Lecture 3

Point spread function, Convolution, Correlation, Filtering

E. Orlova, BBK

- Point spread function
- Concept of convolution
- Relation to Fourier transform
- Correlation
- Filtering
- Introduction to contrast transfer

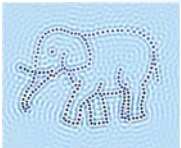


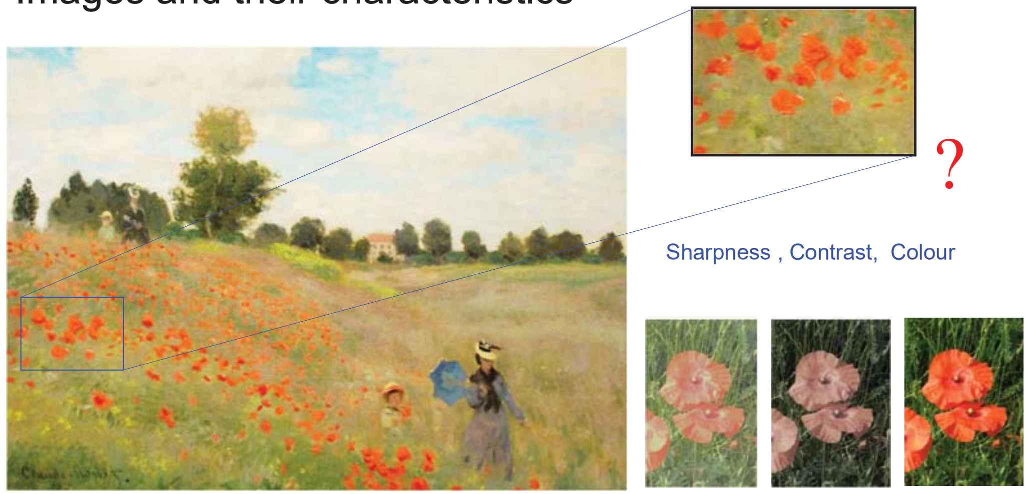
Image processing
for cryo microscopy

Sep 2 - Sept 12, 2019

Practical Course

Birkbeck College
London

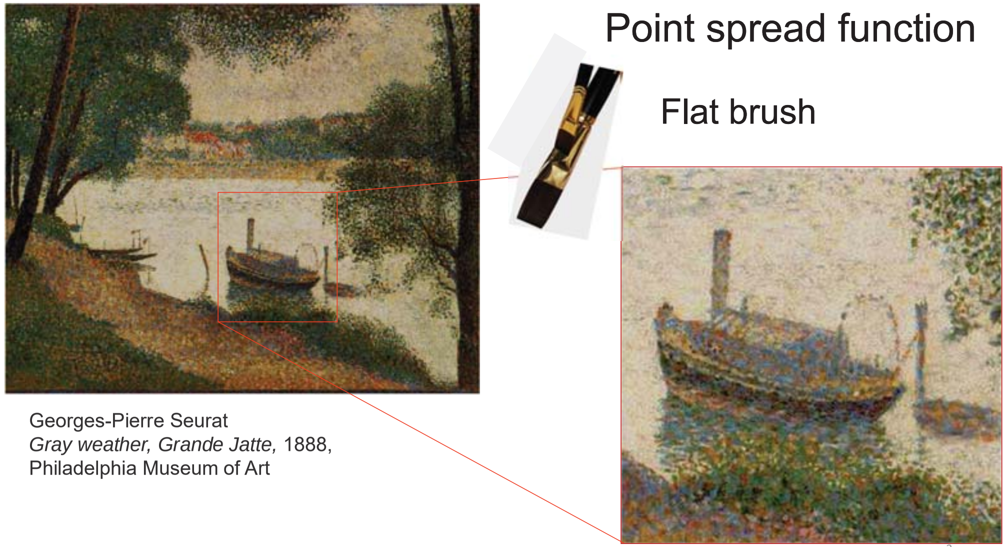
Images and their characteristics



Claude Monet, "Wild poppies", 1873, Musée d'Orsay

Point spread function

Flat brush



Georges-Pierre Seurat
Gray weather, Grande Jatte, 1888,
Philadelphia Museum of Art

