Blacktie Documentation

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Augustine Dunn

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PROJECT SUMMARY

1.1 I want to collaborate with you!

Contact me at wadunn83@gmail.com if you are a Python coder and want to or already have made improvements on this code.

1.2 Introducing Blacktie: a simpler way to do RNA-seq using Tophat, Cufflinks, and CummeRbund

Leveraging multiple fastQ files full of RNA-seq reads into a coherent picture of gene expression and transcript models is a multi-step process. It requires the organization and coordination of many files of different types through many different program calls and output steps. Each step might take hours or days depending on your input data. Then, as you are writing up your work, sometimes weeks/months later, you see that a new version of the programs you use has come out. Do you need to re-run your analysis? What settings DID you use back then?

The Tophat/Cufflinks/CummeRbund group of programs makes quality RNA-seq analysis doable once you understand the process. But what about when its time for you to leave the lab and you need to "train" someone else to repeat your process? It can be a nightmare. Especially if the trainee is not yet comfortable with the command line.

This is why I wrote the Blacktie pipeline software. Its goals are to streamline and simplify the complex task of analyzing full RNA-seq experiments using these programs; to automatically record settings used and program output messages in a way that users can track them to data later; provide a base set of functions and classes that will allow users to create custom pipelines easily by editing a single file (or if they want: writing their own custom scripts).

1.2.1 Some of Blacktie's features include:

- simple installation
- simple command line interface that allows almost ANYBODY to fully automate and reliably repeat their analysis of RNA-seq data with Tophat/Cufflinks/CummeRbund
- · send email updates to the user
- intelligently continue with the analysis if a single run fails
- run multiple, complex tophat/cufflinks experiments at once using a single command
- generates SGE qsub-able scripts for use with a computing cluster
- checks for R installation
- checks for cummeRbund library and walks user through installation if its not installed yet

• automatic preliminary CummeRbund Quality Control, Basic Differential Expression, and Basic Pattern Discovery plots using CummeRbund

Dedicated bioinformatics personnel can be few and far between. Blacktie aims to bring automated, reproducible RNAseq with built-in record keeping to more labs so that your valuable data does not fester on your servers, and you can publish sooner.

1.3 Getting the code

The code is available from the Python Package Index or from its homepage: https://github.com/xguse/blacktie

Visit Installation for more detailed instructions on getting and building the package.

1.4 Issue tracking

If you find issues, bugs, or have feature requests, please go here to submit them: https://github.com/xguse/blacktie/issues

1.5 Blacktie Poster

To credit the use of blacktie please cite the poster using the DOI link provided.

Introducing Blacktie: a simpler way to do RNA-seq using Tophat/Cufflinks/CummeRbund. Augustine Dunn. figshare. http://dx.doi.org/10.6084/m9.figshare.714149

CHAPTER

TWO

INSTALLATION

2.1 Requirements

The following python modules must be installed for blacktie to function properly:

Mako>=0.7.3 PyYAML>=3.10

The following modules will provide useful but optional functionality:

```
pprocess>=0.5
rpy2
```

2.2 Installing the latest version from the git repository

Note: Git is a very useful tool to have installed and to know how to use. Learn more here and try it out here.

Clone the repo: \$ git clone git://github.com/xguse/blacktie.git Install with any unmet requirements using pip: \$ [sudo] pip install -r blacktie/requirements.txt blacktie Install using standard setup.py script:

\$ cd blacktie
\$ [sudo] python setup.py install

2.3 Use pip to obtain the package from PyPI

\$ [sudo] pip install blacktie Mako PyYAML pprocess

2.4 Installing without using git or pip for the download

After installing the requirements:

```
$ wget https://github.com/xguse/blacktie/archive/master.zip
$ unzip master.zip
$ cd blacktie-master
$ [sudo] python setup.py install
```

2.5 Test to see whether the install worked

To test whether your installation was successful, open a new terminal session and type the following command.

