

Computation of melting temperature of nucleic acid duplexes with **rmelting**

Aravind, J.¹ and Krishna, G. K.²

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1. Division of Germplasm Conservation, ICAR-National Bureau of Plant Genetic Resources, New Delhi.
 2. Division of Crop Physiology, ICAR-Indian Agricultural Research Institute, New Delhi.
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1 Introduction

The R package `rmelting` is an interface to the **MELTING 5** program (Le Novère, 2001; Dumasseau et al., 2012) to compute melting temperatures of nucleic acid duplexes (DNA/DNA, DNA/RNA, RNA/RNA or 2'-O-MeRNA/RNA) along with other thermodynamic parameters such as hybridisation enthalpy and entropy.

Melting temperatures are computed by Nearest-neighbour methods for short sequences or approximative estimation formulae for long sequences. Apart from these, multiple corrections are available to take into account the presence of Cations (Na, Tris, K and Mg) or denaturing agents (DMSO and formamide).



2 Installation

The package can be installed from Bioconductor as follows.

```
if (!"BiocManager" %in% rownames(installed.packages()))
  install.packages("BiocManager")
BiocManager::install("rmelting")
```

The development version can be installed from github as follows.

```
if (!require('devtools')) install.packages('devtools')
devtools::install_github("aravind-j/rmelting")
```

Then the package can be loaded as follows.

```
library(rmelting)
```

3 Basic usage

Melting temperatures are computed in **rmelting** through the core function **melting** which takes a number of arguments (see **?melting**). The following are the essential arguments which are mandatory for computation.

- **sequence**
 - 5' to 3' sequence of one strand of the nucleic acid duplex as a character string. Recognises A, C, G, T, U, I, X_C, X_T, A*, AL, TL, GL and CL (**Table 1**). U and T are not considered identical.

Table 1: Recognized sequences

| Code | Type |
|------|---------------------|
| A | Adenine |
| C | Cytosine |
| G | Guanine |
| T | Thymine |
| U | Uracil |
| I | Inosine |
| X_C | Trans azobenzenes |
| X_T | Cis azobenzenes |
| A* | Hydroxyadenine |
| AL | Locked nucleic acid |
| TL | " |
| GL | " |
| CL | " |

- **Comp.sequence**
 - Mandatory if there are mismatches, inosine(s) or hydroxyadenine(s) between the two strands. If not specified, it is computed as the complement of **sequence**. Self-complementarity in **sequence** is detected

even though there may be (are) dangling end(s) and `comp.sequence` is computed.

- `nucleic.acid.conc`
 - In molar concentration (M or mol L⁻¹).
- `Na.conc`, `Mg.conc`, `Tris.conc`, `K.conc`
 - At least one cation (Na, Mg, Tris, K) concentration is mandatory, the other agents(dNTP, DMSO, formamide) are optional.
- `hybridisation.type`
 - The possible options for hybridisation type are as follows (**Table 2**).

Table 2: Hybridisation type options

| Option | Sequence | Complementary sequence |
|----------------------|----------------|------------------------|
| <code>dnadna</code> | DNA | DNA |
| <code>rnarna</code> | RNA | RNA |
| <code>dnarna</code> | DNA | RNA |
| <code>rnadna</code> | RNA | DNA |
| <code>mrnarna</code> | 2-o-methyl RNA | RNA |
| <code>rnamrna</code> | RNA | 2-o-methyl RNA |

With these arguments, the melting temperature can be computed as follows.

```
melting(sequence = "CAGTGAGACAGCAATGGTCG", nucleic.acid.conc = 2e-06,
        hybridisation.type = "dnadna", Na.conc = 1)
```

```
## [1] 73.35168
```

Only the melting temperature is given as a console output. However, the output can be assigned to an object which contains the details of the environment, options and the thermodynamics results as a list.

```
# Get output as list
out <- melting(sequence = "CAGTGAGACAGCAATGGTCG", nucleic.acid.conc = 2e-06,
               hybridisation.type = "dnadna", Na.conc = 1)
# Environment output
out$Environment
```

```
## $Sequence
## [1] "CAGTGAGACAGCAATGGTCG"
##
## $`Complementary sequence`
## [1] "GTCACTCTGTCGTTACCAGC"
##
## $`Nucleic acid concentration (M)`
## [1] 2e-06
##
## $`Hybridization type`
```

```
## [1] "dnadna"
##
## $`Na concentration (M)`
## [1] 1
##
## $`Mg concentration (M)`
## [1] 0
##
## $`Tris concentration (M)`
## [1] 0
##
## $`K concentration (M)`
## [1] 0
##
## $`dNTP concentration (M)`
## [1] 0
##
## $`DMSO concentration (%)`
## [1] 0
##
## $`Formamide concentration (M or %)`
## [1] 0
##
## $`Self complementarity`
## [1] FALSE
##
## $`Correction factor`
## [1] 4
##
# Options used
out$options

## $`Approximative formula`
## [1] NA
##
## $`Nearest neighbour model`
## [1] NA
##
## $`GU model`
## [1] NA
##
## $`Single mismatch model`
## [1] NA
##
## $`Tandem mismatch model`
## [1] NA
```

```
##
## `$Single dangling end model`
## [1] NA
##
## `$Double dangling end model`
## [1] NA
##
## `$Long dangling end model`
## [1] NA
##
## `$Internal loop model`
## [1] NA
##
## `$Single bulge loop model`
## [1] NA
##
## `$Long bulge loop model`
## [1] NA
##
## `$CNG repeats model`
## [1] NA
##
## `$Inosine bases model`
## [1] NA
##
## `$Hydroxyadenine bases model`
## [1] NA
##
## `$Azobenzenes model`
## [1] NA
##
## `$Locked nucleic acids model`
## [1] NA
##
## `$Ion correction method`
## [1] NA
##
## `$Na equivalence correction method`
## [1] NA
##
## `$DMSO correction method`
## [1] NA
##
## `$Formamide correction method`
## [1] NA
##
```

```
## $Mode
## [1] NA
# Thermodynamics results
out$results

## $`Enthalpy (cal)`
## [1] -159000
##
## $`Entropy (cal)`
## [1] -430
##
## $`Enthalpy (J)`
## [1] -664620
##
## $`Entropy (J)`
## [1] -1797.4
##
## $`Melting temperature (C)`
## [1] 73.35168
```

The command for the MELTING 5 java version is saved as an attribute in the list `out` and can be retrieved as follows.

```
# Command for MELTING 5
attributes(out)$command

## [1] "-S CAGTGAGACAGCAATGGTCG -H dnadna -P 2e-06 -E Na=1 -T 60"
```

4 Melting temperature computation

Melting temperature is computed by either approximative or nearest neighbour methods according to the length of the oligonucleotide sequences. For longer sequences (longer than the threshold value, the threshold value set by `size.threshold` with the default value 60) approximative method is used, while for others, nearest neighbour method is used.

4.1 Approximative methods

The approximative method for computation can be specified by the argument `method.approx`. The available methods are given in **Table 3**.

Table 3: Details of approximative methods

| Formula | Type | Limits/Remarks | Reference |
|---------|------|----------------|---------------------|
| ahs01 | DNA | No mismatch | Ahsen et al. (2001) |

| Formula | Type | Limits/Remarks | Reference |
|---------------------|---------|---|---|
| che93 | DNA | No mismatch; Na=0, Mg=0.0015, Tris=0.01, K=0.05 | Marmur and Doty (1962) |
| che93corr | DNA | No mismatch; Na=0, Mg=0.0015, Tris=0.01, K=0.05 | Marmur and Doty (1962) |
| schdot | DNA | No mismatch | Wetmur (1991), Marmur and Doty (1962), Chester and Marshak (1993), Schildkraut and Lifson (1965), Wahl et al. (1987), Britten et al. (1974), Hall et al. (1980) |
| owe69 | DNA | No mismatch | Owen et al. (1969), Frank-Kamenetskii (1971), Blake (1996), Blake and Delcourt (1998) |
| san98 | DNA | No mismatch | SantaLucia (1998), Ahsen et al. (2001) |
| wetdna91* | DNA | | Wetmur (1991) |
| wetrna91* | RNA | | Wetmur (1991) |
| wetdnarna91* | DNA/RNA | | Wetmur (1991) |

* Default method for computation.

Examples

```

DNA:TCTAATGTGCTGTTAGATGTATCCAGAGATAGCCGAGCATAACTTCAACACACGAGACGTTGATTGGATTTAACCAT
|||||
DNA:AGATTACACGACAATCTACATAGGTCTCTATCGGCTCGTATTTGAAGTTGTGTGCTCTGCAACTAACCTAAATTGGTA
|||||

RNA:UUAUUCUCCGUCAUCUUUAAGCCGUGGAGAGACUGUAGACUUGAACAGGGGUAAGCGGAGGCACGUAGGAUUCACAUC
|||||
RNA:AAUUAGAGGCAGUAGAAAUCGGCACCUCUCUGACAUCUGAACUUGUCCCAUUCGCCUCCGUGCAUCCUAGUGUAG
|||||

DNA:TCTAATGTGCTGTTAGATGTATCCAGAGATAGCCGAGCATAACTTCAACACACGAGACGTTGATTGGATTTAACCAT
|||||
RNA:AGAUUACACGACAAUCUACAUAGGUCUCUAUCGGCUCGUAAUUGAAGUUGUGUGCUCUGCAACUAACCUAAAUUGUA

```



```

# Long Nucleotide sequence
DNAseq <- c("TCTAATGTGCTGTTAGATGTATCCAGAGATAGCCGAGCATAACTTCAACACACGAGACGTTGATTGGATTTAACCATAG")
RNAseq <- c("UUAUUCUCCGUCAUCUUUAAGCCGUGGAGAGACUGUAGACUUGAACAGGGGUAAGCGGAGGCACGUAGGAUUCACAUCAU")

# Approximative method - default (DNA/DNA)
melting(sequence = DNAseq, nucleic.acid.conc = 2e-06,
         hybridisation.type = "dnadna", Na.conc = 1)

## [1] 87.82455

# Approximative method - wetdna91 (DNA/DNA)
melting(sequence = DNAseq, nucleic.acid.conc = 2e-06,
         hybridisation.type = "dnadna", Na.conc = 1,
         method.approx = "wetdna91")

## [1] 87.82455

# Approximative method - ahs01 (DNA/DNA)
melting(sequence = DNAseq, nucleic.acid.conc = 2e-06,
         hybridisation.type = "dnadna", Na.conc = 1,
         method.approx = "ahs01")

## [1] 87.325

# Approximative method - che93 (DNA/DNA)
melting(sequence = DNAseq, nucleic.acid.conc = 2e-06,
         hybridisation.type = "dnadna", Na.conc = 1,
         method.approx = "che93")

## [1] 77.575

# Approximative method - che93corr (DNA/DNA)
melting(sequence = DNAseq, nucleic.acid.conc = 2e-06,
         hybridisation.type = "dnadna", Na.conc = 1,
         method.approx = "che93corr")

## [1] 79.0125

# Approximative method - schdot (DNA/DNA)
melting(sequence = DNAseq, nucleic.acid.conc = 2e-06,
         hybridisation.type = "dnadna", Na.conc = 1,
         method.approx = "schdot")

## [1] 89.4625

# Approximative method - owe69 (DNA/DNA)
melting(sequence = DNAseq, nucleic.acid.conc = 2e-06,
         hybridisation.type = "dnadna", Na.conc = 1,
         method.approx = "owe69")

```

```
## [1] 100.96
# Approximative method - san98 (DNA/DNA)
melting(sequence = DNaseq, nucleic.acid.conc = 2e-06,
        hybridisation.type = "dnadna", Na.conc = 1,
        method.approx = "san98")
```

```
## [1] 86.9
# Approximative method - default (RNA/RNA)
melting(sequence = RNaseq, nucleic.acid.conc = 2e-06,
        hybridisation.type = "rnarna", Na.conc = 1)
```

```
## [1] 101.1745
# Approximative method - wetrna91 (RNA/RNA)
melting(sequence = RNaseq, nucleic.acid.conc = 2e-06,
        hybridisation.type = "rnarna", Na.conc = 1,
        method.approx = "wetrna91")
```

```
## [1] 101.1745
# Approximative method - wetdnarna91 (DNA/RNA)
melting(sequence = DNaseq, nucleic.acid.conc = 2e-06,
        hybridisation.type = "dnarna", Na.conc = 1)
```

```
## [1] 88.92455
# Approximative method - wetdnarna91 (DNA/RNA)
melting(sequence = DNaseq, nucleic.acid.conc = 2e-06,
        hybridisation.type = "dnarna", Na.conc = 1,
        method.approx = "wetdnarna91")
```

```
## [1] 88.92455
```

4.2 Nearest neighbour methods

4.2.1 Perfectly matching sequences

The nearest neighbour model for computation in case of perfectly matching sequences can be specified by the argument `method.nn`. The available methods are given in **Table 4**.

