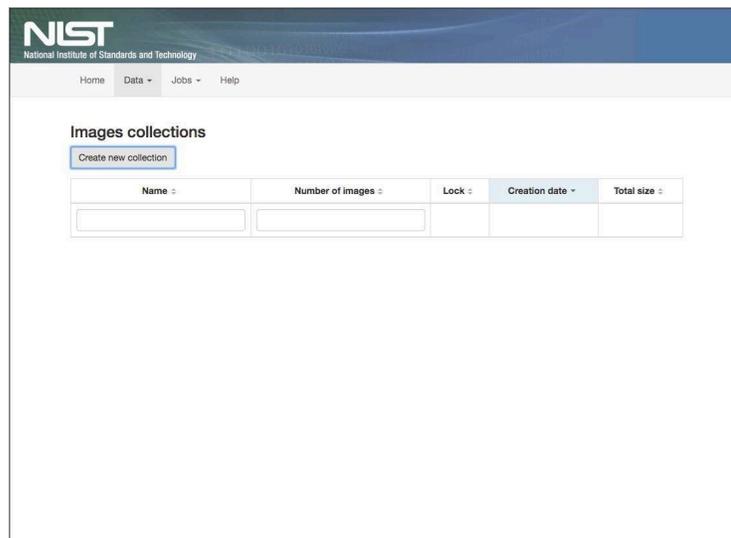


## Web Image Processing Usage and Help

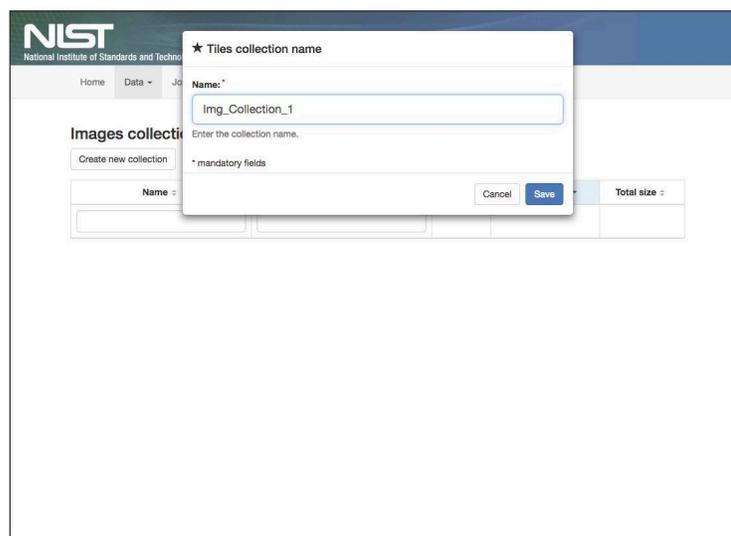
The interface consists of two drop-down menus denoted as "Data" and "Jobs". All input, intermediate and final results are accessible via the "Data" menu while all computations are accessible via the "Jobs" menu. The following are guidelines for a common workflow of steps.

