

# RTCA transformation: Discussion of transformation methods of RTCA data

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May 27, 2021

## Abstract

The xCELLigence™ System provides a dimensionless parameter Cell Index (CI) to reflect the biological status of the monitored cells. In this vignette we discuss the concept of the cell index briefly, and introduces several transformation approaches to allow statistical inferences of the RTCA assays.

## 1 Introduction

To demonstrate different types of transformation algorithms implemented in the *RTCA* package, we first load required Bioconductor packages.

```
> library(xtable)
> library(RTCA)
```

And we load a dataset (with its annotation) provided with the package as example:

```
> ofile <- system.file("extdata/testOutput.csv", package="RTCA")
> pfile <- system.file("extdata/testOutputPhenoData.csv", package="RTCA")
> pData <- read.csv(pfile, sep="\t", row.names="Well")
> metaData <- data.frame(labelDescription=c(
+   "Rack number",
+   "siRNA catalogue number",
+   "siRNA gene symbol",
+   "siRNA EntrezGene ID",
+   "siRNA targeting accession"
+ ))
> phData <- new("AnnotatedDataFrame", data=pData, varMetadata=metaData)
> x <- parseRTCA(ofile, phenoData=phData)
```

The object *x* is an instance of the *RTCA* class, which extends the *ExpressionSet* class structure in the *Biobase* package. It contains the raw data of 96 samples in 242 time points.

## 2 Manipulation

### 2.1 Smooth Transformation

*Smooth Transformation* smoothes the RTCA curves by fitting a cubic smoothing spline. It provides more 'flat' data compared to the raw values due to the smoothing. While it is useful for visualization, the smooth transformation must be used with care when modelling or statistical procedures are performed later.

The transformation is performed with the following syntax:

```
> xSmooth <- smoothTransform(x)
```

### 2.2 Interpolation Transformation

The *RTCA* device can record the cell index at irregular time intervals. For example, two sampling time points per hour at the first 48 hour and one time point per hour at the later 48 hour. Some algorithms and time-series model, however, requires data points distributed with regular time intervals. The *interpolation transformation*, as its name suggests, interpolates the RTCA-readout to regular intervals specified by the user. Several methods could be chosen for the interpolation, with the linear interpolation as the default method.

Similarly as the smooth transformation, the interpolation can be called as easily as the following example shows:

```
> xInter <- interpolationTransform(x)
```

## 3 Methods

### 3.1 Derivative Transformation

The *derivative transformation* calculates the growth rate of the RTCA-readouts by its first derivative against time. As an alternative to the ratio normalization proposed by the device provider, the derivative transformation is independent of the choice of the normalising time point, which has to be given manually by the user and thereby introduces subjectivity in the analysis.

An example of the derivative transformation:

```
> xDeriv <- derivativeTransform(x)
```

### 3.2 Relative Growth Rate Transformation

The *relative growth rate transformation* is a derivative from the simple *derivative transformation* by dividing the first derivative with the raw value at that time point. It is analogue

to the *relative growth rate* known in the population genetics where the mathematic model assumes that the growth rate is proportional to the population size.

This feature is by now experimental and we refer interested users to the manual page of the function:

```
> xRgr <- rgrTransform(x)
```

## 4 Session Info

The script runs within the following session:

- R version 4.1.0 (2021-05-18), x86\_64-w64-mingw32
- Locale: LC\_COLLATE=C, LC\_CTYPE=English\_United States.1252, LC\_MONETARY=English\_United States.1252, LC\_NUMERIC=C, LC\_TIME=English\_United States.1252
- Running under: Windows Server x64 (build 17763)
- Matrix products: default
- Base packages: base, datasets, grDevices, graphics, methods, parallel, stats, utils
- Other packages: Biobase 2.53.0, BiocGenerics 0.39.0, RColorBrewer 1.1-2, RTCA 1.45.0, gtools 3.8.2, xtable 1.8-4
- Loaded via a namespace (and not attached): compiler 4.1.0, tools 4.1.0