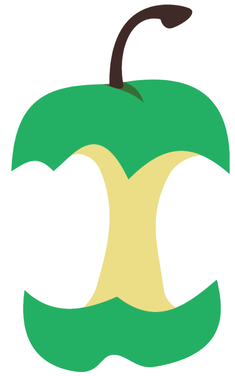
# nf-core/bytesize



#31

## Troubleshooting a failed pipeline

Phil Ewels, SciLifeLab Sweden

# Because at some point, things will go wrong..

1. Start small
2. Categorise the type of error
3. Read the log, check the work directory
4. Check Slack, Google, ask for help
5. Report a bug

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## Start small

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```
nextflow run nf-core/xxx -profile test,docker
```

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## Start small

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Check the basics

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- ✅ Nextflow up to date (`nextflow -self-update`)
- ✅ Haven't run out of disk space
- ✅ Docker demon is running *(if you're using Docker)*
- ✅ ...

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1. Start small
2. Check the basics
3. Check the troubleshooting docs: https://nf-co.re/usage/troubleshooting
4.
5.

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2 Categorise the type of error

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⚠️ *Warning:*

*Note that just because Nextflow reports a particular tool failed, this does not necessarily mean it's an issue with the tool itself.*

1. Categorise the type of error
2. Before the first process
3. First process
4. During run
5. Problem with outputs

1

# Categorise the type of error

2

## Before the first process

```
N E X T F L O W  ~  version 0.27.3
Launching `./main.nf` [prickly_snyder] - revision: bb0fa33a13
ERROR ~ Unknown config attribute: projectDir -- check config file:
nextflow.config

null

 -- Check '.nextflow.log' file for details
```

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❌ Nextflow not up to date

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```
* The pipeline
  https://doi.org/10.5281/zenodo.1400710

* The nf-core framework
  https://doi.org/10.1038/s41587-020-0439-x

* Software dependencies
  https://github.com/nf-core/rnaseq/blob/master/CITATIONS.md
-----------------------------------------------------
WARN: ================================================================================
  Both '--gtf' and '--gff' parameters have been provided.
  Using GTF file as priority.
================================================================================
WARN: ================================================================================
  When using '--additional_fasta <FASTA_FILE>' the aligner index will not
  be re-built with the transgenes incorporated by default since you have
  already provided an index via '--star_index <INDEX>'.

  Set '--additional_fasta <FASTA_FILE> --star_index false --save_reference' to
  re-build the index with transgenes included and the index will be saved in
  'results/genome/index/star/' for re-use with '--star_index'.

  Ignore this warning if you know that the index already contains transgenes.

  Please see:
  https://github.com/nf-core/rnaseq/issues/556
================================================================================
Module compilation error
- file : /Users/phil/GitHub/nf-core/rnaseq/./workflows/../modules/local/bedtools_genomecov.nf
- cause: end of line reached within a simple string 'x' or "x" or /x/;
   solution: for multi-line literals, use triple quotes '''x''' or """x""" or /x/ or $/x/$ @ line 6, column 108.
   ularity_pull_docker_container ?
                                 ^

1 error
```

❌ Nextflow not up to date

```
N E X T F L O W  ~  version 20.07.0
Launching `./main.nf` [furious_hamilton] - revision: bb0fa33a13
------------------------------------------------------

                                          ,--./,-.
          ___     __   __   __   ___     /,-._.--~'
    |\ | |__  __ /  ` /  \ |__) |__         }  {
    | \| |       \__, \__/ |  \ |___     \`-._,-`-,
                                          `._,._,'

    nf-core/rnaseq v3.6dev
------------------------------------------------------
Core Nextflow options
  runName                   : furious_hamilton
  containerEngine           : docker
  launchDir                 : /Users/phil/GitHub/nf-core/rnaseq
  workDir                   : /Users/phil/GitHub/nf-core/rnaseq/work
  projectDir                : /Users/phil/GitHub/nf-core/rnaseq
  userName                  : phil
  profile                   : test,docker
  configFiles               : /Users/phil/GitHub/nf-core/rnaseq/nextflow.config

Input/output options
  input                     : https://raw.githubusercontent.com/nf-core/test-datasets/rnaseq/samplesheet/v3.4/samplesheet_test.csv

UMI options
  umitools_bc_pattern       : NNNN

Read filtering options
  bbsplit_fasta_list        : https://github.com/nf-core/test-datasets/raw/rnaseq/reference/bbsplit_fasta_list.txt
  skip_bbsplit              : false

Reference genome options
  fasta                     : https://github.com/nf-core/test-datasets/raw/rnaseq/reference/genome.fa
  gtf                       : https://github.com/nf-core/test-datasets/raw/rnaseq/reference/genes.gtf.gz
  gff                       : https://github.com/nf-core/test-datasets/raw/rnaseq/reference/genes.gff.gz
  transcript_fasta          : https://github.com/nf-core/test-datasets/raw/rnaseq/reference/transcriptome.fasta
```

