

2020-07-29

# MISO LIMS Training

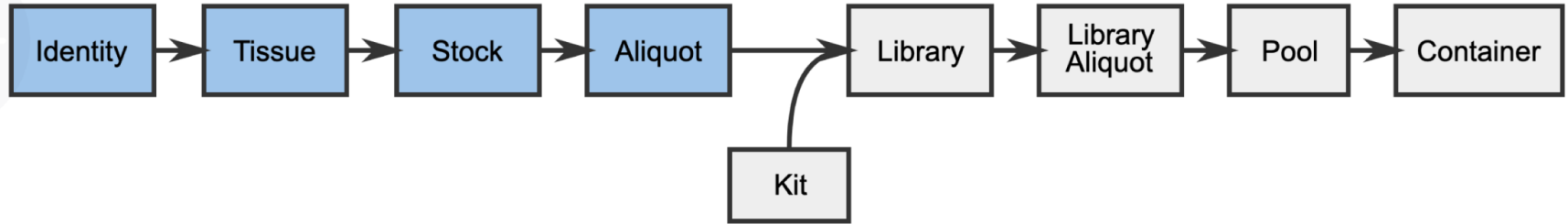
## Libraries

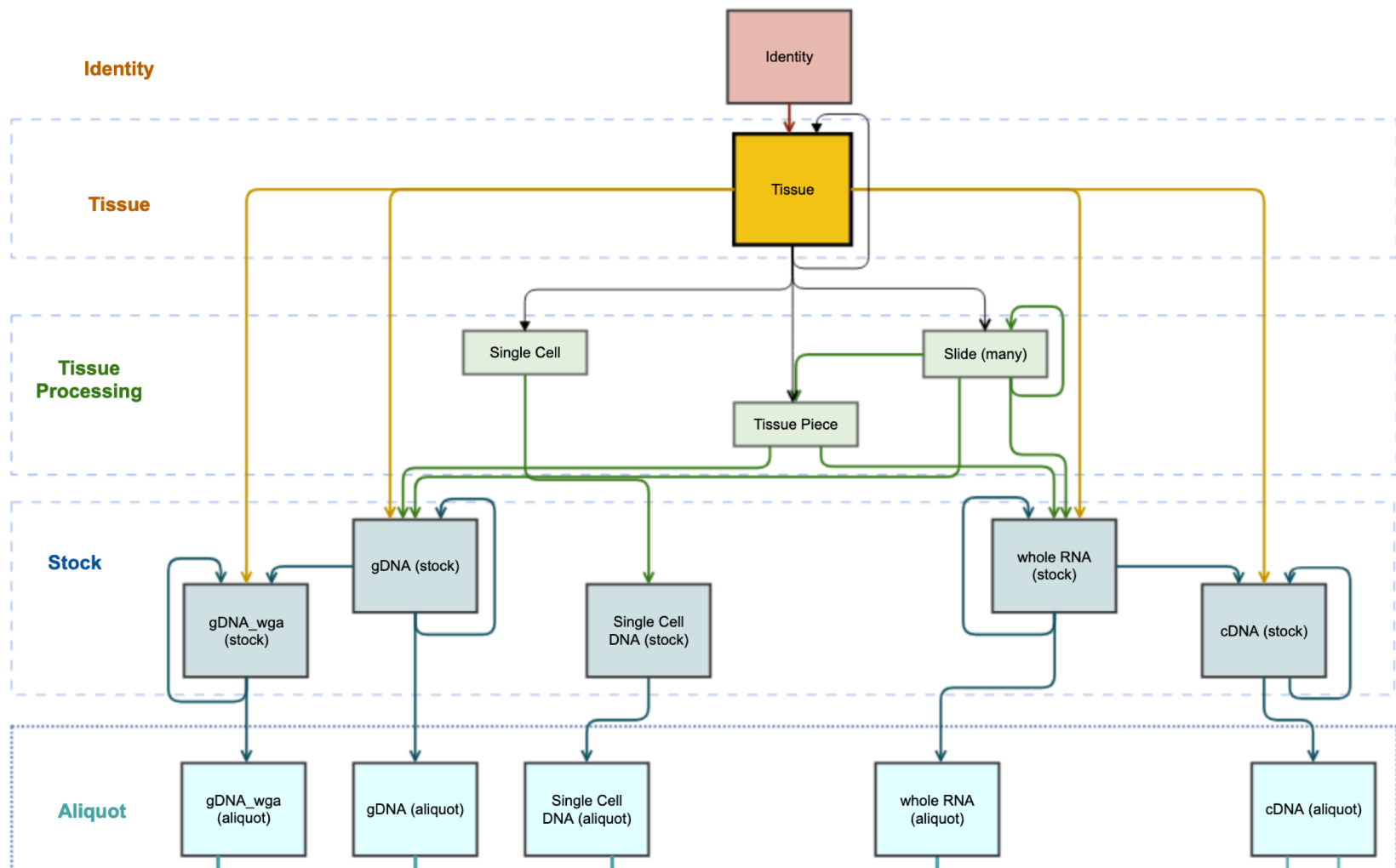


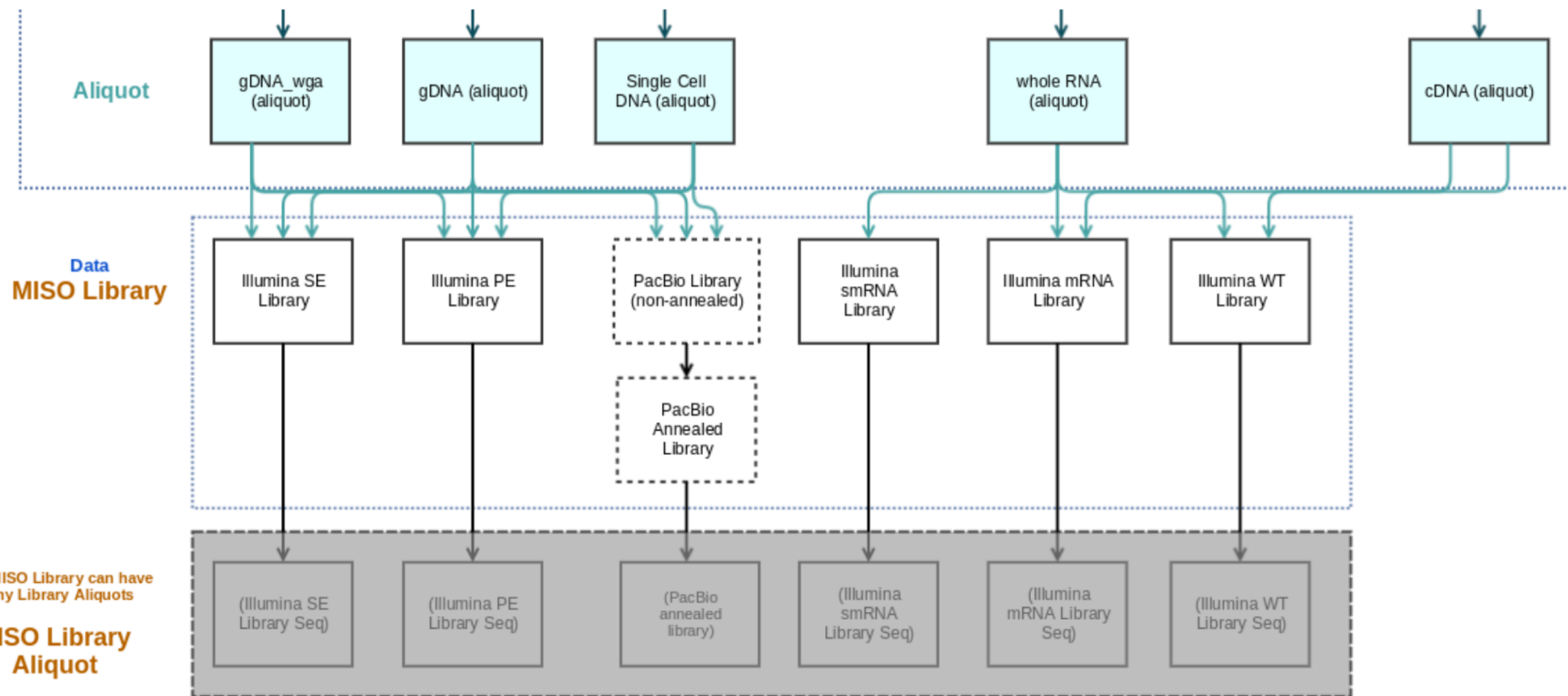
# Outline

- Libraries
  - Processing samples into libraries
  - Receiving libraries
- Library aliquots
  - Processing libraries into library aliquots
- Pools
- Pool Orders
- Sequencing Orders
- Index Distance Tool
- Index Search Tool

