Package 'eDNAfuns'

October 1, 2025

Type Package

Title Working with Metabarcoding Data in a Tidy Format

Version 0.1.0

Description A series of R functions that come in handy while working with metabarcoding data. The reasoning of doing this is to have the same functions we use all the time stored in a curated, reproducible way.
In a way it is all about putting together the grammar of the 'tidyverse' from Wickham et al.(2019) <doi:10.21105/joss.01686>
with the functions we have used in community ecology compiled in packages like 'vegan' from Dixon (2003) <doi:10.1111/j.1654-1103.2003.tb02228.x>
and 'phyloseq' McMurdie & Holmes (2013) <doi:10.1371/journal.pone.0061217>.
The package includes functions to read sequences from FAST(A/Q) into a tibble ('fasta_reader' and 'fastq_reader'),
to process 'cutadapt' Martin (2011) <doi:10.14806/ej.17.1.200> 'info-file' output. When it comes to sequence counts across samples, the package works with the long format in mind (a three column 'tibble' with Sample, Sequence and counts), with functions to move from there to the wider format.

License GPL (>= 3)

Encoding UTF-8

LazyData true

Suggests insect, testthat, tidyverse, knitr, rmarkdown

Imports dplyr, googlesheets4, phyloseq, purrr, rlang, tibble, tidyr, vegan, ggplot2, vroom, Biostrings, stringr, tidyselect, readr

Depends R (>= 4.1.0)

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Contents

AMPURE . . . 2
ASV_table . . . 3
count_stop_codons . . . 3
eDNAindex . . . 5
example_hashes . . . 6
fasta_reader . . . 6
fasta_writer . . . 7
Index_PCR . . . 8
metadata . . . 8
molarity.data . . . 9
mutation . . . 10
ng2nM . . . 10
OTU_taxonomy . . . 11
plot_seq_len_hist . . . 12
ps . . . 13
read_indexing_PCR . . . 13
read_info_file . . . 15
tally_wide . . . 16
template_PCR . . . 16
test_seqs . . . 17
tibble_to_comm . . . 17
tidy2phyloseq . . . 19
training.ASV.table . . . 20
training.metadata . . . 21
tree . . . 21
UDI_Indices . . . 22
write_indexing_PCR . . . 22

Index **24**

AMPURE *AMPURE*

Description

Description: Placeholder for AMPURE dataset.

Usage

```
AMPURE
```

Format

A tibble/data.frame with X rows and Y columns:

Column1 Description of column1

Column2 Description of column2 ...

Source

Generated by the Author

`ASV_table` *ASV_table*

Description

An abundance table in long format.

Usage

```
ASV_table
```

Format

A data frame with 9 rows and 3 variables:

sample_name Character vector, last digit includes PCR replicate

Hash The sequence found, hashed using sha1 for consistency

nReads Number of times that hash was found in that sample

Details

Contains the number of reads from unique ASVs across different samples.

Source

Generated by the Author

`count_stop_codons` *Count stop codons or return translated sequence*

Description

Given a DNA sequence (either a 'DNAString' object or a character string), this function translates it using a specified genetic code and counts the number of stop codons ('*') in the resulting amino acid sequence. Alternatively, the translated sequence itself can be returned.

Usage

```
count_stop_codons(
  sequence = NULL,
  format = "DNAString",
  codon = 1,
  dictionary = 5,
  return = "count"
)
```

Arguments

`sequence`	A DNA sequence, either as a [Biostrings::DNAString] object or as a character string.
`format`	Input format, either ‘"DNAString"‘ (default) or ‘"character"‘. If ‘"character"‘, the sequence must consist of ‘A‘, ‘C‘, ‘G‘, ‘T‘ only.
`codon`	Integer giving the starting codon position (usually 1, 2, or 3).
`dictionary`	Integer specifying which translation code to use. See [Biostrings::GENETIC_CODE_TABLE] for all options. Common examples:

id	name
1	Standard
2	Vertebrate Mitochondrial
3	Yeast Mitochondrial
4	Mold/Protozoan/Coelenterate/Mycoplasma/Spiroplasma Mitochondrial
5	Invertebrate Mitochondrial
6	Ciliate/Dasycladacean/Hexamita Nuclear
9	Echinoderm/Flatworm Mitochondrial
10	Euplotid Nuclear
11	Bacterial, Archaeal, and Plant Plastid
12	Alternative Yeast Nuclear
13	Ascidian Mitochondrial
14	Alternative Flatworm Mitochondrial
15	Blepharisma Macronuclear
16	Chlorophycean Mitochondrial
21	Trematode Mitochondrial
22	Scenedesmus obliquus Mitochondrial
23	Thraustochytrium Mitochondrial
24	Pterobranchia Mitochondrial
25	Candidate Division SR1 and Gracilibacteria
26	Pachysolen tannophilus Nuclear

`return`	Either ‘"count"‘ (default) to return the number of stop codons, or any other value to return the translated amino acid sequence.

