

# High Performance Raman Spectroscopy

Raman spectroscopy allows for the detection and identification of molecules through their unique vibrational and rotational energy level structure. As opposed to fluorescence methods, which require the addition of a separate fluorescing molecule as a "tag" attached to the actual molecule of interest, Raman spectroscopy allows direct detection of a molecule with no chemical alteration. Another important difference, however, is that the scattered Raman signal (as a percentage of the excitation power) is several orders of magnitude weaker than the corresponding fluorescence signal. Because of this, lasers are typically used as excitation sources to provide high power in a tightly focused spot, and very sensitive detectors are used to detect very faint signal. Excellent filtering is therefore essential to block the very intense laser light while still allowing high transmission of the slightly wavelength-shifted Raman scattered signal.

Semrock stocks the widest selection of Ramanspectroscopy edge filters available, with edge wavelengths from 224 to 1550 nm. These filters are so steep and highly transmitting that they out perform even the leading holographic notch filters, yet are less than half the price. Now you can see the weakest signals closer to the laser line than before. With their deep laser-line blocking, ultra-wide and low-ripple passbands, proven hard-coating reliability, and high laser damage threshold, they offer performance that lasts. To prevent laser light from reaching the detector and drowning out the relatively weak Raman signal, we offer a collection of single-notch and multinotch filters, which block one or more laser lines while transmitting light on both sides. For the most discriminating Raman measurements, eliminate laser spectral noise leakage by cleaning up your laser spectrum with a matched MaxLine<sup>™</sup> laser clean-up filter.





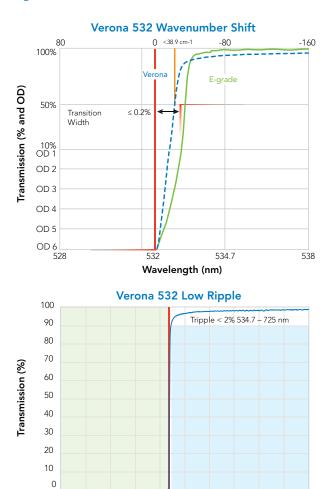


# Verona<sup>™</sup> Raman Filter Family

Semrock's long-pass Verona optical filters provide industrybest solutions for deep blocking close to the laser line for Raman applications. Verona is just the beginning of our evolving Raman product line to match the rapidly developing Raman market. Our understanding of how crucial flat transmission is for the maximal collection of weak Raman signals is why we offer a filter with both low ripple and improved steepness from our RazorEdge™ series, making the Verona family the new gold standard for Raman analysis.

Push the limits of what you can see with your Raman system with Verona Raman filters.

- Transition width\* ≤ 0.2% relative to the edge wavelength while still achieving transmission > 90% and blocking > OD6
- > Steepness improved from our RazorEdge series
- Low ripple to provide the best signal-to-noise ratio and allow for maximal collection of weak Raman spectral features
- > 785 nm, 532 nm, 633 nm, and 488 nm wavelengths are now available
- > Deep blocking of > OD6 at the laser line that eliminates bleed-through of excitation light
- > Hard coated for high laser damage threshold to reduce degradation in performance under ambient conditions
- > With our award-winning, proprietary KolaDeep™ spectral measurement system, we provide the spectral data to prove our superior deep blocking and edge-steepness performance
- > Available in 12.5 mm housed standard sized parts to meet your application needs



#### 504 516 528 540 552 564 576 588 600 Wavelength (nm)

#### 12.5 mm Diameter

Laser Line	Transition Width	Passband	Part Number	Price
785 nm	< 25.9 cm-1	788.9 – 1770.7 nm	VLP02-785-12.5	\$815
532 nm	< 38.9 cm-1	534.7 – 1300 nm	VLP01-532-12.5	\$815
633 nm	< 25.9 cm-1	636.2 - 1427.4 nm	VLP02-633-12.5	\$815
488 nm	< 38.8 cm-1	490.4 – 1100.8 nm	VLP01-488-12.5	\$815

480 492

### Verona Specifications

Property	Specifications	Comments
Edge Steepness (Typical)	0.2% of laser wavelength	Measured from OD = 6 to 50% transmission wavelength
Ripple on passband Transmission	< 2%	
Angle of Incidence	$0.0^{\circ} \pm 2.0^{\circ}$	Range for above optical specifications
Cone Half Angle	0°	Rays uniformly distributed about 0°
Clear Aperture	≥ 10 mm	
Outer Diameter	12.5 mm + 0.0 / – 0.1 mm	
Substrate Thickness	3.0 mm	
Mounted Thickness	5.0 mm	

### **Q** MEASURE DEEPER BLOCKING

## KolaDeep<sup>™</sup> Spectral Measurement System

New fluorescence-based life science and biomedical instrumentation increasingly requires high-performance optical filters with very high blocking (OD) and steep spectral edges that transition between high transmission and high OD. Our IDEX Health & Science Semrock engineers therefore developed the KolaDeep Spectral Measurement System (SMS), a proprietary award-winning platform for proven measurements of the steepest and deepest spectral features of our optical filters. This system is used in routine production to ensure that instruments made from these filters will deliver their intended performance.

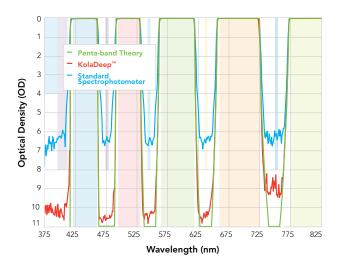
The platform's capabilities include:

- KolaDeep measures blocking to OD 8-9 across wavelength spans in the UV, visible and NIR ranges, and spectral features to OD 11
- KolaDeep resolves edges steeper than 0.2% relative to the edge wavelength from 90% transmission to OD > 7

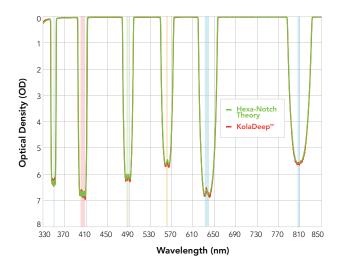
Our engineering team performed extensive qualification testing on KolaDeep to characterize and confirm the wavelength and photometric accuracy of the system

The recent white paper "Advanced Spectral Measurement Systems at IDEX Health & Science Semrock" presents examples and analyses of Semrock filters based on measurements by the KolaDeep SMS; a few are shown here

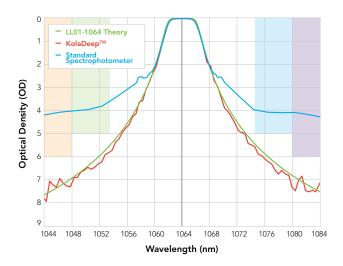
The graph below shows OD measured on a custom pentaband filter. The blocking between passbands is specified to exceed OD 8 to prevent excitation light leakage. Theory and measurement agree to OD > 8, with some blocking reaching from OD > 9 to the noise floor.



The graph below shows spectral performance for a custom hexa-notch filter designed to ensure precise blocking of six laser lines. This customer required assurance that each laser line is repeatably blocked at a specific level. The spectra measured with our KolaDeep SMS show precise matches to the specifications, even in the UV region. The measured blocking (red) matches theory (green).