\$ blacktie

You should see the help text for blacktie and it should look something like this:

```
usage: blacktie [-h] [--version]
                [--prog {tophat,cufflinks,cuffmerge,cuffdiff,cummerbund,all}]
                [--hide-logs] [--no-email]
                [--mode {analyze, dry_run, qsub_script}]
                config_file
This script reads options from a yaml formatted file and organizes the
execution of tophat/cufflinks runs for multiple condition sets.
positional arguments:
config_file Path to a yaml formatted config file containing setup
                       options for the runs.
optional arguments:
-h, --help
                    show this help message and exit
--version
                     Print version number.
--prog {tophat, cufflinks, cuffmerge, cuffdiff, cummerbund, all}
                        Which program do you want to run? (default: tophat)
                     Make your log directories hidden to keep a tidy
--hide-logs
                        'looking' base directory. (default: False)
--no-email
                     Don't send email notifications. (default: False)
--mode {analyze, dry_run, qsub_script}
                        1) 'analyze': run the analysis pipeline. 2) 'dry_run':
                        walk through all steps that would be run and print out
                        the command lines; however, do not send the commands
                        to the system to be run. 3) 'qsub_script': generate
                        bash scripts suitable to be sent to a compute
                        cluster's SGE through the qsub command. (default:
                        analyze)
```

If this worked, great!

GETTING STARTED

Note: Make sure that you have successfully installed the blacktie module before trying the activities below.

To test whether your installation was successful, open a new terminal session and type the following command.

\$ blacktie

You should see the help text for blacktie and it should look something like this:

```
$ blacktie
usage: blacktie [-h] [--version]
    [--prog {tophat,cufflinks,cuffmerge,cuffdiff,all}]
    [--hide-logs] [--no-email]
    [--mode {analyze, dry_run, qsub_script}]
    config_file
This script reads options from a yaml formatted file and organizes the
execution of tophat/cufflinks runs for multiple condition sets.
positional arguments:
    config_file
                          Path to a yaml formatted config file containing setup
        options for the runs.
optional arguments:
    -h, --help
                          show this help message and exit
    --version
                          Print version number.
    --prog {tophat,cufflinks,cuffmerge,cuffdiff,all}
       Which program do you want to run? (default: tophat)
    --hide-logs
                          Make your log directories hidden to keep a tidy
       'looking' base directory. (default: False)
                          Don't send email notifications. (default: False)
    --no-email
    --mode {analyze, dry_run, qsub_script}
        1) 'analyze': run the analysis pipeline. 2) 'dry_run':
        walk through all steps that would be run and print out
        the command lines; however, do not send the commands
        to the system to be run. 3) 'qsub_script': generate
        bash scripts suitable to be sent to a compute
        cluster's SGE through the qsub command. (default:
        analyze)
```

If this worked, great! Let's move on to what all that means.

3.1 The –prog option

This tells blacktie which part of the pipeline you would like to run. Any part can be run individually as long as the correct files exist. You can also run the whole thing from tophat to cuffdiff in one fell swoop if you like!

3.2 The –hide-logs option

This names your log files so that they are hidden in "*nix" systems.

3.3 The –modes option

blacktie can run in three modes. The first, analyze, actually runs the pipeline and does the analyses. However, it can be useful to simply view what WOULD be done to make sure that `blacktie is producing command line calls that match what you expected. For this, use the dry_run mode.

Further, if you are working on a compute cluster running something like a "Sun Grid Engine" (SGE) to which you must submit jobs using qsub, it may not be a good idea to submit a job running all of blacktie as a single qsub job. For this it can be helpful to have blacktie write all of your qsub scripts for you based on a template. Each bash script represents a single program call to the tophat/cufflinks suite.

Note: A starter template for SGE submission can be found here: blacktie/examples/qsub.template. You will want to become familiar with how Mako processes templates if you plan to customize this much.

