Nikon Microscope Parameters (Scope I)

Excitation Filters: anticlockwise, Lambda Wheel A

Filter 1 (Position 0): 420/20 nm -> 410 -430 nm CFP excitation
Filter 2 (Position 1): 546/11 nm mOrange1 excitation
Filter 3 (Position 2): 590/40 nm for Red Photoactivation (previously 340 nm Fura2 excitation).
Filter 4 (Position 3): 660/20 nm for Red Photo-deactivation (previously 380 nm Fura2 excitation 2, broken though).
Filter 5 (Position 4): 495/10x nm YFP excitation
Filter 6 (Position 5): 560/40 nm mCherry1 excitation
Filter 7 (Position 6): 515/10x nm mOrange2 excitation
Filter 8 (Position 7): 580/10 nm mCherry2 excitation
Filter 9 (Position 8): 465/30 nm blue activation
Filter 10 (Position 9): Open (future 760/20 nm red de-activation)

Emission Filters: anticlockwise, Lambda Wheel B

Filter 1 (Position 0): 480/40 nm CFP emission
Filter 2 (Position 1): 535/25 nm YFP emission
Filter 3 (Position 2): 535/40 nm Fura2 emission
Filter 4 (Position 3): 575/20 nm Orange2 emission
Filter 5 (Position 4): 630/20 nm Cherry1 emission
Filter 6 (Position 5): 650/100 nm Cherry2 emission
Filter 7 (Position 6): Closed
Filter 8 (Position 7): Closed
Filter 9 (Position 8): Closed
Filter 10 (Position 9): Open

Dichroic Mirrors:

Position 1: Analysis (DIC) - ANALY
Position 2: 455 dcxru (for CFP and C/Y FRET) – CFPHQ
Position 3: 510dclp (for YFP) – G2B
Position 4: 560 dcxr (for mOrange2 and O/C FRET) - YFPHQ
Position 5: GFP/FITC (full cube) –GFP-L
Position 6: RFP/Tritc (full cube) - TxRed
Backup Dichroic Mirror: 595dclp (for mCherry)

For photo-activation experiments, we switch either position 3 or position 6 to an enhanced silver mirror. We switch back to the original dichroic mirrors right after the experiment.

The arrows of filters should all facing the main body of the scope or dichroic mirror

Nikon Microscope Parameters (Scope II)
(Setting underscored if different from scope I)
Excitation Filters: anticlockwise, Lambda Wheel A

Filter 1 (Position 0): 420/*40* nm -> 400 -440 nm CFP excitation
Filter 2 (Position 1): 630/30 nm
Filter 3 (Position 2): open
Filter 4 (Position 3): 660/20 nm for Red Photo-deactivation.
Filter 5 (Position 4): 495/10x nm YFP excitation
Filter 6 (Position 5): open
Filter 7 (Position 6): 515/10x nm mOrange2 excitation
Filter 8 (Position 7): 580/*20* nm mCherry2 excitation
Filter 9 (Position 8): *470*/30 nm blue activation
Filter 10 (Position 9): *760/20 nm red de-activation*

Emission Filters: anticlockwise, Lambda Wheel B

Filter 1 (Position 0): 480/40 nm CFP emission
Filter 2 (Position 1): 535/*30* nm YFP/Fura2 emission
Filter 3 (Position 2): *open*
Filter 4 (Position 3): 575/20 nm Orange2 emission
Filter 5 (Position 4): 630/20 nm Cherry1 emission
Filter 6 (Position 5): 650/100 nm Cherry2 emission
Filter 7 (Position 6): 700/75 nm
Filter 8 (Position 7): Closed
Filter 9 (Position 8): Closed
Filter 10 (Position 9): Open

Dichroic Mirrors:

Position 1: Analysis (DIC) - ANALY
Position 2: 455 dcxru (for CFP and C/Y FRET) – 455LP (CFPHQ)
Position 3: *595dclp (for mCherry) – 595LP*
Position 4: 560 dcxr (for mOrange2 and O/C FRET) – 560LP (YFPHQ)
Position 5: GFP/FITC (full cube) –GFP-L - 49002
Position 6: RFP/Tritc (full cube) – TxRed - 49008
Backup Dichroic Mirror: silver enhance mirror reflecting > 360 nm (2x)
Backup Dichroic Mirror: T660 lpxrxt

Neutrodensity Filters: (Excitation ND Filter Wheel "C"

POS1 (F2) – 2.0 – 1% Excitation
POS2 (F3) – 1.0 – 10% Excitation
POS3 (F4) – 0.5 – 32%
POS4 (F5) – Open
POS5 (F10) – Open

For photo-activation experiments, we switch either position 3 or position 6 to the silver enhanced mirror. We switch back to the original dichroic mirrors right after the experiment.

The arrows of filters should all facing the main body of the scope or dichroic mirror (Updated on 12/13/2014)