Details

The function uses [Biostrings::translate()] with the specified starting position and genetic code. Translation warnings are suppressed.

Value

If ‘return = "count"‘, an integer giving the number of stop codons. Otherwise, a character string with the translated amino acid sequence.

Author(s)

Ramon Gallego, 2021

Examples

```
if (requireNamespace("Biostrings", quietly = TRUE)) {
  library(Biostrings)
  seq <- DNAString("ATGGCCATTGTAATGGGCCGCTGAAAGGGTGCCCGATAG")
  count_stop_codons(seq, codon = 1, dictionary = 1)
  count_stop_codons(seq, codon = 1, dictionary = 1, return = "translation")
}
```

`eDNAindex` *Create scaled relative proportions of the number of reads of taxa in different samples*

Description

This function takes a long ASV table (tibble) and creates a new column with the relative proportions scaled to their maximum, to avoid the dominance of species with better primer efficiency

Usage

```
eDNAindex(
  tibble,
  Sample_column = Sample,
  OTU_column = Hash,
  Counts_column = nReads,
  Biological_replicate_column = NULL,
  ...
)
```

Arguments

`tibble`	A tibble the ASV table in a long format, with at least three columns, Sample_column, OTU_column, Counts_column
`Sample_column`	The column indicating the sample.
`OTU_column`	The column indicating the OTU/ASV.
`Counts_column`	The column (numeric) with the number of sequences from that OTU in that sample.
`Biological_replicate_column`	The column representing replicate measurements of Sample_column.
`...`	Any extra columns that want to be added to the final dataset (either taxonomical information about OTUs, or metadata information about the samples)

Value

A tibble with at least the columns Sample_column, OTU_column and Normalized.reads

Examples

```
data("training.ASV.table")
eDNAindex(training.ASV.table, Sample_column = Sample_name)
```

`example_hashes` *example_hashes*

Description

Placeholder description for example_hashes dataset.

Usage

```
example_hashes
```

Format

A vector or data frame with X rows/columns

Source

Generated by the Author

`fasta_reader` *Read FASTA or FASTQ files into a tibble*

Description

Functions to read sequence files into a tidy data frame with one row per sequence.

Usage

```
fasta_reader(path_to_fasta)

fastq_reader(path_to_fastq, keepQ = FALSE)
```

Arguments

`path_to_fasta`	Character. Path to a FASTA file.
`path_to_fastq`	Character. Path to a FASTQ file.
`keepQ`	Logical. If 'TRUE', keep a third column with quality scores when reading FASTQ files. Default is 'FALSE'.

Value

- ‘fasta_reader()‘: A tibble with columns: - ‘header‘: sequence identifiers (without the ‘>‘). - ‘seq‘: nucleotide sequences.

- ‘fastq_reader()‘: A tibble with columns: - ‘header‘: sequence identifiers (without the ‘@‘). - ‘seq‘: nucleotide sequences. - ‘Qscores‘ (optional): quality scores, if ‘keepQ = TRUE‘.

Examples

```
fasta_df <- fasta_reader(system.file("extdata", "test.fasta", package="eDNAfuns"))
fastq_df <- fastq_reader(system.file("extdata", "test.fastq", package="eDNAfuns"), keepQ = TRUE)
```

`fasta_writer` *Read FASTA or FASTQ files into a tibble*

Description

Functions to read sequence files into a tidy data frame with one row per sequence.

Usage

```
fasta_writer(df, sequence, header, file.out)

fastq_writer(df, sequence, header, Qscores, file.out)
```

Arguments

`df`	A dataframe where the sequence information is stored, one sequence per row
`sequence`	The name (unquoted) of the column where the sequence information (as characters) is stored
`header`	The name (unquoted) of the column where the header information is stored
`file.out`	Character. Path to the location where to write the file.
`Qscores`	The name (unquoted) of the column where the Quality information (encoded as characters) is stored

Value

- ‘fasta_reader()‘: A tibble with columns: - ‘header‘: sequence identifiers (without the ‘>‘). - ‘seq‘: nucleotide sequences.