Here is what the starter template looks like:

```
#!/bin/bash
   #$ -S /bin/bash
                                                                  # Use a real BASH shell on the worker not
2
   #$ -q ${queues}
                                                                  # What queues do you want to submit to
3
   #$ -M ${email_addy}
                                                                  # Send email updates to this address
4
   #$ -m beas
                                                                  # When to send an email update
5
   #$ -e /data/users/dunnw/logs/${call_id}.e
                                                                  # Write standard error to this file
6
   #$ -o /data/users/dunnw/logs/${call_id}.o
                                                                  # Write standard out to this file
   #$ -N ${job_name}
                                                                  # Name my job this
8
   #$ -R y
                                                                  # Reserve cores for me until there are t.
9
   #$ -pe openmp ${core_range}
                                                                  # Use openmp for multiprocessor use and
10
11
   LD_LIBRARY_PATH="${ld_library_path}$${}{LD_LIBRARY_PATH}"
                                                                 # Make sure worker's LD_LIBRARY_PATH con
12
13
14
   # HPC clusters frequently use a module system to provide system wide access to
15
   # certain programs. The following makes sure that the tools needed are loaded
16
   # for **MY** cluster. You will need alter this to make sure your cluster is set up
17
   # based on its system.
18
19
20
  module load bowtie2/2.0.2
21
  module load tophat/2.0.6
  module load cufflinks/2.0.2
22
   module load samtools/0.1.18
23
24
25
26
   # basic staging stuff
   DATAHOME="${datahome}"
27
```

```
MYSCRATCH="/scratch/$${}{USER}"
28
29
30
   mkdir -p $MYSCRATCH
31
   cd $MYSCRATCH
32
33
34
   # Remind me what will be done
35
   echo ''
36
   echo "${cmd_str}"
37
   echo ''
38
39
   # Run my job
40
   ${cmd_str}
41
42
43
   # Pack up results and send it home to log-in node
44
   tar -zcvf ${call_id}.tar.gz ${out_dir}
45
   cp ${call_id}.tar.gz $${}{DATAHOME}/
46
47
   # Back into the shadows
48
   cd $HOME
40
   rm -rf $MYSCRATCH
```

3.4 The configuration file

The configuration file is a YAML-based document that is where we will store all of the complexity of the options, input and output files of the typical tophat/cufflinks workflow. This way we have though about what we want to do with our RNA-seq data from start to finish before we actually start the analysis. Also, this config file acts as a check on our poor memory. If you get strange results you don't have to worry about whether you entered the samples backwards since you can go back to this config file and see exactly what files and settings were used.

Note: If you are running blacktie in analyze mode, you will have many more files created that document every step of the process where the output files are actually placed as well as central log files.

Here is a dummy example of a config file:

Note: A copy of this file can be found here: blacktie/examples/blacktie_config_example.yaml

```
# The document starts after the '---'
2
   # By the way: everything after a '#' on a line
3
   # will be ignored by the program and acts as a
4
   # comment or note to explain things.
5
6
7
   # run_options is a dictionary that contains variables that will be needed for
8
   # many or all stages of the run
9
   run_options:
10
       base_dir: /path/to/project/base_dir
11
       run_id: False
                                # name your run: if false; uses current date/time for uniqe run_id eve.
12
       bowtie_indexes_dir: /path/to/bowtie2_indexes
13
       email_info:
14
```

```
sender: from_me@gmail.com
15
           to: to_you@email.com
16
           li: /path/to/file/containing/base64_encoded/login_info
                                                                         # base64_encoded pswrd for from_
17
       custom smtp:
18
           host: smtp.gmail.com # or what ever your email smtp server is
19
                                   # or which ever port your smtp server uses
           port: 587
20
21
22
23
   # 'tophat_options':
24
25
   # _____
   # This is a dictionary that contains variables needed for all the tophat runs.
26
   # The names of the key:value combinations are taken directly from the tophat
27
   # option names but have the leading '-' removed.
28
29
   # -o becomes o; --library-type becomes library-type
30
31
32
   # **This is true for the cufflinks, cuffmerge, cuffdiff option dictionaries.**
33
   # 'from_conditions':
34
   # _____
35
   # This is a special value that tells blacktie that you don't want to name a single
36
   # value for this option but would rather set the value individually for each of
37
   # your samples/conditions. If you set the 'o' value here:
38
39
        **all of your different sample results would
   #
40
         be written to the same output directory and
   #
41
          each would overwrite the next!**
   #
42
   # Hence: from_conditions
43
44
45
   # However if you made all of your libraries the same way, things like 'r' and
   # 'mate-std-dev' can be set here to avoid writing the same values over and over
46
   # and perhaps making a mistake or two.
47
48
   # 'positional_args':
40
   # _____
50
   # This is a dictionary inside of the 'tophat_options' dictionary.
51
  # It is where you put the arguments to tophat that do not have 'flags' to make
52
   # their identity explicit like '-o path/to/output_dir' or '--library-type fr-unstranded'
53
54
   # For tophat, these values are
55
         [1] the bowtie index name
   #
56
   #
         [2] the fastq files containing the left_reads
57
58
   #
         [3] the fastq files containing the right_reads
59
   # They will be different for cufflinks, cuffmerge, cuffdiff so consult the
60
   # respective help text or manuals, but you should be fine if you just use what
61
   # I have set up in this file already.
62
63
   tophat_options:
64
65
       o: from_conditions
       library-type: fr-unstranded
66
       p: 6
67
       r: 125
68
       mate-std-dev: 25
69
       G: from_conditions
70
71
       no-coverage-search: True
72
       positional_args:
```

```
bowtie2_index: from_conditions
73
            left_reads: from_conditions
74
            right_reads: from_conditions
75
76
77
    cufflinks_options:
        o: from_conditions
78
        p: 7
79
        GTF-guide: from_conditions # If you want to use annotation as *TRUTH* set this to False and set
80
        GTF: False
                                      # if an option set to false, it will be ommited from the command str.
81
        3-overhang-tolerance: 5000
82
        frag-bias-correct: from_conditions
83
        multi-read-correct: True
84
        upper-quartile-norm: True
85
        positional_args:
86
            accepted_hits: from_conditions
87
88
    cuffmerge_options:
89
        o: from_conditions # output directory
90
        ref-gtf: from_conditions
91
        p: 6
92
        ref-sequence: from_conditions
93
94
        positional_args:
            assembly_list: from_conditions # file with path to cufflinks gtf files to be merged
95
96
   cuffdiff_options:
97
        o: from_conditions
98
        labels: from_conditions
99
        p: 6
100
        time-series: True
101
102
        upper-quartile-norm: True
103
        frag-bias-correct: from_conditions
        multi-read-correct: True
104
        positional_args:
105
            transcripts_gtf: from_conditions
106
            sample_bams: from_conditions
107
108
109
    cummerbund_options:
110
        cuffdiff-dir: from_conditions
111
        gtf-path: from_conditions
112
        out: from_conditions
113
        file-type: pdf
114
115
116
117
    # options for --mode qsub_script
   # If you are not using --mode gsub script, then set all to 'None'
118
   qsub_options:
119
      queues: 'queue1, queue3, queue5'
120
      datahome: '/path/to/baseDirectory/on/cluster/'
