Discrete or Continuous Flow Analysis – Which is Better?

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A wide variety of factors affect the choice of analytical instrument. These include target workload (samples/hour), variety of chemistries, methods required, bench space, staff availability etc. In the following article Lalicia Potter, Technical Sales & Support Director at SEAL Analytical, examines one of the common decisions facing laboratory managers.

Background

The original Continuous flow analyser, the AutoAnalyzer was invented in 1957 by Leonard Skeggs and commercialised by Technicon Corporation. The first applications were for clinical analysis, but methods for industrial analysis soon followed.

The next generation Technicon AutoAnalyzer II (AAII) had an enormous effect on chemical analysis because it enabled laboratories to automate and dramatically increase the numbers of samples that were analysed in a day. In addition, this new automated technology substantially reduced the opportunities for error that existed in traditional manual techniques. The early instruments were able to process 30 - 60 samples per hour and as a result of the success of the AAII, most of the EPA methods for automated colorimetric analysis were based on this instrument.

The Technicon Industrial division, which included the AutoAnalyzer II, was purchased by Bran+Luebbe in 1987 and the AAII was replaced by the next generation AutoAnalyzer 3 (AA3) in 1998.

The TrAAcs800 was the first microflow segmented flow analyser introduced by Technicon. Microflow miniaturised the chemistry pathway, making the internal diameter of the glassware 1mm, rather than the 2mm or larger for standard AA systems. This enabled higher throughput, typically 100 – 120 tests per hour per channel. Microflow segmented flow systems are still the highest capacity nutrient analysers, with a 5 channel system reaching 600 tests/hour.

Discrete Analysers

While discrete analysers are often regarded as 'new' technology in the environmental market, they have actually been around since the early 1980's – in the clinical market. Synermed UK developed some of the first clinical discrete analysers for testing body fluids and generally utilise enzymatic chemistries.

Discrete analysers entered the environmental market in the early 2000's and quickly became popular because this highly automated technique offers low reagent use, low waste generation and true unattended operation. However, the needs of the environmental laboratory are very different to those of the clinical laboratory. Clinical applications generally measure values within a range – 'healthy', 'below' and 'above'.

Discrete analysers minimise operator involvement and can run many different chemistry tests from each sample in the same run.

Accuracy and precision is of course important but it is a different requirement to environmental analysis, where the concentration levels of analytes often approach zero and regulatory authorities are increasingly demanding lower detection limits.

Discrete analysers use a robotic sampling arm to aspirate and dispense precise quantities of sample and reagents in discrete reaction wells for incubation over a pre-programmed period of time, emulating the manual colorimetric methods. Then, depending on the type of analyser, the sample is either read directly by moving the styrene cuvette to the detector, or the sample is transferred into a static optical glass cuvette and the absorbance is read.

The appealing characteristics of discrete analysers include minimal operator involvement and the ability to run many different chemistry tests from each sample in the same run. Analysis is computer controlled, which means that only the tests necessary for each sample will be run. Sample pre-dilution and auto-dilution, auto spiking and auto bracketing of QC's should also feature in the software.

Cross contamination is a potential source of concern and the design of a discrete analyser for environmental applications should take this into consideration - converted clinical analysers would not be suitable.

Washing the fluidic lines between sample and reagent is of course important but another possible source of contamination is the droplets of liquid on the sampling probe. Unless cleaned, these can easily contaminate subsequent samples by dripping over the analyser itself.

A good way to assess the ability of a discrete analyser to conduct problem-free analysis of multiple methods would be to run ammonia (using Phenate), nitrate by cadmium reduction (using ammonium chloride buffer) and low level phenol together in the same run.

The reproducibility and detection limits of discrete analysers depend on a number of factors, including the mixing technique, the washing, the pathlength of the cuvette and the detection system.

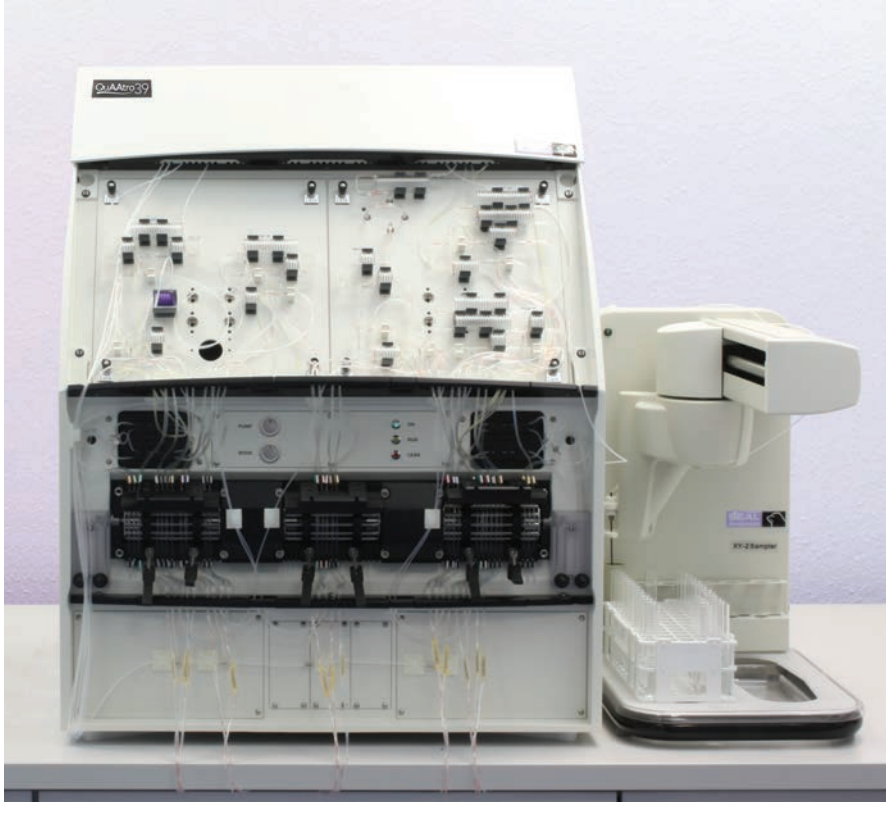
The reproducibility and detection limits of SEAL discrete analysers have been optimised by ensuring that each sample is read in the same optical glass cuvette with a 10mm pathlength. The sample is always read in the same position in front of the detector, which eliminates any potential issues with scratching or reaction well variability that can be found with direct-read systems. Since the liquid is moved and not the tray; fewer moving parts maximises reliability.