### Images/Tiles Upload

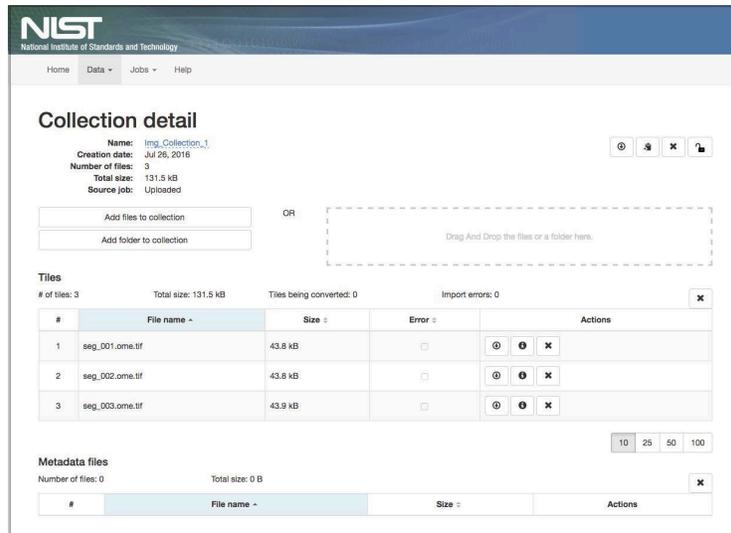
1. Go to Data > Tiles collections



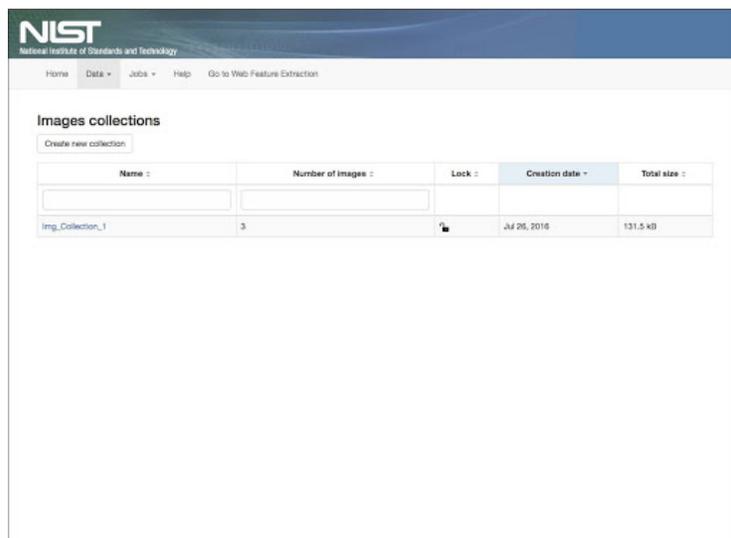
2. Click on "Create new collection"
3. Give it a unique name and click on "Save"



4. Upload the images using either:
  - the "Add files/folder" buttons
  - the drag and drop target



After uploading the images and going back to Tiles Collection, this is what your page should look like:

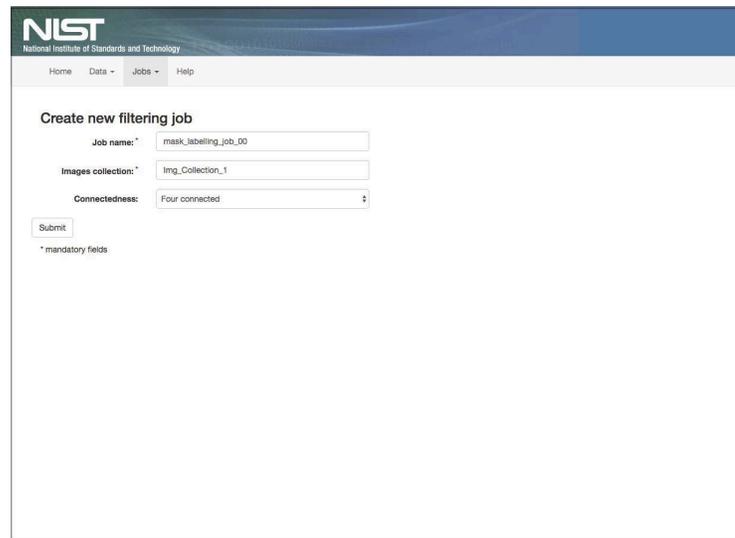


**Note:** File upload can be slow. Once files are uploaded they have to be converted into ome.tif format. We are using [OME](#) to support metadata storage and retrieval along the entire pipeline using a well defined standard format.

File conversion to ome.tif may be slower than the upload.