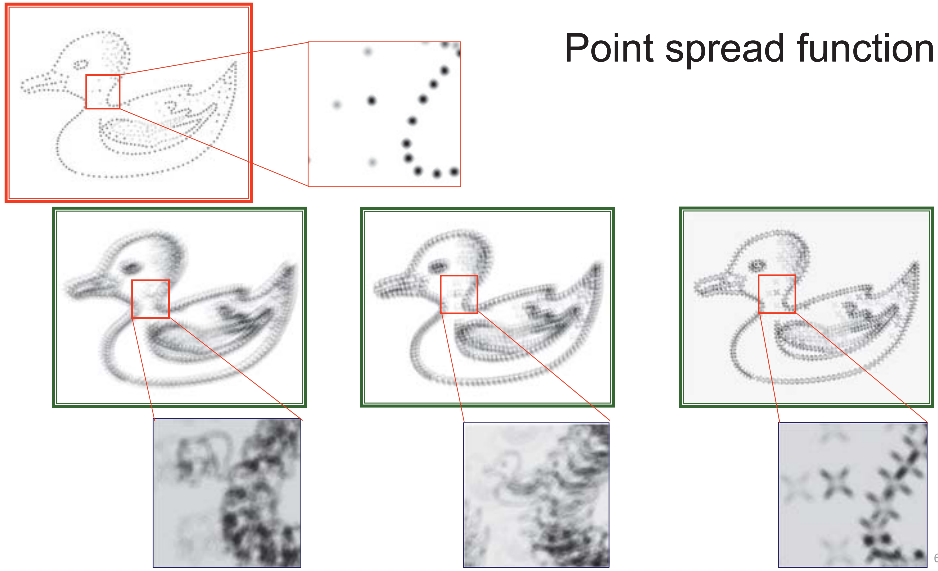
Point spread function

Thin pointed brush

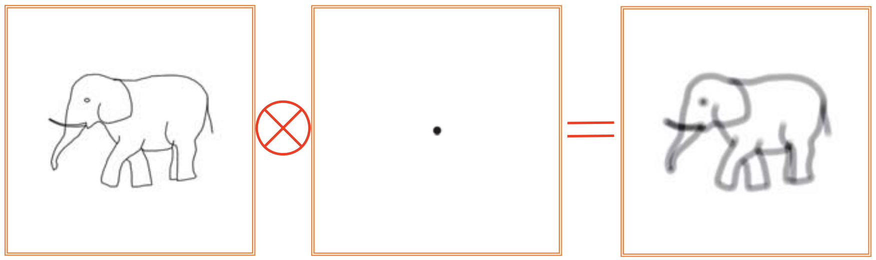
Pieter Bruegel The Elder, 1565
Winter Landscape with Skaters and Bird Trap,
Royal Museums of Fine Arts Belgium

Point spread function – a shape of the brush

Point spread function



Point spread function and convolution



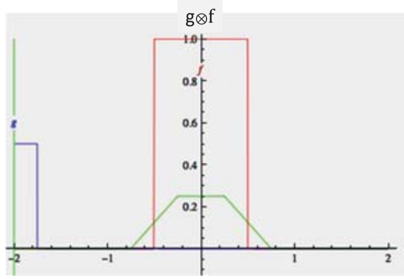
Convolution of two functions

Convolution of two functions *f(x)* and *h(x)* represents a distribution of the one function determined by the second one.

Concept of convolution

$$g(t) = \int h(x) f(t-x) dx = h(x) \otimes f(x)$$

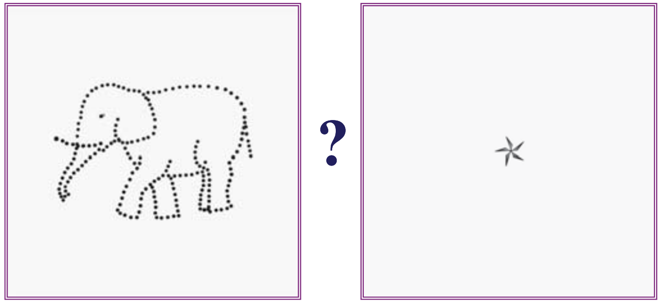
Rectangular functions



Convolution of two functions *f(x)* and *h(x)* represents a distribution of the one function determined by the second one.