121
      core_range: 40-64 # how many cpus do you want
122
      ld_library_path: '' # leave this blank unless you know what it is and need it
123
      template: /path/to/your/altered/version/of/qsub.template
124
125
126
   # 'condition_queue':
127
128
   # _____
129
   # This is a list of info related to each sample/condition contained in your RNA-sequence
130
   # experiment(s)
```

```
131
      'name': the name of this condition program. Usually something like a time-point
132
    #
              ID or treatment type. Should be as short as possible while still being a useful label.
133
    #
134
      'experiment_id': this is how you group different experiments to be included in a
    #
135
                   single cuffmerge/cuffdiff program call. All conditions in a time
    #
136
                   series should share the same 'experiment_id' and be placed in
    #
137
                   'condition_queue' in the order that you want them to be sent to
138
    #
                   cuffdiff.
139
    #
140
141
    #
      'replicate_id': this is how you group data for biological replicates of a single
                   experimental condition experiments to be included in a cuffdiff program
142
    #
                   call. Each replicate of a condition should have a unique 'experiment_id'.
143
    #
144
      'left_reads': a list of the paths to fastq files containing left reads for
145
    #
                     each condition.
146
    #
147
      'right_reads': list of fastqs containing the right mates for the fastqs in
    #
148
    #
                      'left reads'.
149
    #
                       **NOTE** right mate file must be in same order as provided to 'left_reads'
150
151
152
   condition_queue:
153
154
            name: expl_control
            experiment_id: 0
155
            replicate id: 0
156
            left_reads:
157
                 - /path/to/exp1_control/techRep1.left_reads.fastq
158
                 - /path/to/exp1_control/techRep2.left_reads.fastq
159
            right_reads:
160
                 - /path/to/expl_control/techRep1.right_reads.fastq
161
                 - /path/to/expl control/techRep2.right reads.fastg
162
            genome_seq: /path/to/species/genome.fa
163
            gtf_annotation: /path/to/species/annotation.gtf
164
            bowtie2_index: species.bowtie2_index.basename
165
166
167
            name: expl_control
168
            experiment_id: 0
169
            replicate_id: 1
170
            left_reads:
171
                 - /path/to/exp1_control/techRep1.left_reads.fastq
172
                 - /path/to/exp1_control/techRep2.left_reads.fastq
173
            right_reads:
174
                 - /path/to/expl_control/techRep1.right_reads.fastq
175
                 - /path/to/exp1_control/techRep2.right_reads.fastq
176
            genome_seq: /path/to/species/genome.fa
177
178
            gtf_annotation: /path/to/species/annotation.gtf
            bowtie2_index: species.bowtie2_index.basename
179
180
181
            name: expl_treatment
182
            experiment_id: 0
183
            replicate_id: 0
184
            left_reads:
185
                 - /path/to/exp1_treatment/techRep1.left_reads.fastq
186
                 - /path/to/expl_treatment/techRep2.left_reads.fastq
187
            right_reads:
188
```

```
- /path/to/expl_treatment/techRep1.right_reads.fastq
189
                 - /path/to/exp1_treatment/techRep2.right_reads.fastq
190
            genome_seq: /path/to/species/genome.fa
19
            gtf_annotation: /path/to/species/annotation.gtf
192
            bowtie2_index: species.bowtie2_index.basename
193
194
195
            name: exp2_control
196
            experiment_id: 1
197
            replicate_id: 0
198
            left_reads:
199
                 - /path/to/exp2_control/techRep1.left_reads.fastq
200
                 - /path/to/exp2_control/techRep2.left_reads.fastq
201
            right_reads:
202
                 - /path/to/exp2_control/techRep1.right_reads.fastq
203
                 - /path/to/exp2_control/techRep2.right_reads.fastq
204
            genome_seq: /path/to/species/genome.fa
205
            gtf_annotation: /path/to/species/annotation.gtf
206
            bowtie2_index: species.bowtie2_index.basename
207
208
209
            name: exp2_treatment
210
            experiment_id: 1
211
            replicate_id: 0
212
            left_reads:
213
                 - /path/to/exp2_treatment/techRep1.left_reads.fastq
214
                 - /path/to/exp2_treatment/techRep2.left_reads.fastq
215
            right_reads:
216
                 - /path/to/exp2_treatment/techRep1.right_reads.fastq
217
                 - /path/to/exp2_treatment/techRep2.right_reads.fastq
218
            genome_seq: /path/to/species/genome.fa
219
            gtf_annotation: /path/to/species/annotation.gtf
220
            bowtie2_index: species.bowtie2_index.basename
221
222
223
224
    . . .
```