The KolaDeep SMS performs well over the UV and NIR spectral range. An example is shown below for the 1064 nm Semrock MaxLine® laser-line cleanup filter. Measurements (red) confirm the required blocking and agree with the theoretical design (green) to OD > 7. Data (blue) from a superior commercially available system are not accurate beyond OD 3.



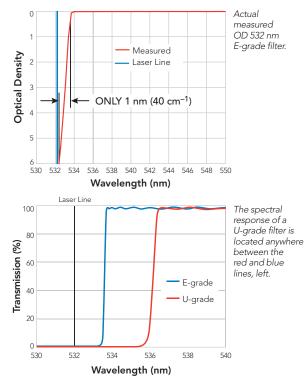
# RazorEdge® Long Wave Pass Raman Edge Filters

Semrock stocks an unsurpassed selection of the highest performance edge filters available for Raman Spectroscopy, with edge wavelengths from 224 to 1319 nm. Now you can see the weakest signals closer to the laser line than you ever have before. With their deep laser-line blocking, ultra-wide and low-ripple passbands, proven hard-coating reliability, and high laser damage threshold, they offer performance that lasts. U.S. Patent No. 7,068,430.

- > The steepest edge filters on the market RazorEdge E-grade filters, see how steep on page R-11
- For long-wave-pass edge filters and normal incidence, see right
- > For short-wave-pass edge filters and normal incidence, see page R-11
- > For ultrasteep 45° beamsplitters, see page R-7
- > For a suitably matched laser-line filter, see page R-5



### ACTUAL MEASURED DATA RAZOREDGE RAMAN FILTER SPECTRA



25 mm and 50 mm Diameters						
Laser Line	Transition Width1	Passband	Part Number	Price		
224.3 nm	< 1920 cm–1	235.0-505.9 nm	LP02-224R-25	\$1,230		
244 nm	< 498 cm–1	247.6-550.4 nm	LP02-244RS-25	\$1,230		
248.6 nm	< 805 cm–1	261.0-560.8 nm	LP02-248RS-25	\$1,230		
257.3 nm	< 385 cm–1	263.0-580.4 nm	LP02-257RU-25	\$1,230		
266.0 nm	< 372 cm–1	272.4-600.0 nm	LP02-266RU-25	\$1,095		
325.0 nm	< 153 cm–1	327.1-733.1 nm	LP03-325RE-25	\$1,095		
	< 305 cm–1	329.2-733.1 nm	LP03-325RU-25	\$745		
355.0 nm	< 140 cm–1	357.3-800.8 nm	LP02-355RE-25	\$1,230		
	< 279 cm–1	359.6-800.8 nm	LP02-355RU-25	\$745		
363.8 nm	< 272 cm–1	368.5-820.6 nm	LP02-364RU-25	\$850		
407.0 nm	< 243 cm–1	412.3-918.0 nm	LP02-407RU-25	\$745		
441.6 nm	< 113 cm–1	444.5-996.1 nm	LP02-442RE-25	\$1,230		
	< 224 cm–1	447.3-996.1 nm	LP02-442RU-25	\$850		
457.9 nm	< 109 cm–1	460.9-1032.9 nm	LP03-458RE-25	\$1,230		
	< 216 cm–1	463.9-1032.9 nm	LP03-458RU-25	\$745		
473.0 nm	< 105 cm–1	476.1-1066.9 nm	LP02-473RE-25	\$1,095		
	< 209 cm–1	479.1-1066.9 nm	LP02-473RU-25	\$745		
488.0 nm	< 102 cm–1	491.2-1100.8 nm	LP02-488RE-25	\$1,095		
	< 203 cm–1	494.3-1100.8 nm	LP02-488RU-25	\$745		
514.5 nm	< 97 cm–1	517.8-1160.5 nm	LP02-514RE-25	\$1,230		
	< 192 cm–1	521.2-1160.5 nm	LP02-514RU-25	\$850		
532.0 nm	< 90 cm–1	535.4-1200.0 nm	LP03-532RE-25	\$1,095		
	< 186 cm–1	538.9-1200.0 nm	LP03-532RU-25	\$745		
561.4 nm	< 89 cm–1	565.0-1266.3 nm	LP02-561RE-25	\$1,230		
	< 176 cm–1	568.7-1266.3 nm	LP02-561RU-25	\$850		
632.8 nm	< 79 cm–1	636.9-1427.4 nm	LP02-633RE-25	\$1,095		
	< 156 cm–1	641.0-1427.4 nm	LP02-633RU-25	\$745		
638 nm	< 78 cm-1	642.1-1439.1 nm	LP02-638RE-25	\$1,230		
	< 155 cm-1	646.3-1439.1 nm	LP02-638RU-25	\$850		
647.1 nm	< 153 cm–1	655.5-1459.6 nm	LP02-647RU-25	\$850		
664.0 nm	< 149 cm–1	672.6-1497.7 nm	LP02-664RU-25	\$850		
671.0 nm	< 147.6 cm–1	679.7-1513.5 nm	LP02-671RU-25	\$745		
780.0 nm	< 127 cm–1	790.1-1759.4 nm	LP02-780RU-25	\$850		
785.0 nm	< 63 cm–1	790.1-1770.7 nm	LP02-785RE-25	\$1,095		
	< 126 cm–1	795.2-1770.7 nm	LP02-785RU-25	\$745		
808.0 nm	< 62 cm–1	813.3-1822.6 nm	LP02-808RE-25	\$1,230		
	< 123 cm–1	818.5-1822.6 nm	LP02-808RU-25	\$850		
830.0 nm	< 60 cm–1	835.4-1872.2 nm	LP02-830RE-25	\$1,230		
	< 119 cm–1	840.8-1872.2 nm	LP02-830RU-25	\$745		
980.0 nm	< 51 cm–1	986.4-2000.0 nm	LP02-980RE-25	\$1,230		
	< 101 cm–1	992.7-2000.0 nm	LP02-980RU-25	\$745		
1064.0	< 47 cm–1	1070.9-2000.0 nm	LP02-1064RE-25	\$1,230		
nm	< 93 cm–1	1077.8-2000.0 nm	LP02-1064RU-25	\$745		
50 mm LWF	<sup>D</sup> Edge Filters		idex-hs.com/sem	rock		

# MaxLine® Laser-line Filters

Semrock MaxLine Laser-line Filters have an unprecedented high transmission exceeding 90% at the laser line, while rapidly rolling off to an optical density (OD) > 5 at wavelengths differing by only 1% from the laser wavelength, and OD > 6 at wavelengths differing by only 1.5% from the laser wavelength. U.S. Patent No. 7,119,960.

