Package 'ProbeDeveloper'

Type Package

Title Develop Hybridization Probes

Version 1.1.1

Date 2026-03-18

Description Hybridization probes for target sequences can be made based on melting temperature value calculated by R package 'TmCalculator' <https://CRAN.R-project.org/package=TmCalculator> and methods extended from Beliveau, B. J.,(2018) <doi:10.1073/pnas.1714530115>, and those hybridization probes can be used to capture specific target regions in fluorescence in situ hybridization and next generation sequence experiments.

License GPL (>= 2)

Imports TmCalculator (>= 1.0.4),Biostrings(>= 2.12.0)

Depends R (>= 2.10)

RoxygenNote 7.3.2

NeedsCompilation no

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Contents

ProbeMake   *Make probes*

Description

Probes are made with a FASTA-formatted input file containing the target sequence. User can specify the allowable ranges of probe length, percent GC content, and adjust melting temperature calculated using nearest neighbor thermodynamics or empirical formulas based on GC content. Candidate probe sequences passing all checks output in BED format.

Usage

```
ProbeMake(
  fafile,
  LN = 90,
  ln = 60,
  TM = 80,
  tm = 60,
  CG = 70,
  cg = 30,
  gap = 0,
  method = c("S2L", "L2S"),
  direction = c("3to5", "5to3"),
  prohibitseq = NULL,
  TmMethod = c("tm_gc", "tm_nn"),
  variant = c("Primer3Plus", "Chester1993", "QuikChange", "Schildkraut1965",
   "Wetmur1991_MELTING", "Wetmur1991_RNA", "Wetmur1991_RNA/DNA", "vonAhsen2001"),
 nn_table = c("DNA_NN_Breslauer_1986", "DNA_NN_Sugimoto_1996", "DNA_NN_Allawi_1998",
    "DNA_NN_SantaLucia_2004", "RNA_NN_Freier_1986", "RNA_NN_Xia_1998",
    "RNA_NN_Chen_2012", "RNA_DNA_NN_Sugimoto_1995"),
  tmm_table = "DNA_TMM_Bommarito_2000",
  imm_table = "DNA_IMM_Peyret_1999",
  de_table = c("DNA_DE_Bommarito_2000", "RNA_DE_Turner_2010"),
  dnac1 = 25,
  dnac2 = 25,
  Na = 0,
  K = 0,
  Tris = 0,
  Mg = 0,
  dNTPs = 0,
 saltcorr = c("Schildkraut2010", "Wetmur1991", "SantaLucia1996", "SantaLucia1998-1",
    "SantaLucia1998-2", "Owczarzy2004", "Owczarzy2008"),
  DMSO = 0,
  fmd = 0,
  DMSOfactor = 0.75,
  fmdfactor = 0.65,
  fmdmethod = c("percent", "molar")
```

)