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# Categorise the type of error

## First process

Remember that you need to tell Nextflow how to handle software dependencies..

```
-profile test,docker
```

No spaces between profile names

# Categorise the type of error

1

## First process

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```
Command exit status:
  127

Command output:
  (empty)

Command error:
  .command.sh: line 3: rsem-prepare-reference: command not found

Work dir:
  /home/lfaller/nextflow/rnaseq/work/f7/b6ef5a3f12f5efbf641f19046aca74

Tip: you can try to figure out what's wrong by changing to the process work dir and showing the script
file named `.command.sh`


Unexpected error [AbortedException]

 -- Check script '/home/lfaller/.nextflow/assets/nf-core/rnaseq/./workflows/rnaseq.nf' at line: 603 or see
'.nextflow.log' file for more details
```

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# Categorise the type of error

1

## First process

2

```
Command exit status:
  127

Command output:
  (empty)

Command error:
  .command.sh: line 3: rsem-prepare-reference: command not found

Work dir:
  /home/lfaller/nextflow/rnaseq/work/f7/b6ef5a3f12f5efbf641f19046aca74

Tip: you can try to figure out what's wrong by changing to the process work dir and showing the script
file named `.command.sh`


Unexpected error [AbortedException]

 -- Check script '/home/lfaller/.nextflow/assets/nf-core/rnaseq/./workflows/rnaseq.nf' at line: 603 or see
'.nextflow.log' file for more details
```

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# Categorise the type of error

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## First process

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```
Error executing process > 'NFCORE_SSDS:SSDS:BEDTOOLS_MAKEWINDOWS (mm10rn6)'

Caused by:
  Failed to submit process to grid scheduler for execution

Command executed:

  sbatch .command.run

Command exit status:
  1

Command output:
  sbatch: error: Invalid --signal specification

Work dir:
  /gpfs/gsfs9/users/RDCOpipes/010101_Fulgent_1010_SE7599/010101_1010_newVersionSS_ssds/
ssds__20211215_155733_rn077168/work/84/3deb9b6cf726a27c167bb775028b09

Tip: view the complete command output by changing to the process work dir and entering the command
`cat .command.out`
```

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3 Read the log, check the work directory

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```
[6:16 PM] Error executing process > 'FASTQC (hct116_h3k4me1_IP_R1_T1)'

Caused by:
  Missing output file(s) `*.{zip,html}` expected by process `FASTQC (hct116_h3k4me1_IP_R1_T1)`

Command executed:

  [ ! -f  hct116_h3k4me1_IP_R1_T1.fastq.gz ] && ln -s hct116_h3k4me1_clean.fastq.gz
hct116_h3k4me1_IP_R1_T1.fastq.gz
  fastqc -q -t 6 hct116_h3k4me1_IP_R1_T1.fastq.gz

Command exit status:
  0

Command output:
  (empty)

Command error:
  WARNING: Your kernel does not support swap limit capabilities or the cgroup is not mounted.
Memory limited without swap.
  Failed to process file hct116_h3k4me1_IP_R1_T1.fastq.gz
  uk.ac.babraham.FastQC.Sequence.SequenceFormatException: Ran out of data in the middle of a
fastq entry.  Your file is probably truncated
              at uk.ac.babraham.FastQC.Sequence.FastQFile.readNext(FastQFile.java:179)
              at uk.ac.babraham.FastQC.Sequence.FastQFile.next(FastQFile.java:125)
              at uk.ac.babraham.FastQC.Analysis.AnalysisRunner.run(AnalysisRunner.java:77)
              at java.base/java.lang.Thread.run(Thread.java:834)
```

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# Where in the pipeline and what type of error

```
[6:16 PM] Error executing process > 'FASTQC (hct116_h3k4me1_IP_R1_T1)'

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Command executed:

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hct116_h3k4me1_IP_R1_T1.fastq.gz
  fastqc -q -t 6 hct116_h3k4me1_IP_R1_T1.fastq.gz