- Highest laser-line transmission stop wasting expensive laser light
- Steepest edges perfect match to RazorEdge® U-grade filters (see page R-8)
- > Ideal complement to StopLine® deep notch filters for fluorescence and other applications
- > Hard dielectric coatings for proven reliability and durability
- For diode lasers, use our MaxDiode™ Laser Clean-up filters



#### Ultraviolet

Wavelength	Guaranteed Transmission	Typical Bandwidth	OD 5 Blue Range (nm)	OD 6 Blue Range (nm)	OD 6 Red Range (nm)	OD 5 Red Range (nm)	12.5 mm Diameter Part Number	25 mm Diameter Part Number
248.6 nm	> 40%	1.7 nm	228.2-246.1	228.7-244.9	252.3-273.5	251.1-279.9	LL01-248-12.5	LL01-248-25
266.0 nm	> 55%	1.9 nm	242.8-263.3	244.7-262.0	270.0-292.6	268.7-302.2	LL01-266-12.5	LL01-266-25
280.0 nm	> 60%	1.5 nm	254.4-277.2	257.6-275.8	282.8-320.4	284.2-308	LL01-280-12.5	LL01-280-25
320.0 nm	> 70%	1.3 nm	286.9-316.8	294.4-315.2	323.2-373.8	324.8-352	LL01-320-12.5	LL01-320-25
325.0 nm	> 80%	1.2 nm	291.0-321.8	299.0-320.1	329.9-357.5	328.3-380.7	LL01-325-12.5	LL01-325-25
355.0 nm	> 80%	1.3 nm	314.8-351.5	326.6-349.7	360.3-390.5	358.6-422.5	LL01-355-12.5	LL01-355-25
360.0 nm	> 85%	1.1 nm	318.7-356.4	331.2-354.6	363.6-429.6	365.4-396	LL01-360-12.5	LL01-360-25

### Visible

Wavelength	Guaranteed Transmission	Typical Bandwidth	OD 5 Blue Range (nm)	OD 6 Blue Range (nm)	OD 6 Red Range (nm)	OD 5 Red Range (nm)	12.5 mm Diameter Part Number	25 mm Diameter Part Number
405.0 nm	> 90%	1.5 nm	353.5-401.0	372.6-398.9	411.1-445.5	409.1-495.3	LL01-405-12.5	LL01-405-25
407.0 nm	> 90%	1.5 nm	355.0-402.9	374.4-400.9	413.1-447.7	411.1-498.3	LL01-407-12.5	LL01-407-25
441.6 nm	> 90%	1.7 nm	381.0-437.2	406.3-435.0	448.2-485.8	446.0-551.1	LL01-442-12.5	LL01-442-25
457.9 nm	> 90%	1.7 nm	393.1-453.3	421.3-451.0	464.8-503.7	462.5-576.7	LL01-458-12.5	LL01-458-25
473.0 nm	> 90%	1.8 nm	404.2-468.3	435.2-465.9	480.1-520.3	477.7-600.9	LL01-473-12.5	LL01-473-25
488.0 nm	> 90%	1.9 nm	415.1-483.1	449.0-480.7	495.3-536.8	492.9-625.3	LL01-488-12.5	LL01-488-25
514.5 nm	> 90%	2.0 nm	434.1-509.4	473.3-506.8	522.2-566.0	519.6-669.5	LL01-514-12.5	LL01-514-25
532.0 nm	> 90%	2.0 nm	446.5-526.7	489.4-524.0	540.0-585.2	537.3-699.4	LL01-532-12.5	LL01-532-25
543.5 nm	> 90%	2.1 nm	454.6-538.1	500.0-535.3	551.7-597.9	548.9-719.5	LL01-543-12.5	LL01-543-25
561.4 nm	> 90%	2.1 nm	467.0-555.8	516.5-553.0	569.8-617.5	567.0-751.2	LL02-561-12.5	LL02-561-25
568.2 nm	> 90%	2.2 nm	471.7-562.5	522.7-559.7	576.7-625.0	573.9-763.4	LL01-568-12.5	LL01-568-25
632.8 nm	> 90%	2.4 nm	515.4-626.5	582.2-623.3	642.3-696.1	639.1-884.7	LL01-633-12.5	LL01-633-25
638.0 nm	> 90%	2.4 nm	518.8-631.6	587-628.4	647.6-701.8	644.4-894.9	LL01-638-12.5	LL01-638-25
647.1 nm	> 90%	2.5 nm	524.8-640.6	595.3-637.4	656.8-711.8	653.6-912.9	LL01-647-12.5	LL01-647-25
671.0 nm	> 90%	2.6 nm	540.4-664.3	617.3-660.9	681.1-738.1	677.7-961.2	LL01-671-12.5	LL01-671-25

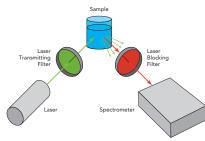
#### **Near-Infrared**

Wavelength	Guaranteed Transmission	Typical Bandwidth	OD 5 Blue Range (nm)	OD 6 Blue Range (nm)	OD 6 Red Range (nm)	OD 5 Red Range (nm)	12.5 mm Diameter Part Number	25 mm Diameter Part Number
780.0 nm	> 90%	3.0 nm	609.0-772.2	717.6-768.3	791.7-858.0	787.8-1201.8	LL01-780-12.5	LL01-780-25
785.0 nm	> 90%	3.0 nm	612.0-777.2	722.2-773.2	796.8-863.5	792.9-1213.8	LL01-785-12.5	LL01-785-25
808.0 nm	> 90%	3.1 nm	625.9-799.9	743.4-795.9	820.1-888.8	816.1-1033.5	LL01-808-12.5	LL01-808-25
810.0 nm	> 90%	3.1 nm	627.1-801.9	745.2-797.9	822.2-891.0	818.1-1143.4	LL01-810-12.5	LL01-810-25
830.0 nm	> 90%	3.2 nm	639.1-821.7	763.6-817.6	842.5-913.0	838.3-1067.9	LL01-830-12.5	LL01-830-25
852.0 nm	> 90%	3.2 nm	652-843.5	783.8-839.2	864.8-937.2	860.5-1106.6	LL01-852-12.5	LL01-852-25
976.0 nm	> 90%	3.7 nm	722.2-966.2	897.9-961.4	990.6-1073.6	985.8-1325.2	LL01-976-12.5	LL01-976-25
980.0 nm	> 90%	3.7 nm	724.4-970.2	901.6-965.3	994.7-1078.0	989.8-1332.6	LL01-980-12.5	LL01-980-25
1030.0 nm	> 90%	3.9 nm	1014.6-1019.7	947.6-1014.6	1045.5-1133	1040.3-1368.2	LL01-1030-12.5	LL01-1030-25
1047.1 nm	> 90%	4.0 nm	963.3-1036.6	963.3-1031.4	1062.8-1151.8	1057.6-1398.6	LL01-1047-12.5	LL01-1047-25
1064.0 nm	> 90%	4.0 nm	978.9-1053.4	978.9-1048.0	1080.0-1170.4	1074.6-1428.9	LL01-1064-12.5	LL01-1064-25
Price	Visit idex-hs.c	om/semrock t	to Search the Ma	axLine Filter Far	nily			

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# **TECHNICAL NOTE**Filter Types for Raman Spectroscopy Applications

Raman spectroscopy is widely used today for applications ranging from industrial process control to laboratory research to bio/ chemical defense measures. Industries that benefit from this highly



specific analysis technique include the chemical, polymer, pharmaceutical, semiconductor, gemology, computer hard disk, and medical fields. In Raman spectroscopy, an intense laser beam is used to create Raman (inelastic) scattered light from a sample under test. The Raman "finger print" is measured by a dispersive or Fourier Transform spectrometer.

