Comparing techniques for flow rate measurement in Liquid Chromatography

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Accurate determination of flow rate is one of the most common, and often overlooked, means of assessing or validating the performance of a pump serving a HPLC, UPLC, Ion-Chromatography or GPC/SEC system. Using the right technique, it is nowadays possible to use flow rate monitoring as a powerful diagnostic tool, allowing fast detection of leaks, faulty check-valves and worn seals.

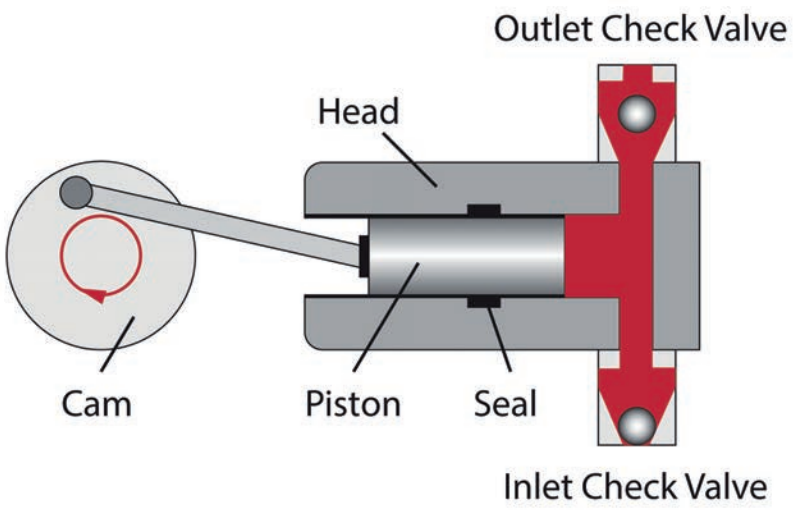
Several different technologies might be considered for liquid chromatographic flow measurement. It can be expected that each technique will have its own pros and cons. Consequently, prior knowledge of the limitations of each particular technique is of great help in better understanding flow measurement results and their positive or negative consequences.

This short paper provides an introductory background to different flow rate monitoring techniques and seeks to explain, for both beginner and expert chromatographers, the value of measurements achievable with each method. With this information our aim has been to provide an independent, informed interpretation of the flow rate monitoring results and how this might lead to higher confidence in performance validation of the liquid chromatography system under test.

Necessity for an accurate determination of flow rate

Modern Liquid Chromatography pumps are incredible instruments, capable of delivery of a constant stream of solvent at very high pressure and with flow rates ranging from a few µL/min to, in the case of preparative systems, several liters per minute. In many liquid chromatography systems, multiple reciprocating pistons are used to achieve the desired pump performance. The value of the flow rate is given, in principle, by the mechanical dimensions of the pump piston and chamber assembly plus the linear velocity of the piston in the chamber.

The concept of relating flow rate only to mechanical dimensions and fluid velocity is perfectly adequate in a perfect world. However, In the real world a number of parameters negatively influence the real flow rate, pressure being one of the most obvious. Pump manufacturers have implemented different ways to correct the real flow rate in order to obtain the desired stream of solvent. This correction, however, is influenced by the general status of the pump itself and on the particular solvent which is delivered. Pump maintenance, which often includes regular replacement of seal rings,

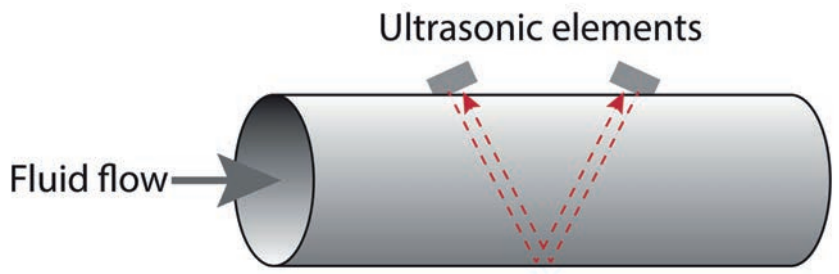


pistons, and check valves, might mitigate the possibility of error, but a difference in real flow rate from the setpoint cannot be excluded completely. Therefore, the performance of your pump must be periodically validated to ensure reliable and accurate flow rate data. In other words, the real flow rate at a specific setpoint must be measured on a regular basis to reveal deviations.

Traditionally, flow rate determination involved manually timed measurement of the volume or weight of the solvent delivered by the pump using a stopwatch, a scale or a graduated cylinder. The necessity for very accurate timing in combination with volume or weight measurement techniques makes both methods impractical for use in modern lab protocols. Using these traditional techniques, it has been found that human error can have a huge impact on the results. As a consequence, using automatic flowmeters, capable of measurement of flow rate without direct human intervention, are nowadays, the well-established flow monitoring instruments of choice. One further advantage of these modern instruments is the much better documentation of the process done and results obtained, which is particularly important in quality sensitive environments in regulated industries.