Todo

Add the slots for custom email server options.

3.5 Using e-mail notifications

Changed in version v0.2.0rc1: any smtp server should now be usable if you code the host and port into the yaml config file. Any email can be used as the recipient. New in version v0.2.0rc1: added --no-email option.

Warning: gmail's 2-step authentication will NOT work. Sorry. I will look into how to deal with that eventually.

You will need to provide your password in order to use the email notifications but it is not a good idea to store human readable passwords lying around your system. So the file that is used to store your password must contain a version of your password that has been encoded in base64. This will scramble your password beyond most people's ability to read it as a password as long as you don't name it something silly like password_file.txt.

The help text for blacktie-encode is:

\$ blacktie-encode -h

usage: blacktie-encode [-h] input_file

This script takes a path to a file where you have placed your password for the email you want blacktie to use as the "sender" in its notification emails. It will replace the file with one containing your password once it has encoded it out of human readable plain-text into seemingly meaningless text. **THIS IS NOT FOOLPROOF:** If someone knows exactly what to look for they might figure it out. ALWAYS use good password practices and never use the same password for multiple important accounts!

positional arguments: input_file Path to a file where you have placed your password for the email you want blacktie to use as the "sender" in its notification emails.

optional arguments: -h, --help show this help message and exit

CHAPTER

FOUR

TUTORIAL

A more detailed tutorial is under development, so watch this space!

FIVE

BLACKTIE AUTO-GENERATED CODE DOCUMENTATION

Todo

DONE Convert docstring style from (given,does,returns) to (:param a: format)

5.1 calls.py

Code defining classes to represent and excute pipeline program calls.

Defines common methods for all program call types.

__init__ (yargs, email_info, run_id, run_logs, conditions, mode='analyze')
initializes a BaseCall object

Parameters

- yargs argument tree generated by parsing the yaml config file
- email_info Bunch() object containing keys: email_from, email_to, email_li
- **run_id** id for the whole set of calls
- run_logs the directory where log file should be put
- conditions one or a list of condition-dictionaries from yargs.condition_queue
- **mode** choices = ['analyze','dry_run','qsub_script']

Returns an initialized BaseCall object

```
_flag_out_dir()
```

renames out directory, prepending 'FAILED' flag: equivalent of mv tophat_Aa0 FAILED.tophat_Aa0

```
build_out_dir_path()
```

builds correct out_dir path based on state of self

Returns out_dir

build_qsub()

Builds and writes this CallObject's qsub script to current working directory using options provided under the "qsub_options" sub-tree in the yaml config file.

construct_options_list()

converts opt_dict into list encoding proper options to send to the current program: saves to self.

execute()

calls correct program, records results, and manages errors

get_condition_id(condition_dict)

Constructs condition ID :param condition_dict: a dictionary containing consition info like name, replicate_id, etc. :returns: an ID used to construct the call_id of a call.

init_log_file()

creates empty log file for this call and stores its path in self.log_file

init_opt_dict()

builds a dict with non-job-specific values set and job-specific values set to False based on option names in the yaml file for this phase

Returns partially populated opt_dict

log_end()

records command string used, program output, and the end of call in self.log_file

 $\log_m g(log_m g=')$

•opens self.log_file

•writes log_msg

•closes self.log_file

log_start()

records start of call in self.log_file

```
notify_end_of_call()
```

sends notification email informing user that self.call_id has exited

notify_start_of_call()

sends notification email informing user that self.call_id has been initiated

purge_progress_bars(stderr_str)

removes the dynamic progress bars included in some output in case user did not turn them off

set_call_id()

builds and stores this call's call ID in self.call_id

```
class blacktie.utils.calls.CuffdiffCall(yargs, email_info, run_id, run_logs, conditions,
```

mode) Manage a single call to cuffdiff and store associated run data.

__init__ (yargs, email_info, run_id, run_logs, conditions, mode)
initializes the CuffdiffCall object

Parameters

- yargs argument tree generated by parsing the yaml config file
- email_info Bunch() object containing keys: email_from, email_to, email_li
- run_id id for the whole set of calls
- run_logs the directory where log file should be put

- conditions one or a list of condition-dictionaries from yargs.condition_queue
- **mode** choices = ['analyze','dry_run','qsub_script']

Returns an initialized CuffdiffCall object

get_bam_path(condition)

Supports self.get_sample_bams().

get_cuffmerge_gtf()

Handles yaml_config.cuffdiff_options.positional_args.transcripts_gtf: from_conditions.

get_genome()

Handles yaml_config.cuffdiff_options.frag-bias-correct: from_conditions.

get_labels()

Handles yaml_config.cuffdiff_options.labels: from_conditions.

get_mask_file()

Handles yaml_config.cuffdiff_options.mask-file: from_conditions.

get out dir()

Handles yaml_config.cuffdiff_options.o: from_conditions.

get_sample_bams()

Handles yaml_config.cuffdiff_options.positional_args.sample_bams: from conditions.

mode)

class blacktie.utils.calls.CufflinksCall (yargs, email_info, run_id, run_logs, conditions,

Manage a single call to cufflinks and store associated run data.

