

Tools for Advanced Use of the frma Package (frmaTools)

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1 Introduction

Frozen RMA (fRMA) is a microarray preprocessing algorithm that allows one to analyze microarrays individually or in small batches and then combine the data for analysis. This is accomplished by utilizing information from the large publicly available microarray databases. In particular, estimates of probe-specific effects and variances are precomputed and frozen. Then, with new data sets, these are used in concert with information from the new array(s) to normalize and summarize the data.

This document describes `frmaTools`, which provides a few advanced usage options above and beyond the basic `frma` functionality. Details of the ideas implemented in the `frma` and `frmaTools` packages can be found in: *Matthew N. McCall, Benjamin M. Bolstad, and Rafael A. Irizarry, “FROZEN ROBUST MULTI-ARRAY ANALYSIS (fRMA)” (May 2009). Johns Hopkins University, Dept. of Biostatistics Working*

Papers. Working Paper 189. Ideally someone interested in using this package would first read that paper and then proceed to the sections below.

2 Creation of Vectors

To create the vectors required by the package **frma**, one needs a large database of biologically diverse samples from a variety of batches. For this reason, we have attempted to create and supply a number of these collections of vectors through packages available on the Bioconductor website.

To create custom vectors of one's own, follow these steps:

1. Download and install **frmaTools**
2. Create a directory and move all the relevant *CEL* files to that directory.
3. If using linux/unix, start R in that directory.
4. If using the Rgui for Microsoft Windows make sure your working directory contains the *CEL* files (use "File -> Change Dir" menu item).
5. Load the library.

```
> library(frmaTools)
```

6. Load data

```
> library(frmaExampleData)
> data(list = "AffyBatch133a")
```

7. Create batch ids based on your arrays, for example:

```
> batch.id <- rep(1:3, each = 3)
```

8. Run **makeVectors**

```
> object <- makeVectors(AffyBatch133a, batch.id)
```

This will create a list with the required vectors:

normVec normalization reference vector

probeVec probe effect vector

probeVarWithin within batch probe variance

probeVarBetween between batch probe variance

probesetSD within probeset standard deviation

3 Platform Conversion

Occasionally it will be beneficial to convert microarray data from one platform to another. This is possible in cases where the probes on one platform are a subset of the probes on another platform. For example, hgu133a is a subset of hgu133atag; therefore, it is possible to convert hgu133atag data to hgu133a data by discarding those probes that are present on hgu133atag but not hgu133a.

Steps to convert an AffyBatch object:

1. Download and install frmaTools

2. Load the library.

```
> library(frmaTools)
```

3. Load the AffyBatch object to convert (for example hgu133atag data)

```
> library(frmaExampleData)
> data(list = "AffyBatch133atag")
```

4. Run convertPlatform (for example convert to hgu133a)

```
> object <- convertPlatform(AffyBatch133atag, "hgu133a")
```

4 HGU133plus2 as HGU133a

It is often beneficial to combine data from two of the most widely used human GeneChip platforms – HGU133a and HGU133plus2. However, the platform conversion method in the previous section does not work in this case because there are a few HGU133a probes that are not on HGU133plus2. Given the demand for this capability, I have created a function specifically designed to preprocess HGU133plus2 data so that it can be compared to preprocessed HGU133a data.

Steps to preprocess HGU133plus2 as HGU133a

1. Download and install frmaTools

2. Load the library.

```
> library(frmaTools)
```

3. Load the AffyBatch object to convert (for example hgu133plus2 data)

```
> library(frmaExampleData)
> data(list = "AffyBatch133plus2")
```

4. Preprocess the data

```
> e <- hgu133plus2ASaFrma(AffyBatch133plus2)
```