**SIMULATOR**

A simulation of fluorescently labeled particles imaged on a confocal microscope can be generated using simul8tr.m, which has the following syntax:

simulation = simul8tr(sizeX,sizeY,sizeT,density,bleachType,bleachDecay,qYield,pixelsize, timesize,PSFType,PSFSize,PSFZ,noBits,diffCoeff,flowX,flowY,flowZ, countingNoise,backgroundNoise);

where:

sizeX and sizeY are the dimensions of the simulation, in pixels.

density is the particle density in particles per um^2.

bleachType determines if the fluorophores bleach, and is either 'none' for no bleaching or'mono' for a monoexponential decay in average intensity.

bleachDecay determines how quickly particles bleach. It is the rate constant for the monoexponential bleaching, in the same units at the time step (usually between 0 and 0.05, given 1 Hz imaging rate).

qYield is the quantum yield of the fluorophores (usually 1).

pixelsize is the size of a pixel, in um (usually ~0.1).

timesize is the time between frames, in seconds (usually ~1).

PSFType gives the shape of the point spread function of the imaging system. It is either 'g'for a 2D Gaussian, or 'a' for an airy disk.

PSFSize is the e^-2 radius, in um, for a Gaussian PSF, or the distance to the first zero of the airy disk, in um.

PSFZ is the size, in um, for the Z dimension of the PSF. For 2D simulations, set this to 0. The PSF in Z is always a Gaussian, regardless of PSFType.

noBits is the number of bits used in the image normalization, imitating a A/D converter (usually 12).

diffCoeff is the diffusion coefficient, in um^2/s.

flowX, flowY, and flowZ are the flow speeds in each of the directions, in um/s.

countingNoise is the noise associated with the PMT amplification electronics (see the August 2005 Costantino BJ paper for details). Usually between 1 and 20.

backgroundNoise is the noise associated with spurious background counts (see the same paper). Usually between 0 and 0.3.

As an example, let's create a simulated image series with the following characteristics: 256 x 256 pixels with 100 images, 10 particles per um^2, 1 s per image, 0.1 um/pixel, particles with a quantum yield of 1, a Gaussian convolving function with an e^-2 radius of 0.4 um, with particles diffusing at 0.01 um^2/s, and no noise

imageSeriesDiff = simul8tr(256,256,100,10,'none',0,1,0.1,1,'g',0.4,0,12,0.01,0,0,0,0,0);

here is example of two species, monomer and tetramer, diffusing at different rates non flowing:

imageSeriesDiff = simul8tr(256,256,100,[1 0.1],'none',0,[1 4],0.1,1,'g',0.4,0,12,[0.1 0.01],[0 0],[0 0],[0 0],0,0);

Use ‘sv’ to display data:

sv(imageSeriesDiff,'c',5)

Flowing tetramer and diffusing monomer:

imageSeriesDiff = simul8tr(256,256,100,[1 0.1],'none',0,[1 4],0.1,1,'g',0.4,0,12,[0.1 0],[0 0.1],[0 0.1],[0 0],0,0);

Biased diffusion of tetramer and diffusing monomer:

imageSeriesDiff = simul8tr(256,256,100,[1 0.1],'none',0,[1 4],0.1,1,'g',0.4,0,12,[0.1 0.01],[0 0.1],[0 0.1],[0 0],0,0);