__init__ (yargs, email_info, run_id, run_logs, conditions, mode)
initializes the CufflinksCall object

Parameters

- yargs argument tree generated by parsing the yaml config file
- email_info Bunch() object containing keys: email_from, email_to, email_li
- run_id id for the whole set of calls
- run_logs the directory where log file should be put
- conditions one or a list of condition-dictionaries from yargs.condition_queue
- **mode** choices = ['analyze','dry_run','qsub_script']

Returns an initialized CufflinksCall object

Todo

DONE add support for -GTF in addition to currently supported -GTF-guide

get_accepted_hits()

```
Handles yaml_config.cufflinks_options.positional_args.accepted_hits: from_conditions.
```

get_bam_path()

Supports self.get_accepted_hits().

get_genome() Handles yaml_config.cufflinks_options.frag-bias-correct: from conditions. get_gtf_anno() Handles yaml_config.cufflinks_options.GTF: from_conditions. get gtf anno guide() Handles yaml_config.cufflinks_options.GTF-guide: from_conditions. get_mask_file() Handles yaml_config.cufflinks_options.mask-file: from_conditions. get_out_dir() Handles yaml_config.cufflinks_options.o: from_conditions. verify_options() Makes sure that conflicting options were not imported from yaml config file. Todo DONE GTF and GTF-guide should not be used together but both can be ommited

Manage a single call to cuffmerge and store associated run data.

__init__ (yargs, email_info, run_id, run_logs, conditions, mode)
initializes the CuffmergeCall object

Parameters

- yargs argument tree generated by parsing the yaml config file
- email_info Bunch() object containing keys: email_from, email_to, email_li
- run_id id for the whole set of calls
- run_logs the directory where log file should be put
- conditions one or a list of condition-dictionaries from yargs.condition_queue
- **mode** choices = ['analyze','dry_run','qsub_script']

Returns an initialized CuffmergeCall object

get_cuffGTF_path(condition)

Supports self.get_cufflinks_gtfs().

get_cufflinks_gtfs()

Handles yaml_config.cuffmerge_options.positional_args.assembly_list: from_conditions.

get_genome()

Handles yaml_config.cuffmerge_options.ref-sequence: from_conditions.

$\texttt{get_gtf}_\texttt{anno}\left(\right)$

Handles yaml_config.cuffmerge_options.ref-gtf: from_conditions.

get_out_dir()

Handles yaml_config.cuffmerge_options.o: from_conditions.

```
class blacktie.utils.calls.CummerbundCall(yargs, email_info, run_id, run_logs, conditions,
```

```
mode)
```

Manage a single call to blacktie-cummerbund script and store associated run data.

__init__ (yargs, email_info, run_id, run_logs, conditions, mode)
initializes the CummerbundCall object

Parameters

- yargs argument tree generated by parsing the yaml config file
- email_info Bunch() object containing keys: email_from, email_to, email_li
- **run_id** id for the whole set of calls
- **run_logs** the directory where log file should be put
- $conditions one \ or \ a \ list \ of \ condition-dictionaries \ from \ yargs.condition_queue$
- **mode** choices = ['analyze','dry_run','qsub_script']

Returns an initialized CummerbundCall object

get_cuffdiff_dir()

Handles yaml_config.cummerbund_options.cuffdiff-dir: from_conditions.

get_cuffmerge_gtf()

Handles yaml_config.cummerbund_options.gtf-path: from_conditions.

get_out_dir()

Handles yaml_config.cummerbund_options.out: from_conditions.

class blacktie.utils.calls.TophatCall (yargs, email_info, run_id, run_logs, conditions, mode)
 Manage a single call to tophat and store associated run data.