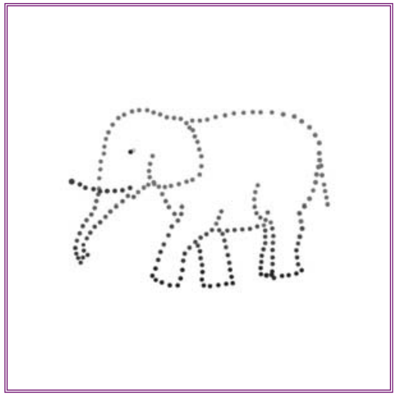
Convolution of two functions

Convolution can be calculated in the real space (space of the image) or using reciprocal space (Fourier space = Fourier transforms)

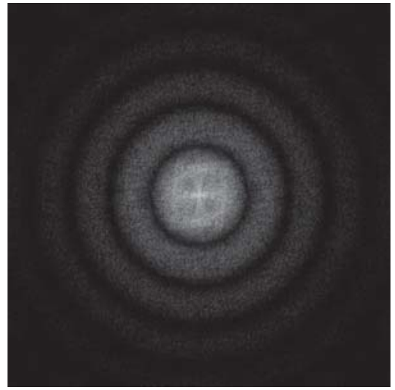


$$F[g(t)] = F[h(x) \otimes f(x)] = F[h(x)]F[f(x)]$$

Function
f (x)

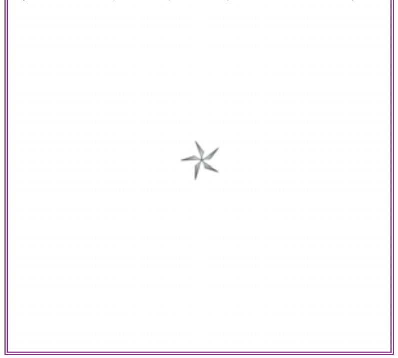


Fourier transform
F[f(x)]

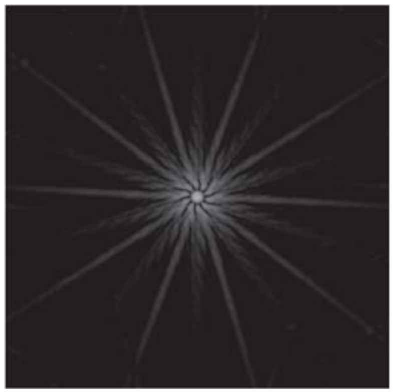


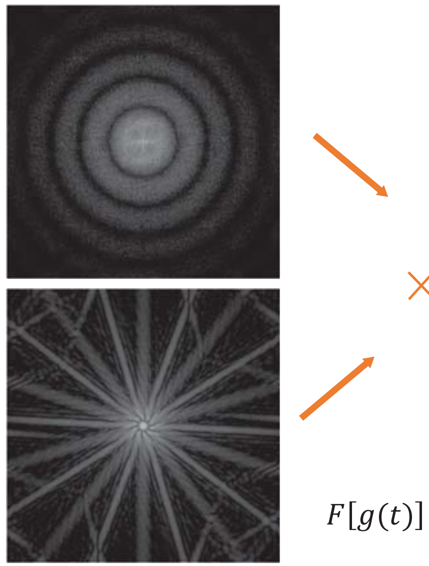
Function
h (x)

(brush shape = point spread function)

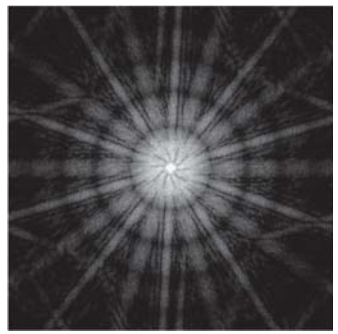


Fourier transform
F[h(x)]



