502.CEL
76424 features used for parameter estimation
Residual value is 0.000000
Background intensities calculated
```

```
Concentrations calculated
Done
------------------
```

where only probes with an intensity up to 400 are used for background calculation and the saturation limit of the Langmuir isotherm is increased to 12000 (default: 10000).

Get PM intensities and corresponding background values:

```
> getIntens(result2files, "AFFX-r2-Ec-bioD-5_at")
```

Probeset	IPM-HG-Focus-1-121502.CEL	I0PM-HG-Focus-1-121502.CEL
AFFX-r2-Ec-bioD-5_at	1384.0	172.92
AFFX-r2-Ec-bioD-5_at	1410.8	144.02
AFFX-r2-Ec-bioD-5_at	2616.3	133.21
AFFX-r2-Ec-bioD-5_at	1404.0	100.46
AFFX-r2-Ec-bioD-5_at	602.5	233.47
AFFX-r2-Ec-bioD-5_at	694.3	204.48
AFFX-r2-Ec-bioD-5_at	1329.8	99.43
AFFX-r2-Ec-bioD-5_at	644.5	124.50
AFFX-r2-Ec-bioD-5_at	628.3	120.37
AFFX-r2-Ec-bioD-5_at	3265.8	184.76
AFFX-r2-Ec-bioD-5_at	1045.5	145.41

Probeset	IPM-HG-Focus-2-121502.CEL	I0PM-HG-Focus-2-121502.CEL
AFFX-r2-Ec-bioD-5_at	1404.39	169.55
AFFX-r2-Ec-bioD-5_at	1351.20	137.58
AFFX-r2-Ec-bioD-5_at	2738.08	134.22
AFFX-r2-Ec-bioD-5_at	1510.84	97.64
AFFX-r2-Ec-bioD-5_at	564.96	234.11
AFFX-r2-Ec-bioD-5_at	665.41	203.08
AFFX-r2-Ec-bioD-5_at	1244.10	95.44
AFFX-r2-Ec-bioD-5_at	569.07	123.70
AFFX-r2-Ec-bioD-5_at	613.21	118.51
AFFX-r2-Ec-bioD-5_at	3275.92	183.89
AFFX-r2-Ec-bioD-5_at	905.84	140.50

- 1st column: Probeset name
- Following columns (read pairwise,i.e. 2 columns per CEL-file): Measured PM intensities `IPM` and background estimates `I0PM`

The function of the background intensity per probe is characterized by its 50 optimized parameters:

```
> getParams(result2files)
```

	HG-Focus-1-121502.CEL	HG-Focus-2-121502.CEL
[1,]	1.0302856768	0.541052675
[2,]	0.0611188541	0.059458658
[3,]	0.0556656444	0.058960849
[4,]	-0.0146940677	-0.013512307
[5,]	-0.0140267281	-0.014351281
[6,]	0.2372035264	0.229545898
[7,]	0.0071061005	0.008841897
[8,]	0.0053782089	0.007204471
[9,]	0.0217924529	0.025816610
[10,]	0.1971714464	-0.295573078
[11,]	0.0812876929	0.079056320
[12,]	0.0721215982	0.069481893
[13,]	-0.0207139593	-0.016798366
[14,]	-0.0102032255	-0.007726154
[15,]	0.3944131736	0.388778215
[16,]	0.0099561961	0.011616956
[17,]	0.0096446474	0.010878876
[18,]	0.0256766106	0.030683355
[19,]	0.2331673766	0.312768882
[20,]	0.1527224605	0.240539062
[21,]	-0.2214175584	-0.107401023
[22,]	-0.1261764284	-0.023923073
[23,]	-0.0767014024	-0.032216816
[24,]	0.1038319120	0.174265887
[25,]	-0.3390762483	-0.253065883
[26,]	-0.2312138499	-0.154092231
[27,]	0.2809728475	0.297399105
[28,]	0.3149427653	0.349188162
[29,]	-0.0422039762	0.012249172
[30,]	0.0031445805	0.045198388
[31,]	0.3047878227	0.342881456
[32,]	0.3456039967	0.401658597
[33,]	-0.0513767293	0.023576924
[34,]	-0.0011116773	0.060619855
[35,]	-0.0048070487	-0.015077861
[36,]	-0.0006150782	-0.011701380
[37,]	0.0245474479	0.011264808
[38,]	0.0160387487	0.003866209
[39,]	0.0166789290	0.008777454
[40,]	0.0035895030	-0.006551098