In this paper, we define a flow meter as a device capable of determining the flow rate (usually as volume unit per time unit) delivered by a pump within the context of an HPLC, UPLC, GPC/SEC or Ion-Chromatography context, without the necessity of human intervention.

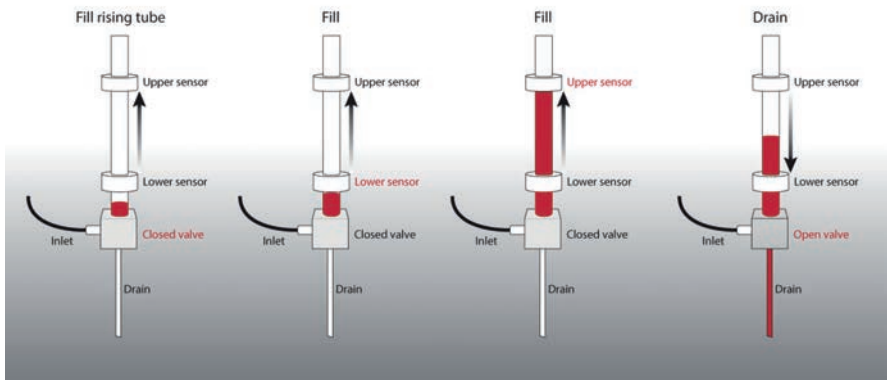
Ultrasonic Flowmeters



Ultrasonic flowmeters are not commonly used in analytical liquid chromatography systems. However, they are used with some preparative scale systems if the dimension and material of the tubing allows. The fundamental principle of ultrasonic flowmeters is based on the measurement of time required by an ultrasound wave to reach two detectors, one placed upstream of the emitters, and the second downstream of the emitter. The measured travel time difference is a function of the flow rate. A similar method of determination is based on the Doppler effect, thus frequency shift.

Employing a ‘clamp-on’ principle, ultrasonic flowmeters are typically relatively small and usefully provide non-invasive measurement. However, limited sensitivity at low flow rates and even more limited resolution, make ultrasonic flowmeters useful as monitoring devices in process applications but much less adequate for the accurate (and therefore high resolution) determination of low flow rates as is required in analytical tasks.

Volumetric Flowmeters



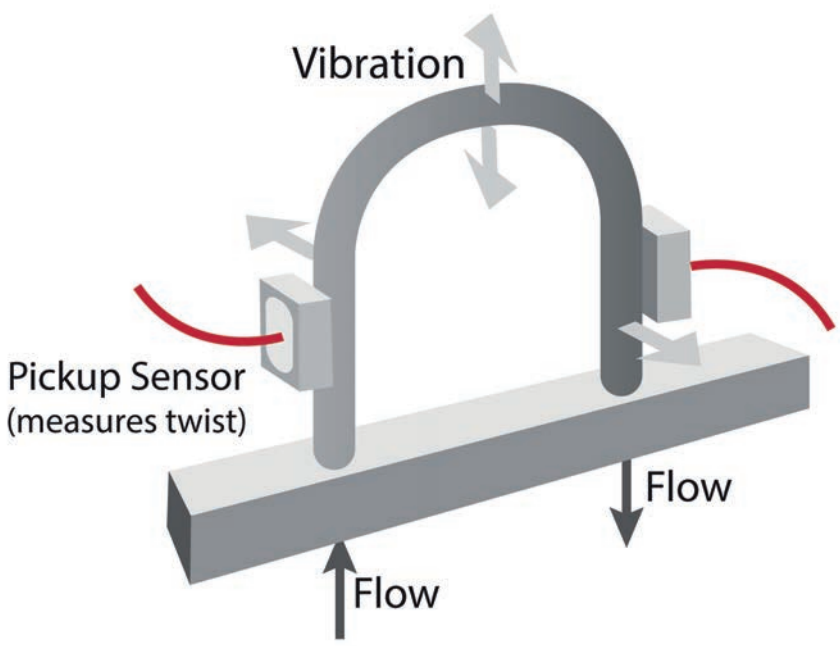
A volumetric Flowmeter is fundamentally the automated version of the classical 'Stopwatch and graduated cylinder' method. A flow meter of this type consists of a tube and two optical level sensors placed at a known distance to each other. The time the solvent front takes to travel from one light trap to the next is a function of the volume of the tube, which is constant, and the flow rate. After each measurement Is completed, the tube has to be automatically voided so that the measurement can be repeated. As such, a volumetric flowmeter is not capable of continuous measurements.

For analytical purposes - volumetric flowmeters must be positioned perfectly vertically and at the end of a liquid chromatography system. As a result, their use is limited to confirmation of performance of the pump, no real diagnostics is possible as each value reported is not related to the previous one.