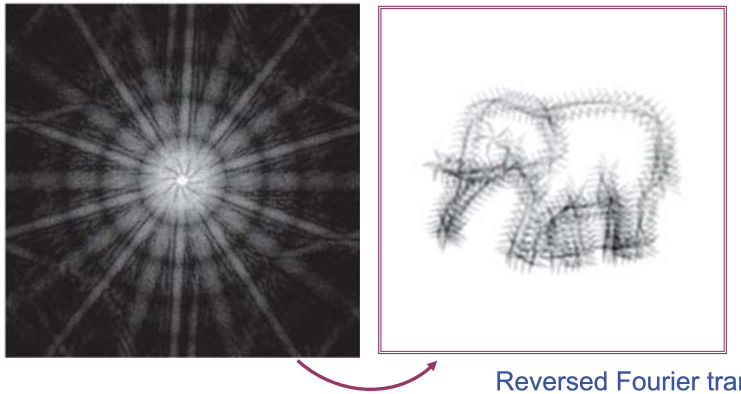


Product of Fourier transforms



$$F[g(t)] = F[h(x) \otimes f(x)] = F[h(x)]F[f(x)]$$

Convolution of two functions

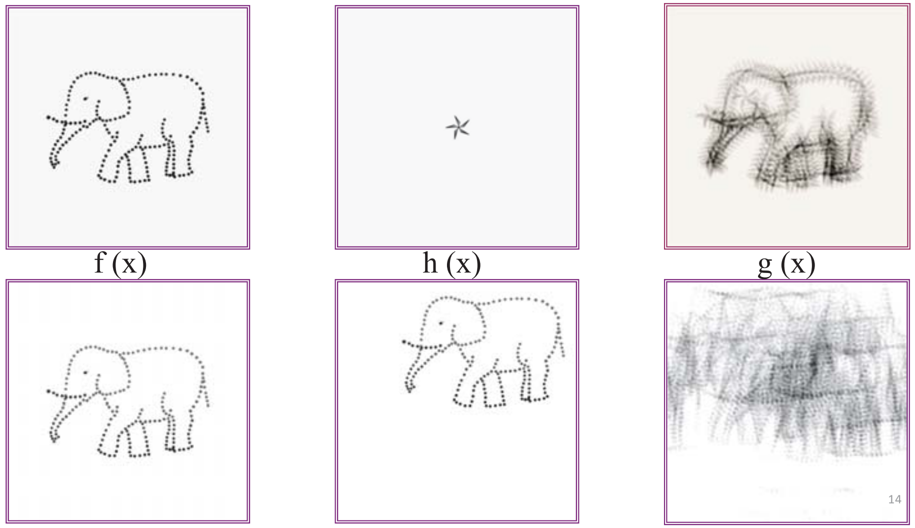


Reversed Fourier transformation

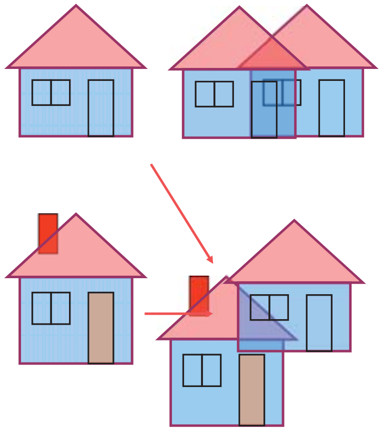
$$g(t) = h(x) \otimes f(x) = F\left[F[h(x)]F[f(x)]\right]^{-1}$$

Convolution of objects

(brush shape = point spread function)