Command exit status:
  0

Command output:
  (empty)

Command error:
  WARNING: Your kernel does not support swap limit capabilities or the cgroup is not mounted.
Memory limited without swap.
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              at java.base/java.lang.Thread.run(Thread.java:834)
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Caused by:
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Command executed:

  [ ! -f  hct116_h3k4me1_IP_R1_T1.fastq.gz ] && ln -s hct116_h3k4me1_clean.fastq.gz
hct116_h3k4me1_IP_R1_T1.fastq.gz
  fastqc -q -t 6 hct116_h3k4me1_IP_R1_T1.fastq.gz

Command exit status:
  0

Command output:
  (empty)

Command error:
  WARNING: Your kernel does not support swap limit capabilities or the cgroup is not mounted.
Memory limited without swap.
  Failed to process file hct116_h3k4me1_IP_R1_T1.fastq.gz
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              at uk.ac.babraham.FastQC.Analysis.AnalysisRunner.run(AnalysisRunner.java:77)
              at java.base/java.lang.Thread.run(Thread.java:834)
```

# Log output from the tool

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```
[6:16 PM] Error executing process > 'FASTQC (hct116_h3k4me1_IP_R1_T1)'

Caused by:
  Missing output file(s) `*.{zip,html}` expected by process `FASTQC (hct116_h3k4me1_IP_R1_T1)`

Command executed:

  [ ! -f  hct116_h3k4me1_IP_R1_T1.fastq.gz ] && ln -s hct116_h3k4me1_clean.fastq.gz
hct116_h3k4me1_IP_R1_T1.fastq.gz
  fastqc -q -t 6 hct116_h3k4me1_IP_R1_T1.fastq.gz

Command exit status:
  0

Command output:
  (empty)

Command error:
  WARNING: Your kernel does not support swap limit capabilities or the cgroup is not mounted.
Memory limited without swap.
  Failed to process file hct116_h3k4me1_IP_R1_T1.fastq.gz
  uk.ac.babraham.FastQC.Sequence.SequenceFormatException: Ran out of data in the middle of a
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              at uk.ac.babraham.FastQC.Analysis.AnalysisRunner.run(AnalysisRunner.java:77)
              at java.base/java.lang.Thread.run(Thread.java:834)
```

# Log output from the tool

```
Error executing process > 'NFCORE_RNASEQ:RNASEQ:ALIGN_STAR:BAM_SORT_SAMTOOLS:SAMTOOLS_SORT
(bln1_t16_rep1)'

Caused by:
  Process `NFCORE_RNASEQ:RNASEQ:ALIGN_STAR:BAM_SORT_SAMTOOLS:SAMTOOLS_SORT (bln1_t16_rep1)`
terminated with an error exit status (1)

Command executed:

  samtools sort  -@ 36 -o bln1_t16_rep1.sorted.bam -T bln1_t16_rep1.sorted
bln1_t16_rep1.Aligned.out.bam
  cat <<-END_VERSIONS > versions.yml
  SAMTOOLS_SORT:
      samtools: $(echo $(samtools --version 2>&1) | sed 's/^.*samtools //; s/Using.*$//')
  END_VERSIONS

Command exit status:
  1

Command output:
  (empty)

Command error:
  [E::bgzf_read_block] Invalid BGZF header at offset 1083189972
  [E::bgzf_read] Read block operation failed with error 6 after 131 of 197 bytes
  samtools sort: truncated file. Aborting

Work dir:
  /lustre/scratch123/tol/teams/tolit/users/ps22/work/eb/4d3c6811645c4730ea79d1bca11ff1

Tip: you can try to figure out what's wrong by changing to the process work dir and showing the
script file named `.command.sh`
```

```
(bln1_t16_rep1)'

Caused by:
  Process `NFCORE_RNASEQ:RNASEQ:ALIGN_STAR:BAM_SORT_SAMTOOLS:SAMTOOLS_SORT (bln1_t16_rep1)`
terminated with an error exit status (1)

Command executed:

  samtools sort  -@ 36 -o bln1_t16_rep1.sorted.bam -T bln1_t16_rep1.sorted
bln1_t16_rep1.Aligned.out.bam
  cat <<-END_VERSIONS > versions.yml
  SAMTOOLS_SORT:
      samtools: $(echo $(samtools --version 2>&1) | sed 's/^.*samtools //; s/Using.*$//')
  END_VERSIONS

Command exit status:
  1

Command output:
  (empty)

Command error:
  [E::bgzf_read_block] Invalid BGZF header at offset 1083189972
  [E::bgzf_read] Read block operation failed with error 6 after 131 of 197 bytes
  samtools sort: truncated file. Aborting

Work dir:
  /lustre/scratch123/tol/teams/tolit/users/ps22/work/eb/4d3c6811645c4730ea79d1bca11ff1

Tip: you can try to figure out what's wrong by changing to the process work dir and showing the
script file named `.command.sh`
```

