Installation

1. Install KNIME Analytics Platform (from thumb drive)
2. Help > Install New Software > Add (> Archive):
   - 00_InstallationFiles/CommunityContributions_trunk.zip
   - https://update.knime.org/community-contributions/trunk
3. Available Software Sites:
   - Enable KNIME AP 3.5 Update Site
   - Enable KNIME Community Contributions (trunk)
4. Select *KNIME Community Contributions (trunk)*
5. Install *KNIME Image Processing* **and** *KNIME Image Processing - OMERO Integration*

OMERO and KNIME Workshop

Stefan Helfrich, Christian Dietz
KNIME

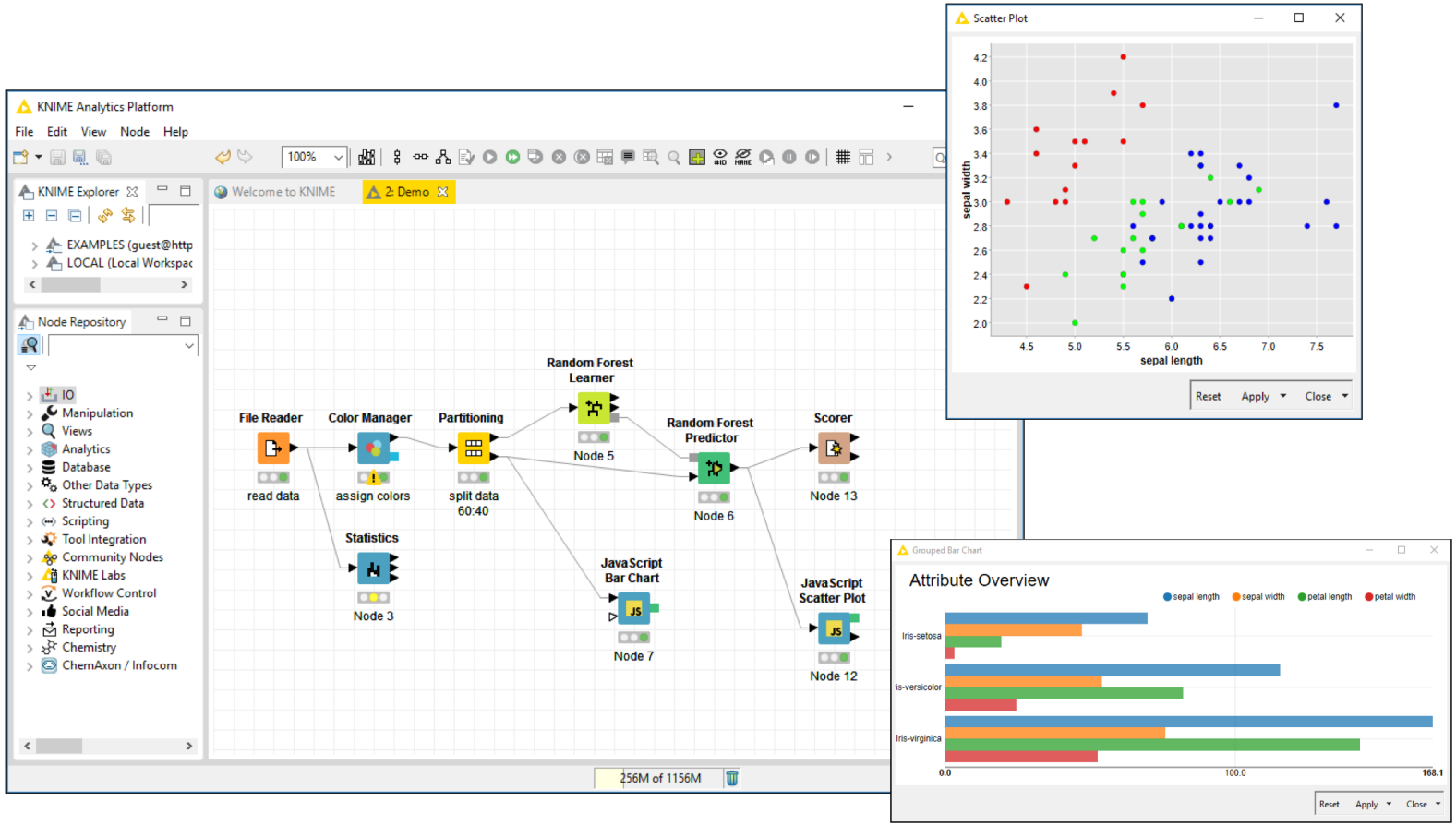
Learning goals

- How to get around KNIME Analytics Platform (AP)
- How to get your images from OMERO into KNIME AP
- How to extract quantitative data from your images
- How to use analytics and visualization functionality in KNIME AP

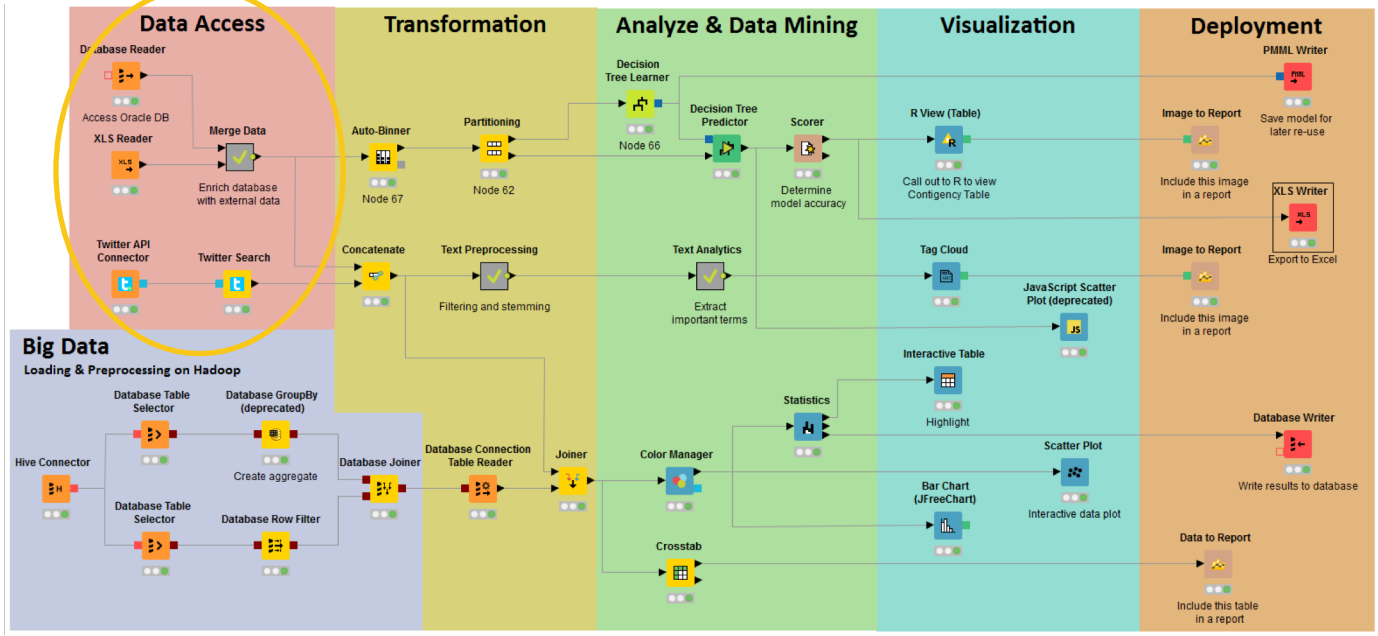
What is KNIME Analytics Platform?