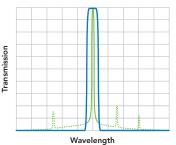
There are three basic types of Raman instrumentation. Raman microscopes, also called micro-Raman spectrophotometers, are larger-scale laboratory analytical instruments for making fast, high-accuracy Raman measurements on very small, specific sample areas. Traditional laboratory Raman spectrometers are primarily used for R&D applications, and range from "home-built" to flexible commercial systems that offer a variety of laser sources, means for holding solid and liquid samples, and different filter and spectrometer types. Finally, a rapidly emerging class of Raman instrumentation is the Raman micro-probe analyzer. These complete, compact and often portable systems are ideal for use in the field or in tight manufacturing and process environments. They utilize a remote probe tip that contains optical filters and lenses, connected to the main unit via optical fiber.

Optical filters are critical components in Raman spectroscopy systems to prevent all undesired light from reaching the spectrometer and swamping the relatively weak Raman signal. Laser Transmitting Filters inserted between the laser and the sample block all undesired light from the laser (such as broadband spontaneous emission or plasma lines) as well as any Raman scattering or fluorescence generated between the laser and the sample (as in a fiber micro-probe system). Laser Blocking Filters inserted between the sample and the spectrometer block the Rayleigh (elastic) scattered light at the laser wavelength.

The illustration above shows a common system layout in which the Raman emission is collected along a separate optical path from the laser excitation path. Systems designed for imaging (e.g., Raman microscopy systems) or with remote fiber probes are often laid out with the excitation and emission paths coincident, so that both may take advantage of the same fiber and lenses (see Technical Note on page R-13).

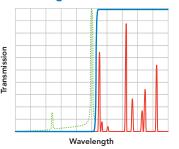
There are three basic types of filters used in systems with separate excitation and emission paths: Laser-line filters, Edge Filters, and Notch Filters. The examples, above right, show how the various filters are used. In these graphs the blue lines represent the filter transmission spectra, the green lines represent the laser spectrum, and the red lines

#### Laser-line Filter



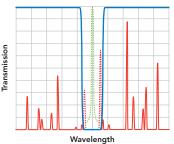
Laser-transmitting filter for both Stokes and Anti-Stokes measurements.

#### LWP Edge Filter



Laser-blocking steep edge filter for superior Stokes measurements.

#### **Notch Filter**



Versatile laser-blocking notch

filter for both Stokes and Anti-Stokes measurements.

as steep as the edge filter or laser-line filter, the ability to get as close to the laser line as those filters allow is lost.

Semrock manufactures high-performance MaxLine® Laserline filters (page R-5), RazorEdge® long-wave-pass and short-wave-pass filters (page R-4), EdgeBasic<sup>™</sup> value longwave-pass filters (page R-8), ultrasteep RazorEdge Dichroic<sup>™</sup> beamsplitter filters (page R-7), and StopLine® notch filters as standard catalog products. Non-standard wavelengths and specifications for these filters are routinely manufactured for volume OEM applications.

represent the Raman signal (not to scale).

Laser-line Filters are ideal for use as Laser Transmitting Filters, and Notch Filters are an obvious choice for Laser Blocking Filters. In systems using these two filter types, both Stokes and Anti-Stokes Raman scattering can be measured simultaneously. However, in many cases Edge Filters provide a superior alternative to notch filters. For example, a long-wave-pass (LWP) Edge Filter used as a Laser Blocking Filter for measuring Stokes scattering offers better transmission, higher laser-line blocking, and the steepest edge performance to see Raman signals extremely close to the laser line. For more details on choosing between edge filters and notch filters, see the Technical Note "Edge Filters vs. Notch Filters for Raman Instrumentation" on page R-12.

In systems with a common excitation and emission path, the laser must be introduced into the path with an optic that also allows the Raman emission to be transmitted to the detection system. A 45° dichroic beamsplitter is needed in this case. If this beamsplitter is not

# RazorEdge® Dichroic<sup>™</sup> Beamsplitters

The unique RazorEdge Dichroic beamsplitters exhibit unparalleled performance. Each filter reflects a standard laser line incident at  $45^{\circ}$  while efficiently passing the longer Raman-shifted wavelengths. They exhibit ultrasteep transition from reflection to transmission, far superior to anything else available on the open market. The guaranteed transition width of < 1% of the laser wavelength for U-grade (regardless of polarization) makes these filters a perfect match to our popular normal-incidence RazorEdge ultrasteep long-wave-pass filters. These beamsplitters are so innovative that they are patent pending.



#### Available as Either Mounted in 25 mm Diameter x 3.5 mm Thick Black-Anodized Aluminum Ring or Unmounted as 25.2 x 35.6 x 1.1 mm or 25.2 x 35.6 x 2.0 mm

Laser Line	Transition Width	Passband	25 mm Mounted Part Number	25.2 x 35.6 x 1.1 mm Part Number	25.2 x 35.6 x 2.0 mm Part Number
488.0 nm	< 203 cm–1	494.3 – 756.4 nm	LPD02-488RU-25	LPD02-488RU-25x36x1.1	LPD02-488RU-25x36x2.0
532.0 nm	< 186 cm–1	538.9 – 824.8 nm	LPD02-532RU-25	LPD02-532RU-25x36x1.1	LPD02-532RU-25x36x2.0
632.8 nm	< 156 cm–1	641.0 – 980.8 nm	LPD02-633RU-25	LPD02-633RU-25x36x1.1	LPD02-633RU-25x36x2.0
785.0 nm	< 126 cm–1	795.2 – 1213.8 nm	LPD02-785RU-25	LPD02-785RU-25x36x1.1	LPD02-785RU-25x36x2.0
830.0 nm	< 119 cm–1	840.8 – 1286.5 nm	LPD02-830RU-25	LPD02-830RU-25x36x1.1	LPD02-830RU-25x36x2.0
1064.0 nm	< 93 cm–1	1077.8 – 1650.8 nm	LPD02-1064RU-25		LPD02-1064RU-25x36x2.0

#### AVAILABLE IN 1.1 mm THICKNESSES FOR MICROSCOPES

SEE SPECTRA GRAPHS AND ASCII DATA ALL OF OUR FILTERS AT WWW.IDEX-HS.COM/SEMROCK

### **Dichroic Beamsplitter Specifications**

Property		Specification	Comment
Edge Steepness (typical)		0.5% of laser wavelength (2.5 nm or 90 cm-1 for 532 nm filter)	Measured from 5% to 50% transmission for light with average polarization
Transition Width	U-grade	< 1% of laser wavelength	Measured from laser wavelength to 50% transmission wavelength for light with average polarization
Reflection at Laser	Wavelength	> 98% (s-polarization) > 90% (p-polarization)	
Average Passband	Transmission	> 93%	Averaged over the Passband (Passband wavelengths detailed above)
Dependence of Wavelength on Angle of Incidence (Edge Shift)		≤ 0.2% / degree	Linear relationship valid between 35° & 55° (see MyLight for actual performance)
Cone Half Angle (for non-collimated light)		< 0.5°	Rays uniformly distributed and centered at 45°
	Clear Aperture	≥ 22 mm	
Size of Round Dichroics	Outer Diameter	25.0 + 0.0 / - 0.1 mm	Black-anodized aluminum ring
D form of too	Overall Thickness	3.5 ± 0.1 mm	Black-anodized aluminum ring
	Clear Aperture	> 80%	Elliptical
Size of Rectangular Dichroics	Size	25.2 mm x 35.6 mm $\pm$ 0.1 mm	
	Unmounted Thickness	1.05 mm ± 0.05 mm	
Wedge Angle		< 20 arcseconds	
Flatness		Reflection of a collimated, Gaussian than one Rayleigh Range of focal shi	laser beam with waist diameter up to 3 mm causes less ift after a focusing lens.