Further to this, the result of each measured flow rate is the integral over the measurement volume, which makes it impossible to determine pulsations or variations in flow with a duty-cycle shorter than the time required to fill the tube. However, this technique has been proven to be very reliable particularly in single solvent (often water) applications. The different surface tensions of other solvents don't allow volumetric tubes to be voided with the same efficacy as with water.

Volumetric flow meters are commonly used in conjunction with HPLC systems as validation tools. It should however be noted, that because this technology is based on the measurement of time required to fill a known constant volume, that the accuracy of results decreases markedly at higher flow rates.

Coriolis Flowmeters

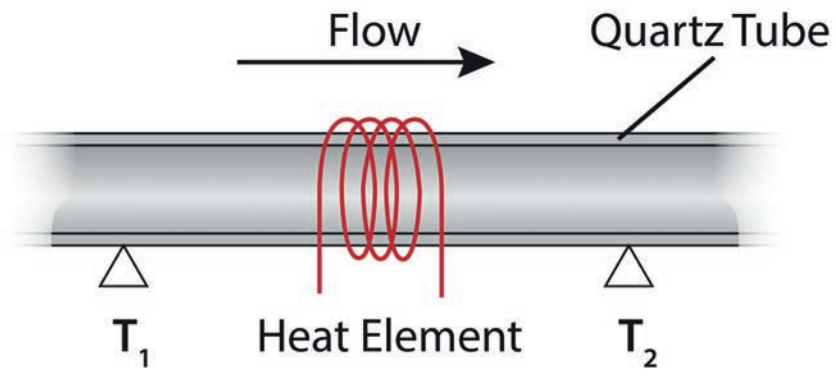


Coriolis flowmeters have found wide use in process applications, mainly supplying information about mass flow of individual reagents. The Coriolis measurement principle is based on detection of the variation in the oscillation frequency of a U-shaped tube through which the solvent travels. It can be demonstrated that the measured change in oscillation frequency can be related to the mass flow of the solvent.

As such, Coriolis flowmeters are by nature mass flowmeters, their data output will be therefore in weight units per time units. This of course represents a strong limitation in the applicability to liquid chromatography systems, where volume flow rate is common.

Devices based on the Coriolis principle are known to be accurate and highly reproducible. However, they are also known to be very slow devices, unable to detect changes in flow rate in the range of a few seconds. These facts combined limits the applicability of Coriolis type flowmeters for use in liquid chromatography applications.

Thermal Flowmeters



Thermal flowmeters were one of the first techniques used for the measurement of flow rate in liquid chromatography. Initially their use was limited because only analogue techniques were available. Developments in microelectronics and microcontrollers, however, have enabled thermal devices to become a very useful method of monitoring liquid chromatography flow rates.

The method is fundamentally based on the measurement of the difference in temperature between two temperature sensors, one placed upstream of a heating element and the second placed downstream of it. For applications ranging from a few microliters per minute up to several millilitres per minute, thermal flowmeters typically employ a quartz flow measurement tube. As all components necessary to flow measurement are located on the outer wall of the quartz tube, and have no contact with the liquid, the technique is non-invasive.

Being non-invasive means that thermal flowmeters have widespread applicability in terms of solvent used and also guarantees the unperturbed operation of the whole liquid chromatography system. Measurement with thermal flowmeters is continuous, allowing use of these devices for real-time monitoring of pump performance. Using modern thermal flowmeters, such as are available from Testa Analytical, it is now possible to interface them directly with chromatography data system allowing storage of flow data along with the chromatograms. This technological advance opens a whole new chapter to the concept of total quality assurance, as each and every chromatogram may now be evaluated under the light of the flow rate delivered by the pump during that one chromatogram.

Conclusion

Although flow rate is a fundamental parameter in any liquid chromatography application, it is one of the most underrated and underestimated measures of the total quality of results obtained with any system. A range of flowmeters are available to the interested user, all of them offering solid answers within the limits of the utilised technology. Ultrasonic flowmeters, although appealing from the viewpoint of ease-of-use, have no real place in chromatography, due to the limited accuracy and resolution of the method. Volumetric flowmeters are a proven solution for aqueous applications where a 'validation' is required and there is no necessity for tracking performance over a longer period of time. Coriolis flowmeters are by nature continuous monitors, their output is however mass flow rate, which is of minor importance to liquid chromatography, where all calculations are based on volumetric flow rate. The necessity of transformation from mass flow to volume flow using density also expresses the practical limitations of this technology, as accurate knowledge of the solvent density at the exact temperature the measurement was done at is required.

Modern thermal flowmeters are proven to provide continuous non-invasive monitoring of liquid chromatography flow rate. When an appropriate standard operating procedure (SOP) is used, thermal flowmeters can be used to deliver representative validation parameters of any liquid chromatography pump.

Available in formats to measure from nanolitres per minute right up to 650 millilitres per minute – thermal flowmeters can supply reliable and decisive information about the confidence level of any chromatogram and help minimise the downtime of a system when used as a fault diagnostic tool.

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