- A tool for data analysis, manipulation, visualization, and reporting
- Based on the graphical programming paradigm
- Provides a diverse array of extensions:
  - Text Mining
  - Network Mining
  - Cheminformatics
  - Image Informatics
  - Many integrations, such as Java, R, Python, Weka, etc.

The KNIME® Analytics Platform



Over 2000 native and embedded nodes included:



Data Access
MySQL, Oracle, ...
SAS, SPSS, ...
Excel, Flat, ...
Hive, Impala, ...
XML, JSON, PMML
Text, Doc, Image, ...
Web Crawlers
Industry Specific
Community / 3rd

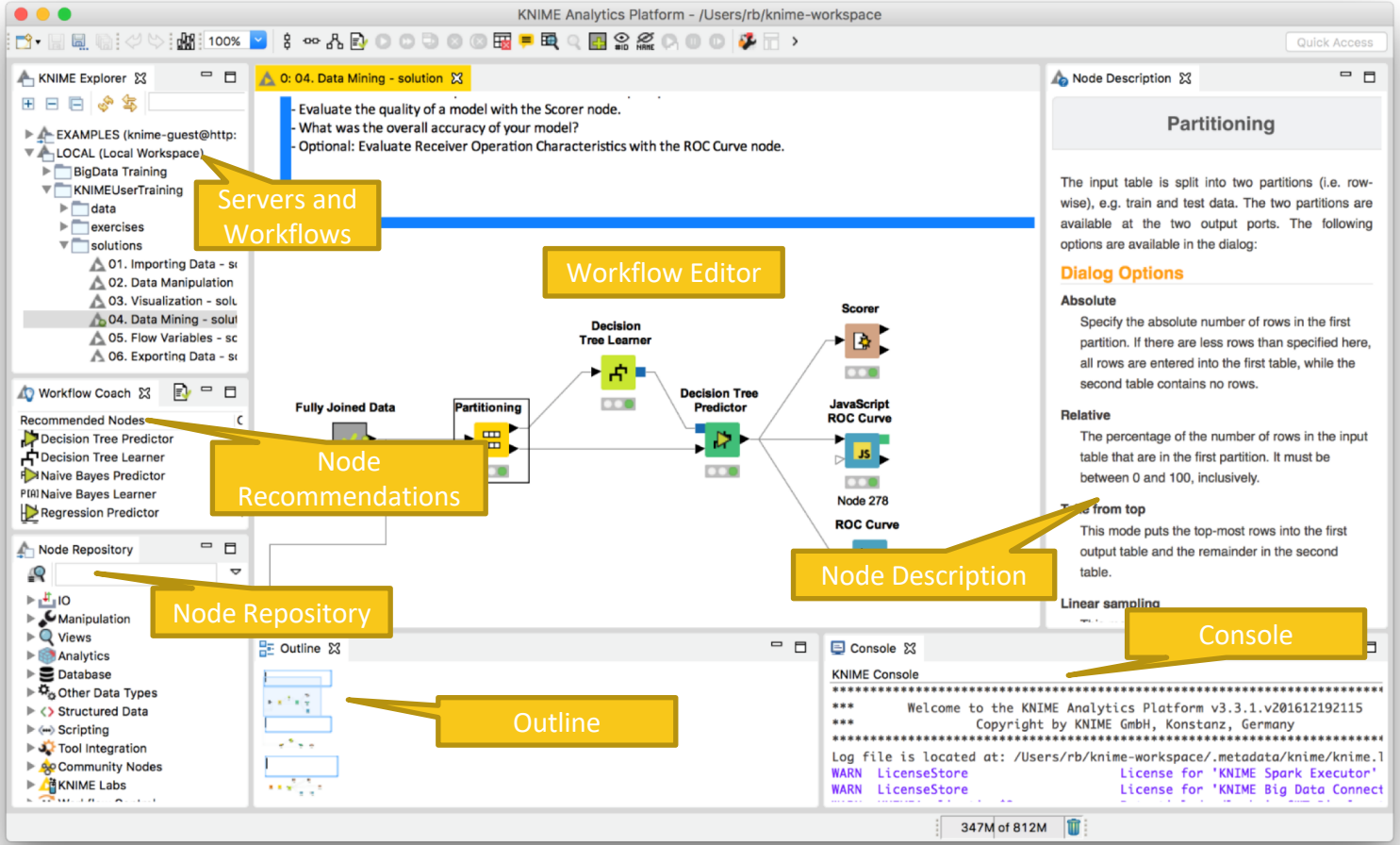
Transformation
Row,
Column
Matrix
Text, Image
Time Series
Java
Python
Community / 3rd

Analysis & Mining
Statistics
Data Mining
Machine Learning
Web Analytics
Text Mining
Network Analysis
Social Media Analysis
R, Weka, Python
Community / 3rd

Visualization
R
JFreeChart
JavaScript
Community / 3rd

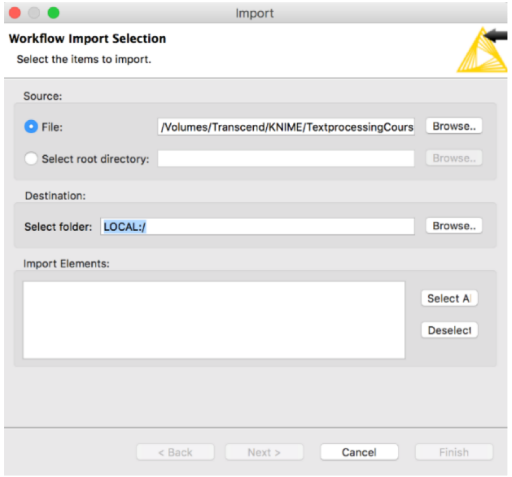
Deployment
via BIRT
PMML
XML, JSON
Databases
Excel, Flat, etc.
Text, Doc, Image
Industry Specific
Community / 3rd