- ‘fastq_reader()‘: A tibble with columns: - ‘header‘: sequence identifiers (without the ‘@‘). - ‘seq‘: nucleotide sequences. - ‘Qscores‘ (optional): quality scores, if ‘keepQ = TRUE‘.

Examples

```
fasta_file <- tempfile(fileext = ".fasta")

fasta_df <- fasta_reader(system.file("extdata", "test.fasta", package="eDNAfuns"))

fasta_writer(fasta_df, sequence=seq,
             header = header,
             file.out = fasta_file)

fastq_file <- tempfile(fileext = ".fastq")

fastq_df <- fastq_reader(system.file("extdata", "test.fastq", package="eDNAfuns"), keepQ = TRUE)

fastq_writer(fastq_df, sequence=seq,
             header = header,Qscores= Qscores,
             file.out = fastq_file)
```

Index_PCR *Index_PCR*

Description

Placeholder description for IndexPCR template web address.

Usage

```
Index_PCR
```

Format

A character vector

Source

Generated by the Author, stored in googledrive

---

metadata *metadata*

Description

Placeholder description for metadata dataset.

Usage

```
metadata
```

Format

A data frame with X rows and Y columns:

sample_name Name of sample

Treatment either control or treatment

Day either 1 or 2

Source

Generated by the Author

`molarity.data` *molarity.data*

Description

Placeholder description for molarity.data dataset.

Usage

```
molarity.data
```

Format

A tibble/data.frame with X rows and Y columns

Sample Name of sample

Molarity in nM

Amp_len length of amplicon in bp

mass in ng/μl

Source

Generated by the Author

mutation *Generate mutated DNA sequences*

Description

This function takes DNA sequences and generates mutated variants. Useful for testing classification algorithms on sequences with either PCR-induced or naturally occurring mutations.

Usage

```
mutation(sequence = NULL, format = "bin", n.mutations = NA, prob.mutation = NA)
```

Arguments

sequence	A list of DNA sequences, either as "DNAbin" objects or character vectors.
format	Character. Format of the input sequences. "bin" for DNAbin, "char" for character vectors.
n.mutations	Integer. Number of mutations to introduce per sequence. Exclusive with prob.mutation.
prob.mutation	Numeric. Probability of mutation per position. Exclusive with n.mutations.

Value

A list of mutated sequences of the same class as the input.

Examples

```
data("test_seqs")
mutation(test_seqs, n.mutations = 2)
mutation(test_seqs, prob.mutation = 0.1)
seqs <- fasta_reader(system.file("extdata", "test.fasta", package="eDNAfuns"))
mutation(seqs$seq, format = "char", n.mutations = 1)
```

ng2nM *functions to translate mass into molarity and vice versa, given we are talking about double stranded DNA it requires two inputs, the mass (or molarity) and the length of the DNA fragment It works with the two most common concentrations used in Molecular Ecology labs ng/μl for mass nM for molarity*

Description

functions to translate mass into molarity and vice versa, given we are talking about double stranded DNA it requires two inputs, the mass (or molarity) and the length of the DNA fragment It works with the two most common concentrations used in Molecular Ecology labs ng/μl for mass nM for molarity

Usage

```
ng2nM(ng, length_amplicon)

nM2ng(nM, length_amplicon)
```

Arguments

ng	Numeric. the concentration in ng per μL
length_amplicon	Integer. The length of the DNA fragment in base pairs.
nM	Numeric. The concentration in nmoles per litre

Value

Numeric. The equivalent concentration in nmoles per litre

Numeric. The equivalent concentration in ng per μL

Examples

```
data("molarity.data")
ng2nM(ng=molarity.data$mass, length_amplicon = molarity.data$Amp_len)


data("molarity.data")
nM2ng(nM=molarity.data$Molarity, length_amplicon = molarity.data$Amp_len)
```

OTU_taxonomy *OTU_taxonomy*

Description

Placeholder description for OTU_taxonomy dataset.

Usage

```
OTU_taxonomy
```

Format

A dataframe of DNA sequences names and their taxonomy, with 3 rows and 7 columns

Hash Sequence identifier

Kingdom taxon info at that rank

Phylum taxon info at that rank

Class taxon info at that rank

Order taxon info at that rank

Family taxon info at that rank

Genus taxon info at that rank

Source

Generated by the Author

`plot_seq_len_hist` *Plot sequence length distribution*

Description

This function takes a tibble produced by ‘fasta_reader()‘ or ‘fastq_reader()‘ and plots the distribution of sequence lengths.