Table 4: Details of nearest neighbour methods for perfectly matching sequences

| Model | Type | Limits/Remarks | Reference |
|---------|------|----------------|------------------------------|
| all197* | DNA | | Allawi and SantaLucia (1997) |
| bre86 | DNA | | Breslauer et al. (1986) |
| san04 | DNA | | SantaLucia and Hicks (2004) |
| san96 | DNA | | SantaLucia et al. (1996) |

| Model | Type | Limits/Remarks | Reference |
|--------|------------------------|---|--------------------------|
| sug96 | DNA | | Sugimoto et al. (1996) |
| tan04 | DNA | | Tanaka et al. (2004) |
| fre86 | RNA | | Freier et al. (1986) |
| xia98* | RNA | | Xia et al. (1998) |
| sug95* | DNA/ RNA | | SantaLucia et al. (1996) |
| tur06* | 2'-O- MeRNA/ RNA | A sodium correction (san04) is automatically applied to convert the entropy (Na = 0.1M) into the entropy (Na = 1M) | Kierzek et al. (2006) |

* Default method for computation.

Examples

```
DNA:CAGTGAGACAGCAATGGTCG
|||||
DNA:GTCACTCTGTCGTTACCAGC
```

```
RNA:CAGUGAGACAGCAAUGGUCG
|||||
RNA:GUCACUCUGUCGUUACCAGC
```

```
DNA:CAGTGAGACAGCAATGGTCG
|||||
RNA:GUCACUCUGUCGUUACCAGC
```

```
# Nearest neighbour method - default (DNA/DNA: No Self-Complimentarity)
melting(sequence = "CAGTGAGACAGCAATGGTCG", nucleic.acid.conc = 2e-06,
         hybridisation.type = "dnadna", Na.conc = 1)
```

```
## [1] 73.35168
```

```
# Nearest neighbour method - all97 (DNA/DNA: No Self-Complimentarity)
melting(sequence = "CAGTGAGACAGCAATGGTCG", nucleic.acid.conc = 2e-06,
         hybridisation.type = "dnadna", Na.conc = 1, method.nn = "all97")
```

```
## [1] 73.35168
```

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```
# Nearest neighbour method - bre86 (DNA/DNA: No Self-Complimentarity)
melting(sequence = "CAGTGAGACAGCAATGGTCG", nucleic.acid.conc = 2e-06,
        hybridisation.type = "dnadna", Na.conc = 1, method.nn = "bre86")

## [1] 83.2203

# Nearest neighbour method - san04 (DNA/DNA: No Self-Complimentarity)
melting(sequence = "CAGTGAGACAGCAATGGTCG", nucleic.acid.conc = 2e-06,
        hybridisation.type = "dnadna", Na.conc = 1, method.nn = "san04")

## [1] 73.30191

# Nearest neighbour method - san96 (DNA/DNA: No Self-Complimentarity)
melting(sequence = "CAGTGAGACAGCAATGGTCG", nucleic.acid.conc = 2e-06,
        hybridisation.type = "dnadna", Na.conc = 1, method.nn = "san96")

## [1] 75.7102

# Nearest neighbour method - sug96 (DNA/DNA: No Self-Complimentarity)
melting(sequence = "CAGTGAGACAGCAATGGTCG", nucleic.acid.conc = 2e-06,
        hybridisation.type = "dnadna", Na.conc = 1, method.nn = "sug96")

## [1] 78.17556

# Nearest neighbour method - tan04 (DNA/DNA: No Self-Complimentarity)
melting(sequence = "CAGTGAGACAGCAATGGTCG", nucleic.acid.conc = 2e-06,
        hybridisation.type = "dnadna", Na.conc = 1, method.nn = "tan04")

## [1] 71.31413

# Nearest neighbour method - default (RNA/RNA: No Self-Complimentarity)
melting(sequence = "CAGUGAGACAGCAAUGGUCG", nucleic.acid.conc = 2e-06,
        hybridisation.type = "rnarna", Na.conc = 1)

## [1] 86.77685

# Nearest neighbour method - xia98 (RNA/RNA: No Self-Complimentarity)
melting(sequence = "CAGUGAGACAGCAAUGGUCG", nucleic.acid.conc = 2e-06,
        hybridisation.type = "rnarna", Na.conc = 1, method.nn = "xia98")

## [1] 86.77685

# Nearest neighbour method - fre86 (RNA/RNA: No Self-Complimentarity)
melting(sequence = "CAGUGAGACAGCAAUGGUCG", nucleic.acid.conc = 2e-06,
        hybridisation.type = "rnarna", Na.conc = 1, method.nn = "fre86")

## [1] 83.81257

# Nearest neighbour method - default (mRNA/RNA: No Self-Complimentarity)
melting(sequence = "CAGUGAGACAGCAAUGGUCG", nucleic.acid.conc = 2e-06,
        hybridisation.type = "mrnarna", Na.conc = 1)
```

```
## [1] 99.01986
# Nearest neighbour method - tur06 (mRNA/RNA: No Self-Complimentarity)
melting(sequence = "CAGUGAGACAGCAAUGGUUCG", nucleic.acid.conc = 2e-06,
        hybridisation.type = "mrnarna", Na.conc = 1, method.nn = "tur06")
```

```
## [1] 99.01986
# Nearest neighbour method - default (DNA/RNA: No Self-Complimentarity)
melting(sequence = "CAGTGAGACAGCAATGGTCG", nucleic.acid.conc = 2e-06,
        hybridisation.type = "dnarna", Na.conc = 1)
```

```
## [1] 66.77049
# Nearest neighbour method - sug95 (DNA/RNA: No Self-Complimentarity)
melting(sequence = "CAGTGAGACAGCAATGGTCG", nucleic.acid.conc = 2e-06,
        hybridisation.type = "dnarna", Na.conc = 1, method.nn = "sug95")
```

```
## [1] 66.77049
```

Self complementarity for perfect matching sequences or sequences with dangling ends is detected automatically. However it can be enforced by the argument `force.self = TRUE`.

Examples

```
DNA:CATATGGCCATATG
      |||||
DNA:GTATACCGGTATAC
```

```
RNA:AUGUACAU
      |||||
RNA:UACAUGUA
```

```
# Nearest neighbour method - default (DNA/DNA: Self-Complimentarity)
melting(sequence = "CATATGGCCATATG", nucleic.acid.conc = 2e-06,
        hybridisation.type = "dnadna", Na.conc = 1)
```

```
## [1] 56.00644
# Nearest neighbour method - all97 (DNA/DNA: Self-Complimentarity)
melting(sequence = "CATATGGCCATATG", nucleic.acid.conc = 2e-06,
        hybridisation.type = "dnadna", Na.conc = 1, method.nn = "all97")
```

```
## [1] 56.00644
# Nearest neighbour method - bre86 (DNA/DNA: Self-Complimentarity)
melting(sequence = "CATATGGCCATATG", nucleic.acid.conc = 2e-06,
        hybridisation.type = "dnadna", Na.conc = 1, method.nn = "bre86")
```

```
## [1] 63.44605
```

```
# Nearest neighbour method - san04 (DNA/DNA: Self-Complimentarity)
melting(sequence = "CATATGGCCATATG", nucleic.acid.conc = 2e-06,
        hybridisation.type = "dnadna", Na.conc = 1, method.nn = "san04")

## [1] 57.80792

# Nearest neighbour method - san96 (DNA/DNA: Self-Complimentarity)
melting(sequence = "CATATGGCCATATG", nucleic.acid.conc = 2e-06,
        hybridisation.type = "dnadna", Na.conc = 1, method.nn = "san96")

## [1] 55.0921

# Nearest neighbour method - sug96 (DNA/DNA: Self-Complimentarity)
melting(sequence = "CATATGGCCATATG", nucleic.acid.conc = 2e-06,
        hybridisation.type = "dnadna", Na.conc = 1, method.nn = "sug96")

## [1] 59.06213

# Nearest neighbour method - tan04 (DNA/DNA: Self-Complimentarity)
melting(sequence = "CATATGGCCATATG", nucleic.acid.conc = 2e-06,
        hybridisation.type = "dnadna", Na.conc = 1, method.nn = "tan04")

## [1] 55.65824

# Nearest neighbour method - default (RNA/RNA: Self-Complimentarity)
melting(sequence = "AUGUACAU", nucleic.acid.conc = 2e-06,
        hybridisation.type = "rnarna", Na.conc = 1)

## [1] 30.27015

# Nearest neighbour method - xia98 (RNA/RNA: Self-Complimentarity)
melting(sequence = "AUGUACAU", nucleic.acid.conc = 2e-06,
        hybridisation.type = "rnarna", Na.conc = 1, method.nn = "xia98")

## [1] 30.27015

# Nearest neighbour method - fre86 (RNA/RNA: Self-Complimentarity)
melting(sequence = "AUGUACAU", nucleic.acid.conc = 2e-06,
        hybridisation.type = "rnarna", Na.conc = 1, method.nn = "fre86")

## [1] 31.48175
```

4.2.2 GU wobble base pairs effect

The nearest neighbour model for computation in case of sequences with GU wobble base pairs can be specified by the argument `method.GU`. The available methods are given in **Table 5**.

Table 5: Details of methods for sequences with GU wobble base pairs

| Model | Type | Limits/Remarks | Reference |
|--------|------|----------------|-----------------------|
| tur99 | RNA | | Mathews et al. (1999) |
| ser12* | RNA | | Chen et al. (2012) |

* Default method for computation.

Examples

```
RNA:CCAGCGUCCU
      |||||
RNA:GGTCGCAGGA
```

```
# GU wobble base pairs effect - default (RNA/RNA)
melting(sequence = "CCAGCGUCCU", nucleic.acid.conc = 0.0001,
        hybridisation.type = "rnarna", Na.conc = 1)

## [1] 79.46955

# GU wobble base pairs effect - ser12 (RNA/RNA)
melting(sequence = "CCAGCGUCCU", nucleic.acid.conc = 0.0001,
        hybridisation.type = "rnarna", Na.conc = 1, method.GU = "ser12")

## [1] 79.46955

# GU wobble base pairs effect - tur99 (RNA/RNA)
melting(sequence = "CCAGCGUCCU", nucleic.acid.conc = 0.0001,
        hybridisation.type = "rnarna", Na.conc = 1, method.GU = "tur99")

## [1] 79.46955
```

4.2.3 Single mismatch effect

The nearest neighbour model for computation in case of sequences with a single mismatch can be specified by the argument `method.singleMM`. The available methods are given in **Table 6**.

Table 6: Details of methods for sequences with single mismatch

| Model | Type | Limits/Remarks | Reference |
|------------|---------|----------------|--|
| allsanpey* | DNA | | Allawi and SantaLucia (1997), Allawi and SantaLucia (1998a), Allawi and SantaLucia (1998b), Allawi and SantaLucia (1998c), Peyret et al. (1999) |
| wat11* | DNA/RNA | | Watkins et al. (2011) |

| Model | Type | Limits/Remarks | Reference |
|--------|------|-------------------------------------|-------------------------|
| tur06 | RNA | | Lu et al. (2006) |
| zno07* | RNA | | Davis and Znosko (2007) |
| zno08 | RNA | At least one adjacent GU base pair. | Davis and Znosko (2008) |

* Default method for computation.

Examples

```
DNA:CAACTTGATATTAATA
      ||||| |||||
DNA:GTTGAACTCTAATTAT
```

```
RNA:GACAGGCUG
      ||| |||
RNA:CUGUGCGAC
```

```
DNA:CCATAACTACC
      ||| |||||
RNA:GGUAAUGAUGG
```

```
# Single mismatch effect - default (DNA/DNA)
melting(sequence = "CAACTTGATATTAATA", comp.sequence = "GTTGAACTCTAATTAT",
        nucleic.acid.conc = 0.0004, hybridisation.type = "dnadna", Na.conc = 1)
```

```
## [1] 51.97499
```

```
# Single mismatch effect - allsanpey (DNA/DNA)
melting(sequence = "CAACTTGATATTAATA", comp.sequence = "GTTGAACTCTAATTAT",
        nucleic.acid.conc = 0.0004, hybridisation.type = "dnadna",
        Na.conc = 1, method.singleMM = "allsanpey")
```

```
## [1] 51.97499
```

```
# Single mismatch effect - default (RNA/RNA)
melting(sequence = "GACAGGCUG", comp.sequence = "CUGUGCGAC",
        nucleic.acid.conc = 0.0001, hybridisation.type = "rnarna", Na.conc = 1)
```

```
## [1] 54.40363
```

```
# Single mismatch effect - zno07 (RNA/RNA)
melting(sequence = "GACAGGCUG", comp.sequence = "CUGUGCGAC",
        nucleic.acid.conc = 0.0001, hybridisation.type = "rnarna",
        Na.conc = 1, method.singleMM = "zno07")
```



```
## [1] 54.40363
# Single mismatch effect - zno08 (RNA/RNA)
melting(sequence = "CAGUACGUC", comp.sequence = "GUCGGGCAG",
        nucleic.acid.conc = 0.0001, hybridisation.type = "rnarna",
        Na.conc = 1, method.singleMM = "zno08")

## [1] 38.26298
# Single mismatch effect - tur06 (RNA/RNA)
melting(sequence = "GACAGGCUG", comp.sequence = "CUGUGCGAC",
        nucleic.acid.conc = 0.0001, hybridisation.type = "rnarna",
        Na.conc = 1, method.singleMM = "tur06")

## [1] 58.27825
# Single mismatch effect - default (DNA/RNA)
melting(sequence = "CCATAACTACC", comp.sequence = "GGUAAUGAUGG",
        nucleic.acid.conc = 0.0001, hybridisation.type = "dnarna", Na.conc = 1)