# EdgeBasic<sup>™</sup> Long / Short Wave Pass Filters

EdgeBasic long wave pass and short wave pass filters offer a superb combination of performance and value for applications in Raman spectroscopy and fluorescence imaging and measurements. This group of filters is ideal for specific Raman applications that do not require measuring the smallest possible Raman shifts, yet demand exceptional laser-line blocking and high transmission over a range of Raman lines.

- > Deep laser-line blocking for maximum laser rejection (OD > 6)
- > Extended short-wavelength blocking (LWP) for high-fidelity fluorescence imaging
- > High signal transmission to detect the weakest signals (> 98% typical)
- > Proven no burn-out durability for lasting and reliable performance
- > For the ultimate performance, upgrade to state-of-the-art RazorEdge® Raman filters

#### **EdgeBasic Long Wave Pass**

	Laser Wavele	ength Range			
Nominal Laser Wavelength	short	long	Passband	Part Number	Price
325 nm	325.0 nm	325.0 nm	334.1 – 900.0 nm	BLP01-325R-25	\$455
355 nm	355.0 nm	355.0 nm	364.9 – 900.0 nm	BLP01-355R-25	\$455
363.8 nm	363.8 nm	363.8 nm	374.0 – 900.0 nm	BLP01-364R-25	\$455
405 nm	400.0 nm	410.0 nm	421.5 – 900.0 nm	BLP01-405R-25	\$405
441.6 nm	441.6 nm	441.6 nm	454.0 – 900.0 nm	BLP01-442R-25	\$405
457.9 nm	439.0 nm	457.9 nm	470.7 – 900.0 nm	BLP01-458R-25	\$405
473 nm	473.0 nm	473.0 nm	486.2 – 900.0 nm	BLP01-473R-25	\$405
488 nm	486.0 nm	491.0 nm	504.7 – 900.0 nm	BLP01-488R-25	\$405
514.5 nm	505.0 nm	515.0 nm	529.4 – 900.0 nm	BLP01-514R-25	\$405
532 nm	532.0 nm	532.0 nm	546.9 – 900.0 nm	BLP01-532R-25	\$405
561.4 nm	561.4 nm	561.4 nm	577.1 – 900.0 nm	BLP02-561R-25	\$405
568.2 nm	561.4 nm	568.2 nm	584.1 – 900.0 nm	BLP01-568R-25	\$405
594 nm	593.5 nm	594.3 nm	610.9 – 900.0 nm	BLP01-594R-25	\$405
632.8 nm	632.8 nm	632.8 nm	650.5 – 1200.0 nm	BLP01-633R-25	\$405
635 nm	632.8 nm	642.0 nm	660.0 – 1200.0 nm	BLP01-635R-25	\$405
647.1 nm	647.1 nm	647.1 nm	665.2 – 1200.0 nm	BLP01-647R-25	\$405
664 nm	664.0 nm	664.0 nm	682.6 – 1200.0 nm	BLP01-664R-25	\$405
785 nm	780.0 nm	790.0 nm	812.1 – 1200.0 nm	BLP01-785R-25	\$405
808 nm	808.0 nm	808.0 nm	830.6 – 1600.0 nm	BLP01-808R-25	\$405
830 nm	830.0 nm	830.0 nm	853.2 – 1600.0 nm	BLP01-830R-25	\$405
980 nm	980.0 nm	980.0 nm	1007.4 – 1600.0 nm	BLP01-980R-25	\$455
1064 nm	1064.0 nm	1064.0 nm	1093.8 – 1600.0 nm	BLP01-1064R-25	\$455
1319 nm	1319.0 nm	1319.0 nm	1355.9 – 2000.0 nm	BLP02-1319R-25	\$455
1550 mm	1550.0 nm	1550.0 nm	1593.4 – 2000.0 nm	BLP01-1550R-25	\$455

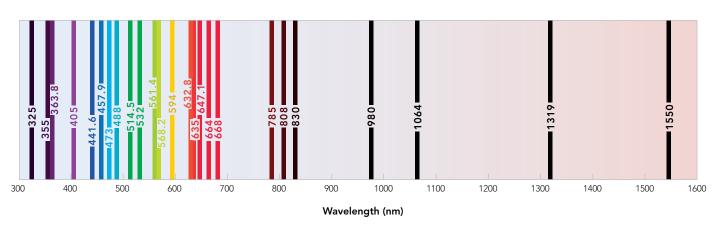
#### **Short Wave Pass**

	Laser Wavele	ength Range			
Nominal Laser Wavelength	short	long	Passband	Part Number	Price
532 nm	532.0 nm	532.0 nm	350.0 – 517.1 nm	BSP01-532R-25	\$455
632.8 nm	632.8 nm	647.1 nm	350.0 – 615.1 nm	BSP01-633R-25	\$455
785 nm	780.0 nm	790.0 nm	350.0 – 758.2 nm	BSP01-785R-25	\$455





# EdgeBasic<sup>™</sup> Long / Short Wave Pass Filters



#### Longpass specifications

Property	Value	Comments
Edge Steepness (typical)	1.5% of λlong	Measured from OD 6 to 50%
Transition Width	< 2.5% of λlong	From λlong to the 50% transmission wavelength
Blocking at Laser Wavelengths	ODabs > 6 from 80% of λshort to λlong ODavg > 5 from 270 nm to 80% of λshort (λs ≤ 1064 nm) ODavg > 5 from 800 nm to 80% of λshort (λs > 1064 nm)	OD = - log10 (transmission)
Guaranteed Transmission	> 93%	Averaged over the passband

### **Shortpass Specifications**

Property	Value	Comments
Edge Steepness (typical)	1.5% of λshort	Measured from OD 6 to 50%
Transition Width	< 2.5% of λshort	From 50% transmission wavelength to $\lambda$ short
Blocking at Laser Wavelengths	ODabs > 6 from λshort to 120% of λlong ODavg > 5 from 120% of λlong to 750 nm ODavg > 4 from 750 nm to 925 nm ODavg > 3 from 925 nm to 1200 nm	OD = - log10 (transmission)
Guaranteed Transmission	> 93%	Averaged over the passband >400nm

### **Common Specifications**

Property	Value	Comments
Guaranteed Transmission	> 93%	Averaged over the passband for Shortpass > 80% 350 – 400nm
Typical Transmission	> 98%	Averaged over the passband
Angle of Incidence	0.0° ± 2.0°	Range for above optical specifications
Cone Half Angle	< 5°	Rays uniformly distributed about 0°
Angle Tuning Range	- 0.3% of Laser Wavelength	Wavelength "blue shift" increasing angle from $0^\circ$ to $8^\circ$
Substrate Material	Low-autofluorescence optical quality glass	
Substrate Thickness	$2.0 \pm 0.1 \text{ mm}$	
Clear Aperture	> 22 mm	
Outer Diameter	25.0 + 0.0 / - 0.1 mm	Black-anodized aluminum ring
Overall Thickness	3.5 ± 0.1 mm	Black-anodized aluminum ring
Beam Deviation	< 10 arc seconds	
Surface Quality	60-40 scratch-dig	
Filter Orientation	Arrow on ring indicates preferred direction of propagation of light	

## **PRODUCT NOTE** Edge Steepness and Transition Width

Semrock edge filters, including our steepest RazorEdge<sup>®</sup> Raman filters as well as our EdgeBasic<sup>™</sup> filters for applicationspecific Raman systems and fluorescence imaging, are specified with a guaranteed "Transition Width."

