SFA

Advantages of Segmented-Flow Analysis

There are two kinds of Continuous-Flow Analysis

Segmented-Flow Analysis (SFA)

The reaction stream is segmented with bubbles of air or nitrogen to reduce inter-sample dispersion.

Flow Injection Analysis (FIA)

There are no air bubbles in the reaction stream. The dispersion in the system is used to mix the samples and reagents.

SEAL Analytical continuous-flow analyzers use SFA technology because the bubbles bring important advantages to routine analysis.



Maximum sensitivity

Two factors contribute to sensitivity

- Complete reaction between reagents and sample
- Measuring the final reaction mixture at maximum concentration

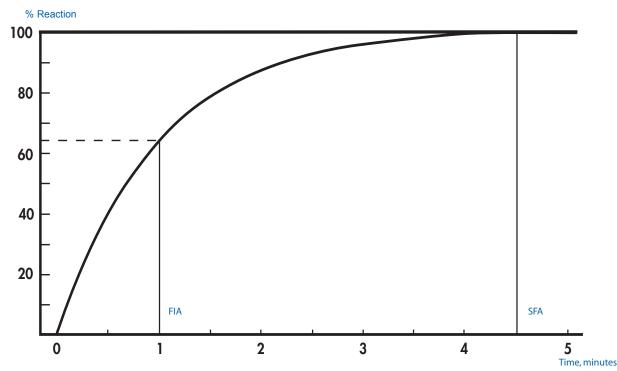
Complete reaction

If a time delay is necessary, the reaction stream can be delayed or heated. Because SFA can automate longer, hotter reactions, it allows slow reactions to proceed to completion as shown in the diagram.

20 min	1 min
95 °C	60 °C

The advantages of complete reaction are

- □ Sensitivity is the maximum possible.
- Small variations in reaction conditions such as changes in flow rate or temperature do not affect method sensitivity.
- When the method sensitivity remains constant, results stay within specification for longer and recalibration is necessary less often.



Reaction rate graph for a method requiring 4 minutes for complete reaction.

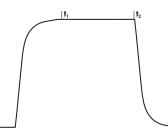
Sample concentration in the detector reaches a constant, maximum value

The output from a SFA system is a peak with a flat plateau. The plateau represents a constant "steady state" value in the flowcell at maximum concentration. The sample result is calculated from the peak height, which is averaged from readings on the plateau.

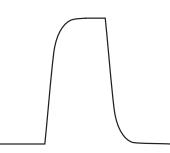
In contrast, the sample peak from a FIA system only reaches maximum concentration for an instant.

The "steady state" SFA peaks maximize sensitivity in two ways.

- ☐ The sample concentration in the flowcell reaches its maximum value, undiluted by wash or carrier solution.
- Maximum concentration is maintained for long enough to obtain an accurate reading.



Detector signal from a sample measured for a long time. Between t1 and t2 the concentration in the flowcell is constant.



Sample peak from a normal SFA analysis. The sampling time is long enough to allow several seconds steady state at maximum concentration.

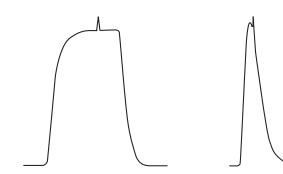
Sample peak from a normal FIA analysis.

Advantage 3

No interference from air bubbles

Degassing from samples or reagents can introduce unwanted air bubbles into the reaction stream. Whereas SFA systems are designed to work with bubbles in the reaction stream, air bubbles interfere with FIA in two ways.

- □ The pressure inside a FIA system is high, because 2-4 ml/min are pumped through tubing 0.8-1 mm internal diameter. Compression of the air bubble disrupts the regular flow which is necessary for reproducible results.
- As shown in the figures below, an air spike is easily separated from the height measurement of a SFA peak's flat plateau, but cannot be easily removed from a FIA peak, whether the area or height is measured.



SFA peak with interference from an air bubble in a reagent. The spike is eliminated from the average reading for the peak plateau and does not affect the result.

The same spike superimposed on a FIA peak affects measurement of height or area.

Low detection limit

Even at very low concentrations, maximum concentration in the flowcell is maintained for several seconds in an SFA system, so the signal can be distinguished from background noise. In contrast, the maximum concentration of a FIA peak exists for only an instant. Combined with the maximum sensitivity available from complete reaction, this results in a lower method detection limit for the SFA system.