## [1] 40.32976
# Single mismatch effect - wat11 (DNA/RNA)
melting(sequence = "CCATAACTACC", comp.sequence = "GGUAAUGAUGG",
        nucleic.acid.conc = 0.0001, hybridisation.type = "dnarna",
        Na.conc = 1, method.singleMM = "wat11")

## [1] 40.32976
```

4.2.4 Tandem mismatches effect

The nearest neighbour model for computation in case of sequences with tandem mismatches can be specified by the argument `method.tandemMM`. The available methods are given in **Table 7**.

Table 7: Details of methods for sequences with tandem mismatches

| Model | Type | Limits/Remarks | Reference |
|------------|------|--|---|
| allsanpey* | DNA | Only GT mismatches and TA/TG mismatches. | Allawi and SantaLucia (1997), Allawi and SantaLucia (1998a), Allawi and SantaLucia (1998b), Allawi and SantaLucia (1998c), Peyret et al. (1999) |
| tur99* | RNA | No adjacent GU or UG base pairs. | Mathews et al. (1999), Lu et al. (2006) |

* Default method for computation.

Examples

```

DNA:GACGTTGGAC
    |||  |||
DNA:CTGCGGCCTG

RNA:GAGCGGAG
   ||  ||
RNA:CUCCACUC

# Tandem mismatches effect - default (DNA/DNA)
melting(sequence = "GACGTTGGAC", comp.sequence = "CTGCGGCCTG",
         nucleic.acid.conc = 0.0004, hybridisation.type = "dnadna", Na.conc = 1)

## [1] 50.20175

# Tandem mismatches effect - allsanpey (DNA/DNA)
melting(sequence = "GACGTTGGAC", comp.sequence = "CTGCGGCCTG",
         nucleic.acid.conc = 0.0004, hybridisation.type = "dnadna",
         Na.conc = 1, method.tandemMM = "allsanpey")

## [1] 50.20175

# Tandem mismatches effect - default (RNA/RNA)
melting(sequence = "GAGCGGAG", comp.sequence = "CUCCACUC",
         nucleic.acid.conc = 0.0001, hybridisation.type = "rnarna", Na.conc = 1)

## [1] 21.07224

# Tandem mismatches effect - tur06 (RNA/RNA)
melting(sequence = "GAGCGGAG", comp.sequence = "CUCCACUC",
         nucleic.acid.conc = 0.0001, hybridisation.type = "rnarna",
         Na.conc = 1, method.tandemMM = "tur99")

## [1] 21.07224

```

4.2.5 Single dangling end effect

The nearest neighbour model for computation in case of sequences with a single dangling end can be specified by the argument `method.single.dangle`. The available methods are given in **Table 8**.

Table 8: Details of methods for sequences with single dangling end

| Model | Type | Limits/Remarks | Reference |
|--------|------|----------------|-------------------------|
| bom00* | DNA | | Bommarito et al. (2000) |

| Model | Type | Limits/Remarks | Reference |
|----------|------|--|---|
| sugdna02 | DNA | Only terminal poly A self complementary sequences. | Ohmichi et al. (2002) |
| sugrna02 | RNA | Only terminal poly A self complementary sequences. | Ohmichi et al. (2002) |
| ser08* | RNA | Only 3' UA, GU and UG terminal base pairs only 5' UG and GU terminal base pairs. | O'Toole et al. (2006), Miller et al. (2008) |

* Default method for computation.

Examples

DNA: -GTAGCTACA

|||||||

DNA: ACATCGATG-

RNA: -GGCGCUG

|||||||

RNA: CCGCGAC

DNA: -GGCGCUG

|||||||

RNA: CCGCGAC

```
# Single dangling end effect - default (DNA/DNA)
melting(sequence = "-GTAGCTACA",
         nucleic.acid.conc = 0.0004, hybridisation.type = "dnadna",
         Na.conc = 1)
```

```
## [1] 52.58935
```

```
# Single dangling end effect - bom00 (DNA/DNA)
melting(sequence = "-GTAGCTACA",
         nucleic.acid.conc = 0.0004, hybridisation.type = "dnadna",
         Na.conc = 1, method.single.dangle = "bom00")
```

```
## [1] 52.58935
```

```
# Single dangling end effect - sugdna02 (DNA/DNA)
melting(sequence = "-GTAGCTACA",
```

```

nucleic.acid.conc = 0.0004, hybridisation.type = "dnadna",
Na.conc = 1, method.single.dangle = "sugdna02")

## [1] 50.78548
# Single dangling end effect - default (RNA/RNA)
melting(sequence = "-GGCGCUG",
nucleic.acid.conc = 0.0001, hybridisation.type = "rnarna",
Na.conc = 1)

## [1] 65.7647
# Single dangling end effect - ser08 (RNA/RNA)
melting(sequence = "-GGCGCUG",
nucleic.acid.conc = 0.0001, hybridisation.type = "rnarna",
Na.conc = 1, method.single.dangle = "ser08")

## [1] 65.7647
# Single dangling end effect - sugrna02 (RNA/RNA)
melting(sequence = "-GGCGCUG",
nucleic.acid.conc = 0.0001, hybridisation.type = "rnarna",
Na.conc = 1, method.single.dangle = "sugrna02")

## [1] 65.7647

```

4.2.6 Double dangling end effect

The nearest neighbour model for computation in case of sequences with a double or secondary dangling ends can be specified by the argument `method.double.dangle`. The available methods are given in **Table 9**.

Table 9: Details of methods for sequences with double dangling ends

| Model | Type | Limits/Remarks | Reference |
|-----------|------|--|-----------------------|
| sugdna02* | DNA | Only terminal poly A self complementary sequences. | Ohmichi et al. (2002) |
| sugrna02 | RNA | Only terminal poly A self complementary sequences. | Ohmichi et al. (2002) |
| ser05 | RNA | Depends on the available thermodynamic parameters for single dangling end. | O'Toole et al. (2005) |
| ser06* | RNA | | O'Toole et al. (2006) |

* Default method for computation.

Examples

DNA:--ATGCATAA

|||||

DNA:AATACGTA--

RNA:--AUGCAUAA

|||||

RNA:AAUACGUA--

```
# Double dangling end effect - default (DNA/DNA)
melting(sequence = "--ATGCATAA",
        nucleic.acid.conc = 0.0004, hybridisation.type = "dnadna",
        Na.conc = 1)
```

```
## [1] 44.88615
```

```
# Double dangling end effect - sugdna02 (DNA/DNA)
melting(sequence = "--ATGCATAA",
        nucleic.acid.conc = 0.0004, hybridisation.type = "dnadna",
        Na.conc = 1, method.double.dangle = "sugdna02")
```

```
## [1] 44.88615
```

```
# Double dangling end effect - default (RNA/RNA)
melting(sequence = "--AUGCAUAA",
        nucleic.acid.conc = 0.0001, hybridisation.type = "rnarna",
        Na.conc = 1)
```

```
## [1] 42.79724
```

```
# Double dangling end effect - ser06 (RNA/RNA)
melting(sequence = "--AUGCAUAA",
        nucleic.acid.conc = 0.0001, hybridisation.type = "rnarna",
        Na.conc = 1, method.double.dangle = "ser06")
```

```
## [1] 42.79724
```

```
# Double dangling end effect - sugrna02 (RNA/RNA)
melting(sequence = "--AUGCAUAA",
        nucleic.acid.conc = 0.0001, hybridisation.type = "rnarna",
        Na.conc = 1, method.double.dangle = "sugrna02")
```

```
## [1] 41.82788
```

```
# Double dangling end effect - ser05 (RNA/RNA)
melting(sequence = "--AUGCAUAA",
```

```
nucleic.acid.conc = 0.0001, hybridisation.type = "rnarna",
Na.conc = 1, method.double.dangle = "ser05")
```

```
## [1] 42.78815
```

4.2.7 Long dangling end effect

The nearest neighbour model for computation in case of sequences with a double or secondary dangling ends can be specified by the argument `method.long.dangle`. The available methods are given in **Table 10**.

Table 10: Details of methods for sequences with long dangling ends

| Model | Type | Limits/Remarks | Reference |
|-----------|------|--|-----------------------|
| sugdna02* | DNA | Only terminal poly A self complementary sequences. | Ohmichi et al. (2002) |
| sugrna02* | RNA | Only terminal poly A self complementary sequences. | Ohmichi et al. (2002) |

* Default method for computation.

Examples

DNA:----GCATATGCAAAA

|||||||

DNA:AAAACGTATACG----

RNA:AAAAGCAUAUGC----

|||||||

RNA:----CGUAUACGAAAA

```
# Long dangling end effect - default (DNA/DNA)
melting(sequence = "----GCATATGCAAAA",
        nucleic.acid.conc = 0.0004, hybridisation.type = "dnadna",
        Na.conc = 1)
```

```
## [1] 55.69854
```

```
# Long dangling end effect - sugdna02 (DNA/DNA)
melting(sequence = "----GCATATGCAAAA",
        nucleic.acid.conc = 0.0004, hybridisation.type = "dnadna",
        Na.conc = 1, method.long.dangle = "sugdna02")
```

```
## [1] 55.69854
```

```
# Long dangling end effect - default (RNA/RNA)
melting(sequence = "AAAAGCAUAUGC----",
        nucleic.acid.conc = 0.0001, hybridisation.type = "rnarna",
        Na.conc = 1)
```

```
## [1] 57.21314
```

```
# Long dangling end effect - sugrna02 (RNA/RNA)
melting(sequence = "AAAAGCAUAUGC----",
        nucleic.acid.conc = 0.0001, hybridisation.type = "rnarna",
        Na.conc = 1, method.long.dangle = "sugrna02")
```

```
## [1] 57.21314
```

4.2.8 Internal loop effect

The nearest neighbour model for computation in case of sequences with an internal loop (more than two adjacent mismatches) can be specified by the argument `method.internal.loop`. The available methods are given in **Table 11**.

Table 11: Details of methods for sequences with internal loops

| Model | Type | Limits/Remarks | Reference |
|--------|------|--|-----------------------------|
| san04* | DNA | Missing asymmetry penalty. Not tested with experimental results. | SantaLucia and Hicks (2004) |
| tur06 | RNA | Not tested with experimental results. | Lu et al. (2006) |
| zno07* | RNA | Only for 1x2 loop. | Badhwar et al. (2007) |

* Default method for computation.

Examples

```
DNA:GCGATTGGCACTTTGGTGAAC
      |||||      |||||
DNA:CGCTACATATGAAACCACTTG

RNA:GACAC-GCUG
      ||||  |||
RNA:CUGUAUCGAC
```

```
# Internal loop effect - default (DNA/DNA)
melting(sequence = "GCGATTGGCACTTTGGTGAAC", comp.sequence = "CGCTACATATGAAACCACTTG",
        nucleic.acid.conc = 0.0001, hybridisation.type = "dnadna",
        Na.conc = 1)
```

```
## [1] 84.09052
```

```
# Internal loop effect - san04 (DNA/DNA)
melting(sequence = "GCGATTGGCACTTTGGTGAAC", comp.sequence = "CGCTACATATGAAACCACTTG",
        nucleic.acid.conc = 0.0001, hybridisation.type = "dnadna",
        Na.conc = 1, method.internal.loop = "san04")
```

```
## [1] 84.09052
```

```
# Internal loop effect - default (RNA/RNA)
melting(sequence = "GACAC-GCUG", comp.sequence = "CUGUAUCGAC",
        nucleic.acid.conc = 0.0001, hybridisation.type = "rnarna",
        Na.conc = 1)
```

```
## [1] 45.98713
```

```
# Internal loop effect - zno07 (RNA/RNA)
melting(sequence = "GACAC-GCUG", comp.sequence = "CUGUAUCGAC",
        nucleic.acid.conc = 0.0001, hybridisation.type = "rnarna",
        Na.conc = 1, method.internal.loop = "zno07")
```

```
## [1] 40.49012
```

```
# Internal loop effect - tur06 (RNA/RNA)
melting(sequence = "GACAC-GCUG", comp.sequence = "CUGUAUCGAC",
        nucleic.acid.conc = 0.0001, hybridisation.type = "rnarna",
        Na.conc = 1, method.internal.loop = "tur06")
```

```
## [1] 45.98713
```

4.2.9 Single bulge loop effect

The nearest neighbour model for computation in case of sequences with a single bulge loop can be specified by the argument `method.single.bulge.loop`. The available methods are given in **Table 12**.

Table 12: Details of methods for sequences with single bulge loop

| Model | Type | Limits/Remarks | Reference |
|--------|------|-----------------------------|-----------------------------|
| tan04* | DNA | | Tan and Chen (2007) |
| san04 | DNA | Missing closing AT penalty. | SantaLucia and Hicks (2004) |

| Model | Type | Limits/Remarks | Reference |
|--------|------|---|---------------------|
| ser07 | RNA | Less reliable results. Some missing parameters. | Blose et al. (2007) |
| tur06* | RNA | | Lu et al. (2006) |

* Default method for computation.

Examples

DNA:TCGATTAGCGACACAGG

||||| |||||

DNA:AGCTAATC-CTGTGTCC

RNA:GACUCUGUC

|||| |||

RNA:CUGA-ACAG

Single bulge loop effect - default (DNA/DNA)

```
melting(sequence = "TCGATTAGCGACACAGG", comp.sequence = "AGCTAATC-CTGTGTCC",
        nucleic.acid.conc = 0.0001, hybridisation.type = "dnadna",
        Na.conc = 1)
```

[1] 71.12754

Single bulge loop effect - tan04 (DNA/DNA)