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# Where the work directory is on the disk

# Anatomy of a work directory..

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`.command.out` - STDOUT from tool

`.command.err` - STDERR from tool

`.command.log` - STDOUT and STDERR from tool

`.command.begin` - Created as soon as the job launches

`.exitcode` - Created when the job ends, with exit code

`.command.trace` - Logs of compute resource usage

`.command.run` - Wrapper script used to run the job

`.command.sh` - Process command used for this task

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4 Search Slack, Google, ask for help

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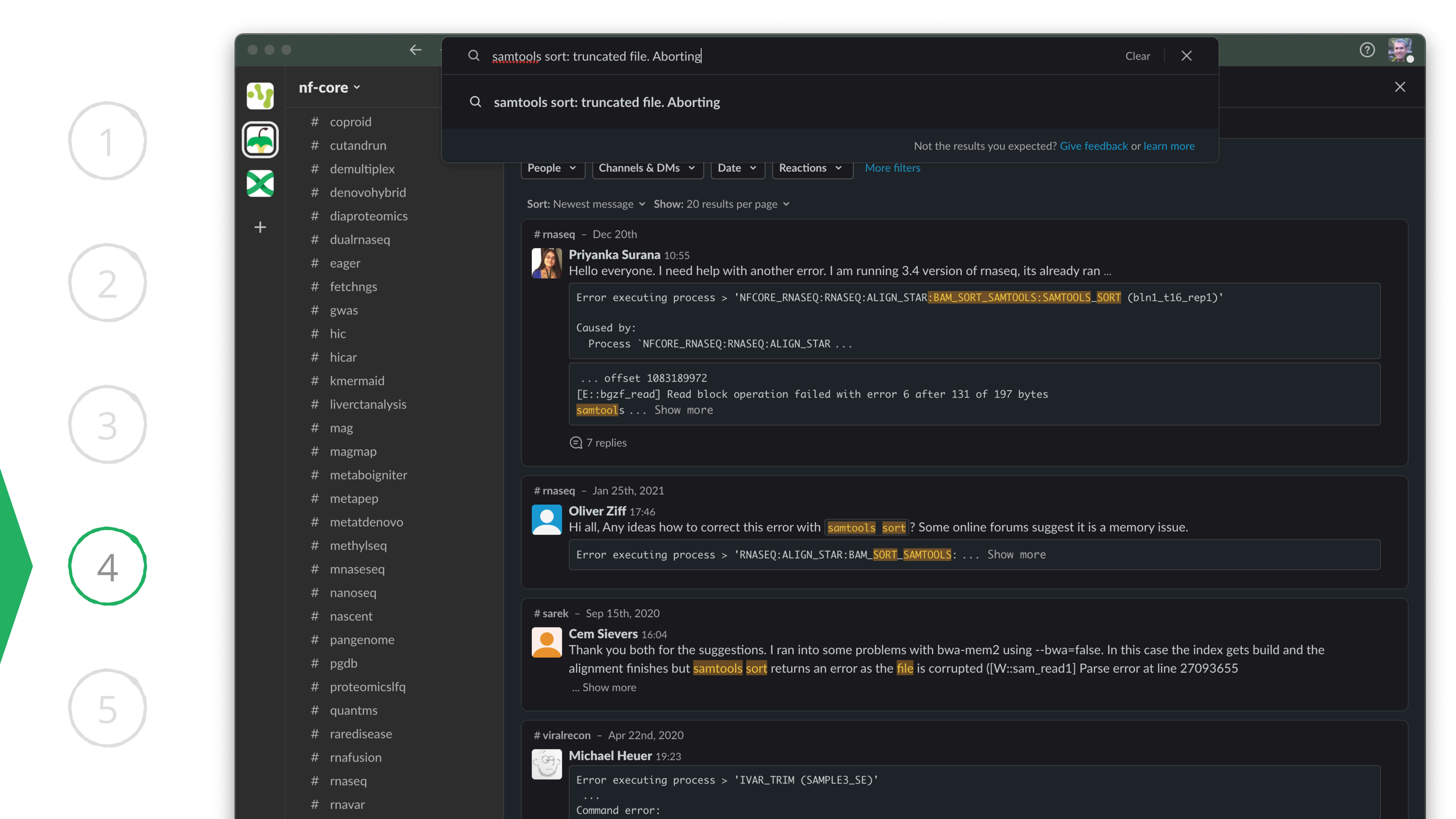
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samtools sort: truncated file. Aborting
Clear
samtools sort: truncated file. Aborting
Not the results you expected? Give feedback or learn more
nf-core
# coproid
# cutandrun
# demultiplex
# denovohybrid
# diaproteomics
# dualrnaseq
# eager
# fetchngs
# gwas
# hic
# hicar
# kmermaid
# liverctanalysis
# mag
# magmap
# metaboigniter
# metapep
# metatdenovo
# methylseq
# mnaseseq
# nanoseq
# nascent
# pangenome
# pgdb
# proteomicslfq
# quantms
# raredisease
# rnafusion
# rnaseq
# rnavar
People