## Mask Labelling

1. Go to Jobs > Mask labelling jobs
2. Click on "Create new job"
3. Give it a unique job name
4. Specify the image collection
5. Specify the connectedness (four or height)



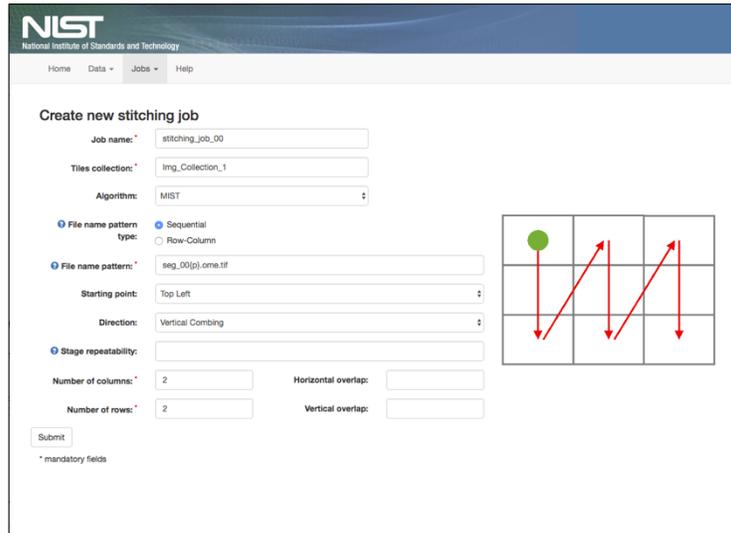
The screenshot shows the NIST National Institute of Standards and Technology website. The header includes the NIST logo and navigation links for Home, Data, Jobs, and Help. The main content area is titled "Create new filtering job" and contains three input fields: "Job name:" with the value "mask\_labelling\_job\_00", "Images collection:" with the value "Img\_Collection\_1", and "Connectedness:" with the value "Four connected". A "Submit" button is located below the fields. A note at the bottom left of the form states "\* mandatory fields".

This step is used to label mask images (such as the ones generated by segmentations jobs) for use by feature extraction software.

## Stitching

1. Go to Jobs > Stitching jobs
2. Click on "Create new job"
3. Give it a unique job name
4. Specify which image collection to use
5. Setup the parameters
  - MIST
  - Stage metadata

Must wait for tiles to be uploaded and converted to ome.tif before launching.  
Job name is used later to specify which stitching vector to use. Need to launch 1 stitching per collection.



The screenshot shows the NIST National Institute of Standards and Technology website interface for creating a new stitching job. The form includes the following fields and options:

- Job name:** stitching\_job\_00
- Tiles collection:** img\_Collection\_1
- Algorithm:** MIST
- File name pattern type:** Sequential (selected), Row-Column
- File name pattern:** seq\_00(p).ome.tif
- Starting point:** Top Left
- Direction:** Vertical Combing
- Stage repeatability:** (empty field)
- Number of columns:** 2
- Horizontal overlap:** (empty field)
- Number of rows:** 2
- Vertical overlap:** (empty field)

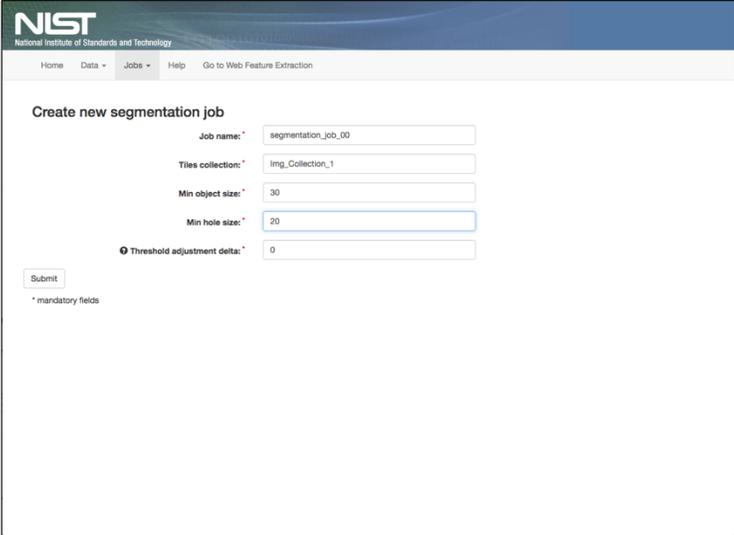
A diagram on the right side of the form illustrates the stitching process on a 2x3 grid. A green dot is located in the top-left cell. Red arrows indicate the stitching path: a vertical arrow pointing down from the green dot, a diagonal arrow pointing down-right to the bottom-right cell, a vertical arrow pointing down from the bottom-right cell, a diagonal arrow pointing down-left to the bottom-middle cell, a vertical arrow pointing down from the bottom-middle cell, and a diagonal arrow pointing down-left to the bottom-left cell.