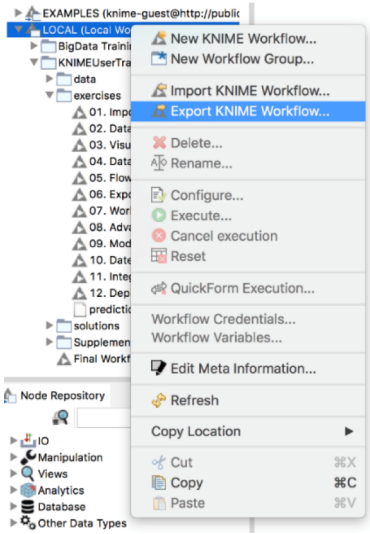
The KNIME Workbench

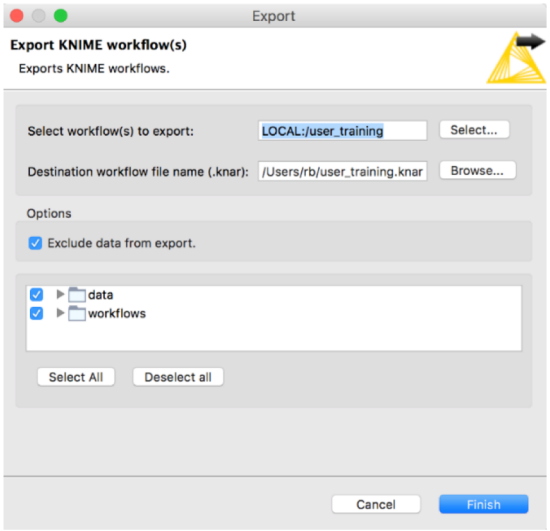


Creating New Workflows, Importing and Exporting

- Right-click anywhere in KNIME Explorer to create a new workflow or workflow group or to import a workflow
- Right-click on workflow or workflow group to export the selected workflow





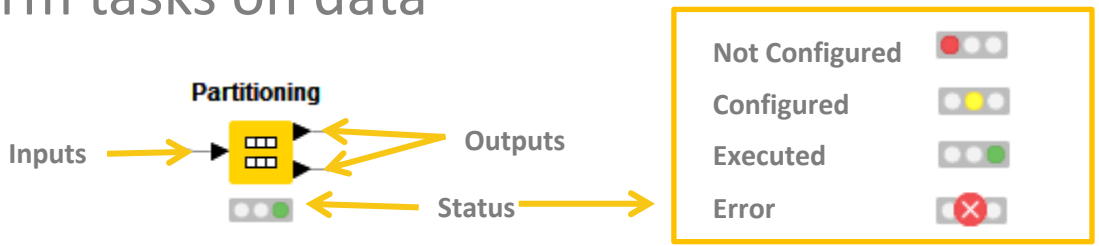


Exercise 00 – Importing the Exercises

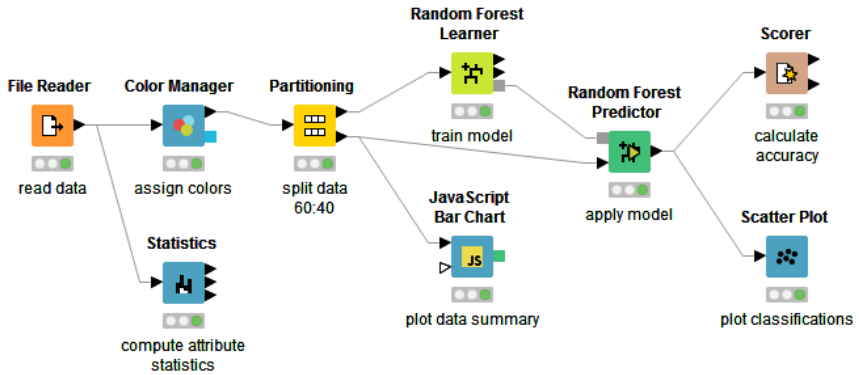
- File > Import KNIME Workflow > Browse
  - 01_KNIME-OMERO/OME-UGM-2018.knar

Visual KNIME Workflows

NODES perform tasks on data



Nodes are combined to create **WORKFLOWS**



More on Nodes…

A node can have 3 states:

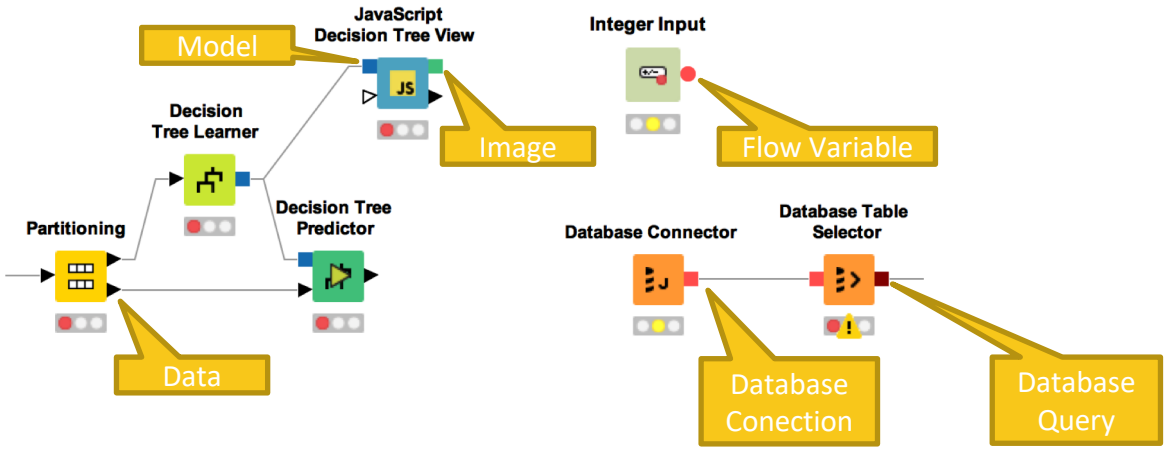
Not Configured:
The node is not yet configured and cannot be executed with its current settings.

Configured:
The node has been set up correctly, and may be executed at any time

Executed:
The node has been successfully executed. Results may be viewed and used in downstream nodes.