Channels & DMs
Date
Reactions
More filters
Sort: Newest message
Show: 20 results per page
# rnaseq – Dec 20th
Priyanka Surana 10:55
Hello everyone. I need help with another error. I am running 3.4 version of rnaseq, its already ran ...
Error executing process > 'NFCORE_RNASEQ:RNASEQ:ALIGN_STAR:BAM_SORT_SAMTOOLS:SAMTOOLS_SORT (bln1_t16_rep1)'
Caused by:
Process `NFCORE_RNASEQ:RNASEQ:ALIGN_STAR ...
... offset 1083189972
[E::bgzf_read] Read block operation failed with error 6 after 131 of 197 bytes
samtools ... Show more
7 replies
# rnaseq – Jan 25th, 2021
Oliver Ziff 17:46
Hi all, Any ideas how to correct this error with samtools sort ? Some online forums suggest it is a memory issue.
Error executing process > 'RNASEQ:ALIGN_STAR:BAM_SORT_SAMTOOLS: ... Show more
# sarek – Sep 15th, 2020
Cem Sievers 16:04
Thank you both for the suggestions. I ran into some problems with bwa-mem2 using --bwa=false. In this case the index gets build and the alignment finishes but samtools sort returns an error as the file is corrupted ([W::sam_read1] Parse error at line 27093655
... Show more
# viralrecon – Apr 22nd, 2020
Michael Heuer 19:23
Error executing process > 'IVAR_TRIM (SAMPLE3_SE)'
...
Command error:

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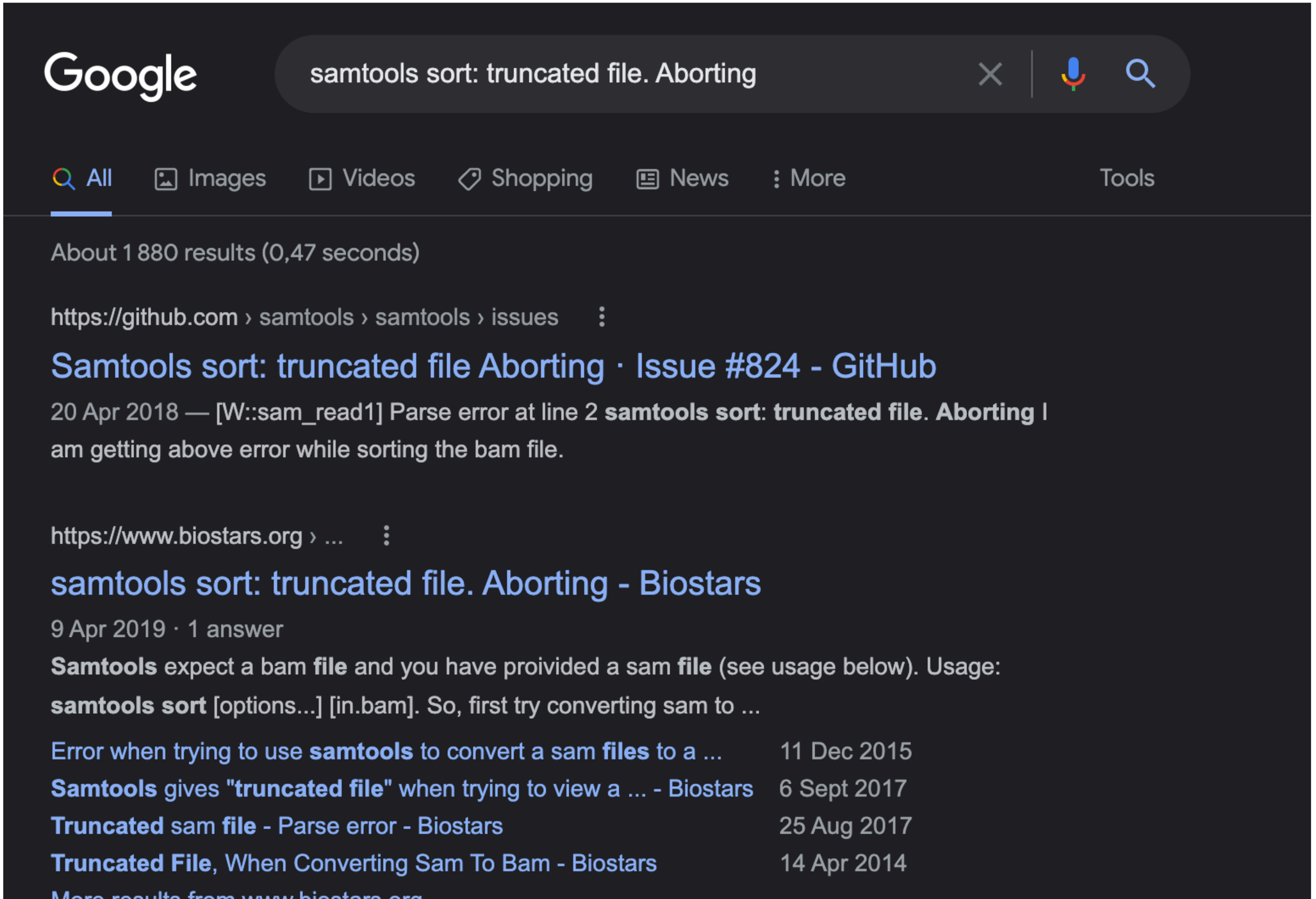
Google
samtools sort: truncated file. Aborting
All
Images
Videos
Shopping
News
More
Tools
About 1 880 results (0,47 seconds)
https://github.com › samtools › samtools › issues
Samtools sort: truncated file Aborting · Issue #824 - GitHub
20 Apr 2018 — [W::sam_read1] Parse error at line 2 samtools sort: truncated file. Aborting I am getting above error while sorting the bam file.
https://www.biostars.org › ...
samtools sort: truncated file. Aborting - Biostars
9 Apr 2019 · 1 answer
Samtools expect a bam file and you have proivided a sam file (see usage below). Usage: samtools sort [options...] [in.bam]. So, first try converting sam to ...
Error when trying to use samtools to convert a sam files to a ... 11 Dec 2015
Samtools gives "truncated file" when trying to view a ... - Biostars 6 Sept 2017
Truncated sam file - Parse error - Biostars 25 Aug 2017
Truncated File, When Converting Sam To Bam - Biostars 14 Apr 2014