Submit

\* mandatory fields

## Segmentation

1. Go to Jobs > Segmentation jobs
2. Click on "Create new job"
3. Give it a unique job name
4. Specify the image collection to segment
5. Specify EGT parameters



The screenshot shows the NIST National Institute of Standards and Technology website. The navigation bar includes Home, Data, Jobs, and Help, with a link to Go to Web Feature Extraction. The main content area is titled "Create new segmentation job" and contains the following form fields:

- Job name: segmentation\_job\_00
- Tiles collection: img\_collection\_1
- Min object size: 30
- Min hole size: 20
- Threshold adjustment delta: 0

A "Submit" button is located below the form fields. A note at the bottom left of the form area states "\* mandatory fields".

Only required if you want to flat-field correct images and view the segmentation mask in the final visualization.

Segmentation is done per image collection.

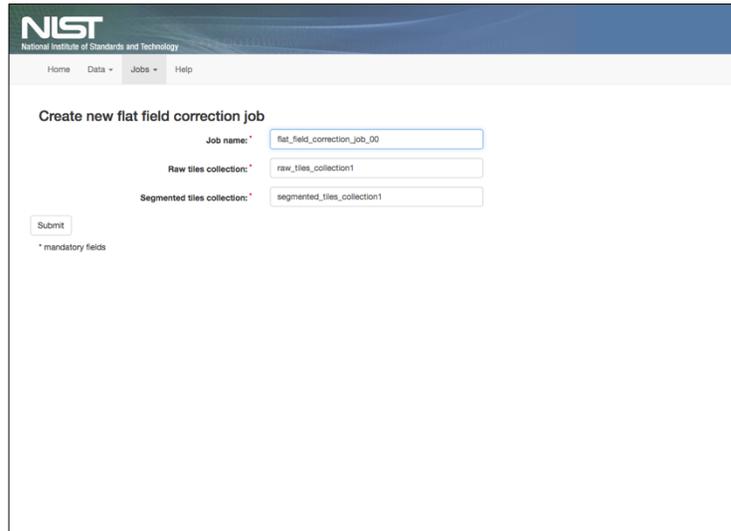
Segmentation job will create a new tile collection to hold the results. Collection will be named using the segmentation job name.

## Flat Field Correction

1. Go to Jobs > Flat field correction jobs
2. Click on "Create new job"
3. Give it a unique job name
4. Specify the Raw Images
5. Specify the Segmented Images

If you want to flat-field correct any image collection you must have previously generated segmentation masks for that collection. Job will create a new tile collection to hold the results.

If the tiles names of the selected raw and segmented tiles collection do not match, you will need to provide the patterns of each collection. The system will try to guess the patterns automatically but you should double check.



The screenshot shows the NIST National Institute of Standards and Technology website. The navigation menu includes Home, Data, Jobs, and Help. The main content area is titled "Create new flat field correction job". It contains three input fields, each with a red asterisk indicating it is a mandatory field:

- Job name:** flat\_field\_correction\_job\_00
- Raw tiles collection:** raw\_tiles\_collection1
- Segmented tiles collection:** segmented\_tiles\_collection1

Below the input fields is a "Submit" button and a note: "\* mandatory fields".

