

# New Technology Solves the Limitations of the Automatic Solid Phase Extraction coupled to Chromatographic Techniques

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## Introduction

Solid Phase Extraction (SPE) has long been the gold standard for laboratory sample clean-up but was considered cumbersome and prone to human error. Moreover, a major advantage of the SPE is its ability to automation which gives increased capacity of samples, less total time of analysis and greater precision and accuracy. Automation of SPE to date has required complex systems that automate sample preparation off-line or add unnecessary expense if integrated on the front end of an analytical instrument.

This poster presents a new technology for the coupling of SPE with chromatographs, which allows the user automate the SPE using only specific cartridges and an usual autosampler. The automation of SPE online with chromatography, in this case, is cost effective, and achieved with the instrumentation present in the laboratory through an easy step-by-step.

The concept is so simple: an autosampler syringe acts as the solvent reservoir for a SPE cartridge or syringe filter. The cartridge can be transported by the robot anywhere in the working envelope. By piercing a septum attached to the top of the cartridge, the analytical syringe can pressurize the cartridge to pass a fluid across the extraction bed or membrane filter in a highly controlled manner. Each sample is self contained for improved efficiency and productivity, so errors can be identified before a whole batch of samples has been processed. Likewise, chemistries can be screened quickly and easily.

## Instrument Top Sample Prep (ITSP™)

Traditional SPE cartridges require expensive complex automation, and extensive method development is often needed. So, while SPE is generally regarded as the “best” method of sample prep, it is often the last choice as it is labour intensive and costly.

ITSP™ (ITSP Solutions, Inc.) is a consumable cartridge designed to automate filtration and SPE that overcomes the shortcomings of on-line concentration listed below:

- Expensive "up-front" costs for equipment acquisition and setup
- Potential carryover risk where a trapping column is needed, for example in non-laminar flow column switching
- Complex hardware and software setup
- Extensive effort to validate trap column regeneration between samples

ITSP's solution of automated SPE removes these issues and actually offers precision with micro-scale solid phase extraction unachievable by manual techniques while also eliminating the massive expense of other in-line/on-line sample preparation products.

## Using ITSP™ on the Thermo Scientific TriPlus RSH Autosampler

ITSP integrates into the Operating Systems of most analytical instruments and prepares samples while the analytical instrument is analyzing a prior prepared sample.

Throughout this poster the Thermo Scientific TriPlus RSH autosampler will be mentioned. The TriPlus RSH can move the ITSP™ cartridge anywhere on the deck which allows significant flexibility in sample prep automation. The ITSP™ cartridge requires a minimal amount of hardware modification to enable a standard TriPlus RSH to prepare samples for GC applications. Modifying a TriPlus RSH does not take away any prior functionality, but simply adds the ability for the TriPlus RSH to perform ITSP filtration and SPE methods in addition to any previous methods it was performing.



Thermo Scientific TriPlus RSH Autosampler

## Explanation of the ITSP Cartridge

The ITSP cartridge combines several features from the familiar manual SPE column and vacuum manifold into a proprietary cartridge designed to miniaturize and automate sample prep methods. The cutaway drawing in Figure 1 illustrates the cartridge construction. The needle of the syringe is inserted through the septum. The gripping action of the septum allows the upper chamber of the cartridge to be pressurized so that solvents can be forced through the retaining membranes and the sorbent bed. Instead of a vacuum manifold, ITSP uses the syringe plunger to provide hydraulic force in either direction across the sorbent bed or filter membrane.

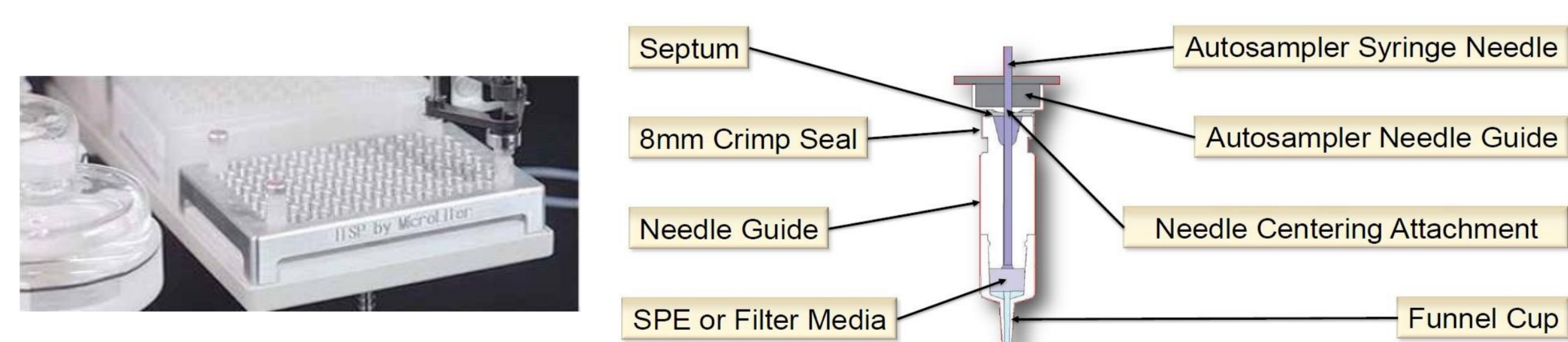


Figure 1: ITSP Device Components.