Usage

```
plot_seq_len_hist(Hash_tibble, length_col, binwidth = 1, label_interval = 5)
```

Arguments

`Hash_tibble`	A tibble containing a numeric column with sequence lengths.
`length_col`	The column of ‘Hash_tibble‘ containing sequence lengths (unquoted).
`binwidth`	Width of histogram bins. Default = 1.
`label_interval`	Interval for x-axis labels. Default = 5.

Value

A ggplot object.

Examples

```
data("example_hashes")
plot_seq_len_hist(example_hashes, seq_len)
```

ps *ps*

Description

Placeholder description for phyloseq object.

Usage

```
ps
```

Format

Formal class 'phyloseq' with five slots

Source

Generated by the Author

read_indexing_PCR *Read Indexing PCR Spreadsheet*

Description

Reads data from a Google Sheets-based indexing PCR template. You can access the template here, create a copy in your Google Drive, and then create copies for each of your multiplexing PCR experiments.

Reads data from a Google Sheets-based PCR template. You can access the template here, create your own copy, and then create one copy per PCR reaction you want to keep track of.

Usage

```
read_indexing_PCR(ss)

read_step1_PCR(ss, trim = TRUE, name = TRUE)
```

Arguments

ss	Google Sheet ID or URL of the indexing PCR spreadsheet.
trim	Logical. If TRUE, removes rows where 'Sample' is NA (default: TRUE).
name	Logical. If TRUE, adds the spreadsheet name as a 'PCR' column in the sample sheet (default: TRUE).

Details

The function extracts sample information across all plates and returns a tidy dataframe.

Captures reagent mix, cycling conditions, and sample information.

Value

A tibble with columns:

Well Well position

Sample Sample name

Barcode Barcode assigned to the sample

Set Barcode set identifier

A named list with three elements:

PCR_mix tibble of reagents and volumes.

Cycling tibble of PCR cycling conditions.

Samples tibble of samples with columns 'Well', 'Sample', 'Success', 'Notes', and optionally 'PCR'.

See Also

[read_step1_PCR()] for reading normal PCR spreadsheets.

[read_indexing_PCR()] for reading multiplexing PCR spreadsheets.

Examples

```
# Examples are not executed because the function requires identification in Google
## Not run:
data("Index_PCR")
read_indexing_PCR(Index_PCR)

## End(Not run)

# Examples are not executed because the function requires identification in Google
## Not run:
data("template_PCR")
read_step1_PCR(template_PCR)

## End(Not run)
```

`read_info_file` *Read cutadapt info files*

Description

Read the ‘–info-file‘ output generated by cutadapt for adapter trimming. Column structure differs depending on whether the input came from Nanopore (ONT) or Illumina sequencing.

Usage

```
read_info_file(file, delim = "\t", col_select = NULL, ...)
```

Arguments

`file`	Path to the cutadapt info file (TSV).
`delim`	Field delimiter (default = "\t").
`col_select`	Optional tidyselect specification of which columns to read.
`...`	Additional arguments passed to [vroom::vroom()].

Details

- ‘read_info_file()‘ (Illumina) returns columns: ‘Seq.id‘, ‘n_errors‘, ‘start_adap‘, ‘end_adap‘, ‘seq_before_adap‘, ‘matching_seq‘, ‘seq_after_adap‘, ‘adap_name‘, ‘QScores_seq_before‘, ‘QScores_matching‘, ‘QScores_after‘.

Value

A tibble with the parsed ’cutadapt’ information. Column names are standardized.

Examples

```
test_info <- system.file("extdata", "cutadapt_info_illumina.txt", package="eDNAfuns")
df_illumina <- read_info_file(test_info)
```

tally_wide *Create contingency tables with two variables*

Description

This function takes a tibble and create human readable contingency tables from two variables, either by showing number of cases in each combination or weighted by the sum of a numerical variable

Usage

```
tally_wide(tibble, rows, cols, wt = NULL, ...)
```

Arguments

tibble	A tibble containing at least two columns
rows	The column with the levels included as rows in the final table.
cols	The column with the levels included as columns in the final table.
wt	The column (numeric) whose values to add in order to fill the cells. If wt = NULL (the default), counts are returned instead of weighted sums.
...	Any parameters that can be passed to tally_wide 'values_fill' is a useful one

Value

A tibble

Examples

```
 df <- tibble::tibble(
  group = c("A", "A", "B", "B", "B"),
    outcome = c("yes", "no", "yes", "yes", "no")
 )
tally_wide(df, rows = group, cols = outcome)
```

template_PCR *template_PCR*

Description

Placeholder description for Normal PCR template web address.