```
[41,]          0.0348306540           0.023749048
[42,]          0.0241440887           0.013852678
[43,]         -0.0117930683          -0.017866206
[44,]         -0.0143402270          -0.021632141
[45,]          0.0120752954           0.003427732
[46,]          0.0056486030          -0.002038731
[47,]         -0.0097682383          -0.017395112
[48,]         -0.0131986028          -0.022041660
[49,]          0.0151881593           0.004857502
[50,]          0.0079928288          -0.001574064
```

To obtain the concentrations (or expression levels) per probeset, use

```
> getConcs(result2files, "AFFX-r2-Ec-bioD-5_at")

genes                   HG-Focus-1-121502.CEL HG-Focus-2-121502.CEL
  AFFX-r2-Ec-bioD-5_at              21.12639              20.49479
```

Use [to subset the results on one or more probesets

```
> res_1 <- result["AFFX-r2-Ec-bioD-5_at"]
> res_1

                     Intensities
Probeset              IPM-HG-Focus-1-121502.CEL IOPM-HG-Focus-1-121502.CEL
  AFFX-r2-Ec-bioD-5_at                    1384.0                      162.38
  AFFX-r2-Ec-bioD-5_at                    1410.8                      136.65
  AFFX-r2-Ec-bioD-5_at                    2616.3                      127.28
  AFFX-r2-Ec-bioD-5_at                    1404.0                       96.57
  AFFX-r2-Ec-bioD-5_at                     602.5                      213.75
  AFFX-r2-Ec-bioD-5_at                     694.3                      190.60
  AFFX-r2-Ec-bioD-5_at                    1329.8                       96.07
  AFFX-r2-Ec-bioD-5_at                     644.5                      120.89
  AFFX-r2-Ec-bioD-5_at                     628.3                      114.54
  AFFX-r2-Ec-bioD-5_at                    3265.8                      172.70
  AFFX-r2-Ec-bioD-5_at                    1045.5                      137.40
[1] 26.19
      HG-Focus-1-121502.CEL
 [1,]              1.096441
 [2,]              0.058993
 [3,]              0.053599
 [4,]             -0.014431
 [5,]             -0.014164
 [6,]              0.232392
```

```
 [7,]  0.006346
 [8,]  0.005312
 [9,]  0.022097
[10,]  0.271669
[11,]  0.077861
[12,]  0.067479
[13,] -0.020032
[14,] -0.009434
[15,]  0.392230
[16,]  0.008602
[17,]  0.009852
[18,]  0.024806
[19,]  0.225314
[20,]  0.159353
[21,] -0.200488
[22,] -0.114104
[23,] -0.081428
[24,]  0.105911
[25,] -0.324216
[26,] -0.225752
[27,]  0.263247
[28,]  0.305862
[29,] -0.039616
[30,]  0.000477
[31,]  0.290430
[32,]  0.339369
[33,] -0.043217
[34,]  0.001141
[35,] -0.004699
[36,] -0.001445
[37,]  0.022734
[38,]  0.014921
[39,]  0.016642
[40,]  0.002982
[41,]  0.033382
[42,]  0.023536
[43,] -0.010844
[44,] -0.014001
[45,]  0.011557
[46,]  0.005621
[47,] -0.009108
[48,] -0.013074
```

```
[49,]              0.014150
[50,]              0.007584
[1] 10000
```

```
> res_2 <- result[c("AFFX-r2-Ec-bioD-5_at", "207218_at")]
> res_2
```

```
                     Intensities
Probeset              IPM-HG-Focus-1-121502.CEL IOPM-HG-Focus-1-121502.CEL
  AFFX-r2-Ec-bioD-5_at                    1384.0                      162.38
  AFFX-r2-Ec-bioD-5_at                    1410.8                      136.65
  AFFX-r2-Ec-bioD-5_at                    2616.3                      127.28
  AFFX-r2-Ec-bioD-5_at                    1404.0                       96.57
  AFFX-r2-Ec-bioD-5_at                     602.5                      213.75
  AFFX-r2-Ec-bioD-5_at                     694.3                      190.60
  AFFX-r2-Ec-bioD-5_at                    1329.8                       96.07
  AFFX-r2-Ec-bioD-5_at                     644.5                      120.89
  AFFX-r2-Ec-bioD-5_at                     628.3                      114.54
  AFFX-r2-Ec-bioD-5_at                    3265.8                      172.70
  AFFX-r2-Ec-bioD-5_at                    1045.5                      137.40
  207218_at                                 73.5                       92.20
  207218_at                                133.0                       97.29
  207218_at                                 60.8                       57.94
  207218_at                                191.5                      154.48
  207218_at                                 62.5                       56.48
  207218_at                                 82.5                       85.11
  207218_at                                 63.8                       66.62
  207218_at                                 58.8                       72.79
  207218_at                                 97.0                       70.24
  207218_at                                 83.5                       84.93
  207218_at                                 83.8                       57.54
AFFX-r2-Ec-bioD-5_at            207218_at
             26.1909                0.5886
      HG-Focus-1-121502.CEL
 [1,]              1.096441
 [2,]              0.058993
 [3,]              0.053599
 [4,]             -0.014431
 [5,]             -0.014164
 [6,]              0.232392
 [7,]              0.006346
 [8,]              0.005312
 [9,]              0.022097
```