Arguments

fafile	Input file with a FASTA format read by function readDNAStringSet in R package 'Biostrings'
LN	The maximum allowed probe length, default is 90
ln	The minimum allowed probe length, default is 60
TM	The maximum allowed melting temperature, default is 80
tm	The minimum allowed melting temperature, default is 60
CG	The maximum allowed percent GC content, default is 70
cg	The minimum allowed percent GC content, default is 30
gap	The minimum gap between adjacent probes, default is 0
method	'S2L' is used to design probe extending from minimal length to the maximum until passing all checks, conversely 'L2S' make probe from maximal length to the minimum. Default is 'S2L'
direction	Design probes from 3 to 5 end or from 5 to 3 end of target sequence, default is '3to5'
prohibitseq	Prohibited sequence list, e.g prohibitseq=c("GGGGG","CCCCC"), default is NULL
TmMethod	The method used to calculate Tm, 'tm_nn' and 'tm_gc' can be seleted
variant	Empirical constants coefficient with 8 variant for 'tm_gc' method: Chester1993, QuikChange, Schildkraut1965, Wetmur1991_MELTING, Wetmur1991_RNA, Wetmur1991_RNA/DNA, Primer3Plus and vonAhsen2001
nn_table	Thermodynamic nearest-neighbor parameters for different nucleic acid hybridizations. Eight parameter sets are available, organized by hybridization type:
	DNA/DNA hybridizations: - "DNA_NN_Breslauer_1986": Original DNA/DNA parameters - "DNA_NN_Sugimoto_1996": Improved DNA/DNA parameters - "DNA_NN_Allawi_1998": DNA/DNA parameters with internal mismatch corrections - "DNA_NN_SantaLucia_2004": Updated DNA/DNA parameters
	RNA/RNA hybridizations: - "RNA_NN_Freier_1986": Original RNA/RNA parameters - "RNA_NN_Xia_1998": Improved RNA/RNA parameters - "RNA_NN_Chen_2012": Updated RNA/RNA parameters with GU pair corrections
	RNA/DNA hybridizations: - "RNA_DNA_NN_Sugimoto_1995": RNA/DNA hybridization parameters
tmm_table	Thermodynamic parameters for terminal mismatches. Default: "DNA_TMM_Bommarito_2000" These parameters account for mismatches at the ends of the duplex.
imm_table	Thermodynamic parameters for internal mismatches. Default: "DNA_IMM_Peyret_1999" These parameters account for mismatches within the duplex, including inosine mismatches.
de_table	Thermodynamic parameters for dangling ends. Default: "DNA_DE_Bommarito_2000" Available options: - "DNA_DE_Bommarito_2000": Parameters for DNA dangling ends - "RNA_DE_Turner_2010": Parameters for RNA dangling ends

dnac1	Concentration of the higher concentrated strand [nM]. Typically this will be the primer (for PCR) or the probe. Default: 25
dnac2	Concentration of the lower concentrated strand [nM]. Default: 25
Na	Millimolar concentration of Na, default is 0
K	Millimolar concentration of K, default is 0
Tris	Millimolar concentration of Tris, default is 0
Mg	Millimolar concentration of Mg, default is 0
dNTPs	Millimolar concentration of dNTPs, default is 0
saltcorr	Salt correction method. Options are "Schildkraut2010", "Wetmur1991","SantaLucia1996","SantaLucia19 1","Owczarzy2004","Owczarzy2008". Note that "SantaLucia1998-2" is not available for this function.
DMSO	Percent of DMSO
fmd	Formamide concentration in percentage (fmdmethod="percent") or molar (fmdmethod="molar"). Default is 0.
DMSOfactor	Coeffecient of Tm decreases per percent DMSO. Default=0.75 von Ahsen N (2001) <PMID:11673362>. Other published values are 0.5, 0.6 and 0.675.
fmdfactor	Coeffecient of Tm decrease per percent formamide. Default=0.65. Several papers report factors between 0.6 and 0.72.
fmdmethod	"percent" method for formamide concentration in percentage and "molar" for formamide concentration in molar

Value

Returns a bed file in the format TargetID <tab> Chr <tab> Start <tab> End <tab> Sequence <tab> Tm <tab> GC

Author(s)

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Examples

```
## Not run:
data(samplefa)
ProbeMake(samplefa,LN=90,ln=60,TM=80,tm=70,CG=80,cg=20,TmMethod="tm_nn",Na=50)

## End(Not run)
```

---

samplefa   *sample data for target sequence region with class 'DNAStringSet'*

Description

sample data read by function readDNAStringSet in R package 'Biostrings' from fasta format file, there are two target sequence resion in this data

Usage

```
data("samplefa")
```

Format

Formal class 'DNAStringSet' [package "Biostrings"] with 5 slots

sample data read by function readDNAStringSet in R package 'Biostrings' from fasta format file, which is from ncbiRefSeq database for Homo Sapiens with referece genome version hg19

Examples

```
data(samplefa)
```

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