## Intensity Scaling

1. Go to Jobs > Intensity scaling jobs
2. Click on "Create new job"
3. Give it a unique job name
4. Specify the image collection
5. Specify re-scaling type. *Use truncation unless there is a compelling reason not to.*

Job will create a new tile collection to hold the results. Web browsers only support viewing 8bpp images. If no intensity rescaling is done the system will map the entire 16 bit range [0-65535] into the 8 bit range [0-255]. Each image collection you wish to view must be rescaled independently. There are two types of intensity scaling:

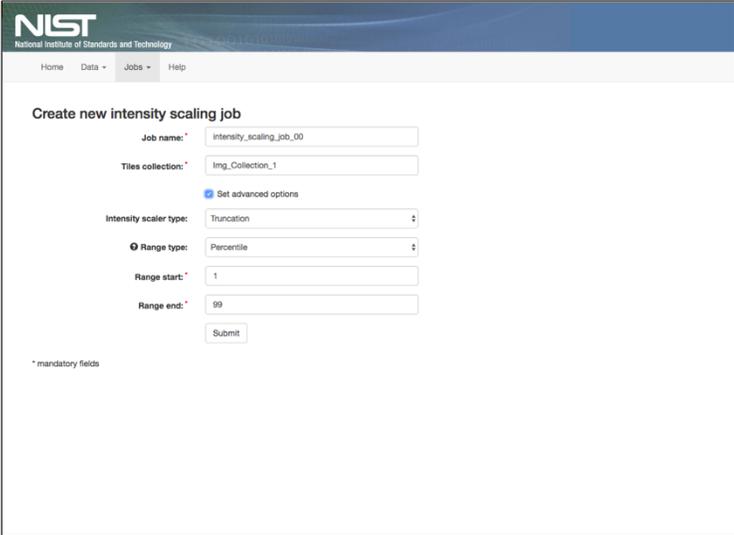
### Truncation (preferred)

By default will saturate the bottom and top 1% of intensities, linearly mapping intensities into that range. This is the same as Fiji/ImageJ auto-contrast.

If range start and end are specified those values are used instead of computing range start as 1st percentile and end as 99th percentile.

### Gamma Correction

This performs a non-linear (exponential) rescaling. By default start and end are 1st percentile and 99th percentile.



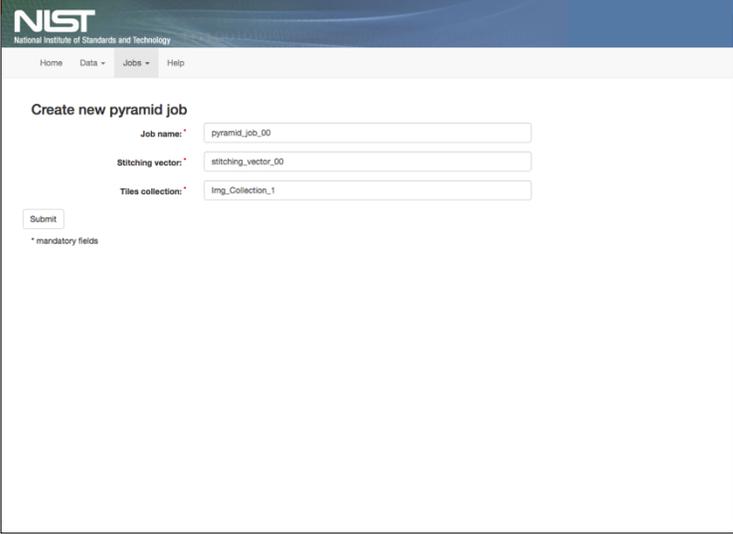
The screenshot shows the NIST National Institute of Standards and Technology website. The navigation bar includes Home, Data, Jobs, and Help. The main content area is titled "Create new intensity scaling job". It contains several input fields: "Job name:" with the value "intensity\_scaling\_job\_00", "Tiles collection:" with the value "Img\_Collection\_1", and a checked checkbox for "Set advanced options". Below this, there are three dropdown menus: "Intensity scaler type:" set to "Truncation", "Range type:" set to "Percentile", and "Range start:" set to "1". The "Range end:" field is set to "99". A "Submit" button is located at the bottom right of the form. A small asterisk and the text "\* mandatory fields" are visible at the bottom left of the form area.