```
melting(sequence = "TCGATTAGCGACACAGG", comp.sequence = "AGCTAATC-CTGTGTCC",
        nucleic.acid.conc = 0.0001, hybridisation.type = "dnadna",
        Na.conc = 1, method.single.bulge.loop = "tan04")
```

[1] 71.12754

Single bulge loop effect - san04 (DNA/DNA)

```
melting(sequence = "TCGATTAGCGACACAGG", comp.sequence = "AGCTAATC-CTGTGTCC",
        nucleic.acid.conc = 0.0001, hybridisation.type = "dnadna",
        Na.conc = 1, method.single.bulge.loop = "san04")
```

[1] 62.0496

Single bulge loop effect - default (RNA/RNA)

```
melting(sequence = "GACUCUGUC", comp.sequence = "CUGA-ACAG",
        nucleic.acid.conc = 0.0001, hybridisation.type = "rnarna",
        Na.conc = 1)
```

[1] 39.47787

```
# Single bulge loop effect - tur06 (RNA/RNA)
melting(sequence = "GACUCUGUC", comp.sequence = "CUGA-ACAG",
        nucleic.acid.conc = 0.0001, hybridisation.type = "rnarna",
        Na.conc = 1, method.single.bulge.loop = "tur06")
```

```
## [1] 39.47787
```

```
# Single bulge loop effect - ser07 (RNA/RNA)
melting(sequence = "GACUCUGUC", comp.sequence = "CUGA-ACAG",
        nucleic.acid.conc = 0.0001, hybridisation.type = "rnarna",
        Na.conc = 1, method.single.bulge.loop = "ser07")
```

```
## [1] 31.42849
```

4.2.10 Long bulge loop effect

The nearest neighbour model for computation in case of sequences with long bulge loop can be specified by the argument `method.long.bulge.loop`. The available methods are given in **Table 13**.

Table 13: Details of methods for sequences with long bulge loop

| Model | Type | Limits/Remarks | Reference |
|--------|------|---------------------------------------|---|
| san04* | DNA | Missing closing AT penalty. | SantaLucia and Hicks (2004) |
| tur06* | RNA | Not tested with experimental results. | Mathews et al. (1999), Lu et al. (2006) |

* Default method for computation.

Examples

```
DNA: ATATGACGCCACAGCG
      ||||  |||||
DNA: TATAC---GGTGTGCGC

RNA: AUAUGACGCCACAGCG
      ||||  |||||
RNA: UAUAC---GGUGUCGC
```

```
# Long bulge loop effect - default (DNA/DNA)
melting(sequence = "ATATGACGCCACAGCG", comp.sequence = "TATAC---GGTGTGCGC",
        nucleic.acid.conc = 0.0001, hybridisation.type = "dnadna",
        Na.conc = 1)
```

```
## [1] 51.7104
```

```
# Long bulge loop effect - san04 (DNA/DNA)
```

```
melting(sequence = "ATATGACGCCACAGCG", comp.sequence = "TATAC---GGTGTTCGC",
        nucleic.acid.conc = 0.0001, hybridisation.type = "dnadna",
        Na.conc = 1, method.long.bulge.loop = "san04")
```

```
## [1] 51.7104
```

```
# Long bulge loop effect - default (RNA/RNA)
```

```
melting(sequence = "AUAUGACGCCACAGCG", comp.sequence = "UAUAC---GGUGUCGC",
        nucleic.acid.conc = 0.0001, hybridisation.type = "rnarna",
        Na.conc = 1)
```

```
## [1] 66.0497
```

```
# Long bulge loop effect - tur06 (RNA/RNA)
```

```
melting(sequence = "AUAUGACGCCACAGCG", comp.sequence = "UAUAC---GGUGUCGC",
        nucleic.acid.conc = 0.0001, hybridisation.type = "rnarna",
        Na.conc = 1, method.long.bulge.loop = "tur06")
```

```
## [1] 66.0497
```

4.2.11 CNG repeats effect

The nearest neighbour model for computation in case of sequences with CNG repeats can be specified by the argument `method.CNG`. The available methods are given in **Table 14**.

Table 14: Details of methods for sequences with CNG repeats

| Model | Type | Limits/Remarks | Reference |
|--------|------|---|---------------------|
| bro05* | RNA | Self complementary sequences. 2 to 7 CNG repeats. | Broda et al. (2005) |

* Default method for computation.

Examples

```
RNA:GCGGCGGCGGC
```

```
|||||
```

```
RNA:CGCCGCCGCCG
```

```
# CNG repeats effect - default (RNA/RNA)
```

```
melting(sequence = "GCGGCGGCGGC",
```

```
nucleic.acid.conc = 0.0001, hybridisation.type = "rnarna",
Na.conc = 1)

## [1] 94.25719

# CNG repeats effect - bro05 (RNA/RNA)
melting(sequence = "GCGGCGGCGGC",
nucleic.acid.conc = 0.0001, hybridisation.type = "rnarna",
Na.conc = 1, method.CNG = "bro05")

## [1] 94.25719
```

4.2.12 Inosine bases effect

The nearest neighbour model for computation in case of sequences with inosine bases (I) can be specified by the argument `method.inosine`. The available methods are given in **Table 15**.

Table 15: Details of methods for sequences with inosine bases

| Model | Type | Limits/Remarks | Reference |
|--------|------|--|-------------------------------|
| san05* | DNA | Missing parameters for tandem base pairs containing inosine bases. | Watkins and SantaLucia (2005) |
| zno07* | RNA | Only IU base pairs. | Wright et al. (2007) |

* Default method for computation.

Examples

```
DNA:CCGICTGTIGCG
    ||| ||| |||
DNA:GGCCGACACCGC

RNA:GCAICGC
   ||| |||
RNA:CGUUGCG

# Inosine bases effect - default (DNA/DNA)
melting(sequence = "CCGICTGTIGCG", comp.sequence = "GGCCGACACCGC",
nucleic.acid.conc = 0.0001, hybridisation.type = "dnadna",
Na.conc = 1)

## [1] 65.36853
```

```
# Inosine bases effect - san05 (DNA/DNA)
melting(sequence = "CCGICTGTIGCG", comp.sequence = "GGCCGACACCGC",
        nucleic.acid.conc = 0.0001, hybridisation.type = "dnadna",
        Na.conc = 1, method.inosine = "san05")
```

```
## [1] 65.36853
```

```
# Inosine bases effect - default (RNA/RNA)
melting(sequence = "GCAICGC", comp.sequence = "CGUUGCG",
        nucleic.acid.conc = 0.0001, hybridisation.type = "rnarna",
        Na.conc = 1)
```

```
## [1] 46.75042
```

```
# Inosine bases effect - zno07 (RNA/RNA)
melting(sequence = "GCAICGC", comp.sequence = "CGUUGCG",
        nucleic.acid.conc = 0.0001, hybridisation.type = "rnarna",
        Na.conc = 1, method.inosine = "zno07")
```

```
## [1] 46.75042
```

4.2.13 Hydroxyadenine bases effect

The nearest neighbour model for computation in case of sequences with hydroxyadenine bases can be specified by the argument `method.hydroxyadenine`. The available methods are given in **Table 16**.

Table 16: Details of methods for sequences with hydroxyadenine bases

| Model | Type | Limits/Remarks | Reference |
|--------|------|---|------------------------|
| sug01* | DNA | Only 5' GA*C 3' and 5' TA*A 3' contexts. | Kawakami et al. (2001) |

* Default method for computation.

Examples

```
      *
DNA:AGAAATGACACGGTG
    |||||
DNA:TCTTTACCGTGCCAC
```

```
# Hydroxyadenine bases effect - default (DNA/DNA)
melting(sequence = "AGAAATGA*ACCGTG", comp.sequence = "TCTTTACCGTGCCAC",
        nucleic.acid.conc = 0.0001, hybridisation.type = "dnadna",
        Na.conc = 1)
```

```
## [1] 68.46041
```

```
# Hydroxyadenine bases effect - sug01 (DNA/DNA)
melting(sequence = "AGAAATGA*CACGGTG", comp.sequence = "TCTTTACCGTGCCAC",
        nucleic.acid.conc = 0.0001, hybridisation.type = "dnadna",
        Na.conc = 1, method.hydroxyadenine = "sug01")
```

```
## [1] 68.46041
```

4.2.14 Azobenzenes effect

The nearest neighbour model for computation in case of sequences with azobenzenes (X_T for trans azobenzenes and X_C for cis azobenzenes) can be specified by the argument `method.azobenzenes`. The available methods are given in **Table 17**.

Table 17: Details of methods for sequences with azobenzenes

| Model | Type | Limits/Remarks | Reference |
|--------|------|--|-----------------------|
| asa05* | DNA | Less reliable results when the number of cis azobenzene increases. | Asanuma et al. (2005) |

* Default method for computation.

Examples

```
      C   C   C   C   C
DNA:CTXTTAAXGAAGXGAGAXTATAXCC
    || |||| |||| |||| |||| ||
DNA:GA AATT CTTC CTCT ATAT GG
```

```
# Azobenzenes effect - default (DNA/DNA)
melting(sequence = "CTX_CTTAAX_CGAAGX_CGAGAX_CTATAX_CCC",
        nucleic.acid.conc = 0.0001, hybridisation.type = "dnadna",
        Na.conc = 1)
```

```
## [1] 47.85385
```

```
# Azobenzenes effect - asa05 (DNA/DNA)
melting(sequence = "CTX_CTTAAX_CGAAGX_CGAGAX_CTATAX_CCC",
        nucleic.acid.conc = 0.0001, hybridisation.type = "dnadna",
        Na.conc = 1, method.azobenzenes = "asa05")
```

```
## [1] 47.85385
```

4.2.15 Single Locked nucleic acid effect

The nearest neighbour model for computation in case of sequences with single locked nucleic acids can be specified by the argument `method.locked`. The available methods are given in **Table 18**.

Table 18: Details of methods for sequences with single locked nucleic acids

| Model | Type | Limits/Remarks | Reference |
|--------|------|----------------|------------------------|
| mct04 | DNA | | McTigue et al. (2004) |
| owc11* | DNA | | Owczarzy et al. (2011) |

* Default method for computation.

Examples

```

      L
DNA:CCATTGCTACC
      |||||
DNA:GGTAACGATGG

# Single locked nucleic acids effect - default (DNA/DNA)
melting(sequence = "CCATTGCTACC",
        nucleic.acid.conc = 0.0001, hybridisation.type = "dnadna",
        Na.conc = 1)

## [1] 63.48299

# Single locked nucleic acids effect - mct04 (DNA/DNA)
melting(sequence = "CCATTGCTACC",
        nucleic.acid.conc = 0.0001, hybridisation.type = "dnadna",
        Na.conc = 1, method.locked = "mct04")

## [1] 63.61426

# Single locked nucleic acids effect - owc11 (DNA/DNA)
melting(sequence = "CCATTGCTACC",
        nucleic.acid.conc = 0.0001, hybridisation.type = "dnadna",
        Na.conc = 1, method.locked = "owc11")

## [1] 63.48299

```

4.3 Consecutive Locked nucleic acids effect

The nearest neighbour model for computation in case of sequences with consecutive locked nucleic acids can be specified by the argument `method.consecutive.locked`. The available methods are given in **Table 19**.

Table 19: Details of methods for sequences with single locked nucleic acids

| Model | Type | Limits/Remarks | Reference |
|--------|------|----------------|------------------------|
| owc11* | DNA | | Owczarzy et al. (2011) |

* Default method for computation.

Examples

```

      LL
DNA:GACC
      ||||
DNA:CTGG

# Consecutive locked nucleic acids effect - default (DNA/DNA)
melting(sequence = "GALCLC",
        nucleic.acid.conc = 0.0001, hybridisation.type = "dnadna",
        Na.conc = 1)

## [1] 12.94323

# Consecutive locked nucleic acids effect - owc11 (DNA/DNA)
melting(sequence = "GALCLC",
        nucleic.acid.conc = 0.0001, hybridisation.type = "dnadna",
        Na.conc = 1, method.consecutive.locked = "owc11")

## [1] 12.94323

```

4.4 Consecutive Locked nucleic acids with a single mismatch effect

The nearest neighbour model for computation in case of sequences with consecutive locked nucleic acids with single mismatch can be specified by the argument `method.consecutive.locked.singleMM`. The available methods are given in **Table 20**.

Table 20: Details of methods for sequences with single locked nucleic acids

| Model | Type | Limits/Remarks | Reference |
|--------|------|----------------|------------------------|
| owc11* | DNA | | Owczarzy et al. (2011) |

* Default method for computation.

Examples

```

      LLL
DNA:GACGC
  || ||
DNA:CTTCG

# Consecutive locked nucleic acids effect - default (DNA/DNA)
melting(sequence = "GALCLGLC", comp.sequence = "CTTCG",
        nucleic.acid.conc = 0.0001, hybridisation.type = "dnadna",
        Na.conc = 1)

## [1] 0.2520424

# Consecutive locked nucleic acids effect - owc11 (DNA/DNA)
melting(sequence = "GALCLGLC", comp.sequence = "CTTCG",
        nucleic.acid.conc = 0.0001, hybridisation.type = "dnadna",
        Na.conc = 1, method.consecutive.locked.singleMM = "owc11")