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# Ask for help

- Pick the correct Slack channel to post in
- Provide as much information as you can
  - As a minimum, the command and configs you used
  - Use a thread under your message if in doubt
- Use markdown code blocks
- Narrow the issue down as much as possible before asking
- Explain the steps to reproduce if possible

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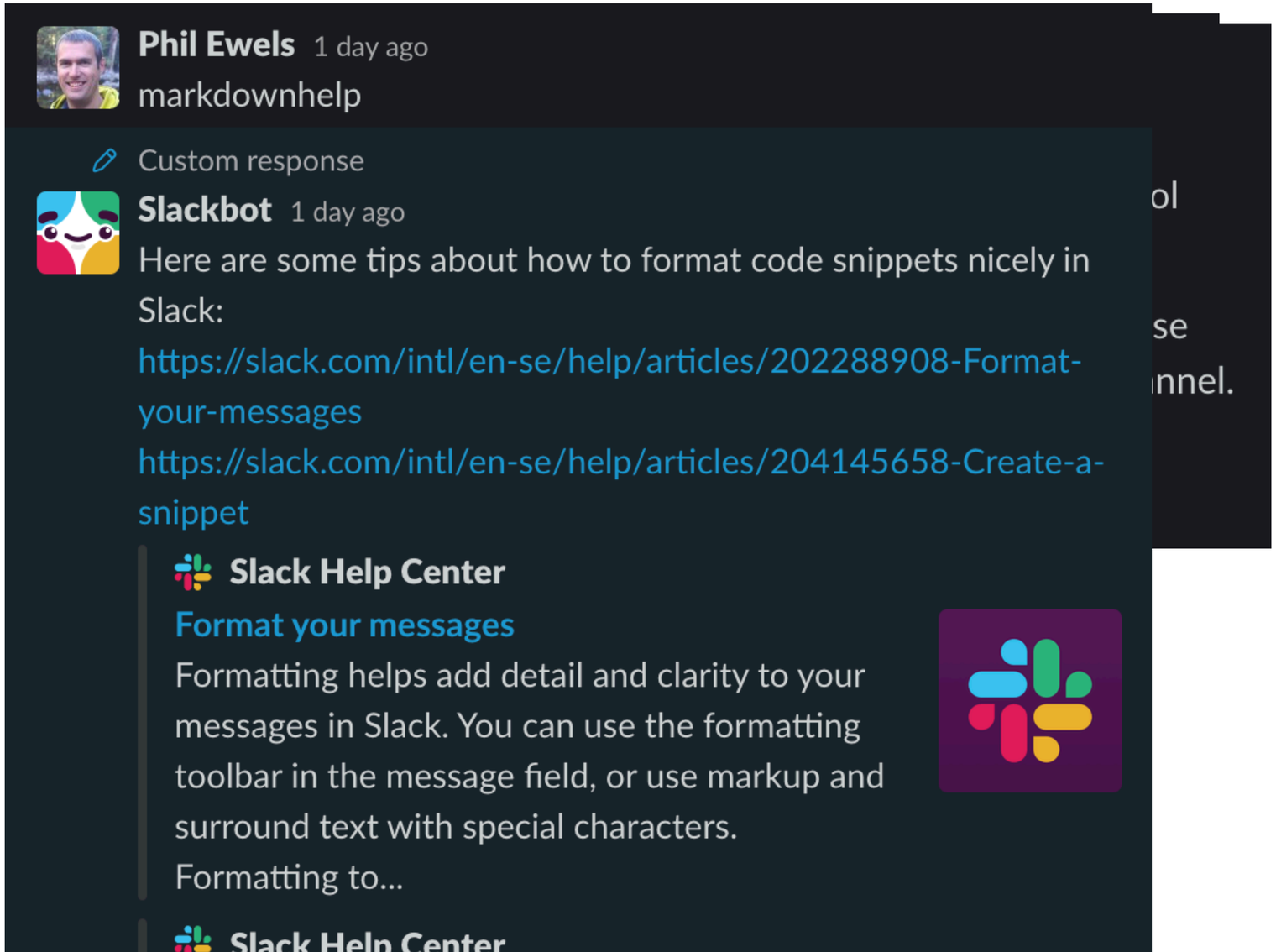
# Ask for help



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5 Report a bug

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nf-core / rnaseq Public

Watch 83 | Fork 394 | Star 409

Code | Issues 28 | Pull requests 3 | Discussions | Actions | Security | ...

**Bug report**
Report something that is broken or incorrect — Get started

**Feature request**
Suggest an idea for the nf-core/rnaseq pipeline — Get started

**Join nf-core**
Please join the nf-core community here — Open

**Slack #rnaseq channel**
Discussion about the nf-core/rnaseq pipeline — Open

Don't see your issue here? Open a blank issue.

Edit templates

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# Issue: Bug report

Report something that is broken or incorrect. If this doesn't look right, choose a different type.

Title

Before you post this issue, please check the documentation:

- nf-core website: troubleshooting
- nf-core/rnaseq pipeline documentation

## Description of the bug *

A clear and concise description of what the bug is.

## Command used and terminal output

Steps to reproduce the behaviour. Please paste the command you used to launch the pipeline and the output from your terminal.

```
$ nextflow run ...

Some output where something broke
```

## Relevant files

Please drag and drop the relevant files here. Create a `.zip` archive if the extension is not allowed.
Your verbose log file `.nextflow.log` is often useful *(this is a hidden file in the directory where you launched the pipeline)* as well as custom Nextflow configuration files.

**Assignees**

No one—assign yourself

**Labels**

bug

**Projects**

None yet

**Milestone**

No milestone

**Linked pull requests**

Successfully merging a pull request may close this issue.

