HPLC Separations with Sharper Peaks - Remote UV Detection

Interview conducted by Jake Wilkinson Oct 6 2016

Placing UV detectors far away from a chromatographic column can be problematic. The increased extra column volume can cause band broadening, reducing the efficiency of a separation. Also, experiments involving radioactive tags can result in contamination issues. To solve this problems (and more) KNAUER have developed an external detector flow cell, which reads the UV absorption remotely from the device via a fiber optic cable.

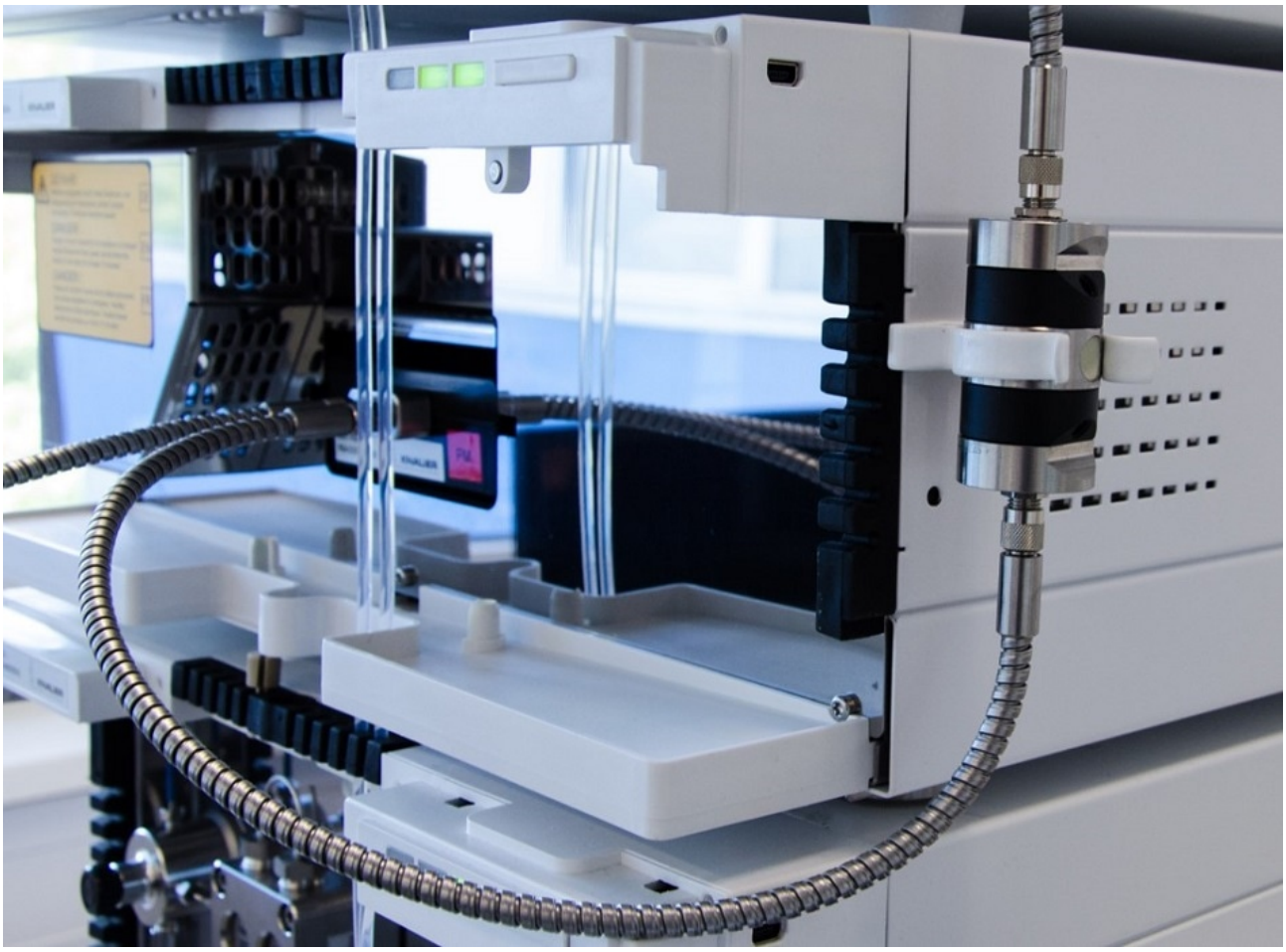
AZoM spoke to Kate Monks, a product manager at KNAUER about their external flow cells, the different types of experiments they facilitate and the range of detectors that they offer.