## [1] 0.2520424

```

5 Corrections

Once the melting temperature is computed, a correction is applied to it according to the concentration of nucleic acids, cations and/or denaturing agents.

5.1 Nucleic acid concentration

For self complementary sequences (auto detected or specified by `force.self`) it is 1. Otherwise it is 4 if the both strands are present in equivalent amount and 1 if one strand is in excess.

5.2 Ion corrections

Melting temperature is computed initially for $[\text{Na}^+] = 1 \text{ M}$, after which a correction for the presence of cations ($[\text{Na}^+]$, $[\text{K}^+]$, $[\text{Tris}^+]$ and $[\text{Mg}^+]$) is applied either directly on the computed melting temperature or on the computed entropy.

The correction methods for cation concentration can be specified by the argument `correction.ion`.

5.2.1 Sodium corrections

The available correction methods for sodium concentration are given in **Table 21**.

Table 21: Details of the corrections for sodium concentration

| Correction | Type | Limits/Remarks | Reference |
|------------|-----------------------|---|--|
| ahs01 | DNA | Na>0. | Ahsen et al. (2001) |
| kam71 | DNA | Na>0; Na>=0.069; Na<=1.02. | Frank-Kamenetskii (1971) |
| marschdot | DNA | Na>=0.069; Na<=1.02. | Marmur and Doty (1962), Blake and Delcourt (1998) |
| owc1904 | DNA | Na>0. (equation 19) | Owczarzy et al. (2004) |
| owc2004 | DNA | Na>0. (equation 20) | Owczarzy et al. (2004) |
| owc2104 | DNA | Na>0. (equation 21) | Owczarzy et al. (2004) |
| owc2204* | DNA | Na>0. (equation 22) | Owczarzy et al. (2004) |
| san96 | DNA | Na>=0.1. | SantaLucia et al. (1996) |
| san04 | DNA | Na>=0.05; Na<=1.1; Oligonucleotides inferior to 16 bases. | SantaLucia and Hicks (2004), SantaLucia (1998) |
| schlif | DNA | Na>=0.07; Na<=0.12. | Schildkraut and Lifson (1965) |
| tanna06 | DNA | Na>=0.001; Na<=1. | Tan and Chen (2006) |
| tanna07* | RNA or 2'-O-MeRNA/RNA | Na>=0.003; Na<=1. | Tan and Chen (2007) |
| wet91 | RNA, DNA and RNA/DNA | Na>0. | Wetmur (1991) |

* Default method for computation.

```
# Na correction - default (DNA/DNA)
melting(sequence = "CCAGCCAGTCTCTCC",
         nucleic.acid.conc = 0.000002, hybridisation.type = "dnadna",
         Na.conc = 0.069)
```

```
## [1] 56.70492
```

```
# Na correction - owc2204 (DNA/DNA)
melting(sequence = "CCAGCCAGTCTCTCC",
         nucleic.acid.conc = 0.000002, hybridisation.type = "dnadna",
```

```
Na.conc = 0.069, correction.ion = "owc2204")

## [1] 56.70492
# Na correction - ahs01 (DNA/DNA)
melting(sequence = "CCAGCCAGTCTCTCC",
        nucleic.acid.conc = 0.000002, hybridisation.type = "dnadna",
        Na.conc = 0.069, correction.ion = "ahs01")

## [1] 54.1569
# Na correction - kam71 (DNA/DNA)
melting(sequence = "CCAGCCAGTCTCTCC",
        nucleic.acid.conc = 0.000002, hybridisation.type = "dnadna",
        Na.conc = 0.069, correction.ion = "kam71")

## [1] 51.72963
# Na correction - marschdot (DNA/DNA)
melting(sequence = "CCAGCCAGTCTCTCC",
        nucleic.acid.conc = 0.000002, hybridisation.type = "dnadna",
        Na.conc = 0.069, correction.ion = "marschdot")

## [1] 49.18075
# Na correction - owc1904 (DNA/DNA)
melting(sequence = "CCAGCCAGTCTCTCC",
        nucleic.acid.conc = 0.000002, hybridisation.type = "dnadna",
        Na.conc = 0.069, correction.ion = "owc1904")

## [1] 56.18571
# Na correction - owc2004 (DNA/DNA)
melting(sequence = "CCAGCCAGTCTCTCC",
        nucleic.acid.conc = 0.000002, hybridisation.type = "dnadna",
        Na.conc = 0.069, correction.ion = "owc2004")

## [1] 56.67553
# Na correction - owc2104 (DNA/DNA)
melting(sequence = "CCAGCCAGTCTCTCC",
        nucleic.acid.conc = 0.000002, hybridisation.type = "dnadna",
        Na.conc = 0.069, correction.ion = "owc2104")

## [1] 56.63967
# Na correction - san96 (DNA/DNA)
melting(sequence = "CCAGCCAGTCTCTCC",
        nucleic.acid.conc = 0.000002, hybridisation.type = "dnadna",
        Na.conc = 0.069, correction.ion = "san96")
```

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```
## [1] 53.01651
```

```
# Na correction - san04 (DNA/DNA)
melting(sequence = "CCAGCCAGTCTCTCC",
        nucleic.acid.conc = 0.000002, hybridisation.type = "dnadna",
        Na.conc = 0.069, correction.ion = "san04")
```

```
## [1] 54.15157
```

```
# Na correction - schlif (DNA/DNA)
melting(sequence = "CCAGCCAGTCTCTCC",
        nucleic.acid.conc = 0.000002, hybridisation.type = "dnadna",
        Na.conc = 0.069, correction.ion = "schlif")
```

```
## [1] 48.25579
```

```
# Na correction - tanna06 (DNA/DNA)
melting(sequence = "CCAGCCAGTCTCTCC",
        nucleic.acid.conc = 0.000002, hybridisation.type = "dnadna",
        Na.conc = 0.069, correction.ion = "tanna06")
```

```
## [1] 55.26711
```

```
# Na correction - wet91 (DNA/DNA)
melting(sequence = "CCAGCCAGTCTCTCC",
        nucleic.acid.conc = 0.000002, hybridisation.type = "dnadna",
        Na.conc = 0.069, correction.ion = "wet91")
```

```
## [1] 51.74573
```

```
# Na correction - default (RNA/RNA)
melting(sequence = "CCAGCCAGUCUCUCC",
        nucleic.acid.conc = 0.000002, hybridisation.type = "rnarna",
        Na.conc = 0.069)
```

```
## [1] 75.1552
```

```
# Na correction - tanna07 (RNA/RNA)
melting(sequence = "CCAGCCAGUCUCUCC",
        nucleic.acid.conc = 0.000002, hybridisation.type = "rnarna",
        Na.conc = 0.069, correction.ion = "tanna07")
```

```
## [1] 75.1552
```

```
# Na correction - wet91 (RNA/RNA)
melting(sequence = "CCAGCCAGUCUCUCC",
        nucleic.acid.conc = 0.000002, hybridisation.type = "rnarna",
        Na.conc = 0.069, correction.ion = "wet91")
```

```
## [1] 69.55572
```

```

# Na correction - default (mRNA/RNA)
melting(sequence = "UACGCGUCAAUAACGCUA",
        nucleic.acid.conc = 0.000002, hybridisation.type = "mrnarna",
        Na.conc = 0.069)

## [1] 81.57763

# Na correction - tanna07 (mRNA/RNA)
melting(sequence = "UACGCGUCAAUAACGCUA",
        nucleic.acid.conc = 0.000002, hybridisation.type = "mrnarna",
        Na.conc = 0.069, correction.ion = "tanna07")

## [1] 81.57763

# Na correction - default (DNA/RNA)
melting(sequence = "CCAGCCAGTCTCTCC",
        nucleic.acid.conc = 0.000002, hybridisation.type = "dnarna",
        Na.conc = 0.069)

## [1] 62.08869

# Na correction - wet91 (DNA/RNA)
melting(sequence = "CCAGCCAGTCTCTCC",
        nucleic.acid.conc = 0.000002, hybridisation.type = "dnarna",
        Na.conc = 0.069, correction.ion = "wet91")

## [1] 62.08869

```

5.2.2 Magnesium corrections

The available correction methods for magnesium concentration are given in **Table 22**.

Table 22: Details of the corrections for magnesium concentration

| Correction | Type | Limits/Remarks | Reference |
|------------|---------------------------|---|------------------------|
| owcmg08* | DNA | Mg \geq 0.0005; Mg \leq 0.6. | Owczarzy et al. (2008) |
| tanmg06 | DNA | Mg \geq 0.0001; Mg \leq 1; Oligomer length superior to 6 base pairs. | Tan and Chen (2006) |
| tanmg07* | RNA or 2'-O- MeRNA/RNA | Mg \geq 0.1; Mg \leq 0.3. | Tan and Chen (2007) |

* Default method for computation.

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```
# Mg correction - default (DNA/DNA)
melting(sequence = "CAGCCTCGTCGCAGC",
        nucleic.acid.conc = 0.000002, hybridisation.type = "dnadna",
        Mg.conc = 0.0015)
```

```
## [1] 65.52043
```

```
# Mg correction - owcmg08 (DNA/DNA)
melting(sequence = "CAGCCTCGTCGCAGC",
        nucleic.acid.conc = 0.000002, hybridisation.type = "dnadna",
        Mg.conc = 0.0015, correction.ion = "owcmg08")
```

```
## [1] 65.52043
```

```
# Mg correction - tanmg06 (DNA/DNA)
melting(sequence = "CAGCCTCGTCGCAGC",
        nucleic.acid.conc = 0.000002, hybridisation.type = "dnadna",
        Mg.conc = 0.0015, correction.ion = "tanmg06")
```

```
## [1] 64.88082
```

```
# Mg correction - default (RNA/RNA)
melting(sequence = "CAGCCUCGUCGCAGC",
        nucleic.acid.conc = 0.000002, hybridisation.type = "rnarna",
        Mg.conc = 0.0015)
```

```
## [1] 82.0796
```

```
# Mg correction - tanmg07 (RNA/RNA)
melting(sequence = "CAGCCUCGUCGCAGC",
        nucleic.acid.conc = 0.000002, hybridisation.type = "rnarna",
        Mg.conc = 0.0015, correction.ion = "tanmg07")
```

```
## [1] 82.0796
```

```
# Mg correction - default (mRNA/RNA)
melting(sequence = "UACGCGUCAUAACGCUA",
        nucleic.acid.conc = 0.000002, hybridisation.type = "mrnarna",
        Mg.conc = 0.0015)
```

```
## [1] 90.06842
```

```
# Mg correction - tanmg07 (mRNA/RNA)
melting(sequence = "UACGCGUCAUAACGCUA",
        nucleic.acid.conc = 0.000002, hybridisation.type = "mrnarna",
        Mg.conc = 0.0015, correction.ion = "tanmg07")
```

```
## [1] 90.06842
```

5.2.3 Mixed Sodium and Magnesium corrections

The available correction methods for mixed sodium magnesium concentration are given in **Table 23**.

Table 23: Details of the corrections for mixed sodium and magnesium concentration

| Correction | Type | Limits/Remarks | Reference |
|------------|-----------------------------------|--|------------------------|
| owcmix08* | DNA | Mg \geq 0.0005; Mg \leq 0.6; Na+K+Tris/2 $>$ 0. | Owczarzy et al. (2008) |
| tanmix07 | DNA, RNA or 2'-O- MeRNA/RNA | Mg \geq 0.1; Mg \leq 0.3; Na+K+Tris/2 \geq 0.1; Na+K+Tris/2 \leq 0.3. | Tan and Chen (2007) |

* Default method for computation.

```
# Mixed Na & Mg correction - default (DNA/DNA)
melting(sequence = "CAGCCTCGTCGCAGC",
        nucleic.acid.conc = 0.000002, hybridisation.type = "dnadna",
        Na.conc = 0.069, Mg.conc = 0.0015)

## [1] 65.83371

# Mixed Na & Mg correction - owcmix08 (DNA/DNA)
melting(sequence = "CAGCCTCGTCGCAGC",
        nucleic.acid.conc = 0.000002, hybridisation.type = "dnadna",
        Na.conc = 0.069, Mg.conc = 0.0015, correction.ion = "owcmix08")

## [1] 65.83371

# Mixed Na & Mg correction - tanmix07 (DNA/DNA)
melting(sequence = "CAGCCTCGTCGCAGC",
        nucleic.acid.conc = 0.000002, hybridisation.type = "dnadna",
        Na.conc = 0.069, Mg.conc = 0.0015, correction.ion = "tanmix07")

## [1] 63.21723

# Mixed Na & Mg correction - default (RNA/RNA)
melting(sequence = "CAGCCUCGUCGCAGC",
        nucleic.acid.conc = 0.000002, hybridisation.type = "rnarna",
        Na.conc = 0.069, Mg.conc = 0.0015)

## [1] 79.40119
```

```
# Mixed Na & Mg correction - tanmix07 (RNA/RNA)
melting(sequence = "CAGCCUGUCGACGC",
        nucleic.acid.conc = 0.000002, hybridisation.type = "rnarna",
        Na.conc = 0.069, Mg.conc = 0.0015, correction.ion = "tanmix07")

## [1] 79.40119

# Mixed Na & Mg correction - default (mRNA/RNA)
melting(sequence = "UACGCGUCAUAACGCUA",
        nucleic.acid.conc = 0.000002, hybridisation.type = "mrnarna",
        Na.conc = 0.069, Mg.conc = 0.0015)

## [1] 96.46186

# Mixed Na & Mg correction - tanmix07 (mRNA/RNA)
melting(sequence = "UACGCGUCAUAACGCUA",
        nucleic.acid.conc = 0.000002, hybridisation.type = "mrnarna",
        Na.conc = 0.069, Mg.conc = 0.0015, correction.ion = "tanmix07")

## [1] 96.46186
```

The ion correction by Owczarzy et al. (2008) is used by default according to the $\frac{[\text{Mg}^{2+}]^{0.5}}{[\text{Mon}^+]}$ ratio, where $[\text{Mon}^+] = [\text{Na}^+] + [\text{Tris}^+] + [\text{K}^+]$.

If,

- $[\text{K}^+] = 0$, default sodium correction is used;
- $\text{Ratio} < 0.22$, default sodium correction is used;
- $0.22 \leq \text{Ratio} < 6$, default mixed Na and Mg correction is used and
- $\text{Ratio} \geq 6$, default magnesium correction is used.

Note that $[\text{Tris}^+]$ is about half of the total tris buffer concentration.

5.2.4 Sodium equivalent concentration methods

The available correction methods for mixed sodium magnesium concentration are given in **Table 24**.

Table 24: Details of the methods for computation of sodium equivalent concentration in the presence of other ions

| Correction | Type | Limits/Remarks | Reference |
|------------|------|----------------|---------------------|
| ahs01* | DNA | | Ahsen et al. (2001) |
| mit96 | DNA | | Mitsuhashi (1996) |
| pey00 | DNA | | Peyret (2000) |

* Default method for computation.