**Transition Width** = maximum allowed spectral width between the laser line (where OD > 6) and the 50% transmission point

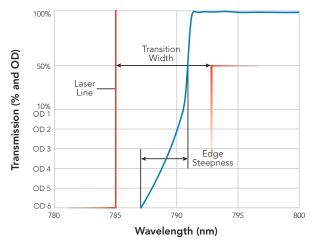
Any given filter can also be described by its "Edge Steepness," which is the actual steepness of the filter, regardless of the precise wavelength placement of the edge.

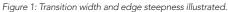
**Edge Steepness** =actual steepness of a filter measured from the OD 6 point to the 50% transmission point

Figure 1 illustrates Transition Width and Edge Steepness for an edge filter designed to block the 785 nm laser line (example shows a U-grade RazorEdge filter). The table below lists the guaranteed Transition Width and typical Edge Steepness (for 25 mm diameter parts) for Semrock edge filters.

All Verona and RazorEdge filters provide exceptional steepness to allow measurement of signals very close to the blocked laser line with high signal-to-noise ratio. However, the state-of-the-art E-grade RazorEdge filters take closeness to an extreme level.

Figure 2 illustrates that U-grade RazorEdge filters have a transition width that is 1% of the laser wavelength. E-grade filters have a transition width that is twice as narrow, or 0.5% of the laser line!





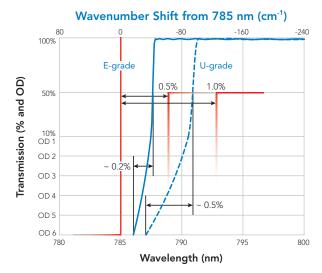


Figure 2: Transition widths and edge steepnesses for LP02-785RE and LP02-785RU filters (see page R-11).

Edge Filter Type	Guaranteed Transition Width (% of laser wavelength)	Typical Edge Steepness (% of laser wavelength)
Verona	Refer to page R-2	
RazorEdge E-grade	< 0.5% (< 90 cm-1 for 532)	0.2% (1.1 nm for 532)
RazorEdge U-grade	< 1.0% (< 186 cm-1 for 532)	0.5% (2.7 nm for 532)
EdgeBasic	< 2.5% (< 458 cm-1 for 532)	1.5% (8.0 nm for 532)
* except UV filters		

# RazorEdge® Short Wave Pass Raman Edge Filters

These unique filters are ideal for Anti-Stokes Raman applications. An addition to the popular high-performance RazorEdge family of steep edge filters, these short-wavepass filters are designed to attenuate a designated laser-line by six orders of magnitude, and yet maintain a typical edge steepness of only 0.5% of the laser wavelength. Both short and long-wave-pass RazorEdge filters are perfectly matched to Semrock's popular MaxLine<sup>®</sup> laser-line cleanup filters. U.S. Patent No. 7,068,430



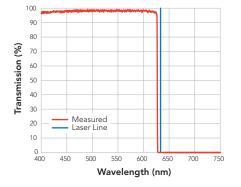
#### 25 mm and 50 mm Diameters

Laser Line	Transition Width	Passband	Part Number	Price
532.0 nm	< 186 cm–1	350.0 – 525.2 nm	SP01-532RU-25	\$855
561.4 nm	< 176 cm–1	400.0 – 554.1 nm	SP01-561RU-25	\$745
632.8 nm	< 160 cm–1	372.0 – 624.6 nm	SP01-633RU-25	\$855
785.0 nm	< 129 cm–1	400.0 – 774.8 nm	SP01-785RU-25	\$745
50 mm SWP Edge			idex-hs.com/semr	ock

Filters

SEE SPECTRA GRAPHS AND ASCII DATA FOR ALL OF OUR FILTERS AT WWW.IDEX-HS.COM/SEMROCK

### **ACTUAL MEASURED DATA** FROM A 632.8 nm **RAZOREDGE FILTER**



### PRODUCT NOTE

### RazorEdge and MaxLine® are a Perfect Match

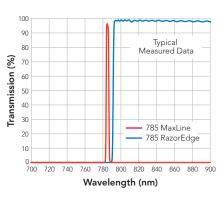
The MaxLine (see page R-6) and RazorEdge U-grade (see page R-9) filters make an ideal filter pair for applications like Raman spectroscopy – they fit together like hand-in-glove. The MaxLine filter spectrally "cleans up" the excitation laser light before it reaches the sample under test – allowing only the desired laser line to reach the sample - and then the RazorEdge filter removes the laser line from the light scattered off the sample, while efficiently transmitting desired light at wavelengths very close to the laser line.

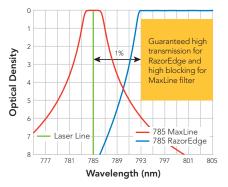
Typical measured spectral curves of 785 nm filters on a linear transmission plot demonstrate how the incredibly steep edges and high transmission exhibited by both of these filters allow them to be spectrally positioned very close together, while still maintaining complementary transmission and blocking characteristics.

The optical density plot illustrates the complementary nature of these filters on a logarithmic scale using the theoretical design spectral curves. The MaxLine filter provides very high transmission (> 90%) of light immediately in the vicinity of the laser line, and then rapidly rolls off to achieve very high blocking (> OD 5) at wavelengths within 1% of the laser line. The RazorEdge filter provides extremely high blocking (> OD 6) of the laser line itself, and then rapidly climbs to

achieve very high transmission (> 90%) of the desired signal light at wavelengths only 1% away from the laser line.

If you are currently using an E-grade RazorEdge filter and need a laser clean-up filter, please contact Semrock.





# Edge Filters vs. Notch Filters for Raman Instrumentation

### RazorEdge® Filter Advantages:

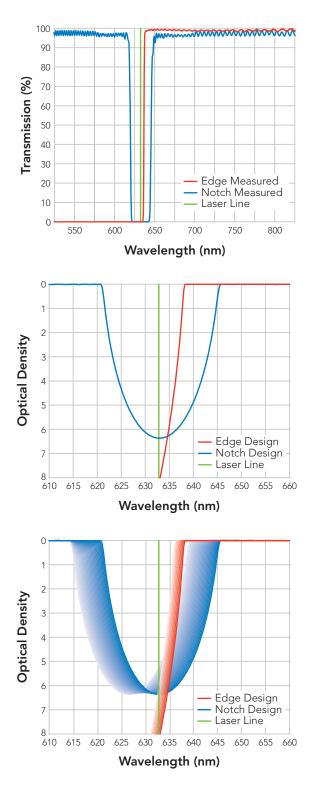
- Steepest possible edge for looking at the smallest Stokes shifts
- Largest blocking of the laser line for maximum laser rejection

### StopLine® Notch Filter Advantages:

- Measure stokes and anti-stokes signals simultaneously
- > Greater angle-tunability and bandwidth for use with variable laser lines

The graph on the top illustrates the ability of a long-wave-pass (LWP) filter to get extremely close to the laser line. The graph in the center compares the steepness of an edge filter to that of a notch filter. A steeper edge means a narrower transition width from the laser line to the high-transmission region of the filter. With transition widths guaranteed to be below 1% of the laser wavelength (on Semrock U-grade edge filters), these filters don't need to be angle-tuned!