How does the setup of an HPLC system with an external detector differ from conventional HPLC systems?

In most HPLC systems you find in the lab, the detector and flow cell are snuggled inside the instrument, sometimes not even visible without removing the instrument's front panel or cover.

KNAUER external flow cells are different: they are physically separated from the detector, connected via fiber optic cables.

This enables the measurement of UV/Vis absorption in flow-through mode remotely from the instrument. The detection principle is the same in both setups: light from the detector's lamp or lamps passes through the flow cell and falls on the detecting element allowing us to register a signal. The light path for external flow cells is just longer.

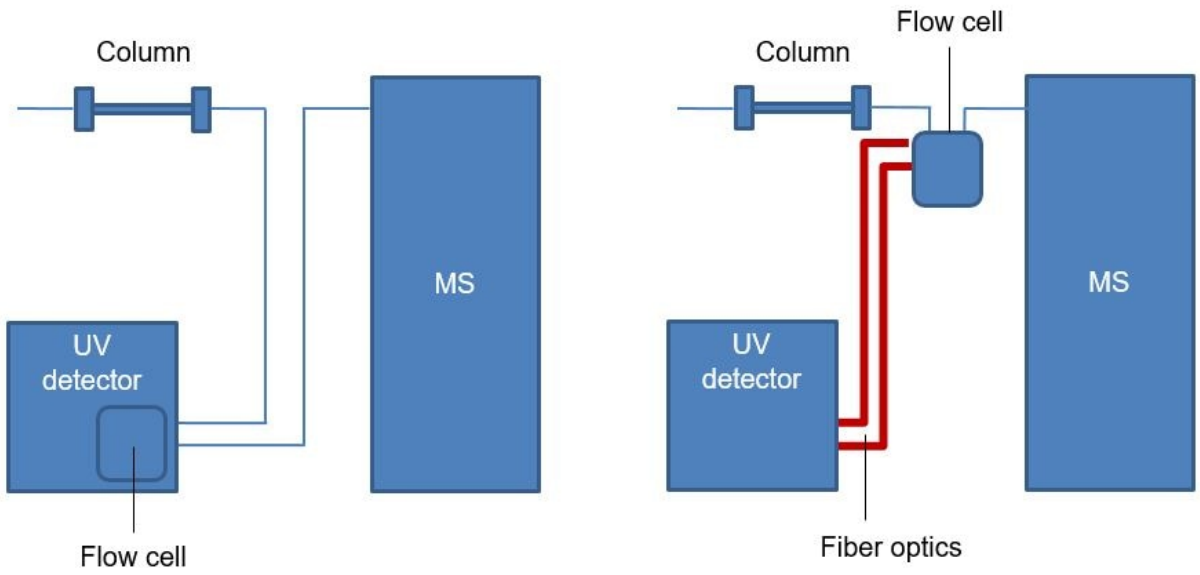


External detector flow cells sit outside of the chromatographic column (on the right of the system in this image) and receive UV absorption data via a fiberoptic cable linked to column itself.

Does remote detection result in better separations? If so, why?

That depends on the application. For instance low-flow separations (nano or micro HPLC) are greatly dependent on extra column volumes.

This is the volume between the points of injection and detection, excluding the stationary phase part of the column. Through fiber optic technology we can place the micro or nano flow cell as close as possible to the HPLC column outlet. This way we can minimize the extra column volume and reduce undesirable peak broadening. This can make or break a separation.