```
# Na equivalent concentration method - default (DNA/DNA)
melting(sequence = "CAGCCTCGTCGCAGC",
        nucleic.acid.conc = 0.000002, hybridisation.type = "dnadna",
        Na.conc = 0.069, Mg.conc = 0.0015)
```

```
## [1] 65.83371
```

```
# Na equivalent concentration method - ahs01 (DNA/DNA)
melting(sequence = "CAGCCTCGTCGCAGC",
        nucleic.acid.conc = 0.000002, hybridisation.type = "dnadna",
        Na.conc = 0.069, Mg.conc = 0.0015, method.Naeq = "ahs01")
```

```
## [1] 65.83371
```

```
# Na equivalent concentration method - mit96 (DNA/DNA)
melting(sequence = "CAGCCTCGTCGCAGC",
        nucleic.acid.conc = 0.000002, hybridisation.type = "dnadna",
        Na.conc = 0.069, Mg.conc = 0.0015, method.Naeq = "mit96")
```

```
## [1] 65.83371
```

```
# Na equivalent concentration method - pey00 (DNA/DNA)
melting(sequence = "CAGCCTCGTCGCAGC",
        nucleic.acid.conc = 0.000002, hybridisation.type = "dnadna",
        Na.conc = 0.069, Mg.conc = 0.0015, method.Naeq = "pey00")
```

```
## [1] 65.83371
```

5.3 Denaturing agent corrections

These include melting temperature corrections for concentration of formamide and DMSO.

5.3.1 DMSO corrections

The available correction methods for DMSO concentration are given in **Table 25**.

Table 25: Details of the corrections for DMSO concentration

| Correction | Type | Limits/Remarks | Reference |
|------------|------|---------------------------------------|------------------------|
| ahs01* | DNA | Not tested with experimental results. | Ahsen et al. (2001) |
| cu176 | DNA | Not tested with experimental results. | Cullen and Bick (1976) |

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| Correction | Type | Limits/Remarks | Reference |
|------------|------|---------------------------------------|--------------------------|
| esc80 | DNA | Not tested with experimental results. | Escara and Hutton (1980) |
| mus81 | DNA | Not tested with experimental results. | Musielski et al. (1981) |

* Default method for computation.

```
# DMSO correction - default (DNA/DNA)
melting(sequence = "CAGCCTCGTCGCAGC",
        nucleic.acid.conc = 0.000002, hybridisation.type = "dnadna",
        Na.conc = 1, DMSO.conc = 10)
```

```
## [1] 65.40154
```

```
# DMSO correction - ahs01 (DNA/DNA)
melting(sequence = "CAGCCTCGTCGCAGC",
        nucleic.acid.conc = 0.000002, hybridisation.type = "dnadna",
        Na.conc = 1, DMSO.conc = 10, correction.DMSO = "ahs01")
```

```
## [1] 65.40154
```

```
# DMSO correction - cul76 (DNA/DNA)
melting(sequence = "CAGCCTCGTCGCAGC",
        nucleic.acid.conc = 0.000002, hybridisation.type = "dnadna",
        Na.conc = 1, DMSO.conc = 10, correction.DMSO = "cul76")
```

```
## [1] 67.90154
```

```
# DMSO correction - esc80 (DNA/DNA)
melting(sequence = "CAGCCTCGTCGCAGC",
        nucleic.acid.conc = 0.000002, hybridisation.type = "dnadna",
        Na.conc = 1, DMSO.conc = 10, correction.DMSO = "esc80")
```

```
## [1] 66.15154
```

```
# DMSO correction - mus80 (DNA/DNA)
melting(sequence = "CAGCCTCGTCGCAGC",
        nucleic.acid.conc = 0.000002, hybridisation.type = "dnadna",
        Na.conc = 1, DMSO.conc = 10, correction.DMSO = "mus81")
```

```
## [1] 66.90154
```

5.3.2 Formamide corrections

The available correction methods for formamide concentration are given in **Table 26**.

Table 26: Details of the corrections for formamide concentration

| Correction | Type | Limits/Remarks | Reference |
|------------|------|--|---|
| bla96* | DNA | With formamide concentration in mol/L. | Blake (1996) |
| lincorr | DNA | With a % of formamide volume. | McConaughy et al. (1969), Record (1967), Casey and Davidson (1977), Hutton (1977) |

* Default method for computation.

```
# Formamide correction - default (DNA/DNA)
melting(sequence = "CAGCCTCGTCGCAGC",
         nucleic.acid.conc = 0.000002, hybridisation.type = "dnadna",
         Na.conc = 1, formamide.conc = 0.06)

## [1] 72.74867

# Formamide correction - bla96 (DNA/DNA)
melting(sequence = "CAGCCTCGTCGCAGC",
         nucleic.acid.conc = 0.000002, hybridisation.type = "dnadna",
         Na.conc = 1, formamide.conc = 0.06, correction.formamide = "bla96")

## [1] 72.74867

# Formamide correction - lincorr (DNA/DNA)
melting(sequence = "CAGCCTCGTCGCAGC",
         nucleic.acid.conc = 0.000002, hybridisation.type = "dnadna",
         Na.conc = 1, formamide.conc = 10, correction.formamide = "lincorr")

## [1] 66.40154
```

6 Equivalent options in MELTING 5

The options in MELTING 5 command line equivalent to the arguments in `rmelting` are given in **Table 27**.

Table 27: Arguments in `rmelting` and their equivalent options in MELTING 5 command line.

| <code>rmelting</code> | MELTING 5 (command line) |
|---|--------------------------|
| <code>sequence</code> | <code>-S</code> |
| <code>comp.sequence</code> | <code>-C</code> |
| <code>nucleic.acid.conc</code> | <code>-P</code> |
| <code>hybridisation.type</code> | <code>-H</code> |
| <code>Na.conc</code> | <code>-E</code> |
| <code>Mg.conc</code> | <code>-E</code> |
| <code>Tris.conc</code> | <code>-E</code> |
| <code>K.conc</code> | <code>-E</code> |
| <code>dNTP.conc</code> | <code>-E</code> |
| <code>DMSO.conc</code> | <code>-E</code> |
| <code>formamide.conc</code> | <code>-E</code> |
| <code>size.threshold</code> | <code>-T</code> |
| <code>self</code> | <code>-self</code> |
| <code>correction.factor</code> | <code>-F</code> |
| <code>method.approx</code> | <code>-am</code> |
| <code>method.nn</code> | <code>-nn</code> |
| <code>method.GU</code> | <code>-GU</code> |
| <code>method.singleMM</code> | <code>-sinMM</code> |
| <code>method.tandemMM</code> | <code>-tanMM</code> |
| <code>method.single.dangle</code> | <code>-sinDE</code> |
| <code>method.double.dangle</code> | <code>-secDE</code> |
| <code>method.long.dangle</code> | <code>-lonDE</code> |
| <code>method.internal.loop</code> | <code>-intLP</code> |
| <code>method.single.bulge.loop</code> | <code>-sinBU</code> |
| <code>method.long.bulge.loop</code> | <code>-lonBU</code> |
| <code>method.CNG</code> | <code>-CNG</code> |
| <code>method.inosine</code> | <code>-ino</code> |
| <code>method.hydroxyadenine</code> | <code>-ha</code> |
| <code>method.azobenzenes</code> | <code>-azo</code> |
| <code>method.locked</code> | <code>-lck</code> |
| <code>method.consecutive.locked</code> | <code>-tanLck</code> |
| <code>method.consecutive.locked.singleMM</code> | <code>-sinMMLck</code> |
| <code>correction.ion</code> | <code>-ion</code> |
| <code>method.Naeq</code> | <code>-naeq</code> |

7 Batch computation

Melting temperature for multiple nucleic acid duplexes can be computed using the `meltingBatch` function.

```
sequence <- c("CAAAAAG", "CAAAAAG", "TTTATAATAAA", "CCATCGCTACC",
              "CAAAACAAAG", "CCATTGCTACC", "CAAAAAAAG", "GTGAAC", "AAAAAAA",
              "CAACTTGATATTATTA", "CAAATAAAG", "GCGAGC", "GGGACC",
```

```

"CAAAGAAAG", "CTGACAAGTGTC", "GCGAAAAGCG")

meltingBatch(sequence, nucleic.acid.conc = 0.0004,
             hybridisation.type = "dnadna", Na.conc = 1)

##      Environment.Sequence Environment.Complementary sequence
## [1,] "CAAAAAG"          "GTTTTTC"
## [2,] "CAAAAAG"          "GTTTTTC"
## [3,] "TTTATAATAAA"      "AAAATATTATTT"
## [4,] "CCATCGCTACC"      "GGTAGCGATGG"
## [5,] "CAAACAAAG"        "GTTTGTTTC"
## [6,] "CCATTGCTACC"      "GGTAACGATGG"
## [7,] "CAAAAAAAG"        "GTTTTTTTC"
## [8,] "GTGAAC"           "CACTTG"
## [9,] "AAAAAAA"          "TTTTTTT"
## [10,] "CAACTTGATATTATTA" "GTTGAACTATAATAAT"
## [11,] "CAAATAAAG"        "GTTTATTTTC"
## [12,] "GCGAGC"           "CGCTCG"
## [13,] "GGGACC"           "CCCTGG"
## [14,] "CAAAGAAAG"        "GTTTCTTTC"
## [15,] "CTGACAAGTGTC"     "GACTGTTTCACAG"
## [16,] "GCGAAAAGCG"       "CGCTTTTCGC"
##      Environment.Nucleic acid concentration (M) Environment.Hybridization type
## [1,] "4e-04"          "dnadna"
## [2,] "4e-04"          "dnadna"
## [3,] "4e-04"          "dnadna"
## [4,] "4e-04"          "dnadna"
## [5,] "4e-04"          "dnadna"
## [6,] "4e-04"          "dnadna"
## [7,] "4e-04"          "dnadna"
## [8,] "4e-04"          "dnadna"
## [9,] "4e-04"          "dnadna"
## [10,] "4e-04"         "dnadna"
## [11,] "4e-04"         "dnadna"
## [12,] "4e-04"         "dnadna"
## [13,] "4e-04"         "dnadna"
## [14,] "4e-04"         "dnadna"
## [15,] "4e-04"         "dnadna"
## [16,] "4e-04"         "dnadna"
##      Environment.Na concentration (M) Environment.Mg concentration (M)
## [1,] "1"              "0"
## [2,] "1"              "0"
## [3,] "1"              "0"
## [4,] "1"              "0"
## [5,] "1"              "0"

```

```
## [6,] "1" "0"
## [7,] "1" "0"
## [8,] "1" "0"
## [9,] "1" "0"
## [10,] "1" "0"
## [11,] "1" "0"
## [12,] "1" "0"
## [13,] "1" "0"
## [14,] "1" "0"
## [15,] "1" "0"
## [16,] "1" "0"
## Environment.Tris concentration (M) Environment.K concentration (M)
## [1,] "0" "0"
## [2,] "0" "0"
## [3,] "0" "0"
## [4,] "0" "0"
## [5,] "0" "0"
## [6,] "0" "0"
## [7,] "0" "0"
## [8,] "0" "0"
## [9,] "0" "0"
## [10,] "0" "0"
## [11,] "0" "0"
## [12,] "0" "0"
## [13,] "0" "0"
## [14,] "0" "0"
## [15,] "0" "0"
## [16,] "0" "0"
## Environment.dNTP concentration (M) Environment.DMSO concentration (%)
## [1,] "0" "0"
## [2,] "0" "0"
## [3,] "0" "0"
## [4,] "0" "0"
## [5,] "0" "0"
## [6,] "0" "0"
## [7,] "0" "0"
## [8,] "0" "0"
## [9,] "0" "0"
## [10,] "0" "0"
## [11,] "0" "0"
## [12,] "0" "0"
## [13,] "0" "0"
## [14,] "0" "0"
## [15,] "0" "0"
## [16,] "0" "0"
## Environment.Formamide concentration (M or %)
```

