## 2 – Point processing

- <u>Image histogram</u>: open image **Oxford.tif** and type **h** (= *Analyze*>*Histogram*). Make sure you understand the meaning of the displayed parameters: Count, Mean, StDev, etc.
- Click on the buttons List, Log (logarithmic histogram) and Live (make a selection on the image and move it around; the histogram changes correspondingly).
- <u>Brightness & contrast adjustment</u>: Open image **Venice\_lc.tif** and then type **SHIFT** + **c** (= *Image>Adjust>Brightness/Contrast*).
- Adjust brightness or contrast automatically (Auto button); reset the image to its original view.
- Only the displayed intensities are changed, not the pixel values (check this with **h**); in order to change the pixel values, you should click on the Apply button.
- Adjustment works by displaying all intensity values lower than *min* as 0 (= black) and all intensity values higher than *max* as *Max intensity* (= white). Values between *min* and *max* are linearly mapped into values between 0 and *Max intensity*. In our case, auto adjustment mapping function is the following one:  $89 \dots 168 \rightarrow 0 \dots 255$ .



- For a non-linear input  $\rightarrow$  output values mapping you can use e.g. Gamma function (*Process>Math>Gamma*):  $V_{out} = V_{in}^{\gamma}$
- Experiment with shifting brightness or contrast sliders left or right; what is happening?
- The brightness adjustment simply adds or subtracts a constant value to every pixel leading to a histogram shift (left or right), but the distribution itself remains unchanged. Verify this with **h**.
- <u>Contrast enhancement</u>: use image **Venice\_lc.tif** again and select *Process>Enhance Contrast*.
- This dialog has several options:
  - Neither Normalize nor Equalize is selected: the display is adjusted in a way, that the given number of pixels becomes saturated; the intensity values in the image are not changed.

- Normalize is selected: a contrast stretching (normalization) is done. The result is very similar to that obtained with automatic brightness/contrast adjustment (see above); the intensity values are now changed.
- Equalize Histogram is selected: histogram equalization is performed; the other input in the dialog (Normalize) is ignored.
- <u>Arithmetic operations</u>: these can be performed via *Process>Math* for adding (subtracting, multiplying, etc.) a constant to each image pixel or *Process>Image Calculator* for adding (subtracting, multiplying, etc.) two images. Use the latter command for detecting differences between the images **SchoolObjects.tif** and **SchoolObjects\_2.tif** by subtracting the first one from the second one.
- <u>Intensity thresholding:</u> Open image **Blobs.tif**. To define a global threshold value, type **SHIFT** + t (= *Image*>*Adjust*>*Threshold*), choose one of the available algorithms and click on Apply. Thresholded image is now in a form suitable for *Particle analysis* (see <a href="https://sites.google.com/site/obdelavadigitalnihslik/primeri-uporabe/analiza-delcev">https://sites.google.com/site/obdelavadigitalnihslik/primeri-uporabe/analiza-delcev</a>)
- To see how severe is the effect of a particular thresholding method (i.e. the threshold value choice) on an (astronomical) image, select *Plugins*<*Examples*<*Macros*<*AutoThresholdingDemo*. These algorithms are accessible via *Image*<*Adjust*<*AutoThreshold*.
  - Global thresholding is not always possible (check with **h** (=*Analyze*>*Histogram*)) see the figure below!

Histogram = discrete frequency distribution, i.e. distribution of intensity values



Suitable for global thresholding

Adaptive thresholding needed

- Using **Oxford.tif** image, try various adaptive (local) thresholding algorithms: *Image* <*Adjust*<*Auto Threshold* (Web: <u>http://fiji.sc/wiki/index.php/Auto\_Local\_Threshold</u>)
- Note that here the thresholding operation is performed examining a (small) window around a particular pixel, e.g. 7x7 region, rather than the whole image.

## Literature

- Volker Baecker: Workshop Image processing and analysis with ImageJ and MRI Cell Image Analyzer. Web: <u>http://www.mri.cnrs.fr/datas/fichiers/articles/60/183.pdf</u>
- Computer Assisted Image Analysis: Course at the Centre for Image Analysis, Swedish University of Agricultural Sciences, Uppsala University. Web: http://www.it.uu.se/edu/course/homepage/bild1/vt08/schedule.html