```
[10,]  0.271669
[11,]  0.077861
[12,]  0.067479
[13,] -0.020032
[14,] -0.009434
[15,]  0.392230
[16,]  0.008602
[17,]  0.009852
[18,]  0.024806
[19,]  0.225314
[20,]  0.159353
[21,] -0.200488
[22,] -0.114104
[23,] -0.081428
[24,]  0.105911
[25,] -0.324216
[26,] -0.225752
[27,]  0.263247
[28,]  0.305862
[29,] -0.039616
[30,]  0.000477
[31,]  0.290430
[32,]  0.339369
[33,] -0.043217
[34,]  0.001141
[35,] -0.004699
[36,] -0.001445
[37,]  0.022734
[38,]  0.014921
[39,]  0.016642
[40,]  0.002982
[41,]  0.033382
[42,]  0.023536
[43,] -0.010844
[44,] -0.014001
[45,]  0.011557
[46,]  0.005621
[47,] -0.009108
[48,] -0.013074
[49,]  0.014150
[50,]  0.007584
[1] 10000
```

and/or on one or more files:

```
> res2_1 <- result2files["AFFX-r2-Ec-bioD-5_at"]
> res2_1

Probeset               IPM-HG-Focus-1-121502.CEL IOPM-HG-Focus-1-121502.CEL
  AFFX-r2-Ec-bioD-5_at                    1384.0                      172.92
  AFFX-r2-Ec-bioD-5_at                    1410.8                      144.02
  AFFX-r2-Ec-bioD-5_at                    2616.3                      133.21
  AFFX-r2-Ec-bioD-5_at                    1404.0                      100.46
  AFFX-r2-Ec-bioD-5_at                     602.5                      233.47
  AFFX-r2-Ec-bioD-5_at                     694.3                      204.48
  AFFX-r2-Ec-bioD-5_at                    1329.8                       99.43
  AFFX-r2-Ec-bioD-5_at                     644.5                      124.50
  AFFX-r2-Ec-bioD-5_at                     628.3                      120.37
  AFFX-r2-Ec-bioD-5_at                    3265.8                      184.76
  AFFX-r2-Ec-bioD-5_at                    1045.5                      145.41

Probeset               IPM-HG-Focus-2-121502.CEL IOPM-HG-Focus-2-121502.CEL
  AFFX-r2-Ec-bioD-5_at                    1404.4                      169.55
  AFFX-r2-Ec-bioD-5_at                    1351.2                      137.58
  AFFX-r2-Ec-bioD-5_at                    2738.1                      134.22
  AFFX-r2-Ec-bioD-5_at                    1510.8                       97.64
  AFFX-r2-Ec-bioD-5_at                     565.0                      234.11
  AFFX-r2-Ec-bioD-5_at                     665.4                      203.08
  AFFX-r2-Ec-bioD-5_at                    1244.1                       95.44
  AFFX-r2-Ec-bioD-5_at                     569.1                      123.70
  AFFX-r2-Ec-bioD-5_at                     613.2                      118.51
  AFFX-r2-Ec-bioD-5_at                    3275.9                      183.89
  AFFX-r2-Ec-bioD-5_at                     905.8                      140.50
HG-Focus-1-121502.CEL HG-Focus-2-121502.CEL
                21.13                 20.49
      HG-Focus-1-121502.CEL HG-Focus-2-121502.CEL
 [1,]             1.0302857             0.541053
 [2,]             0.0611189             0.059459
 [3,]             0.0556656             0.058961
 [4,]            -0.0146941            -0.013512
 [5,]            -0.0140267            -0.014351
 [6,]             0.2372035             0.229546
 [7,]             0.0071061             0.008842
 [8,]             0.0053782             0.007204
 [9,]             0.0217925             0.025817
[10,]             0.1971714            -0.295573
```