```
## [1,] "0"
## [2,] "0"
## [3,] "0"
## [4,] "0"
## [5,] "0"
## [6,] "0"
## [7,] "0"
## [8,] "0"
## [9,] "0"
## [10,] "0"
## [11,] "0"
## [12,] "0"
## [13,] "0"
## [14,] "0"
## [15,] "0"
## [16,] "0"
##      Environment.Self complementarity Environment.Correction factor
## [1,] "FALSE"                                "4"
## [2,] "FALSE"                                "4"
## [3,] "FALSE"                                "4"
## [4,] "FALSE"                                "4"
## [5,] "FALSE"                                "4"
## [6,] "FALSE"                                "4"
## [7,] "FALSE"                                "4"
## [8,] "FALSE"                                "4"
## [9,] "FALSE"                                "4"
## [10,] "FALSE"                               "4"
## [11,] "FALSE"                               "4"
## [12,] "FALSE"                               "4"
## [13,] "FALSE"                               "4"
## [14,] "FALSE"                               "4"
## [15,] "FALSE"                               "4"
## [16,] "FALSE"                               "4"
##      Options.Approximative formula Options.Nearest neighbour model
## [1,] NA                                NA
## [2,] NA                                NA
## [3,] NA                                NA
## [4,] NA                                NA
## [5,] NA                                NA
## [6,] NA                                NA
## [7,] NA                                NA
## [8,] NA                                NA
## [9,] NA                                NA
## [10,] NA                               NA
## [11,] NA                               NA
## [12,] NA                               NA
```

```
## [13,] NA NA
## [14,] NA NA
## [15,] NA NA
## [16,] NA NA
## Options.GU model Options.Single mismatch model
## [1,] NA NA
## [2,] NA NA
## [3,] NA NA
## [4,] NA NA
## [5,] NA NA
## [6,] NA NA
## [7,] NA NA
## [8,] NA NA
## [9,] NA NA
## [10,] NA NA
## [11,] NA NA
## [12,] NA NA
## [13,] NA NA
## [14,] NA NA
## [15,] NA NA
## [16,] NA NA
## Options.Tandem mismatch model Options.Single dangling end model
## [1,] NA NA
## [2,] NA NA
## [3,] NA NA
## [4,] NA NA
## [5,] NA NA
## [6,] NA NA
## [7,] NA NA
## [8,] NA NA
## [9,] NA NA
## [10,] NA NA
## [11,] NA NA
## [12,] NA NA
## [13,] NA NA
## [14,] NA NA
## [15,] NA NA
## [16,] NA NA
## Options.Double dangling end model Options.Long dangling end model
## [1,] NA NA
## [2,] NA NA
## [3,] NA NA
## [4,] NA NA
## [5,] NA NA
## [6,] NA NA
## [7,] NA NA
```

```

## [8,] NA NA
## [9,] NA NA
## [10,] NA NA
## [11,] NA NA
## [12,] NA NA
## [13,] NA NA
## [14,] NA NA
## [15,] NA NA
## [16,] NA NA
## Options.Internal loop model Options.Single bulge loop model
## [1,] NA NA
## [2,] NA NA
## [3,] NA NA
## [4,] NA NA
## [5,] NA NA
## [6,] NA NA
## [7,] NA NA
## [8,] NA NA
## [9,] NA NA
## [10,] NA NA
## [11,] NA NA
## [12,] NA NA
## [13,] NA NA
## [14,] NA NA
## [15,] NA NA
## [16,] NA NA
## Options.Long bulge loop model Options.CNG repeats model
## [1,] NA NA
## [2,] NA NA
## [3,] NA NA
## [4,] NA NA
## [5,] NA NA
## [6,] NA NA
## [7,] NA NA
## [8,] NA NA
## [9,] NA NA
## [10,] NA NA
## [11,] NA NA
## [12,] NA NA
## [13,] NA NA
## [14,] NA NA
## [15,] NA NA
## [16,] NA NA
## Options.Inosine bases model Options.Hydroxyadenine bases model
## [1,] NA NA
## [2,] NA NA

```

```
## [3,] NA NA
## [4,] NA NA
## [5,] NA NA
## [6,] NA NA
## [7,] NA NA
## [8,] NA NA
## [9,] NA NA
## [10,] NA NA
## [11,] NA NA
## [12,] NA NA
## [13,] NA NA
## [14,] NA NA
## [15,] NA NA
## [16,] NA NA
## Options.Azobenzenes model Options.Locked nucleic acids model
## [1,] NA NA
## [2,] NA NA
## [3,] NA NA
## [4,] NA NA
## [5,] NA NA
## [6,] NA NA
## [7,] NA NA
## [8,] NA NA
## [9,] NA NA
## [10,] NA NA
## [11,] NA NA
## [12,] NA NA
## [13,] NA NA
## [14,] NA NA
## [15,] NA NA
## [16,] NA NA
## Options.Ion correction method Options.Na equivalence correction method
## [1,] NA NA
## [2,] NA NA
## [3,] NA NA
## [4,] NA NA
## [5,] NA NA
## [6,] NA NA
## [7,] NA NA
## [8,] NA NA
## [9,] NA NA
## [10,] NA NA
## [11,] NA NA
## [12,] NA NA
## [13,] NA NA
## [14,] NA NA
```

```

## [15,] NA NA
## [16,] NA NA
## Options.DMSO correction method Options.Formamide correction method
## [1,] NA NA
## [2,] NA NA
## [3,] NA NA
## [4,] NA NA
## [5,] NA NA
## [6,] NA NA
## [7,] NA NA
## [8,] NA NA
## [9,] NA NA
## [10,] NA NA
## [11,] NA NA
## [12,] NA NA
## [13,] NA NA
## [14,] NA NA
## [15,] NA NA
## [16,] NA NA
## Options.Mode Results.Enthalpy (cal) Results.Entropy (cal)
## [1,] NA "-47700" "-138.1"
## [2,] NA "-55600" "-160.3"
## [3,] NA "-78800" "-229.7"
## [4,] NA "-83500" "-227"
## [5,] NA "-64600" "-183.2"
## [6,] NA "-81100" "-222.5"
## [7,] NA "-63500" "-182.5"
## [8,] NA "-41200" "-117.5"
## [9,] NA "-50700" "-147.2"
## [10,] NA "-113800" "-323.6"
## [11,] NA "-62100" "-179.8"
## [12,] NA "-46000" "-124.8"
## [13,] NA "-40400" "-109.9"
## [14,] NA "-63700" "-181.3"
## [15,] NA "-90400" "-249.5"
## [16,] NA "-80300" "-218.6"
## Results.Enthalpy (J) Results.Entropy (J) Results.Melting temperature (C)
## [1,] "-199386" "-577.258" "31.7814953255144"
## [2,] "-232408" "-670.054" "38.1103863719918"
## [3,] "-329384" "-960.146" "44.5553259145469"
## [4,] "-349030" "-948.86" "67.2098590318261"
## [5,] "-270028" "-765.776" "47.400072116762"
## [6,] "-338998" "-930.05" "63.6040550863501"
## [7,] "-265430" "-762.85" "43.0400604037136"
## [8,] "-172216" "-491.15" "30.1735038367475"
## [9,] "-211926" "-615.296" "33.1415226764116"

```

```
## [10,] "-475684"          "-1352.648"          "59.6680431282422"
## [11,] "-259578"          "-751.564"           "40.2828264688437"
## [12,] "-192280"          "-521.664"           "48.2393469411973"
## [13,] "-168872"          "-459.382"           "41.9123740666287"
## [14,] "-266266"          "-757.834"           "45.9425910944819"
## [15,] "-377872"          "-1042.91"           "64.379329012421"
## [16,] "-335654"          "-913.748"           "65.7707030297917"
##      Message
## [1,] NA
## [2,] NA
## [3,] NA
## [4,] NA
## [5,] NA
## [6,] NA
## [7,] NA
## [8,] NA
## [9,] NA
## [10,] NA
## [11,] NA
## [12,] NA
## [13,] NA
## [14,] NA
## [15,] NA
## [16,] NA
```

Complementary sequences are computed by default, but need to be specified in case of mismatches, inosine(s) or hydroxyadenine(s) between the two strands.

```
seq <- c("GCAUACG", "CAGUAGGUC", "CGCUCGC", "GAGUGGAG", "GACAGGCUC",
         "CAGUACGUC", "GACAUCUG", "GACCACUG", "CAGAAUGUC", "GCGUCGC",
         "CGUCCGG", "GACUCUCUG", "CAGCUGGUC", "GACUAGCUG", "CUCUGCUC",
         "GCGUCCG", "GUCCGCG", "CGAUCAC", "GACUACCUG", "GACGAUCUG")

comp.seq <- c("CGUUUGC", "GUCGGCCAG", "GCGUGCG", "CUCUUCUC", "CUGUGCGAC",
              "GUCGGGCAG", "CUGUUGGAC", "CUGGGGGAC", "GUCUGGCAG", "CGCUGCG",
              "GCUGGCC", "CUGAUAGAC", "GUCGUUCAG", "CUGAGCGAC", "GAGUUGAG",
              "CGCUGGC", "CUGGCGC", "GCUUGUG", "CUGAGGGAC", "CUGCCAGAC")

meltingBatch(sequence = seq, comp.seq = comp.seq, nucleic.acid.conc = 0.0004,
              hybridisation.type = "rnarna", Na.conc = 1,
              method.singleMM = "tur06")
```

```
##      Environment.Sequence Environment.Complementary sequence
## [1,] "GCAUACG"          "CGUUUGC"
## [2,] "CAGUAGGUC"        "GUCGGCCAG"
## [3,] "CGCUCGC"          "GCGUGCG"
## [4,] "GAGUGGAG"         "CUCUUCUC"
```

```

## [5,] "GACAGGCUG"      "CUGUGCGAC"
## [6,] "CAGUACGUC"      "GUCGGGCAG"
## [7,] "GACAUCCUG"      "CUGUUGGAC"
## [8,] "GACCACCUG"      "CUGGGGGAC"
## [9,] "CAGAAUGUC"      "GUCUGGCAG"
## [10,] "GCGUCGC"       "CGCUGCG"
## [11,] "CGUCCGG"       "GCUGGCC"
## [12,] "GACUCUCUG"     "CUGAUAGAC"
## [13,] "CAGCUGGUC"     "GUCGUUCAG"
## [14,] "GACUAGCUG"     "CUGAGCGAC"
## [15,] "CUCUGCUC"      "GAGUUGAG"
## [16,] "GCGUCCG"       "CGCUGGC"
## [17,] "GUCCGCG"       "CUGGCGC"
## [18,] "CGAUCAC"       "GCUUGUG"
## [19,] "GACUACCUG"     "CUGAGGGAC"
## [20,] "GACGAUCUG"     "CUGCCAGAC"
##      Environment.Nucleic acid concentration (M) Environment.Hybridization type
## [1,] "4e-04"                                     "rna"
## [2,] "4e-04"                                     "rna"
## [3,] "4e-04"                                     "rna"
## [4,] "4e-04"                                     "rna"
## [5,] "4e-04"                                     "rna"
## [6,] "4e-04"                                     "rna"
## [7,] "4e-04"                                     "rna"
## [8,] "4e-04"                                     "rna"
## [9,] "4e-04"                                     "rna"
## [10,] "4e-04"                                    "rna"
## [11,] "4e-04"                                    "rna"
## [12,] "4e-04"                                    "rna"
## [13,] "4e-04"                                    "rna"
## [14,] "4e-04"                                    "rna"
## [15,] "4e-04"                                    "rna"
## [16,] "4e-04"                                    "rna"
## [17,] "4e-04"                                    "rna"
## [18,] "4e-04"                                    "rna"
## [19,] "4e-04"                                    "rna"
## [20,] "4e-04"                                    "rna"
##      Environment.Na concentration (M) Environment.Mg concentration (M)
## [1,] "1"                                     "0"
## [2,] "1"                                     "0"
## [3,] "1"                                     "0"
## [4,] "1"                                     "0"
## [5,] "1"                                     "0"
## [6,] "1"                                     "0"
## [7,] "1"                                     "0"
## [8,] "1"                                     "0"

```

```
## [9,] "1" "0"
## [10,] "1" "0"
## [11,] "1" "0"
## [12,] "1" "0"
## [13,] "1" "0"
## [14,] "1" "0"
## [15,] "1" "0"
## [16,] "1" "0"
## [17,] "1" "0"
## [18,] "1" "0"
## [19,] "1" "0"
## [20,] "1" "0"
## Environment.Tris concentration (M) Environment.K concentration (M)
## [1,] "0" "0"
## [2,] "0" "0"
## [3,] "0" "0"
## [4,] "0" "0"
## [5,] "0" "0"
## [6,] "0" "0"
## [7,] "0" "0"
## [8,] "0" "0"
## [9,] "0" "0"
## [10,] "0" "0"
## [11,] "0" "0"
## [12,] "0" "0"
## [13,] "0" "0"
## [14,] "0" "0"
## [15,] "0" "0"
## [16,] "0" "0"
## [17,] "0" "0"
## [18,] "0" "0"
## [19,] "0" "0"
## [20,] "0" "0"
## Environment.dNTP concentration (M) Environment.DMSO concentration (%)
## [1,] "0" "0"
## [2,] "0" "0"
## [3,] "0" "0"
## [4,] "0" "0"
## [5,] "0" "0"
## [6,] "0" "0"
## [7,] "0" "0"
## [8,] "0" "0"
## [9,] "0" "0"
## [10,] "0" "0"
## [11,] "0" "0"
## [12,] "0" "0"
```