## Pyramid Building

1. Go to Jobs > Pyramid jobs
2. Click on "Create new job"
3. Give it a unique job name
4. Specify the image collection
5. Specify the stitching vector. *The stitching vector must have been generated by a previously completed stitching job.*

Before images can be viewed, individual image tiles need to be converted into pyramids that can be viewed in the browser.

If tiles names in tiles collection and stitching vector differ, you must specify the tiles pattern. The system will try to guess the patterns automatically but you should double check.



The screenshot shows the NIST National Institute of Standards and Technology website. The navigation menu includes Home, Data, Jobs, and Help. The main content area is titled "Create new pyramid job" and contains three mandatory input fields: "Job name:" with the value "pyramid\_job\_00", "Stitching vector:" with the value "stitching\_vector\_00", and "Tiles collection:" with the value "img\_collection\_1". A "Submit" button is located below the fields. A note at the bottom left indicates "\* mandatory fields".

## Deep Zoom Visualization

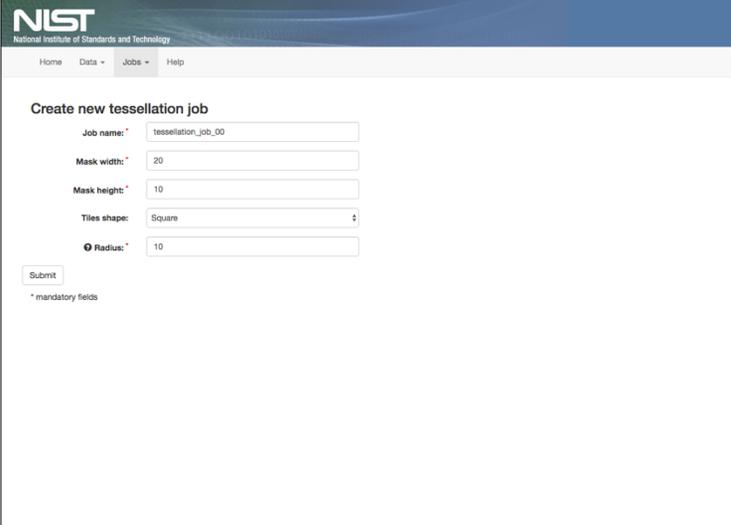
1. Go to Data > Visualization
2. Click on "Create new visualization"
3. Give it the visualization a unique name
4. At this point create one or more groups
5. Give the group a label and click "+"
6. Add layers (pyramids) to the group with "+". (*For example, add Transmitted, Excitation, Segmentation (if pyramid was built from the masks)*)

This step must be executed once all pyramids have been built. Multiple pyramids can be combined into a single visualization with multiple layers.

## Image Tessellation

1. Go to Jobs > Tessellation jobs
2. Click on "Create new job"
3. Give it a unique job name
4. Specify the mask width of the final tessellation image
5. Specify the mask height of the final tessellation image
6. Specify the shape in a tessellation (square or hexagon)
7. Specify the radius of the shape in a tessellation
  - For square shapes, the radius is one half of its width.
  - For hexagonal shapes, the radius is the distance between the center and the summit.

This step is independent of all other steps. It is executed to generate a mask subdividing a segmentation result. Tessellation masks are used together with segmentation masks to compute spatially local image features for studying spatial heterogeneity.



The screenshot shows the NIST National Institute of Standards and Technology website. The header includes the NIST logo and navigation links for Home, Data, Jobs, and Help. The main content area is titled "Create new tessellation job" and contains several input fields:

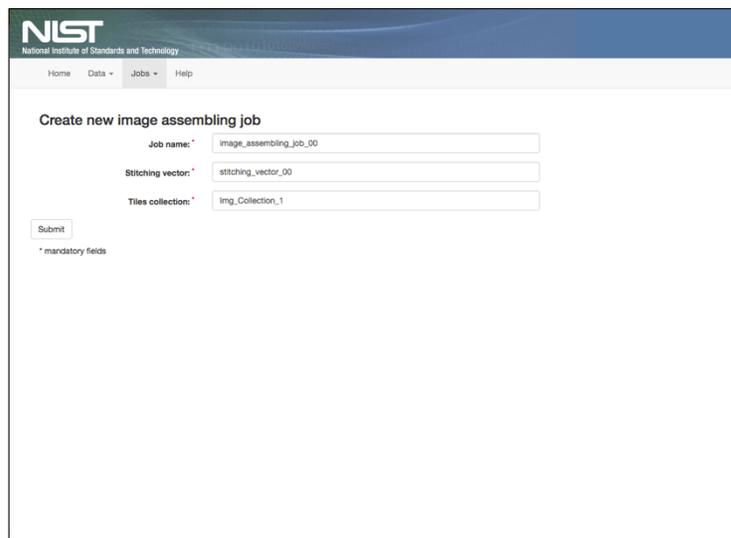
- Job name:** A text input field containing "tessellation\_job\_00".
- Mask width:** A text input field containing "20".
- Mask height:** A text input field containing "10".
- Tiles shape:** A dropdown menu with "Square" selected.
- Radius:** A text input field containing "10".

Below the input fields is a "Submit" button and a note: "\* mandatory fields".

## Image Assembling

1. Go to Jobs > Image Assembling jobs
2. Click on "Create new job"
3. Give it a unique job name
4. Specify the stitching vector
5. Specify the image tile collection

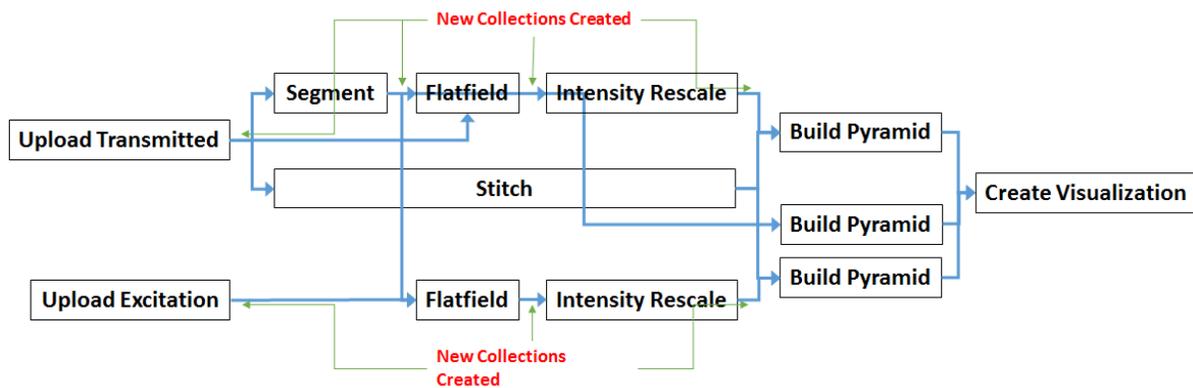
This step is similar to the pyramid building step but instead of building a pyramid, it builds big images. Those big images can then be used to compute features using the feature extraction software.



The screenshot shows the NIST National Institute of Standards and Technology website. The header includes the NIST logo and navigation links for Home, Data, Jobs, and Help. The main content area is titled "Create new image assembling job" and contains three mandatory input fields: "Job name:" with the value "image\_assembling\_job\_00", "Stitching vector:" with the value "stitching\_vector\_00", and "Tiles collection:" with the value "img\_Collection\_1". A "Submit" button is located below the fields, and a note indicates that the asterisk (\*) denotes mandatory fields.

## Example Workflow

The figure below illustrate possible workflows of computational steps that have to be executed in order to visually inspect two image collections of overlapping fields of views (FOV). The web image processing always includes stitching, pyramid building, and visualization creation. If the images have pixel depth more than 8 bits per pixel (BPP) then intensity rescaling has to be executed. Flat field correction and segmentation steps are optional. However, they are important for quantitative analyses.



***Example of a workflow using the Web Image Processing system to process two image collections (transmitted and excitation channels) acquired at the same time (the channels are registered).***