Correlation of objects is a measure of their similarity



acf – auto correlation function

acf =1

ccf – cross correlation function

ccf < 1

Correlation function

$$ccf = \int g(x)h(x-t)dx$$

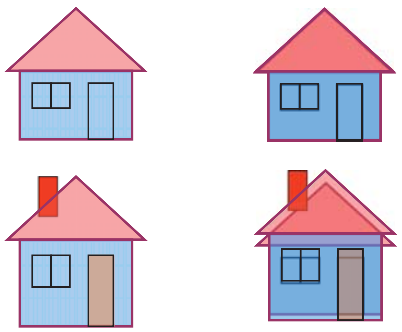
Auto-correlation function

$$acf = \int g(x)g(x-t)dx$$

Cross-correlation function (normalised)

$$ccf = \frac{\int g(x)h(x-t)dx}{\sqrt{\int g^2(x)dx \int h^2(x)dx}}$$

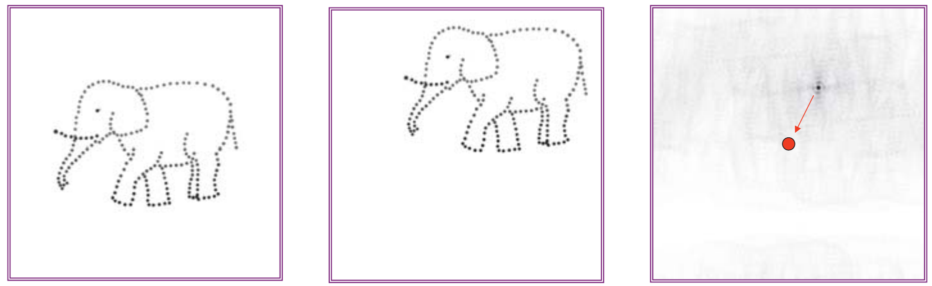
Correlation of objects is a measure of their similarity



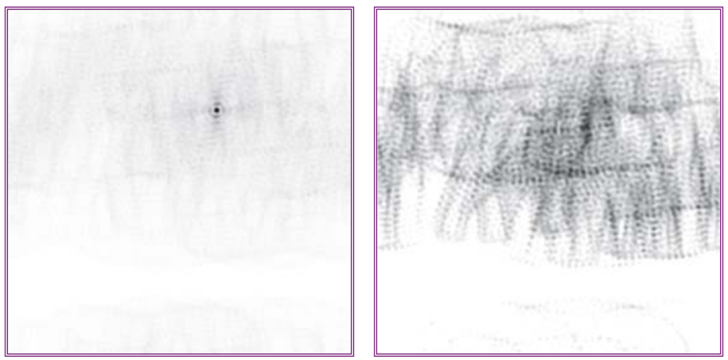
Fourier transform is used in cross-correlation analysis as a method of fast calculation. Calculation of the correlation function can be done using product of conjugate Fourier transforms .

Correlation of two objects

Position of the highest peak indicates on the shift required for the best alignment. The height of the peak shows a level of similarity