The graph on the bottom shows the relative tuning ranges that can be achieved for edge filters and notch filters. Semrock edge filters can be tuned up to 0.3% of the laser wavelength. The filters shift toward shorter wavelengths as the angle of incidence is increased from 0° to about 8°. Semrock notch filters can be tuned up to 1.0% of the laser wavelength. These filters also shift toward shorter wavelengths as the angle of incidence is increased from 0° up to about 14°.



# RazorEdge® Common Specifications

#### **RazorEdge Specifications**

Properties apply to all long-wave-pass and short-wave-pass edge filters unless otherwise noted

Property		Specification	Comment
Edge Steepness (typical)	E-grade U-grade	0.2% of laser wavelength 0.5% of laser wavelength	Measured from OD 6 to 50%; Up to 0.8% for 248-300 nm filters and 3.3% for 224 nm filter
Blocking at Laser Wavelength		>6 OD	OD = - log10 (transmission)
Transition Width	E-grade U-grade	< 0.5% of laser wavelength < 1% of laser wavelength	Measured from laser wavelength to 50% transmission wavelength; < 4.5% for 224 nm filter
Guaranteed Passband Transmission		> 93%	Except > 90% for 224 – 325 nm filters; Averaged over the Passband
Typical Passband Transmission		> 98%	
Angle of Incidence		$0.0^{\circ} \pm 2.0^{\circ}$	Range for above optical specifications
Cone Half Angle		< 5°	Rays uniformly distributed about 0°
Angle Tuning Range1		-0.3% of Laser Wavelength	Wavelength "blue shift" attained by increasing angle from 0° to $8^\circ$
Laser Damage Threshold		0.5 J/cm2 @ 266 nm 1 J/cm2 @ 532 nm	10 ns pulse width Tested for 266 and 532 nm filters only (see page 109)
Clear Aperture		> 22 mm (or > 45 mm)	
Outer Diameter		25.0 + 0.0 / -0.1 mm (or 50.0 + 0.0 / -0.1 nm)	Black-anodized aluminum ring
Substrate Thickness		2.0 mm	
Overall Thickness		3.5 ± 0.1 mm	Black-anodized aluminum ring (thickness measured unmounted)
Beam Deviation		< 10 arcseconds	
1 For small angles (in degrees),	the wavele	ength shift near the laser wavel	length is DI (nm) = $-5.0 \times 10-5 \times IL \times q^2$ and the wavenumber shift is

1 For small angles (in degrees), the wavelength shift near the laser wavelength is Dl (nm) =  $-5.0 \times 10-5 \times 1L \times q^2$  and the wavenumber shift is D(wavenumbers) (cm-1) =  $500 \times q^2 / IL$ , where IL (in nm) is the laser wavelength.

#### General Specifications (all RazorEdge filters)

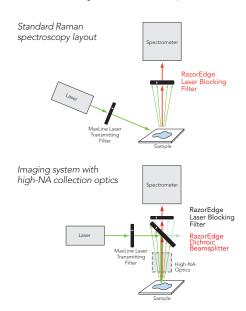
Property	Specification	Comment
Coating Type	"Hard" ion-beam-sputter	red
Reliability and Durability		d-coated technology with epoxy-free, single-substrate construction for Edge filters are rigorously tested and proven to MIL-STD-810F and MIL-C- andards.
Transmitted Wavefront Error	$<\lambda$ / 4 RMS at $\lambda$ = 633 nn	n Peak-to-valley error < 5 x RMS value measured within clear aperture
Surface Quality	60-40 scratch-dig	
Temperature Dependence	< 5 ppm / °C	
Substrate Material	Ultra-low autofluorescenc fused silica	ie de la constant de
Filter Orientation		v on ring indicates preferred direction of propagation of transmitted light. reflective coating side should face toward light source and sample.

### TECHNICAL NOTE

## RazorEdge Filter Layouts

Only the unique RazorEdge Dichroic beamsplitter reflects a standard laser line incident at 45° while transmitting longer Raman-shifted wavelengths with an ultrasteep transition far superior to anything else available on the open market. The guaranteed transition width of < 1% of the laser wavelength for U-grade (regardless of polarization) makes these filters a perfect match to our popular normal-incidence RazorEdge ultrasteep long-wave-pass filters.

In order for the two-filter configuration to work, the 45° beamsplitter must be as steep as the laser-blocking filter. Traditionally thin-film filters could not achieve very steep edges at 45° because ofthe "polarization splitting" problem – the edge position tends to be different for different polarizations of light. However, through continued innovation in thin-film filter technology, Semrock has been able to achieve ultrasteep 45° beamsplitters with the same steepness of our renowned RazorEdge laser-blocking filters: the transition from the laser line to the passband of the filter is guaranteed to be less than 1% of the laser wavelength (for U-grade filters).



## 

### **CALE NOTE** Filter Spectra at Non-normal Angles of Incidence

While most applications call for optical filters to be used at normal incidence, it is important to understand how the spectral properties of different types of filters change when using these filters a non-normal angles of incidence (AOI). There are two main effects exhibited by all filter spectrum as the angle is increased from normal:

- 1. The features of the spectrum shift to shorter wavelengths.
- 2. Two distinct spectra emerge one for s-polarized light and one for p-polarized light.

### RazorEdge® Edge Filters

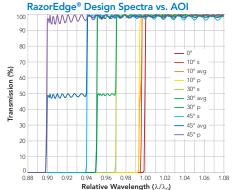
The graph in Figure 1 shows a series of spectra derived from a typical RazorEdge long-wave-pass (LWP) filter design, and can be applied to any of the RazorEdge edge filters. Here, the resulting wavelength  $\lambda$  is compared to the wavelength  $\lambda$ 0 of the edge location at normal incidence. As the angle increases from normal incidence, the filter edge shifts toward shorter wavelengths and the edges associated with s- and p-polarized light shift by different amounts. For LWP filters, the response of p-polarized light shifts more than s-polarized light. The opposite is true for SWP filters. This polarization splitting associated wavelength.

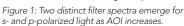
causes the unpolarized spectrum to show a "shelf" near the 50% transmission point, but note that the edge steepness remains intact, even for polarized light.

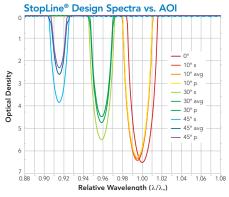
The shift of any spectral feature of a Semrock filter can be calculated using the MyLight tool on the website-based product page. For example, on the product for the 785 nm RazorEdge long pass filter at 785 nm, click on the MyLight link in the green box. Visit **idex-hs.com/resources/toolsdrawings/mylight-tutorial** for a tutorial for how to use MyLight. Also note that any catalog filter can also be modeled at specific AOI, CHA, and Polarization at **idex-hs.com/SearchLight.** 

### MaxLine® Laser-line Filters

Varying filter designs will respond to changes in AOI in different ways, but still show marked differences between s- and p-polarized light. The spectra of a MaxLine laser-line filter (Figure 2) shows that as the angle increases from normal incidence, the center wavelength shifts toward shorter wavelengths and the bandwidth broadens slightly for p-polarized light while narrowing for s-polarized light. The most striking feature is the decrease in transmission for s-polarized









light, while the p-polarized light maintains high transmission. Use the MyLight Tool described above to model and download spectra of AOI-shifted MaxLine filters, or model them in **SearchLight**.