```
## [13,] "0" "0"
## [14,] "0" "0"
## [15,] "0" "0"
## [16,] "0" "0"
## [17,] "0" "0"
## [18,] "0" "0"
## [19,] "0" "0"
## [20,] "0" "0"
##      Environment.Formamide concentration (M or %)
## [1,] "0"
## [2,] "0"
## [3,] "0"
## [4,] "0"
## [5,] "0"
## [6,] "0"
## [7,] "0"
## [8,] "0"
## [9,] "0"
## [10,] "0"
## [11,] "0"
## [12,] "0"
## [13,] "0"
## [14,] "0"
## [15,] "0"
## [16,] "0"
## [17,] "0"
## [18,] "0"
## [19,] "0"
## [20,] "0"
##      Environment.Self complementarity Environment.Correction factor
## [1,] "FALSE" "4"
## [2,] "FALSE" "4"
## [3,] "FALSE" "4"
## [4,] "FALSE" "4"
## [5,] "FALSE" "4"
## [6,] "FALSE" "4"
## [7,] "FALSE" "4"
## [8,] "FALSE" "4"
## [9,] "FALSE" "4"
## [10,] "FALSE" "4"
## [11,] "FALSE" "4"
## [12,] "FALSE" "4"
## [13,] "FALSE" "4"
## [14,] "FALSE" "4"
## [15,] "FALSE" "4"
## [16,] "FALSE" "4"
```

```
## [17,] "FALSE" "4"
## [18,] "FALSE" "4"
## [19,] "FALSE" "4"
## [20,] "FALSE" "4"
## Options.Approximative formula Options.Nearest neighbour model
## [1,] NA NA
## [2,] NA NA
## [3,] NA NA
## [4,] NA NA
## [5,] NA NA
## [6,] NA NA
## [7,] NA NA
## [8,] NA NA
## [9,] NA NA
## [10,] NA NA
## [11,] NA NA
## [12,] NA NA
## [13,] NA NA
## [14,] NA NA
## [15,] NA NA
## [16,] NA NA
## [17,] NA NA
## [18,] NA NA
## [19,] NA NA
## [20,] NA NA
## Options.GU model Options.Single mismatch model
## [1,] NA NA
## [2,] NA NA
## [3,] NA NA
## [4,] NA NA
## [5,] NA NA
## [6,] NA NA
## [7,] NA NA
## [8,] NA NA
## [9,] NA NA
## [10,] NA NA
## [11,] NA NA
## [12,] NA NA
## [13,] NA NA
## [14,] NA NA
## [15,] NA NA
## [16,] NA NA
## [17,] NA NA
## [18,] NA NA
## [19,] NA NA
## [20,] NA NA
```



```
##      Options.Tandem mismatch model Options.Single dangling end model
## [1,] NA                          NA
## [2,] NA                          NA
## [3,] NA                          NA
## [4,] NA                          NA
## [5,] NA                          NA
## [6,] NA                          NA
## [7,] NA                          NA
## [8,] NA                          NA
## [9,] NA                          NA
## [10,] NA                         NA
## [11,] NA                         NA
## [12,] NA                         NA
## [13,] NA                         NA
## [14,] NA                         NA
## [15,] NA                         NA
## [16,] NA                         NA
## [17,] NA                         NA
## [18,] NA                         NA
## [19,] NA                         NA
## [20,] NA                         NA
##      Options.Double dangling end model Options.Long dangling end model
## [1,] NA                          NA
## [2,] NA                          NA
## [3,] NA                          NA
## [4,] NA                          NA
## [5,] NA                          NA
## [6,] NA                          NA
## [7,] NA                          NA
## [8,] NA                          NA
## [9,] NA                          NA
## [10,] NA                         NA
## [11,] NA                         NA
## [12,] NA                         NA
## [13,] NA                         NA
## [14,] NA                         NA
## [15,] NA                         NA
## [16,] NA                         NA
## [17,] NA                         NA
## [18,] NA                         NA
## [19,] NA                         NA
## [20,] NA                         NA
##      Options.Internal loop model Options.Single bulge loop model
## [1,] NA                          NA
## [2,] NA                          NA
## [3,] NA                          NA
```

```
## [4,] NA NA
## [5,] NA NA
## [6,] NA NA
## [7,] NA NA
## [8,] NA NA
## [9,] NA NA
## [10,] NA NA
## [11,] NA NA
## [12,] NA NA
## [13,] NA NA
## [14,] NA NA
## [15,] NA NA
## [16,] NA NA
## [17,] NA NA
## [18,] NA NA
## [19,] NA NA
## [20,] NA NA
## Options.Long bulge loop model Options.CNG repeats model
## [1,] NA NA
## [2,] NA NA
## [3,] NA NA
## [4,] NA NA
## [5,] NA NA
## [6,] NA NA
## [7,] NA NA
## [8,] NA NA
## [9,] NA NA
## [10,] NA NA
## [11,] NA NA
## [12,] NA NA
## [13,] NA NA
## [14,] NA NA
## [15,] NA NA
## [16,] NA NA
## [17,] NA NA
## [18,] NA NA
## [19,] NA NA
## [20,] NA NA
## Options.Inosine bases model Options.Hydroxyadenine bases model
## [1,] NA NA
## [2,] NA NA
## [3,] NA NA
## [4,] NA NA
## [5,] NA NA
## [6,] NA NA
## [7,] NA NA
```

```

## [8,] NA NA
## [9,] NA NA
## [10,] NA NA
## [11,] NA NA
## [12,] NA NA
## [13,] NA NA
## [14,] NA NA
## [15,] NA NA
## [16,] NA NA
## [17,] NA NA
## [18,] NA NA
## [19,] NA NA
## [20,] NA NA
## Options.Azobenzenes model Options.Locked nucleic acids model
## [1,] NA NA
## [2,] NA NA
## [3,] NA NA
## [4,] NA NA
## [5,] NA NA
## [6,] NA NA
## [7,] NA NA
## [8,] NA NA
## [9,] NA NA
## [10,] NA NA
## [11,] NA NA
## [12,] NA NA
## [13,] NA NA
## [14,] NA NA
## [15,] NA NA
## [16,] NA NA
## [17,] NA NA
## [18,] NA NA
## [19,] NA NA
## [20,] NA NA
## Options.Ion correction method Options.Na equivalence correction method
## [1,] NA NA
## [2,] NA NA
## [3,] NA NA
## [4,] NA NA
## [5,] NA NA
## [6,] NA NA
## [7,] NA NA
## [8,] NA NA
## [9,] NA NA
## [10,] NA NA
## [11,] NA NA

```

--- Computation of melting temperature of nucleic acid duplexes with `rmelting`

```
## [12,] NA NA
## [13,] NA NA
## [14,] NA NA
## [15,] NA NA
## [16,] NA NA
## [17,] NA NA
## [18,] NA NA
## [19,] NA NA
## [20,] NA NA
## Options.DMSO correction method Options.Formamide correction method
## [1,] NA NA
## [2,] NA NA
## [3,] NA NA
## [4,] NA NA
## [5,] NA NA
## [6,] NA NA
## [7,] NA NA
## [8,] NA NA
## [9,] NA NA
## [10,] NA NA
## [11,] NA NA
## [12,] NA NA
## [13,] NA NA
## [14,] NA NA
## [15,] NA NA
## [16,] NA NA
## [17,] NA NA
## [18,] NA NA
## [19,] NA NA
## [20,] NA NA
## Options.Mode Results.Enthalpy (cal) Results.Entropy (cal)
## [1,] NA "-47650" "-138.8"
## [2,] NA "-71130" "-200.5"
## [3,] NA "-57930" "-164.1"
## [4,] NA "-60570" "-176.6"
## [5,] NA "-79870" "-219.9"
## [6,] NA "-68380" "-194.5"
## [7,] NA "-73880" "-208.3"
## [8,] NA "-78430" "-218.3"
## [9,] NA "-59650" "-171.5"
## [10,] NA "-61330" "-173.8"
## [11,] NA "-58350" "-165.4"
## [12,] NA "-64570" "-184.7"
## [13,] NA "-70970" "-200.6"
## [14,] NA "-72010" "-203"
## [15,] NA "-58820" "-171"
```

```

## [16,] NA          "-59840"          "-169.6"
## [17,] NA          "-59840"          "-169.6"
## [18,] NA          "-50210"          "-148.3"
## [19,] NA          "-70520"          "-198.8"
## [20,] NA          "-69730"          "-198.2"
##      Results.Enthalpy (J) Results.Entropy (J) Results.Melting temperature (C)
## [1,] "-199177"          "-580.184"          "30.1048299398322"
## [2,] "-297323.4"        "-838.09"          "51.8989532242754"
## [3,] "-242147.4"        "-685.938"          "44.3989325444856"
## [4,] "-253182.6"        "-738.188"          "37.5791954133529"
## [5,] "-333856.6"        "-919.182"          "62.1162425798375"
## [6,] "-285828.4"        "-813.01"          "48.141439592185"
## [7,] "-308818.4"        "-870.694"          "52.845957204839"
## [8,] "-327837.4"        "-912.494"          "58.2977096620104"
## [9,] "-249337"          "-716.87"          "41.08087522322"
## [10,] "-256359.4"        "-726.484"          "46.0633145887674"
## [11,] "-243903"          "-691.372"          "44.4380466975271"
## [12,] "-269902.6"        "-772.046"          "44.8840464672343"
## [13,] "-296654.6"        "-838.508"          "51.0196486690585"
## [14,] "-301001.8"        "-848.54"          "52.203376709706"
## [15,] "-245867.6"        "-714.78"          "37.5268181873443"
## [16,] "-250131.2"        "-708.928"          "45.2688421309843"
## [17,] "-250131.2"        "-708.928"          "45.2688421309843"
## [18,] "-209877.8"        "-619.894"          "28.1788644808993"
## [19,] "-294773.6"        "-830.984"          "51.6345164549562"
## [20,] "-291471.4"        "-828.476"          "48.8860141674642"
##      Message
## [1,] NA
## [2,] NA
## [3,] NA
## [4,] NA
## [5,] NA
## [6,] NA
## [7,] NA
## [8,] NA
## [9,] NA
## [10,] NA
## [11,] NA
## [12,] NA
## [13,] NA
## [14,] NA
## [15,] NA
## [16,] NA
## [17,] NA
## [18,] NA
## [19,] NA

```

```
## [20,] NA
```

8 Further reading

Further details about algorithm, formulae and methods are available in the [MELTING 5 documentation](#).

9 Citing `rmelting`

```
## To cite the R package 'rmelting' in publications use:
##
## Aravind, J. and Krishna, G. K. (2025).  rmelting: R Interface to
## MELTING 5. R package version 1.27.0,
## https://aravind-j.github.io/rmelting/.
##
## A BibTeX entry for LaTeX users is
##
## @Manual{,
##   title = {rmelting: R Interface to MELTING 5},
##   author = {J. Aravind and G. K. Krishna},
##   year = {2025},
##   note = {R package version 1.27.0 https://aravind-j.github.io/rmelting/},
## }
##
## This free and open-source software implements academic research by the
## authors and co-workers. If you use it, please support the project by
## citing the package.
```

10 Session Info

```
sessionInfo()
```

```
## R Under development (unstable) (2025-10-21 r88958)
## Platform: x86_64-apple-darwin20
## Running under: macOS Ventura 13.7.8
##
## Matrix products: default
## BLAS:   /Library/Frameworks/R.framework/Versions/4.6-x86_64/Resources/lib/libRblas.0
## LAPACK: /Library/Frameworks/R.framework/Versions/4.6-x86_64/Resources/lib/libRlapack
##
## locale:
## [1] C/en_US.UTF-8/en_US.UTF-8/C/en_US.UTF-8/en_US.UTF-8
##
## time zone: America/New_York
```

```
## tzcode source: internal
##
## attached base packages:
## [1] stats      graphics  grDevices  utils      datasets  methods   base
##
## other attached packages:
## [1] rmelting_1.27.0 readxl_1.4.5
##
## loaded via a namespace (and not attached):
## [1] digest_0.6.38      fastmap_1.2.0      xfun_0.54          cellranger_1.1.0
## [5] magrittr_2.0.4     glue_1.8.0         tibble_3.3.0       knitr_1.50
## [9] pkgconfig_2.0.3    htmltools_0.5.8.1 rJava_1.0-11       rmarkdown_2.30
## [13] lifecycle_1.0.4    Rdpack_2.6.4       cli_3.6.5          pander_0.6.6
## [17] vctrs_0.6.5        compiler_4.6.0     rbibutils_2.4      tools_4.6.0
## [21] pillar_1.11.1      evaluate_1.0.5     Rcpp_1.1.0         yaml_2.3.10
## [25] rlang_1.1.6
```

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doi:[10.1021/bi701024w](https://doi.org/10.1021/bi701024w).

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