Inserting and Connecting Nodes

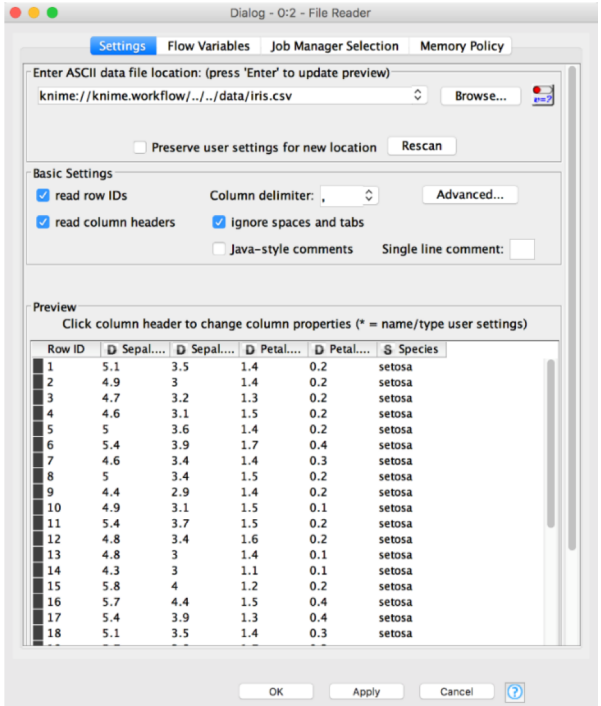
- Insert nodes into workspace by dragging them from the Node Repository or by double-clicking in the Node Repository
- Connect nodes by left-clicking output port of Node A and dragging the cursor to (matching) input port of Node B
- Common port types:



Node Configuration

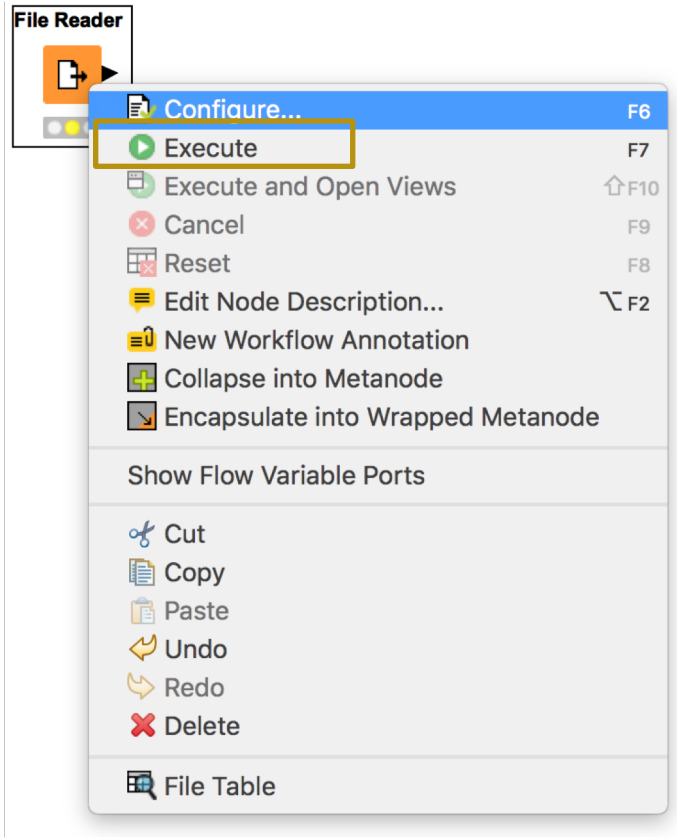
- Most nodes require configuration
- To access a node configuration window:
  - Double-click the node

  OR
  - Right-click > Configure



Node Execution

- Right-click node
- Select Execute in context menu
- If execution is successful, status shows green light
- If execution encounters errors, status shows red light