Correlation of objects   Convolution of objects



$$ccf = \int g(x)h(x-t)dx \qquad g(t) = \int g(x)h(t-x)dx$$

Correlation function

$$ccf = \int g(x)h(x-t)dx$$

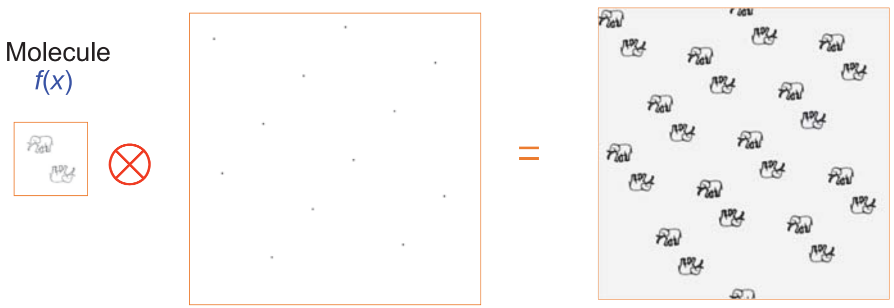
Auto-correlation function

$$acf = \int g(x)g(x-t)dx$$

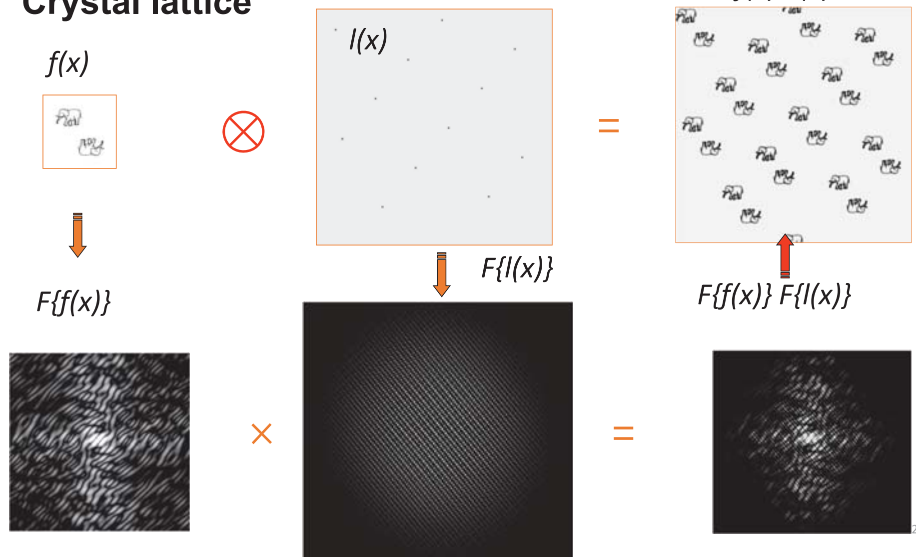
Cross-correlation function (normalised)

$$ccf = \frac{\int g(x)h(x-t)dx}{\sqrt{\int g^2(x)dx \int h^2(x)dx}}$$

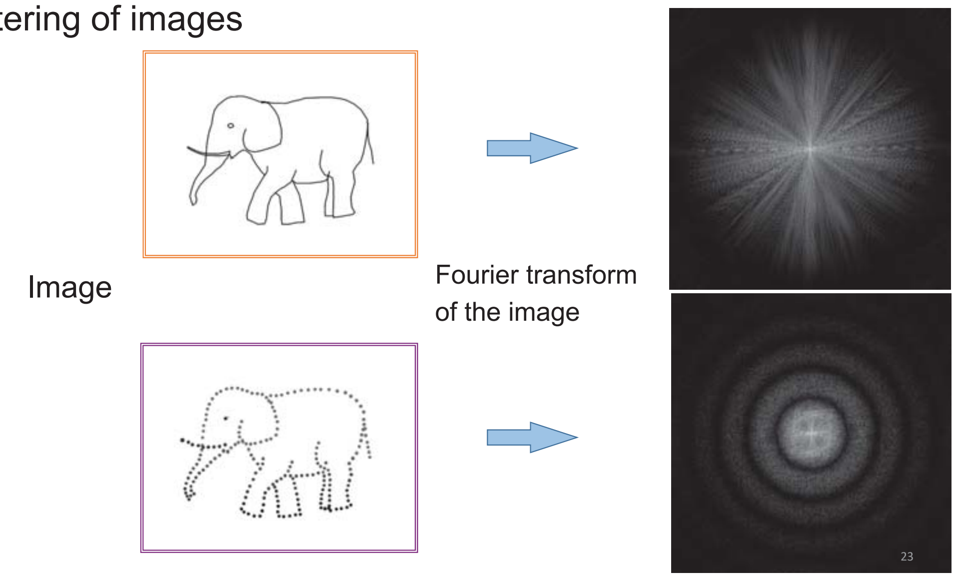
Convolution of a molecule $f(x)$ with a lattice $l(x)$ generates a crystal.