# General MISO Flow







# Libraries

- Libraries are made from aliquot samples for a particular platform (e.g. Illumina)
- There is a library design that decides
  - The selection type
  - The strategy type
- Indices may be added to a library
- Libraries can be created from existing aliquots or received directly
  - When receiving libraries, some sample information is also required

# Working with Libraries

- QC information may be added to a library
- Deleting libraries
  - Creator or MISO administrators can delete
  - Cannot delete if there are library aliquots
- Dropdown options
  - Check Misc. and Institute Defaults menus
  - Ask your MISO administrator to add new items

# Library Aliquots

- A library aliquot is a portion of a library separated for pooling or further processing
- Can be made from libraries, or from other library aliquots
- Some attributes may be changed from the library
  - Concentration
  - Design code (WG, EX, etc.)
  - Insert size



# Pools

- A pool is made up of one or multiple library aliquots that are ready to be loaded into a sequencing container partition for sequencing
- Aliquots may be added at different proportions
- QC information may be added to pools
- A pool containing multiple aliquots is called multiplexed
  - Indices are used to demultiplex the libraries later on
  - Duplicate or near-duplicate indices cause problems
  - MISO may prevent you from creating pools containing problematic indices
  - Index Distance Tool can be used to check index compatibility

# Transfers

- Libraries, library aliquots, and pools can be included in transfers
- Transfers represent a custody change from one lab or group to another
  - Receipt: from an external lab
  - Internal: within the organization
  - Distribution: to an external lab
- Specify
  - Who/where items came from
  - Who/where they were sent
  - Confirmation of receipt
  - Confirmation of QC upon receipt
- Full chain of custody can be tracked

# QCs

- Libraries, library aliquots and pools have QC status
  - Pass/Fail/Unknown
- Additional QC data can be added to libraries and pools
  - Results may be pass/fail or numerical
  - May use instruments, kits, and/or controls

Library Alias	Date	Type	Instrument	Kit	Kit Lot	Result	Units
DEMO_0002_Ab_A_PE_123_WG	2020-07-28 ▼	Average Library Size (bp) (Fragment Analyzer) ▼	Fragment Analyzer ▼	▼		300	bp
DEMO_0002_Ab_A_PE_123_WG	2020-07-28 ▼	Concentration (Fragment Analyzer) ▼	Fragment Analyzer ▼	▼		12.3	ng/ul

# Pool Orders

- A pool order tells the sequencing team that a set of libraries is ready to be pooled and sequenced
- Specifies which library aliquots to include and at what proportions
  - Library aliquots derived from the ones specified are also accepted
- May specify sequencing requirements
- Fulfilling a pool order requires
  - Linking a pool that contains all of the specified aliquots (or derivatives)
  - Linking a sequencing order that matches the sequencing requirements of the pool order (if specified)
- Pool orders are optional – useful if one team hands libraries off to another team for pooling

# Sequencing Orders

- A sequencing order tells the sequencing team that a pool is ready to be sequenced
- Specifies
  - Instrument type
  - Sequencing parameters such as chemistry and read length
  - Number of partitions (lanes) required
- Automatically marked as fulfilled when matching runs are completed

# Index Distance Tool

- Indices are used to identify libraries in a multiplexed pool
- Pools containing duplicate and near-duplicate indices cause analysis problems
- The tool allows you to check indices before pooling or creating pool orders

Indices

```
AAAAAAAA
AAAAAACC
CCCCCCCC
CGCGCGCG
CCCCCCCC
```

Min. Mismatches:

Calculate

Clear

Results

Indices with 2 or fewer mismatches are considered **DUPLICATES**.

Indices were compared at the length of the shortest index, which is 8 bp.

Near match (2): "AAAAAAAA", "AAAAAACC"  
Duplicate: "CCCCCCCC"

# Index Search Tool

- Allows you to identify index families by sequences
- Collaborators may only provide the index sequences
- Collaborators may use different names for the same indices
- Search before adding new index families to prevent duplication

# Exercise

Complete the following tutorials using Chrome or Firefox:

- Tutorial 7: Libraries
- Tutorial 9: Checking Index Distance

<https://miso-lims.github.io/walkthroughs/>

Note: These are the OICR walkthroughs. Other labs may have an alternate site.





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