SFA peak at low concentration. The complete plateau lies above the baseline, and an accurate average can be calculated.



FIA peak at low concentration. Only the small central portion lies above the baseline.

Advantage 5

Stable, robust analysis methods

A "robust" method delivers stable, constant results over a long time and is not affected by small changes or disturbances in the analysis conditions. Several factors contribute to the stability of SFA methods.

Even slow reactions go to completion: changes in reaction time or temperature do not affect the method sensitivity or the sample result.

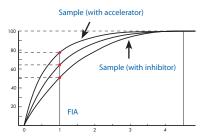
The diagram below shows the effect on method sensitivity of a 2% increase in reaction time. Such a small change can easily occur as the pump warms up during the analysis and flow rates drop. The SFA method sensitivity is not affected: the FIA method sensitivity changes by 4.4%.

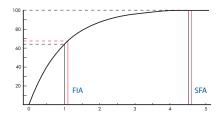
Each peak reading is the average of several at maximum concentration: interferences are easy to identify and separate.

The sample peak from an SFA analysis has a steady-state plateau at maximum concentration. As shown here, this plateau consists of multiple individual data points. The final result is the average of the readings from several separate liquid segments.

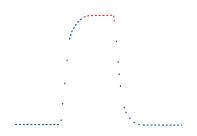
Changes in sample matrix do not affect the result.

This graph shows the effect on the analysis result from samples containing impurities which change the reaction rate.





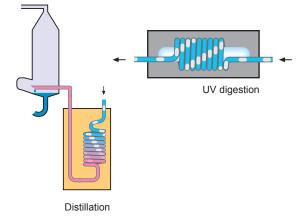
The effect on method sensitivity of a 2% change in reaction time



Sample peak data from an AutoAnalyzer running at 80 samples per hour, taken from the AACE software Retrieve Chart program, showing readings at 2-second intervals. Red dots show which data were examined by the peak reading algorithm. The final result is the best average of 5 from the last 8 points on the plateau.

Complex procedures can be automated

SFA can automate complex and long analytical procedures such as distillation and hydrolysis. Only the SFA digestion procedure is approved in the 2004 draft of the ISO standard method for Total Phosphorus with UV digestion.

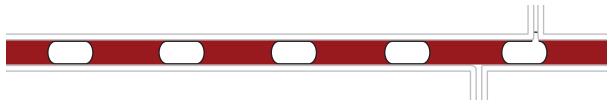


Advantage 7

Easy visual check for correct operation

The individual segments in SFA systems offer two advantages:

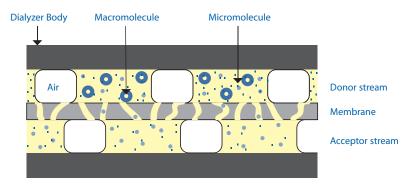
- The segments of liquid and air with a regular bubble pattern make it easy to see that the flow is correct and to identify the source of a problem such as a reagent which has run out or a tube which is blocked.
- The result for each segment is stored by the computer, so a complete run can be replayed for fault finding. The concentration of each segment can be displayed, to provide a validation check on the final result.



Regular bubble pattern. Each liquid segment contains the same proportion of reagents and sample, giving high reproducibility.

Interference from dirty samples is removed

Dialyzers in methods for waste water allow only small ions or molecules to pass through the membran. Large coloured compounds, protein, fat and suspended solids are pumped to waste.



The principle of dialysis

SEAL Analytical is a global company with offices worldwide - contact us at:

SEAL Analytical Limited. 2 Concorde Close Segensworth Fareham Hampshire PO15 5RT United Kingdom Tel: +44 (0) 1489 864400 Fax: +44 (0) 1489 880531 Email: mail@seal-analytical.com SEAL Analytical, Inc. Mequon Technology Center 10520-C Baehr Rd. Mequon, WI 53092 United States Tel: +1 (262) 241 7900 Fax: +1 (262) 241 7970 Email: sales@seal-us.com SEAL Analytical GmbH Werkstrasse 4 22844 Norderstedt Germany

Tel: +49 40 52202 100 Fax: +49 40 52202 473 Email: info.germany@seal-analytical.com



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