**Helpful resources**

Contributing
Code of conduct
GitHub Community Guidelines

Beta You're using an **issue form**, a new type of issue template.

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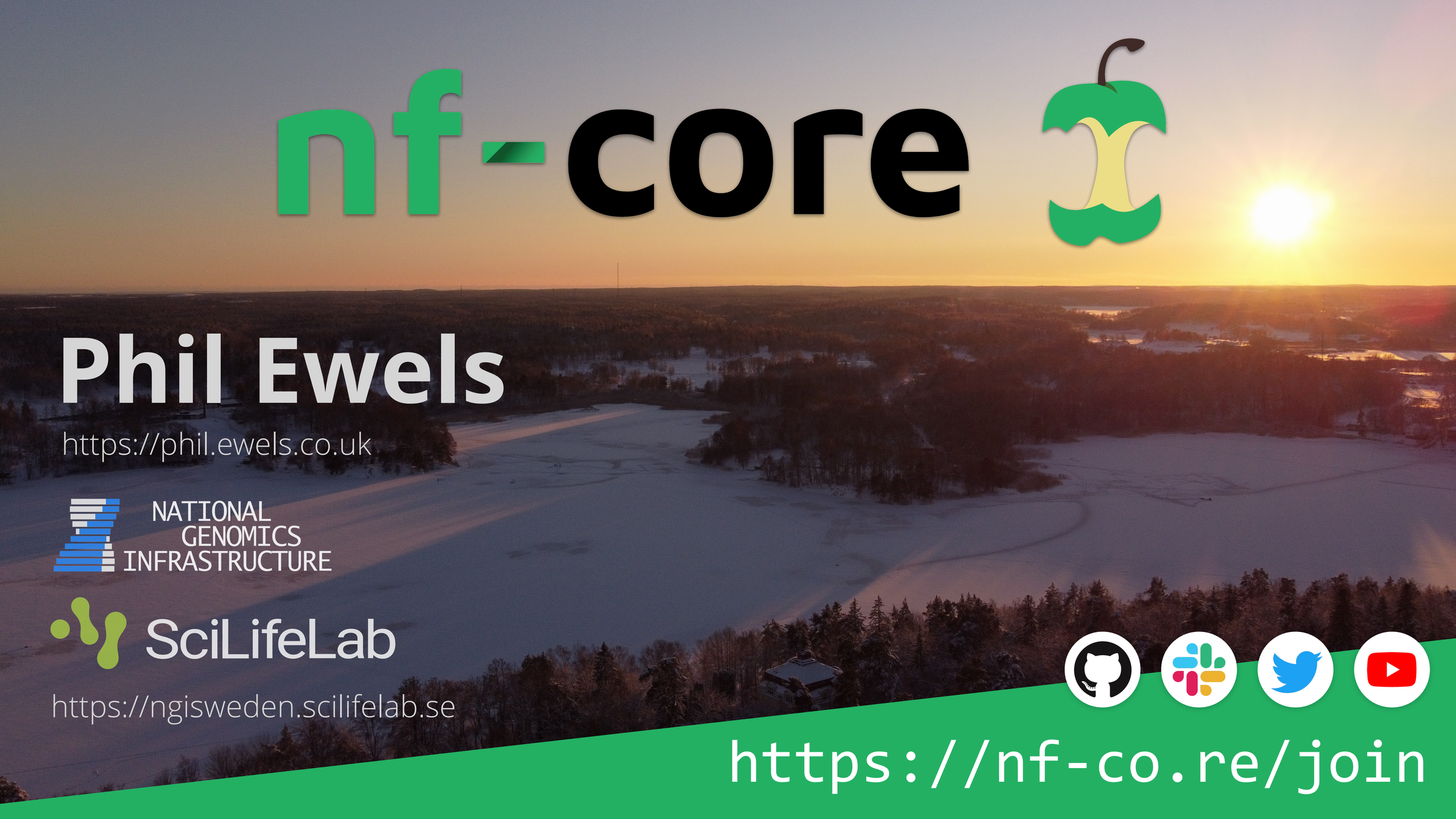
- Fill in the bug issue template
- Narrow the issue down as much as possible before asking
- Explain the steps to reproduce
- If you think you know the solution, please say so
- If you think you can fix the problem, please make a pull request

## Report a bug


nf-core
Phil Ewels
https://phil.ewels.co.uk
NATIONAL GENOMICS INFRASTRUCTURE
SciLifeLab
https://ngisweden.scilifelab.se
https://nf-co.re/join