

# Package ‘nanosttringr’

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**Type** Package

**Title** Performs quality control and data normalization and batch effect correction for NanoString nCounter data

**Version** 0.0.0.9008

**Date** 2015-12-16

**Description** QC and batch adjustment operations for NanoString data.

**Depends** R (>= 3.2.0)

**Imports** dplyr, magrittr, assertthat

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**LazyData** TRUE

**Suggests** epiR, dplyr, testthat, knitr, rmarkdown, pander, stringr, tidyr, ggplot2

**RoxygenNote** 5.0.1

**VignetteBuilder** knitr

**NeedsCompilation** no

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**CCplot***Concordance Correlation Plot*

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**Description**

Plotting function for reliability measure.

**Usage**

```
CCplot(method1, method2, Ptype = "None", metrics = FALSE, xlabel = "",
        ylabel = "", title = "", subtitle = "", xrange = NULL,
        yrange = NULL, MArange = c(-3.5, 5.5))
```

**Arguments**

method1	measurements obtained in batch 1 or using method 1
method2	measurements obtained in batch 2 or using method 2
Ptype	type of plot to be outputted c("scatter", "MAplot")
metrics	if return metrics is set to TRUE, returns Rc, Ca and R2
xlabel	label for x axis
ylabel	label for y axis
title	title for the main plot
subtitle	subtitle of plot
xrange	range of x axis
yrange	range of y axis
MArange	MA range

**Value**

Either a scatterplot or MA plot showing concordance correlation.

**Author(s)**

Aline Talhouk

**Examples**

```
# Simulate normally distributed data
set.seed(12)
a1 <- rnorm(20) + 2
a2 <- a1 + rnorm(20, 0, 0.15)
a3 <- a1 + rnorm(20, 0, 0.15) + 1.4
a4 <- 1.5 * a1 + rnorm(20, 0, 0.15)
a5 <- 1.3 * a1 + rnorm(20, 0, 0.15) + 1
a6 <- a1 + rnorm(20, 0, 0.8)
par(mfrow = c(3, 2), mar = c(5.1, 4.1, 1.5, 1.5))

# Scatterplots
CCplot(a1, a1, Ptype = "scatter", "X", "Y", "Perfect Agreement", subtitle = letters[1])
CCplot(a1, a2, Ptype = "scatter", "X", "Y", "Very Good Agreement", subtitle = letters[2])
```

```

CCplot(a1, a3, Ptype = "scatter", "X", "Y", "Location Shift", subtitle = letters[3])
CCplot(a1, a4, Ptype = "scatter", "X", "Y", "Scale Shift", subtitle = letters[4])
CCplot(a1, a5, Ptype = "scatter", "X", "Y", "Location and Scale Shift", subtitle = letters[5])
CCplot(a1, a6, Ptype = "scatter", "X", "Y", "Measurement Error", subtitle = letters[6])

# MAplots
CCplot(a1, a1, Ptype = "MAplot", "X", "Y", "Perfect Agreement", subtitle = letters[1])
CCplot(a1, a2, Ptype = "MAplot", "X", "Y", "Very Good Agreement", subtitle = letters[2])
CCplot(a1, a3, Ptype = "MAplot", "X", "Y", "Location Shift", subtitle = letters[3])
CCplot(a1, a4, Ptype = "MAplot", "X", "Y", "Scale Shift", subtitle = letters[4])
CCplot(a1, a5, Ptype = "MAplot", "X", "Y", "Location and Scale Shift", subtitle = letters[5])
CCplot(a1, a6, Ptype = "MAplot", "X", "Y", "Measurement Error", subtitle = letters[6])

```

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HKnorm	<i>Normalization to Housekeeping Genes</i>
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## Description

Normalizes the gene expression of NanoString nCounter data to housekeeping genes. This is done by subtracting the average log housekeeping gene expression from the expression level of every gene in each sample.

## Usage

```
HKnorm(raw.data, is.logged = FALSE, corr = 1e-04)
```

## Arguments

raw.data	matrix of raw counts obtained from nCounter (rows are genes). The first three columns must be labeled: c("Code.Class", "Name", "Accession") and contain that information.
corr	small correction to avoid error

## Value

A matrix of log normalized data in the same format but without reference genes.

## Author(s)

Aline Talhouk

## Examples

```
HKnorm(ovd.r)
```

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NanoStringQC

*QC metrics for NanoString Data*


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### Description

Computes and returns NanoString quality control metrics and flags.

### Usage

```
NanoStringQC(raw, exp, detect = 80, sn = 150)
```

### Arguments

raw	matrix of raw counts obtained from nCounter (rows represent genes, columns represent samples). The first three columns must be labeled: c("Code.Class", "Name", "Accession") and contain that information.
exp	matrix of annotations with rows in the same order as the columns of raw. Requires a column labeled "File.Name" with entries corresponding to sample names in raw, also needs columns c("fov.counted", "fov.count", "binding.density"). These fields can be extracted from the nanosttring RCC files.
detect	threshold of percentage of genes expressed over limit of detection (LOD) that we would like to detect (not decimal), defaults to 80 percent.
sn	signal to noise ratio of the housekeeping genes we are willing to tolerate, defaults to 150.

### Value

matrix of annotations updated with normalization parameters

### Author(s)

Aline Talhouk, Derek Chiu

### Examples

```
exp.OVD <-subset(expQC,OVD=="Yes")
expOVD <- NanoStringQC(ovd.r,exp.OVD)
```

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nanosttringr

*nanosttringr: Quality Control and normalization for nanosttringr platform*


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### Description

The nanosttringr package provides a novel set of tools used for quality control purposes in the NanoString platform. It includes batch adjustment using the ratio method thus far.

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refMethod*Reference-based approach for batch adjustment*

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**Description**

Batch adjustment by considering a measure relative to a reference sample

**Usage**

```
refMethod(Y, R1, R2)
```

**Arguments**

Y	data run in first or second batch, samples are rows and genes are columns. If correcting one batch only R1 is needed and would correspond to reference run in the same batch as Y, if calibrating one batch to the other Y represents the data from batch 2 and R1 would be reference run in batch 1 and R2 would be reference from batch 2
R1	reference data run in the first batch
R2	reference data run in the second batch

**Value**

The Y data adjusted calibrated to batch 1 (if two batches are presented) or the data with reference sample expression removed if only one data is provided

**Author(s)**

Aline Talhouk

**Examples**

```
set.seed(12)
A <- matrix(rnorm(120), ncol = 10)
B <- matrix(rnorm(80), ncol = 10)
C <- matrix(rnorm(50), ncol = 10)
refMethod(A, B, C)
```

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