__init___(yargs, email_info, run_id, run_logs, conditions, mode)
initializes the TophatCall object

Parameters

- yargs argument tree generated by parsing the yaml config file
- email_info Bunch() object containing keys: email_from, email_to, email_li
- **run_id** id for the whole set of calls
- **run_logs** the directory where log file should be put
- conditions one or a list of condition-dictionaries from <code>yargs.condition_queue</code>
- **mode** choices = ['analyze','dry_run','qsub_script']

Returns an initialized TophatCall object

get_bt_idx()

Handles yaml_config.tophat_options.positional_args.bowtie2_index: from_conditions.

get_gtf_anno()

Handles yaml_config.tophat_options.G: from_conditions.

get_lt_reads()

Handles yaml_config.tophat_options.positional_args.left_reads: from_conditions.

get_out_dir()

Handles yaml_config.tophat_options.o: from_conditions.

```
get_rt_reads()
    Handles yaml_config.tophat_options.positional_args.right_reads:
    from_conditions.
```

5.2 errors.py

Code defining custom base error classes to provide a foundation for graceful error handling.

```
exception blacktie.utils.errors.BlacktieError
Base class for exceptions in the blacktie package.
```

```
exception blacktie.utils.errors.InvalidFileFormatError
When errors occur due to malformed file formats.
```

exception blacktie.utils.errors.**MissingArgumentError** (*errMsg*) When a required argument is missing from the parsed command line options.

__init__(errMsg)

```
exception blacktie.utils.errors.SanityCheckError
When a 'state check' comes back as conflicting or nonsensical.
```

exception blacktie.utils.errors.**SystemCallError** (*errno*, *strerror*, *filename=None*) Error raised when a problem occurs while attempting to run an external system call.

Attributes:

errno - return code from system call

filename – file in volved if any

strerror - error msg

__init___(errno, strerror, filename=None)

```
exception blacktie.utils.errors.UnexpectedValueError
When values that "should" not be possible happen; like if a variable was changed unexpectedly.
```

5.3 externals.py

Code facilitating the execution of external system calls.

```
blacktie.utils.externals.mkdirp(path)
```

Create new dir while creating any parent dirs in the path as needed.

```
blacktie.utils.externals.runExternalApp (progName, argStr)
Convenience func to handle calling and monitoring output of external programs.
```

Parameters

- progName name of system program command
- argStr string containing command line options for progName

Returns subprocess.communicate object

```
blacktie.utils.externals.whereis(program)
```

returns path of program if it exists in your \$PATH variable or None otherwise

5.4 misc.py

Code facilitating random aspects of this package.

class blacktie.utils.misc.Bunch(*args, **kwds)

A dict like class to facilitate setting and access to tree-like data. Allows access to dictionary keys through 'dot' notation: "yourDict.key = value".

____init___(*args, **kwds)

```
blacktie.utils.misc.bunchify(dict_tree)
```

Traverses a dictionary tree and converts all sub-dictionaries to Bunch() objects.

blacktie.utils.misc.email_notification(sender, to, subject, txt, pw, server_info)

Sends email to recipient using GMAIL server by default but will now accept server_info to customize this.

Parameters

- sender email address of sender
- to email addres of recipient
- subject subject text
- **txt** body text
- pw-password of sender
- server_info dictionary = { 'host':str,'port':int }

Returns None

Todo

DONE make email_notification() adjustable for other email servers

```
blacktie.utils.misc.get_time()
    Return system time formatted as 'YYYY:MM:DD-hh:mm:ss'.
```

```
blacktie.utils.misc.get_version_number(path_to_setup)
Provides access to current version info contained in setup.py
```

```
blacktie.utils.misc.map_condition_groups (yargs)
    creates a Bunch obj groups with key='experiment_id' from yargs, value=list(condition_queue objects with
    'experiment_id')
```

Parameters yargs - argument object generated from the yaml config file

Returns groups

blacktie.utils.misc.whoami() Returns the name of the currently active function.

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