### StopLine® Notch Filters

The StopLine series of filters has a different response to AOI depending on the grade of filter used. For the standard U- and S-grade filters (Figure 3), as the angle is increased from normal incidence, the notch center wavelength shifts to shorter wavelengths, the notch depth decreases, and the notch bandwidth decreases (with a greater decrease for p-polarized light than for s-polarized light).

E-grade filter (Figure 4), which is based on a different type of filter design, shows a large increase in OD for s-polarized light as the AOI increases. As the angle is increased from normal incidence, the notch center wavelength shifts to shorter wavelengths; however, the shift is greater for p-polarized light than it is for s-polarized light.



To learn more about our Semrock notch filters, search for StopLine® Notch Filters at idex-hs.com. Use our MyLight Tool to model and download spectra of AOI-shifted StopLine filters, or model them in **SearchLight**.

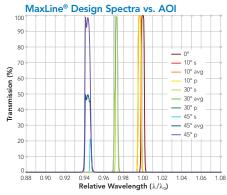


Figure 2: MaxLine filter spectral response to AOI shift.

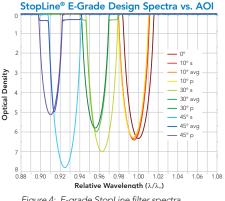


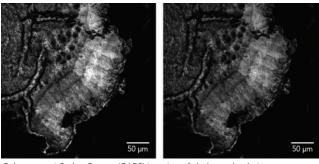
Figure 4: E-grade StopLine filter spectra.

### Ф тесниісац моте Coherent Raman Scattering (CRS, CARS and SRS)

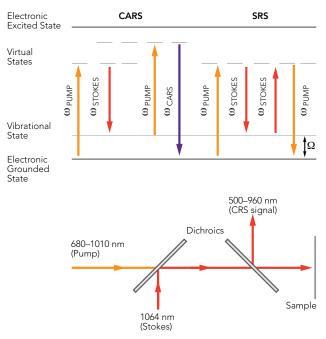
With coherent Raman scattering (CRS) it is possible to perform highly specific, label-free chemical and biological imaging with orders of magnitude higher sensitivity at video-rate speeds compared with traditional Raman imaging. CRS is a nonlinear four-wave mixing process that is used to enhance the weak spontaneous Raman signal associated with specific molecular vibrations. Two different types of CRS that are exploited for chemical and biological imaging are coherent anti-Stokes Raman scattering (CARS) and stimulated Raman scattering (SRS).

In CRS, two lasers are used to excite the sample. The wavelength of a first laser (often a fixed-wavelength, 1064 nm laser) is set at the Stokes frequency,  $\omega_{\text{Stokes}}$ . The wavelength of the second laser is tuned to the pump frequency,  $\omega_{\text{pump}}$ . When the frequency difference  $\omega_{\text{pump}} - \omega_{\text{Stokes}}$  between these two lasers matches an intrinsic molecular vibration of frequency  $\Omega$  both CARS and SRS signals are generated within the sample.

In CARS, the coherent Raman signal is generated at a new, third wavelength, given by the anti-Stokes frequency  $\omega_{\text{CARS}} = 2\omega_{\text{pump}} - \omega_{\text{Stokes}} = \omega_{\text{pump}} + \Omega$ . In SRS there is no signal at a wavelength that is different from the laser excitation wavelengths. Instead, the intensity of the scattered light at the pump wavelength experiences a stimulated Raman loss (SRL), with the intensity of the scattered light at the Stokes wavelength experiencing a stimulated Raman gain (SRG). The key advantage of SRS microscopy over CARS microscopy is that it provides background-free chemical imaging with improved image contrast, both of which are important for biomedical imaging applications where water represents the predominant source of nonresonant background signal in the sample.



Coherent anti-Stokes Raman (CARS) imaging of cholesteryl palmitate. The image on the left was obtained using Semrock filter FF01-625/90. The image on the right was obtained using a fluorescence bandpass filter having a center wavelength of 650 nm and extended blocking. An analysis of the images revealed that the FF01-625/90 filter provided greater than 2.6 times CARS signal. Images courtesy of Prof. Eric Potma (UC Irvine).



Coherent Raman scattering energy diagrams for both CARS and SRS (top), and a schematic of a typical experimental setup (bottom).

### Harmonic Generation Microscopy

Harmonic generation microscopy (HGM) is a label-free imaging technique that uses high-peak power ultrafast lasers to generate appreciable image contrast in biological imaging applications. Harmonic generation microscopy exploits intrinsic energy-conserving second and third order nonlinear optical effects. In second-harmonic generation (SHG) two incident photons interact at the sample to create a single emission photon having twice the energy i.e.,  $2\omega_i = \omega_{SHG}$ . A prerequisite for SHG microscopy is that the sample must exhibit a significant degree of noncentrosymmetric order at the molecular level before an appreciable SHG signal can be generated. In third-harmonic generation (THG), three incident photons interact at the sample to create a single emission photon having three times the energy i.e.,  $3\omega_i = \omega_{THG}$ . Both SHG and THG imaging techniques can be combined with other nonlinear optical imaging (NLO) modalities, such as multiphoton fluorescence and coherent Raman scattering imaging. Such a multimodal approach to biological imaging allows a comprehensive analysis of a wide variety of biological entities, such as individual cells, lipids, collagen fibrils, and the integrity of cell membranes at the same time.

### **CALE NOTE** Ultraviolet (UV) Raman Spectroscopy

Raman spectroscopy measurements generally face two limitations: (1) Raman scattering cross sections are tiny, requiring intense lasers and sensitive detection systems just to achieve enough signal; and (2) the signal-to-noise ratio is further limited by fundamental, intrinsic noise sources like sample autofluorescence. Raman measurements are most commonly performed with green, red, or near-infrared (IR) lasers, largely because of the availability of established lasers and detectors at these wavelengths. However, by measuring Raman spectra in the ultraviolet (UV) wavelength range, both of the above limitations can be substantially alleviated.

Visible and near-IR lasers have photon energies below the first electronic transitions of most molecules. However, when the photon energy of the laser lies within the electronic spectrum of a molecule, as is the case for UV lasers and most molecules, the intensity of Raman-active vibrations can increase by many orders of magnitude – this effect is called "resonanceenhanced Raman scattering." Although UV lasers tend to excite strong autofluorescence, it typically occurs only at wavelengths above about 300 nm, independent of the UV laser wavelength. Since even a 4000 cm<sup>-1</sup> (very large) Stokes shift leads to Raman emission below 300 nm when excited by a common 266 nm laser, autofluorescence simply does not interfere with the Raman signal making high signal-to-noise ratio measurements possible.

An increasing number of compact, affordable, and high-power UV lasers have become widely available, such as quadrupled, diode-pumped Nd:YAG lasers at 266 nm and NeCu hollowcathode metal-ion lasers at 248.6 nm, making ultra-sensitive UV Raman spectroscopy a now widely accessible technique.