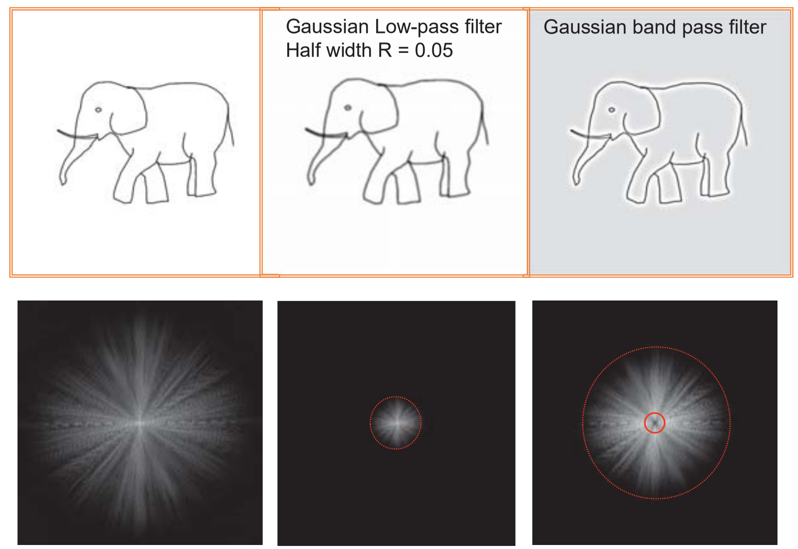


Crystal lattice

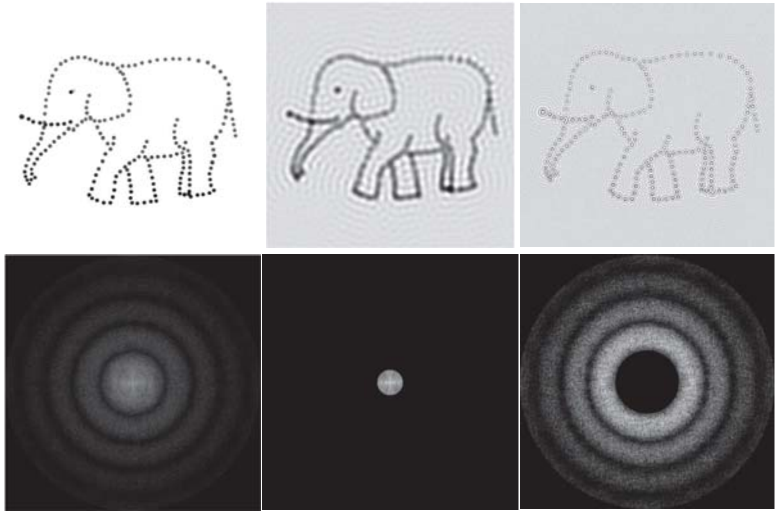


Filtering of images

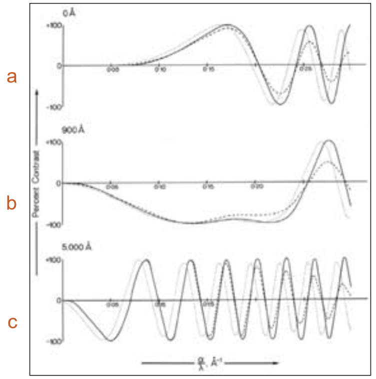




Unfiltered | Low pass filter | High pass filter



Contrast transfer function – CTF of the microscope



a - CTF at medium defocus
b - Scherzer focus, where the CTF is the most flat
c – CTF at large defocus

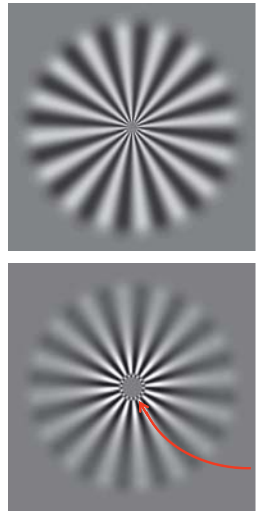
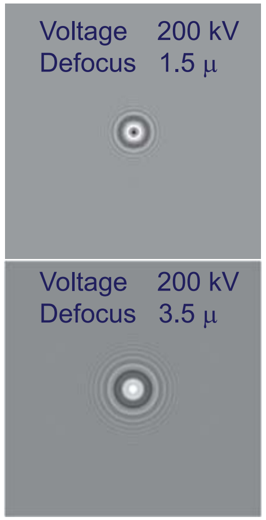