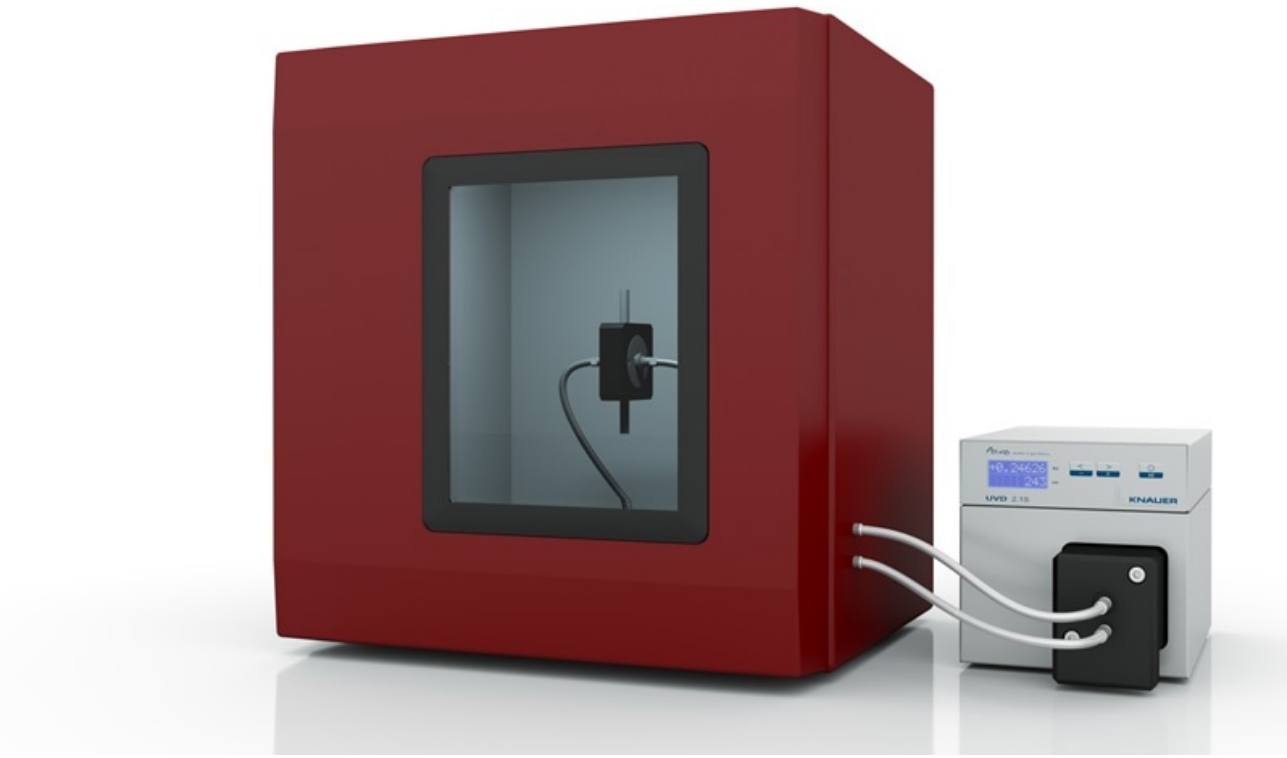
Using an external detector reduces the path length of analytes that have passed through the column reducing the effect of band broadening. When working with micro and nanoscale flow cells this can be the difference between an effective separation and a failure.

Does the use of an external detector allow for any experiments to be undertaken which, using a conventional HPLC system, would be difficult or not possible?

This technology really comes into its own in hazardous environments with radioactive, explosive, toxic or high temperature applications.

For instance many of our customers working with radioactive substances place theKNAUER remote cell inside their dangerous hot cell leaving their detector safely outside. This prevents the detectors getting into contact with contamination, and means you don't become the Hulk when you carry out maintenance.

Another interesting application for this technology is high flow rate LC, where thick capillaries (> 1/8") are used for plumbing the system. Anyone who has plumbed one of these systems will know that bending tubing as thick as your little finger is no easy task. Here, the fiber optic cables can be used to take the flow cell to the flow rather than redirecting the flow through the detector flow cell.