Usage

```
template_PCR
```

Format

A character vector

Source

Generated by the Author, stored in googledrive

`test_seqs` *test_seqs*

Description

Placeholder description for test_seqs dataset.

Usage

```
test_seqs
```

Format

An object of class `DNAbin` of length 3.

Source

Generated by the Author

`tibble_to_comm` *Convert a long tibble to a community matrix*

Description

Converts a long-format table of sequence counts into a wide community matrix (samples × taxa) suitable for vegan or other community ecology tools.

Produces a distance matrix between samples using vegan's `vegdist()`. Optionally applies a data transformation before distance calculation.

Returns the environmental data frame (sample metadata) from a long sequence/OTU tibble by removing taxon and abundance columns.

Usage

```
tibble_to_comm(long.table, taxon, Abundance, sample.name)

tibble_to_dist(
  long.table,
  taxon,
  Abundance,
  sample.name,
  distance = "bray",
  transformation = NULL,
  ...
)

tibble_to_env(long.table, taxon, Abundance, sample.name, ...)
```

Arguments

long.table	A tibble with sample, taxon, abundance, and metadata columns.
taxon	Column containing taxa/OTU IDs (unquoted).
Abundance	Column with counts/abundance values (unquoted).
sample.name	Column with sample IDs (unquoted).
distance	Distance metric to use (default = "bray").
transformation	Optional transformation (e.g. "hellinger", "log"). See vegan::decostand documentation for a great explanation of all transformations
...	Additional metadata columns to retain.

Value

A numeric matrix with taxa as columns and samples as row names.

A dist object.

A tibble of unique sample-level metadata.

Examples

```
data("ASV_table")
 tibble_to_comm(ASV_table,
                taxon = Hash,
                 Abundance = nReads,
                  sample.name = sample_name)
data("ASV_table")
tibble_to_dist(ASV_table,
               taxon = Hash,
               Abundance = nReads,
               sample.name = sample_name,
               distance = "bray",
               transformation = "hellinger")
```

```
data("ASV_table")
data("metadata")
dplyr::inner_join(ASV_table,metadata) |>
tibble_to_env(taxon = Hash,
              Abundance = nReads,
              sample.name = sample_name)
```

`tidy2phyloseq` *Convert a tidy ASV table to a phyloseq object*

Description

This function converts a tidy ASV table, along with optional taxonomy and metadata, into a ‘phyloseq‘ object.

Extracts ASV counts, taxonomy, and metadata from a ‘phyloseq‘ object into tidy data frames.

Usage

```
tidy2phyloseq(
  ASV_table,
  OTU_taxonomy = NULL,
  metadata = NULL,
  Taxa = "sseqid",
  Sample = "sample_name",
  Reads = "nr",
  tree = NULL
)

phyloseq2tidy(phylo_obj, Taxa = "sseqid", Sample = "sample_name", Reads = "nr")
```

Arguments

`ASV_table`	A tidy data.frame/tibble of ASV counts.
`OTU_taxonomy`	A data.frame with OTU taxonomy. Optional.
`metadata`	A data.frame with sample metadata. Optional.
`Taxa`	Column name for OTU IDs in the output (default: "sseqid").
`Sample`	Column name for sample IDs in the output (default: "sample_name").
`Reads`	Column name for read counts in the output (default: "nr").
`tree`	A phylogenetic tree of class ‘phylo‘. Optional.
`phylo_obj`	A phyloseq object.

Value

A ‘phyloseq‘ object combining OTU table, taxonomy, metadata, and optionally a tree.

A list with three tibbles:

ASV_table Long-format tibble of counts: Sample, Taxa, Reads.

taxonomy Taxonomy table as tibble (NULL if none in phyloseq).

metadata Sample metadata as tibble (NULL if none in phyloseq).

Examples

```
data("ASV_table")
data("metadata")
data("OTU_taxonomy")
ps <- tidy2phyloseq(ASV_table = ASV_table,
                    OTU_taxonomy = OTU_taxonomy,
                    metadata = metadata,
                    Taxa = "Hash",
                    Reads = "nReads")


data("ps")
tidy_list <- phyloseq2tidy(ps)
```

`training.ASV.table` *training.ASV.table*

Description

Placeholder description for training.ASV.table dataset.