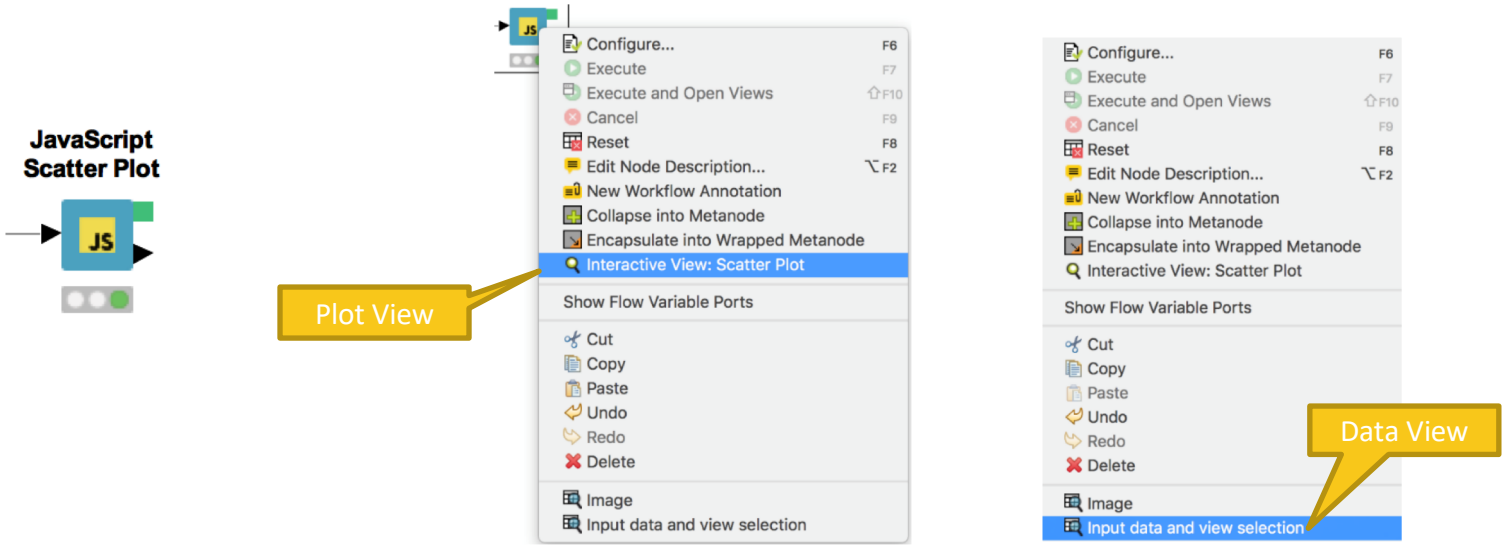


Node Outputs and Views

- Right-click executed node
- Select View option in context menu

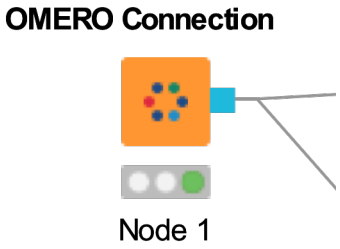
OR

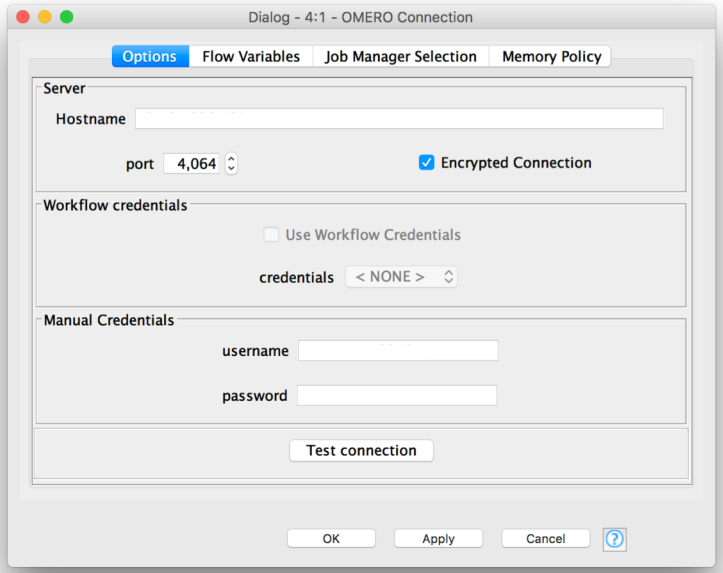
- Select output port (last item) to inspect execution results



New node: OMERO Connection

- Uses server settings (IP address, port) and credentials to establish a connection
- Doesn’t download any images

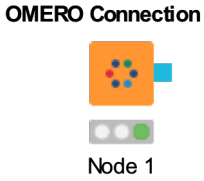




Exercise 01 – Connecting to OMERO

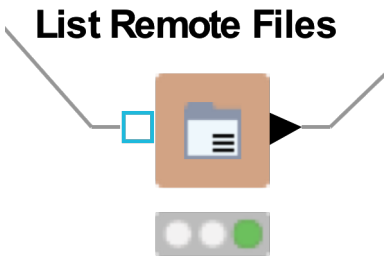
- Create a connection to an OMERO instance:
  - **IP**: …
  - **User**: …
  - **PW**: …
- Which port(s) do you see on the *OMERO Connection* node?

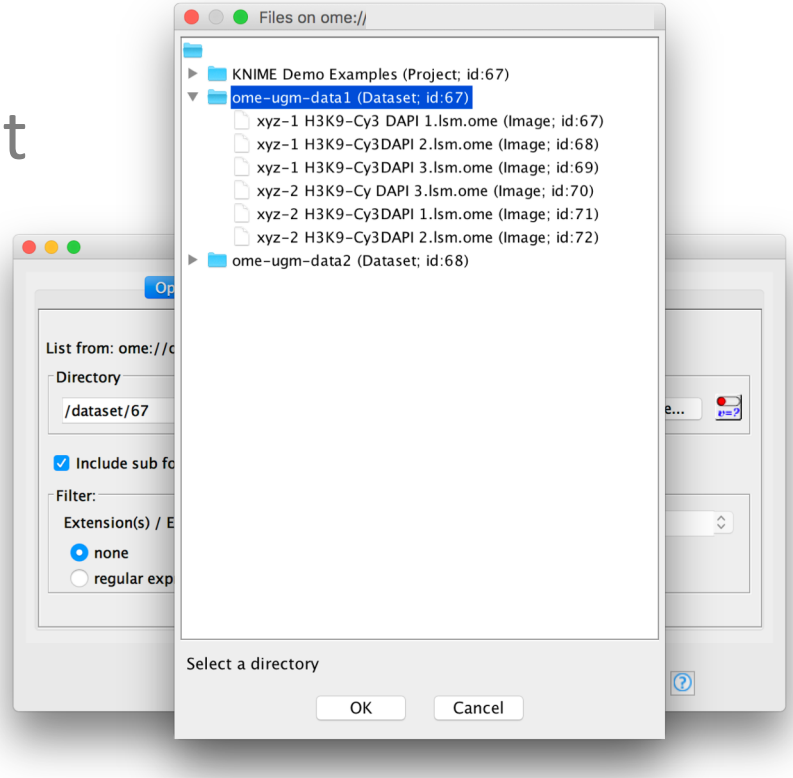
Exercise 01 - Solution



New node: List Remote Files

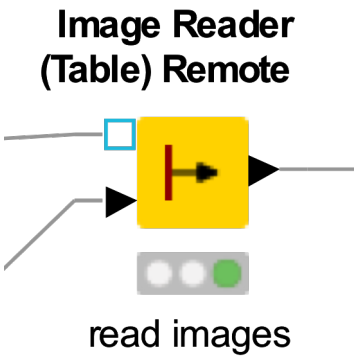
- Creates file locations for a set of files (usually all files in a folder → *List Files*)
- Doesn't load any images yet





New node: Image Reader (Table) Remote

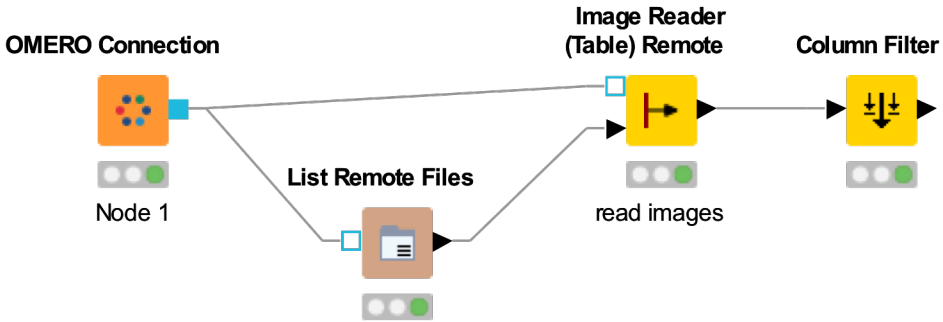
- Input table has to contain file locations
- Uses Bio-Formats to open images and store them into an `Img` column