Most discrete analysers employ miniaturised components to reduce reagent consumption and waste costs. For example, both the AQ1 and AQ2 SEAL analysers use just 20 to 400µl of reagent per sample.

Segmented Flow Autoanalyzers

SFAs have considerably reduced the labour requirement and dramatically increased the workload capability of wet chemistry analysers by automating the entire process of mixing samples with reagents, applying the necessary reaction conditions for an appropriate period, and passing the resulting solution for photometric analysis. The reaction stream in an SFA is segmented with bubbles of air or nitrogen to reduce inter-sample dispersion. These bubbles are introduced at regular intervals forming unique reaction segments which are mixed using glass coils. Glass is preferred because it is inert, stays clean and enables easy visual checks.

Based on the original tried and tested technology of the Technicon/Bran+Luebbe AutoAnalyzer, today's SFAs deliver fast, accurate analysis for enormous numbers of samples; the QuAAtro for example can run up to 600 tests per hour. SFA's are now also highly automated and once the analyser is configured and the reagents and samples are loaded, reliable unattended operation is a major benefit.



SFAs have considerably reduced labour requirements and dramatically increased workload capability by automating the entire analytical process.

A basic SFA system consists of an autosampler, a peristaltic pump, a chemistry manifold, a detector and AACE data acquisition software. Sample and reagents are pumped continuously through the chemistry manifold and air bubbles are introduced at precisely defined intervals, forming unique reaction segments which are mixed using glass coils. With SFA, even slow reactions run to completion and the ratio of sample to reagents in the detector reaches a constant maximum value; the steady-state condition.

SFAs have been developed for running a few parameters on a larger number of samples, and the SEAL SFAs are the system of choice for marine and seawater organisations and anyone running very low nutrient waters. The SEAL AutoAnalyzer 3 and QuAAtro deliver high levels of performance and reproducibility, and are also the systems of choice for tobacco, soil and fertiliser testing around the world. These analysers provide maximum sensitivity by ensuring that the reaction always goes to completion, and with a digital true dual-beam detection system with real time referencing, the highest reproducibility and very lowest detection limits are achieved.

Factors Affecting the Choice of Analyser

As the manufacturer of an instrumentation range that includes both discrete analysers and continuous segmented flow analysers, SEAL Analytical's technical support chemists are often asked which is the better technique. Both offer fast, automated, colorimetric analysis of multiple samples, however, the answer depends on the current and future analytical requirements of the laboratory.

SEAL's discrete analysers employ sample trays and discrete reaction wells in which the colorimetric reaction takes place. In contrast, segmented flow analysers (SFA) employ a continuous flow of samples and reagent, segregated by air bubbles within tubing and mixing coils.

In general terms, discrete analysers are ideal when automation is a priority and/or when many and varied tests are needed on different samples. SFA is ideal when a larger number of samples are to be analysed for a smaller number of chemistries. However, both techniques are flexible, so it is important that expert advice is sought in the choice of analyser and that the instrument is configured to meet the precise needs of the laboratory.

Methods

It is obviously extremely important that analysers, both discrete and SFA, are configured with the most appropriate methods. SEAL therefore continually invests in method development and has an extensive library of over 1000 methods for the AA3, AA1, and QuAAtro SFAs. Applications for the older AutoAnalyzer II and TrAAcs systems are completely transferable to the new systems. The USEPA methodologies for automated colorimetric analysis reference the Technicon AutoAnalyzer II, and the AA3 is the latest version of this instrument.

SEAL provides a wide selection of multi-test manifolds which means that a large number of different parameters can be analysed with a single channel system. In addition, AutoAnalyzer segmented flow methods provide the best reproducibility and the lowest detection limits - crucial in applications such as seawater and low level pollutants such as cyanide and phenol.

SEAL also has USEPA approval for discrete analyser methods, with individual letters of approval for SEAL AQ1 and AQ2 environmental analysis methods. These letters state that the methods are acceptable for both wastewater (NPDES) and drinking water (NPDWR/NSDWR) compliance monitoring. In addition, SEAL's UKAS methods employed by the AQ2 and AQUA 900 discrete analysers are traceable to those methods published in the 'Methods for the Examination of Waters and Associated Materials', produced by the Standing Committee of Analysts in the UK.

In summary, when choosing the most appropriate analytical technique, it is important to consider both the current and likely future needs of the laboratory. However, one of the reasons behind the large numbers of SEAL instruments in laboratories around the globe, is that each analyser has been configured to meet the individual needs of its laboratory. So, it is good practice to contact the technical support team of the instrument provider at an early stage, because if the question is: "Which technique is better," the answer is: "It depends…"