Knauer's external detectors allow high temperature seperations, or seperations with hazardous components, to take place in isolated systems whilst keeping the detector accessible.

Is there any significant loss in accuracy as a result of the fiber optic cable?

As the light path is longer when working with external flow cells in comparison to standard integrated flow cell designs, light intensity is lost along the way. With the KNAUER standard 750 mm fiber optic cables this loss is comparatively low.

The longer the cables, the greater the loss in light reaching the detector element. In the field we have customers using 10 meter long fiber optic cables! In these cases the light intensity loss is considerable. Highly sensitive analytical HLPC separations would suffer from such a light loss, however for concentrated samples, it is enough.

What range of detectors do KNAUER offer for remote HPLC detection?

KNAUER offers a wide range of remote detectors including simple wavelength UV detectors (ideal for monitoring), multi-wavelength UV/Vis detectors and diode array UV/Vis detectors.

For each of these detectors, lots of different flow cells are available. This range enables the same detector to be a great choice for utterly different application fields: from certified

pharmaceutical production with flow rates up to 10 L/min to green nano LC with flow rates down to 1 µL/min.

Standard flow cells are made out of stainless steel, however, recently the demand for bioinert wetted components has greatly risen. For these applications our PEEK or titanium flow cells are ideal.

Can standard KNAUER detectors and flow cells be used for remote HPLC?

KNAUER fiber optic flow cells have two SMA 905 connectors which make it possible to attach the fiber optic cables. Our standard flow cells don't have these connectors, as they are placed directly in the detector's light path. So, it isn't possible here to mix and match; for remote applications you need a remote flow cell and for non-remote or standard applications you need a standard flow cell.

The same can be said for our simple wavelength detectors: there are remote and non-remote versions which can be used according to the required mode of detection. Our diode array and multi-wavelength detectors on the other hand are hybrids: they can be used for remote and standard detection with the aid of an adapter.

For non-remote applications a standard flow cell cartridge can be mounted, and for remote applications a fiber optics adapter cartridge can be mounted. The fiber optics adapter is then connected to the actual flow cell via fiber optic cables.

Do KNAUER offer any advice on integrating remote detectors into existing systems or on which detector would best suit a particular researcher's needs?

Integrating a fiber optics detector supporting external flow cells is easy. The KNAUER instruments can be controlled by a wide range of software platforms, as well as from the front panel (stand-alone operation), via various interfaces, or through analog input/output; allowing it to be integrated into almost any LC system.

My favorite way of integrating these devices into existing systems is via the KNAUER Mobile Control software, installed onto a tablet. The software communicates with the detector via WIFI (or LAN) and serves as a friendly and colorful display, as well as enabling data acquisition.

The detector I find myself recommending to most customers is the small UV detectorAZURA UVD 2.1S; compact enough to fits in your hand and versatile enough to

satisfy analytical to process LC requirements. A dream!

Where can our readers find out more about KNAUER and HPLC using remote detection?

On our website the readers can find product information to both fiber optic detectors and flow cells. The choice is tremendous.

About Dr. Kate Monks

Kathryn studied Chemistry at Universidad de Valencia.

In 2009 she joined the Molnár-Institute for Applied Chromatography in Berlin and in 2012, she finished her Ph.D. in Pharmaceutical Analysis at the Freie Universität Berlin.

Kate joined KNAUER in 2013 as Product Manager for HPLC detectors.

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With more than 130 employees, Knauer is one of the well-established manufacturers of HPLC instruments, SMB systems, and osmometers. Product portfolio includes extremely compact HPLC solutions, UHPLC systems for high-resolution analysis, preparative HPLC instruments, process LC equipment for the purification of substances in the kilogram scale, autosamplers, column thermostats, degassers, detectors, dosing pumps, eluent mixers, flowmeters, LC columns, and accessories and spare parts, amongst others.

The source of Knauer’s success is numerous world’s firsts that have won more than 20

awards for innovation.