Usage

```
training.ASV.table
```

Format

A data frame with 1909 rows and 4 variables:

Sample_name Character vector, last digit includes PCR replicate

Locus Which locus the sequences are from, numeric

Hash The sequence found, hashed using sha1 for consistency

nReads Number of times that hash was found in that sample

Source

Generated by the Author

`training.metadata` *training.metadata*

Description

Placeholder description for training.metadata dataset.

Usage

```
training.metadata
```

Format

A data frame with 95 rows and 8 columns

Sample_name Character vector, last digit includes PCR replicate

eDNA.sample Biological sample

rep PCR replicate

Transect Transect of origin

position Stop in Transect

depth in the water column

lat latitude in decimal degrees

lon longitude in decimal degrees

Source

Generated by the Author

`tree` *tree*

Description

Placeholder description for tree dataset.

Usage

```
tree
```

Format

An object of class phylo or similar

Source

Generated by the Author

UDI_Indices *UDI_Indices*

Description

Placeholder description for UDI_Indices dataset.

Usage

```
UDI_Indices
```

Format

A data frame with 384 rows and 6 columns

Index Character vector

Bases_in_Adapter_i7 Seqs in i7

Bases_for_Sample_Sheet_i7 Seqs to include

Bases_for_Sample_Sheet_i5 Seqs to include in some illumina platoforms

Bases_for_Sample_Sheet_i5_B Seqs to include in other illumina platoforms

Set A, B, C, or D

Source

Generated by the Author, data from Illumina

write_indexing_PCR *Write Indexing PCR Spreadsheet*

Description

Creates a new Google Sheet for indexing PCRs from a template, fills in the sample information, and writes it into the correct ranges of the sheet.

Usage

```
write_indexing_PCR(
  data,
  name,
  ss_template = "1naS-F_dj4SNmND5nJ5TKhMX3TikmRS00ILKjfg_Ucgc"
)
```

Arguments

data	A tibble or dataframe with at least the columns: **Well** Well position **Sample** Sample identifier **Column** Plate column (used to split data across sheet sections)
name	Character. Name for the new Google Sheet that will be created.
ss_template	Character. ID of the template sheet to copy from (default: '"1naS-F_dj4SNmND5nJ5TKhMX3TikmRS00

Details

This function: 1. Creates a new Google Sheet with the given 'name'. 2. Copies the template sheet into it. 3. Splits the input 'data' by plate column. 4. Writes each split dataframe into its designated range of the sheet.

Value

A Google Sheet object (as returned by [googlesheets4::gs4_create()]) with the data written into the correct plate layout.

Examples

```
#Examples are not executed because the function requires identification in Google
## Not run:
my_data <- tibble::tibble(
 Well = c("A1","A2"),
  Sample = c("Sample1","Sample2"),
  Column = c(1,2)
)
write_indexing_PCR(my_data, "PCR_001")

## End(Not run)
```

Index

∗ **datasets**
  AMPURE, 2
  ASV_table, 3
  example_hashes, 6
  Index_PCR, 8
  metadata, 8
  molarity.data, 9
  OTU_taxonomy, 11
  ps, 13
  template_PCR, 16
  test_seqs, 17
  training.ASV.table, 20
  training.metadata, 21
  tree, 21
  UDI_Indices, 22

AMPURE, 2
ASV_table, 3

count_stop_codons, 3

eDNAindex, 5
example_hashes, 6

fasta_reader, 6
fasta_writer, 7
fastq_reader *(*fasta_reader*)*, 6
fastq_writer *(*fasta_writer*)*, 7

Index_PCR, 8

metadata, 8
molarity.data, 9
mutation, 10

ng2nM, 10
nM2ng *(*ng2nM*)*, 10

OTU_taxonomy, 11

phyloseq2tidy *(*tidy2phyloseq*)*, 19
plot_seq_len_hist, 12
ps, 13

read_indexing_PCR, 13
read_info_file, 15
read_step1_PCR *(*read_indexing_PCR*)*, 13

tally_wide, 16
template_PCR, 16
test_seqs, 17
tibble_to_comm, 17
tibble_to_dist *(*tibble_to_comm*)*, 17
tibble_to_env *(*tibble_to_comm*)*, 17
tidy2phyloseq, 19
training.ASV.table, 20
training.metadata, 21
tree, 21

UDI_Indices, 22

write_indexing_PCR, 22