Exercise 02 – Downloading Images

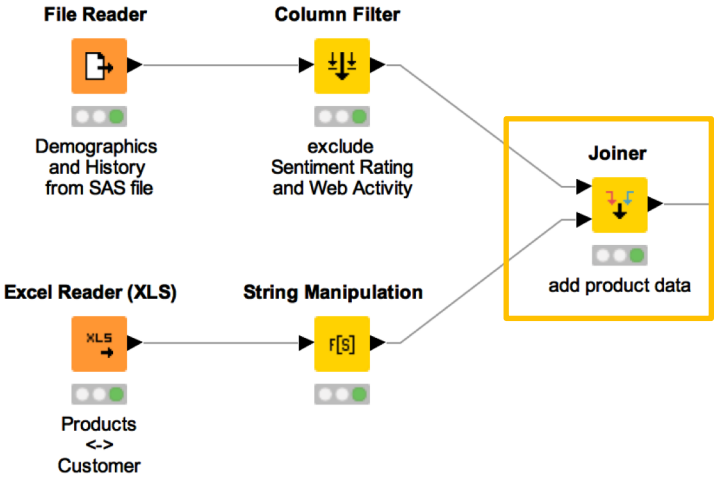
- Use the existing connection to select a dataset from our OMERO instance:
  - *ome-ugm-data1* or *ome-ugm-data2*
- What does the output table look like?
- Download images from the selected dataset and inspect the images

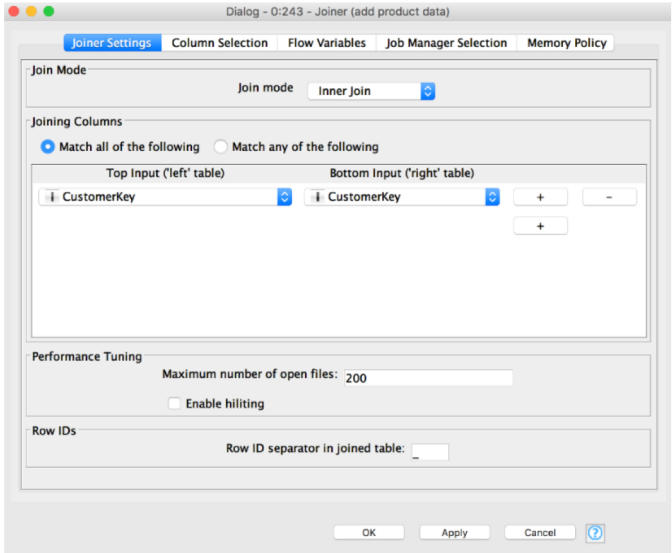
Exercise 02 - Solution



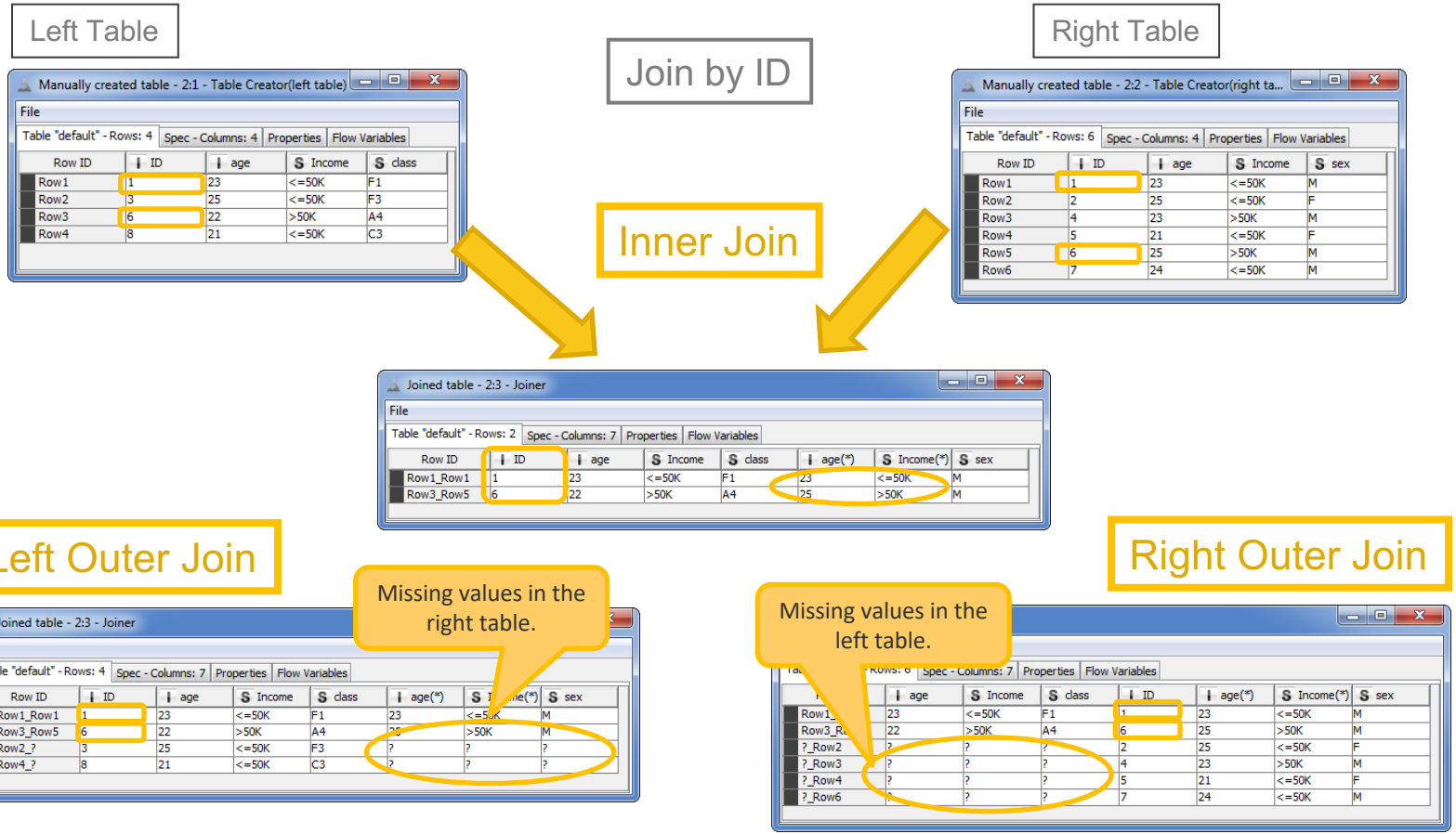
New Node: Joiner

- Combines columns from 2 different tables
- Top port contains “Left” data table
- Bottom port contains the “Right” data table