```
[11,]  0.0812877  0.079056
[12,]  0.0721216  0.069482
[13,] -0.0207140 -0.016798
[14,] -0.0102032 -0.007726
[15,]  0.3944132  0.388778
[16,]  0.0099562  0.011617
[17,]  0.0096446  0.010879
[18,]  0.0256766  0.030683
[19,]  0.2331674  0.312769
[20,]  0.1527225  0.240539
[21,] -0.2214176 -0.107401
[22,] -0.1261764 -0.023923
[23,] -0.0767014 -0.032217
[24,]  0.1038319  0.174266
[25,] -0.3390762 -0.253066
[26,] -0.2312138 -0.154092
[27,]  0.2809728  0.297399
[28,]  0.3149428  0.349188
[29,] -0.0422040  0.012249
[30,]  0.0031446  0.045198
[31,]  0.3047878  0.342881
[32,]  0.3456040  0.401659
[33,] -0.0513767  0.023577
[34,] -0.0011117  0.060620
[35,] -0.0048070 -0.015078
[36,] -0.0006151 -0.011701
[37,]  0.0245474  0.011265
[38,]  0.0160387  0.003866
[39,]  0.0166789  0.008777
[40,]  0.0035895 -0.006551
[41,]  0.0348307  0.023749
[42,]  0.0241441  0.013853
[43,] -0.0117931 -0.017866
[44,] -0.0143402 -0.021632
[45,]  0.0120753  0.003428
[46,]  0.0056486 -0.002039
[47,] -0.0097682 -0.017395
[48,] -0.0131986 -0.022042
[49,]  0.0151882  0.004858
[50,]  0.0079928 -0.001574
[1] 12000
```

```
> res2_2 <- result2files[c("AFFX-r2-Ec-bioD-5_at", "207218_at")]
> res2_2
```

Probeset	IPM-HG-Focus-1-121502.CEL	IOPM-HG-Focus-1-121502.CEL
AFFX-r2-Ec-bioD-5_at	1384.0	172.92
AFFX-r2-Ec-bioD-5_at	1410.8	144.02
AFFX-r2-Ec-bioD-5_at	2616.3	133.21
AFFX-r2-Ec-bioD-5_at	1404.0	100.46
AFFX-r2-Ec-bioD-5_at	602.5	233.47
AFFX-r2-Ec-bioD-5_at	694.3	204.48
AFFX-r2-Ec-bioD-5_at	1329.8	99.43
AFFX-r2-Ec-bioD-5_at	644.5	124.50
AFFX-r2-Ec-bioD-5_at	628.3	120.37
AFFX-r2-Ec-bioD-5_at	3265.8	184.76
AFFX-r2-Ec-bioD-5_at	1045.5	145.41
207218_at	73.5	93.01
207218_at	133.0	98.20
207218_at	60.8	57.84
207218_at	191.5	155.70
207218_at	62.5	56.16
207218_at	82.5	86.50
207218_at	63.8	66.44
207218_at	58.8	73.30
207218_at	97.0	70.12
207218_at	83.5	84.97
207218_at	83.8	57.25

Probeset	IPM-HG-Focus-2-121502.CEL	IOPM-HG-Focus-2-121502.CEL
AFFX-r2-Ec-bioD-5_at	1404.39	169.55
AFFX-r2-Ec-bioD-5_at	1351.20	137.58
AFFX-r2-Ec-bioD-5_at	2738.08	134.22
AFFX-r2-Ec-bioD-5_at	1510.84	97.64
AFFX-r2-Ec-bioD-5_at	564.96	234.11
AFFX-r2-Ec-bioD-5_at	665.41	203.08
AFFX-r2-Ec-bioD-5_at	1244.10	95.44
AFFX-r2-Ec-bioD-5_at	569.07	123.70
AFFX-r2-Ec-bioD-5_at	613.21	118.51
AFFX-r2-Ec-bioD-5_at	3275.92	183.89
AFFX-r2-Ec-bioD-5_at	905.84	140.50
207218_at	84.26	100.53
207218_at	121.49	95.81
207218_at	63.71	56.90
207218_at	197.53	157.08

