



## Planning an experiment? Using a microscopy facility? Creating a publication figure?

The Cellular Imaging Core is a little different from other SWEHSC Cores. We don't actually have a lab full of microscope equipment to call our own. We work closely with you while using and supporting existing University facilities. The difference is the personalized attention that we can give you every step of the way. Let us help you: plan your experiment, capture the images you need with one of the instruments available at the UA (*working with you or training you in how to use the equipment*), analyze the images, teach you how to best prepare the images for a publication or grant figure, and even help you write the methods section if needed. We have over 50 years worth of combined experience in microscopy, including light microscopes of many kinds as well as electron microscopy, image analysis and digital imaging. Access to core services like ours is one of the important benefits of being part of the SWEHSC. Contact us and find out how we can help you!

## Zeiss Multiphoton laser- a reminder

If you need to use this laser (e.g., to excite DAPI, Hoescht, Alexa 350, etc) during your confocal session, you must make arrangements with Doug Cromey ahead of time. Doug and Barb Carolus (*confocal facility manager*) are the only people allowed to turn on and tune the laser, so your scheduled time must be coordinated with their schedules. Users of this laser should be sure to read the list of safety rules that was recently posted in the microscope room.

## A highly recommended graduate class

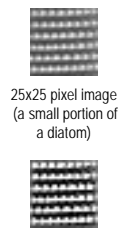
CBA 565A - Fundamentals of Light Microscopy and Electronic Imaging (3 units)

This is a lecture/hands-on course that teaches the essential principles and applications of light microscopy and electronic imaging. By the end of the course you will be able to use and adjust a research microscope for various modes of light microscopy. This class is taught by Dr. David Elliott, Director of the College of Medicine Research Microscopy Service Core. The class meets Wednesdays from 9am-11am; weekly lab exercises will be assigned.

Doug Cromey audited this class last year and rates it two thumbs up.

## How do software "filters" for digital images work?

Underlying the individual pixels in an image on your computer screen are numbers. Each pixel has a number representing its XY location in the matrix, and an intensity number that may be a single value for grayscale images, or three numbers representing the mix of red/green/blue used to create a specific color. Software filters in programs like ImageJ, Photoshop® or PhotoPaint® use mathematical functions called convolution kernels to create the changes we see in the image after the filter has been selected.



16	16	16	15	16	16	15	15	16	16	16	16	16	16	16	16	15	16	16	13	14	14	11	15		
16	35	39	10	25	48	13	42	19	13	38	12	47	6	14	23	5	17	18	12	24	30	19	29	12	
14	44	47	26	37	38	19	49	52	15	51	37	23	23	50	24	3	40	14	13	18	29	10	12	16	
16	4	33	19	14	48	14	22	55	22	27	37	28	20	52	35	36	37	17	12	35	10	1	12	16	
16	28	33	43	34	35	51	37	39	52	42	55	53	24	42	58	22	33	62	4	13	49	7	19	15	
17	42	53	44	17	43	25	11	40	12	0	33	8	12	20	18	22	20	10	21	14	50	15	15	10	
15	42	26	26	46	15	39	46	12	46	48	15	45	48	17	36	30	29	35	23	47	27	13	26	14	
17	9	15	21	1	15	31	11	21	34	16	25	64	17	54	56	21	50	59	40	36	50	23	0	15	
15	35	44	51	52	54	52	46	51	49	51	54	58	54	14	30	54	40	13	38	65	15	25	70	14	
16	1	56	15	26	54	20	72	42	58	61	38	24	20	23	36	30	30	25	9	48	20	18	12		
13	40	14	45	25	12	21	19	19	17	28	21	15	33	37	20	51	35	29	51	14	49	48	4	14	
16	17	18	9	15	23	8	1	16	12	32	8	27	40	20	32	62	36	61	56	52	65	64	25	16	
15	50	45	50	46	43	48	53	50	46	51	52	50	54	54	50	55	7	16	28	27	10	5	23	12	
16	15	64	7	10	59	29	29	19	49	47	3	50	16	7	42	3	17	26	34	26	48	28	5	9	
13	18	39	30	23	22	17	29	40	8	50	50	10	46	48	14	51	50	23	36	13	52	51	8	14	
15	35	5	28	28	4	34	8	10	5	38	7	18	67	11	16	65	37	35	59	52	67	60	30	15	
15	46	52	52	42	55	46	47	38	50	49	51	46	56	52	43	11	41	51	2	14	63	0	19	16	
15	56	11	28	66	13	40	48	27	47	29	68	12	42	19	68	31	29	10	8	25	25	3	24	3	
5	16	37	12	26	43	5	49	50	1	22	28	24	7	33	37	30	11	51	40	19	42	14	40	13	
15	10	25	19	8	22	10	11	9	8	23	15	26	30	60	63	29	68	61	50	67	57	61	51	16	
16	57	51	48	47	48	46	50	46	42	50	49	54	53	55	38	57	12	26	65	14	15	11	14		
15	43	24	31	30	32	26	54	23	13	58	30	7	44	17	15	34	2	32	19	8	46	26	7	6	
12	11	45	37	9	34	42	2	33	44	9	38	30	15	50	44	19	42	42	33	20	53	34	9	11	
15	33	8	1	29	8	0	21	28	1	2	47	4	13	64	11	27	64	35	46	61	29	22	27	16	
16	15	16	16	16	15	15	15	15	15	16	15	16	15	15	16	16	16	16	16	16	16	16	16	16	15

This array of numbers represents the difference in intensities between the 25x25 pixel image and the sharpened image (to the left of this figure) at each individual pixel. Note that some intensity values changed very little, while others were increased by over 60. This great a change in intensity represents roughly 25% of the dynamic range of the entire image (256 intensity levels, or 8 bit).

The convolution kernel adds or subtracts numbers from adjacent pixels to create the effect that the filter is named for. In the case of the sharpening filter used in this example, the effect of the filter is to enhance edges. To accentuate the edges, the filter increases the contrast in areas where there is a strong transition from dark to light.

Software filters are very powerful and can create unexpected artifacts in images. These artifacts can influence the interpretation of image data. As a general rule, we do not recommend the use of software filters on scientific digital images. **Remember that any image processing should always be done on a copy of the original image. Don't overwrite your data!** Any image processing (e.g., filters) that is performed on images in figures needs to be described in the figure legend or the methods section of a paper.