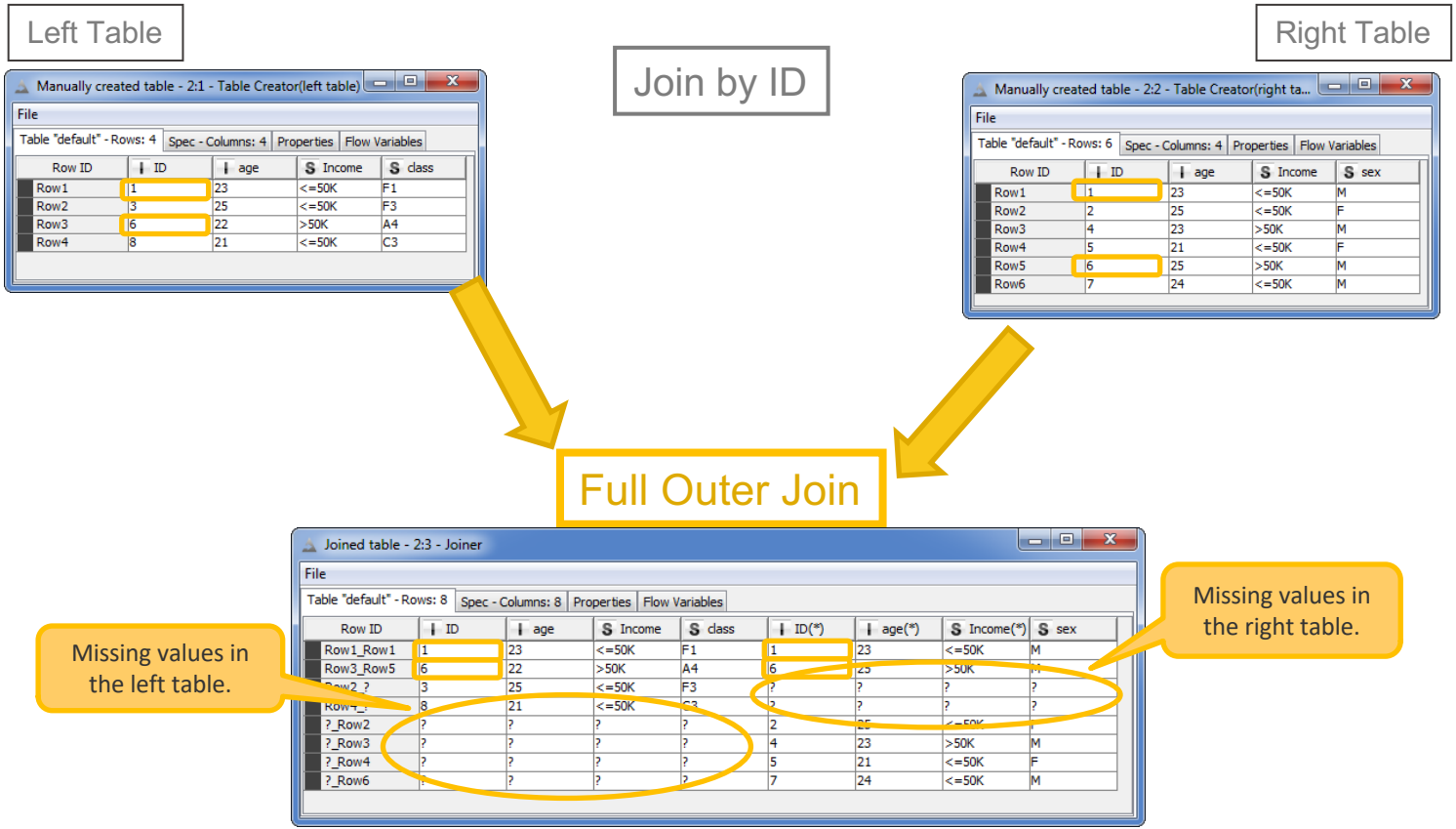




Joining Columns of Data



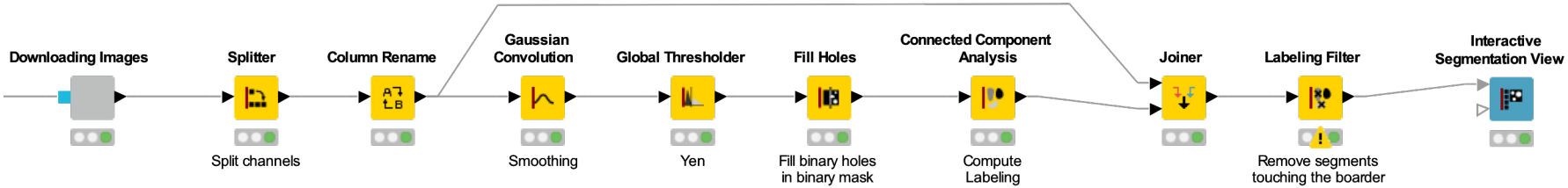
Joining Columns of Data



Exercise 03 – Processing Images

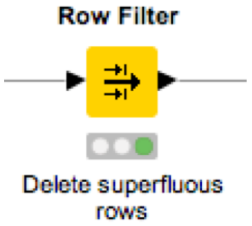
- Segment the first channel of each image:
  - Smoothing (*Gaussian Convolution*), *Global Thresholding* (Yen), *Fill Holes*, and *Connected Component Analysis*
  - Replace the result from the previous step!
- *Join* the resulting Labeling with the input data to recover a raw version of the first channel
- Remove cells that are touching the border image
  - Use *Labeling Filter*