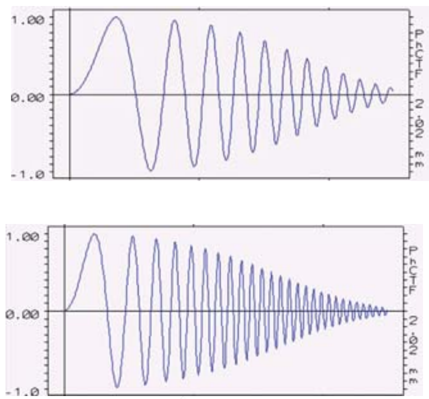
Image that is not affected by CTF

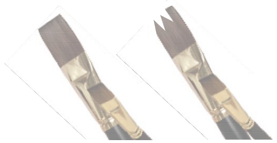
Image affected by CTF: Small details change the contrast

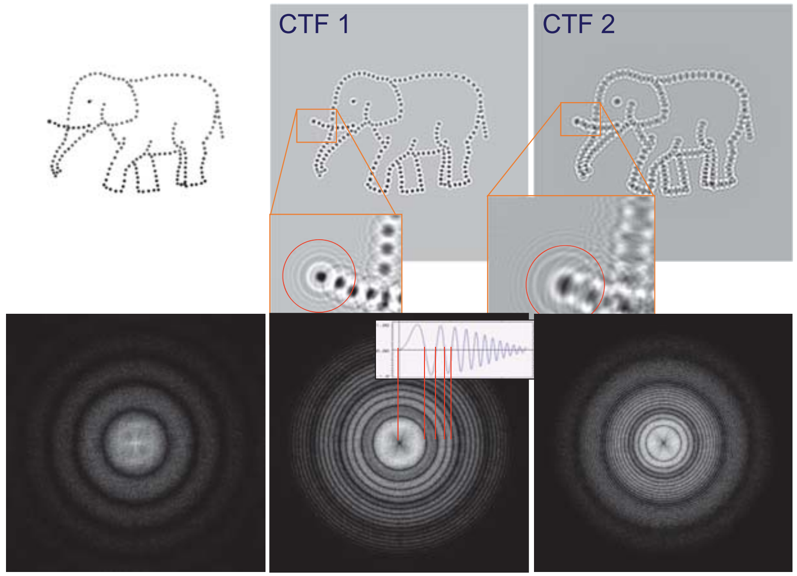
Point spread function



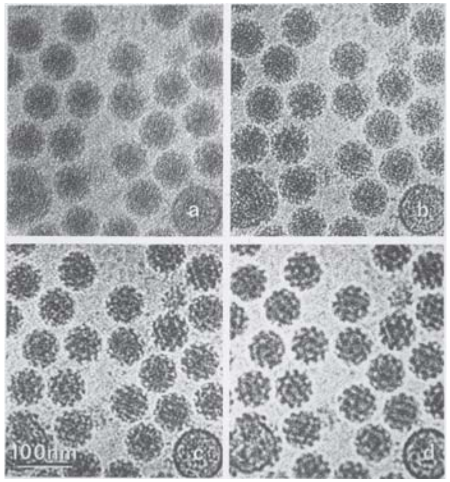
Contrast transfer function







Images at different defocus, Semliki Forest Virus



S. Fuller & B. Gowen, Heidelberg, F20

Reading list

http://fourier.eng.hmc.edu/e101/lectures/handout3/node2.html
https://en.wikipedia.org/wiki/Cross-correlation
https://en.wikipedia.org/wiki/Convolution_theorem

Frank, J. (2006), Three-Dimensional Electron Microscopy of Macromolecular Assemblies (2nd ed.), Oxford: Oxford University Press, ISBN 978-0195182187.

Huang Z., Baldwin P.R., Mullapudi S., Penczek P. (2003) Automated determination of parameters describing power spectra of micrograph images in electron microscopy. J. Struct. Biol. 144 79-94.

Mindell J.A., Grigorieff N. (1997) Accurate determination of local defocus and specimen tilt in electron microscopy. J. Struct. Biol. 142 334-347.

Reimer, L. (2008) Transmission Electron Microscopy – Physics Of Image Formation and Microanalysis. *Springer*: Berlin.

Satya P. Mallick, Bridget Carragher, Clinton S. Potter, David J. Kriegman (2005) ACE: Automated CTF estimation, Ultramicroscopy 104, 8-29.

Spence J.CH. (2003) High resolution microscopy. 3rd ed., Oxford University Press.