```
  207218_at                                54.25                    51.43
  207218_at                                70.12                    84.06
  207218_at                                54.50                    63.19
  207218_at                                51.79                    70.00
  207218_at                                90.01                    66.75
  207218_at                                86.56                    86.43
  207218_at                                80.97                    55.29

genes                    HG-Focus-1-121502.CEL HG-Focus-2-121502.CEL
  AFFX-r2-Ec-bioD-5_at                 21.1264                  20.4948
  207218_at                             0.4973                   0.1623
      HG-Focus-1-121502.CEL HG-Focus-2-121502.CEL
 [1,]             1.0302857              0.541053
 [2,]             0.0611189              0.059459
 [3,]             0.0556656              0.058961
 [4,]            -0.0146941             -0.013512
 [5,]            -0.0140267             -0.014351
 [6,]             0.2372035              0.229546
 [7,]             0.0071061              0.008842
 [8,]             0.0053782              0.007204
 [9,]             0.0217925              0.025817
[10,]             0.1971714             -0.295573
[11,]             0.0812877              0.079056
[12,]             0.0721216              0.069482
[13,]            -0.0207140             -0.016798
[14,]            -0.0102032             -0.007726
[15,]             0.3944132              0.388778
[16,]             0.0099562              0.011617
[17,]             0.0096446              0.010879
[18,]             0.0256766              0.030683
[19,]             0.2331674              0.312769
[20,]             0.1527225              0.240539
[21,]            -0.2214176             -0.107401
[22,]            -0.1261764             -0.023923
[23,]            -0.0767014             -0.032217
[24,]             0.1038319              0.174266
[25,]            -0.3390762             -0.253066
[26,]            -0.2312138             -0.154092
[27,]             0.2809728              0.297399
[28,]             0.3149428              0.349188
[29,]            -0.0422040              0.012249
[30,]             0.0031446              0.045198
```

```
[31,]             0.3047878             0.342881
[32,]             0.3456040             0.401659
[33,]            -0.0513767             0.023577
[34,]            -0.0011117             0.060620
[35,]            -0.0048070            -0.015078
[36,]            -0.0006151            -0.011701
[37,]             0.0245474             0.011265
[38,]             0.0160387             0.003866
[39,]             0.0166789             0.008777
[40,]             0.0035895            -0.006551
[41,]             0.0348307             0.023749
[42,]             0.0241441             0.013853
[43,]            -0.0117931            -0.017866
[44,]            -0.0143402            -0.021632
[45,]             0.0120753             0.003428
[46,]             0.0056486            -0.002039
[47,]            -0.0097682            -0.017395
[48,]            -0.0131986            -0.022042
[49,]             0.0151882             0.004858
[50,]             0.0079928            -0.001574
[1] 12000
```

The output objects are of class `ILM`.

Plot the intensities:

```
> plotIntens(result2files, "AFFX-r2-Ec-bioD-5_at", "HG-Focus-1-121502.CEL")
```

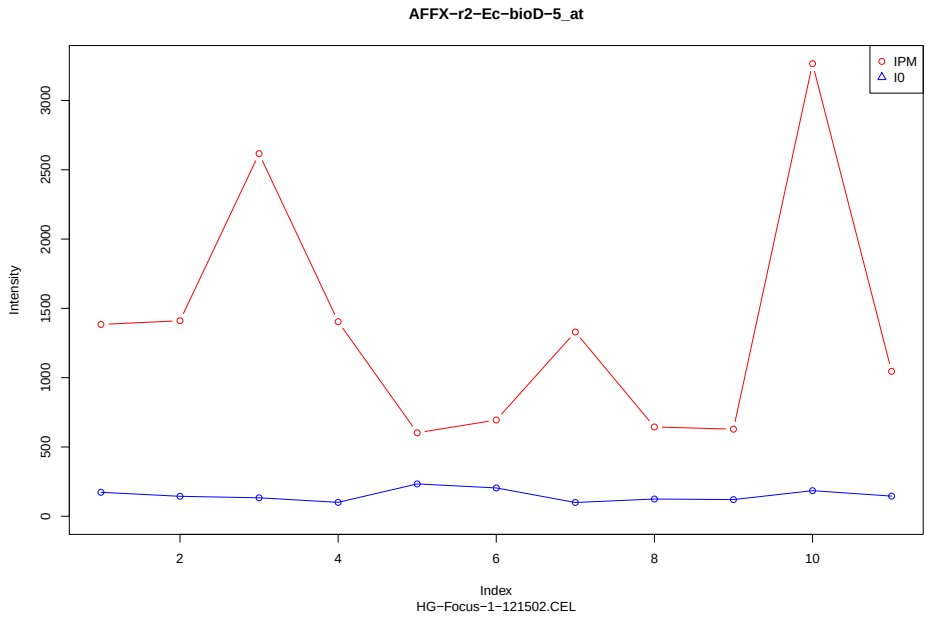

Figure 2: Comparison between PM intensity and the background values

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