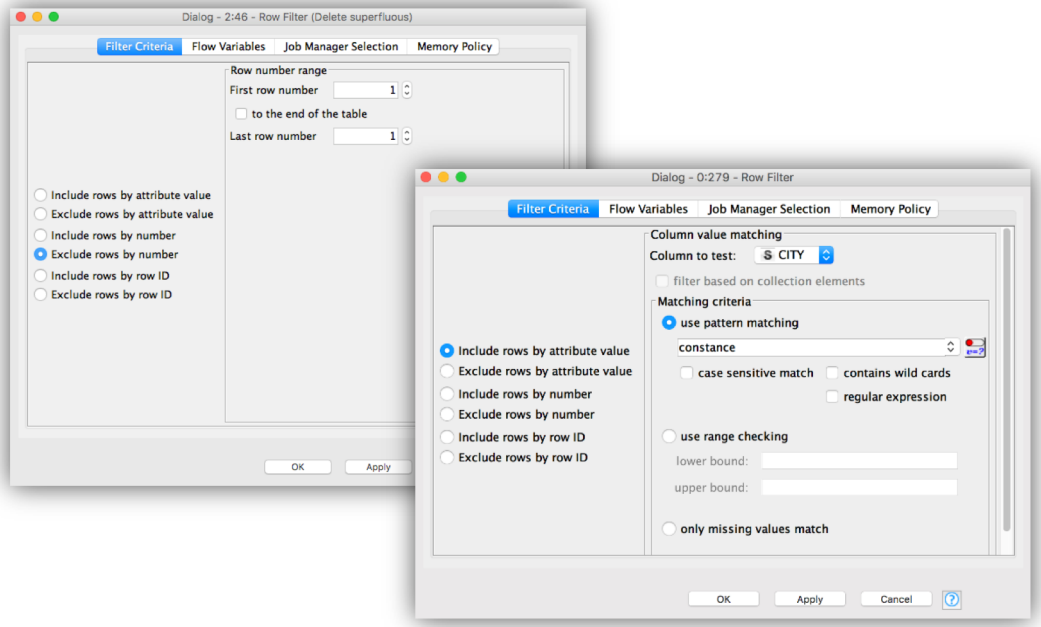
Exercise 03 - Solution



New Node: Row Filter

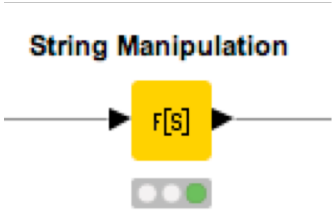
- Row filtering with in- and exclude option according to certain criteria
  - Certain value or pattern in a selectable column
  - Row number
  - Row ID

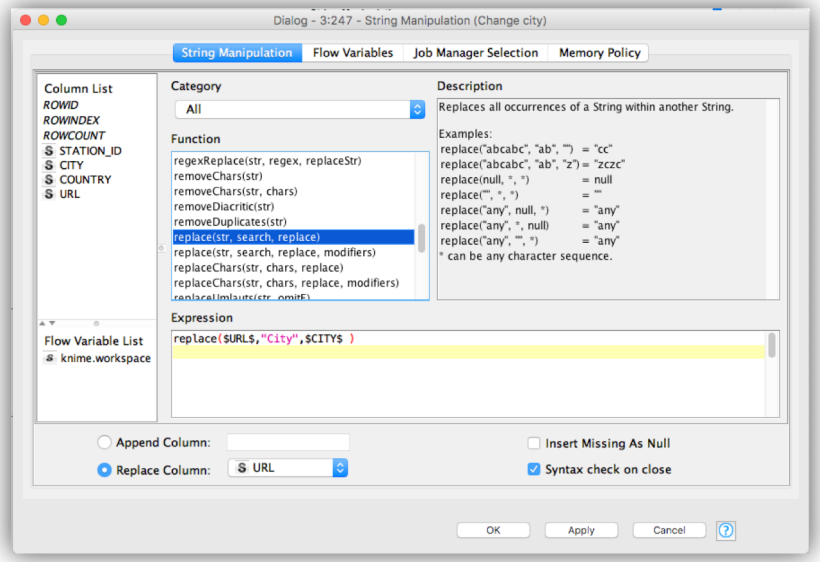




New Node: String Manipulation

- Create and edit values in string columns
  - Clean up capitalization (e.g. Lowercase)
  - Search and Replace, join
- Modify existing strings or create new columns





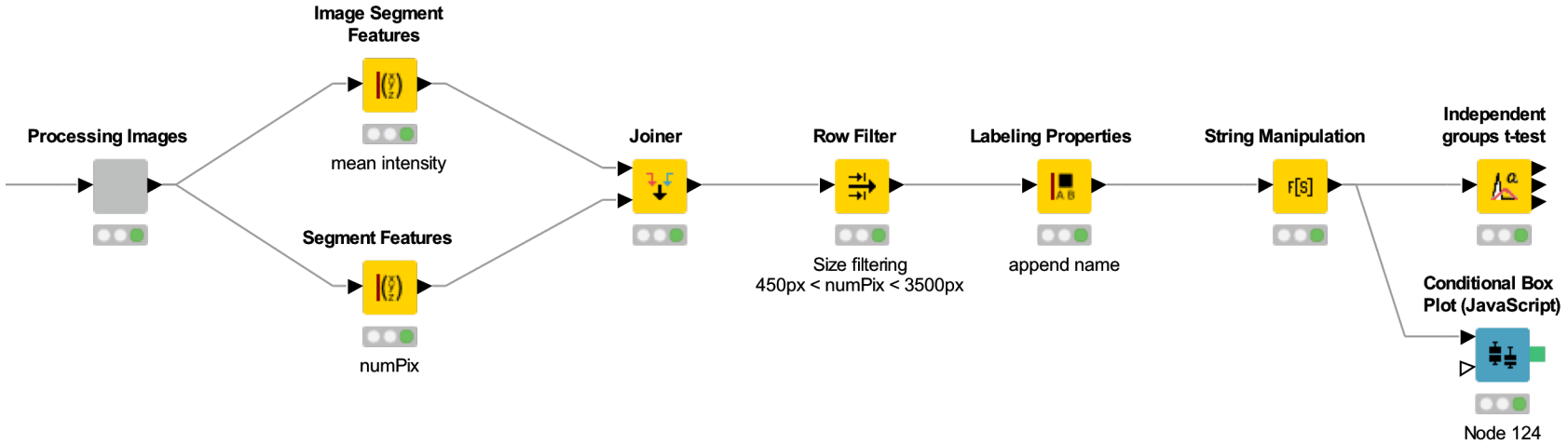
Exercise 04 – Visualizing Results

- Extract mean intensity from 2nd channel for each cell in an image (*Image Segment Features*)
- Extract the cell area (`NumPix`) for each cell in an image (*Segment Features*)
- *Join* the information for each cell
- Filter cells according to their size (*Row Filter*)
- Extract original file name for each cell (*Labeling Properties*)

Exercise 04 – Visualizing Results (continued)

- Extract class (*xyz-1* or *xyz-2*) from Name column
  - *String Manipulation* to extract first 5 characters
- Compute a p-value for the (independent) mean intensities distributions (*Independent groups t-test*)
- Generate a box plot of the NumPix properties (per group):
  - Use the *Conditional Box Plot (JavaScript)*

Exercise 04 - Solution



Additional Resources

- Learning/**LEARNING HUB:**
  https://www.knime.com/learning-hub
- Learning/**NODE GUIDE:**
  https://www.knime.com/nodeguide
- Community/**Forum:**
  https://forum.knime.com/

KNIME TV on **YouTube**
https://www.youtube.com/user/KNIMETV

KNIME Image Processing and the ImageJ Ecosystem