The precision of the TriPlus RSH is such that you can control flow rates at microvolume levels so that you gain a very high degree of precision yet at the same time a high degree of flexibility.

ITSP is bi-directional so filtration of samples that could damage the syringe are also possible using ITSP. The TriPlus RSH dips the cartridge tip into the raw sample and aspirates only the filtrate into the syringe.

The precise movements of the TriPlus RSH allow the cartridge moving around the platform. The needle stays inserted deep in the cartridge while the TriPlus RSH Injection Unit moves to the location where the sample is eluted. The cartridge grips the needle with the septum to keep the cartridge at a precise vertical orientation on the needle. The needle guide is programmed to remain in its position so that the cartridge is not pushed off the needle until it is safe to release it.

The capacity of an ITSP cartridge is not limited by its small size because the analytical syringe of the TriPlus RSH replaces the liquid reservoir of a manual SPE column. ITSP cartridge capacity is determined by a wide variety of factors including:

- Sorbent bed mass
- Affinity of the analytes of interest to the selected sorbent phase
- Affinity of the interference compounds in the sample matrix to the selected sorbent phase
- Size of the syringe times the number of loading cycles in each step of the method

Manual SPE methods are typically scaled down by 10<sup>1</sup> order of magnitude when automated using ITSP. Typical solvent and sample volumes are in the 100µL range using sorbent bed masses from 10 mg to 25 mg.

The cartridges sit in a specially designed Prep Tray over a PTFE waste receptacle that has an attached drain hose so waste can be automatically diverted to an appropriate receptacle.

Most of the SPE steps – conditioning, sample loading, and the column wash after loading – are performed in this position. Once those steps are complete the TriPlus RSH aspirates a volume of elution solvent, pierces the septum and inserts the needle to just above the sorbent; retracts the cartridge into the tower; moves to an open well in the elution tray and dispenses the elution solvent through the sorbent to elute the compounds of interest into the open well. Once the elution is complete the cartridge can be returned to the Prep Tray. The TriPlus RSH can then perform a standard injection cycle on the eluate and starts preparing the next sample while waiting on the GC to perform its analysis cycle.

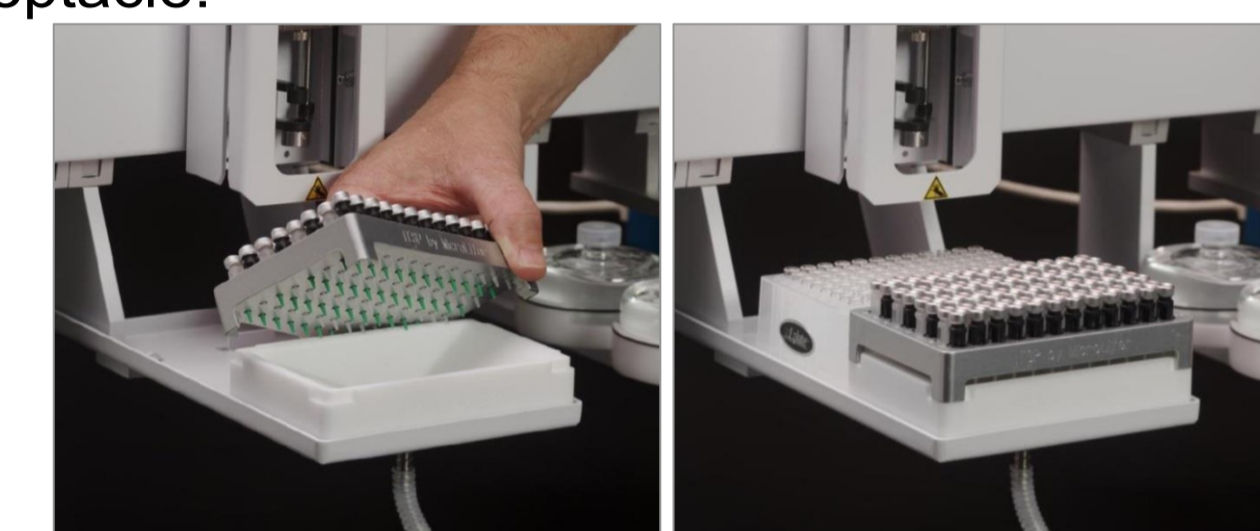


Figure 2: ITSP Prep Waste Station.

## Automated Method Development

A 96 well microplate and a set of macros automates method development based on application notes provided by most sorbent manufacturers. It is also an excellent tool for scaling down methods that can increase the productivity of the lab while at the same time reduces the costs of solvents, raw sample requirements and hazardous waste disposal.

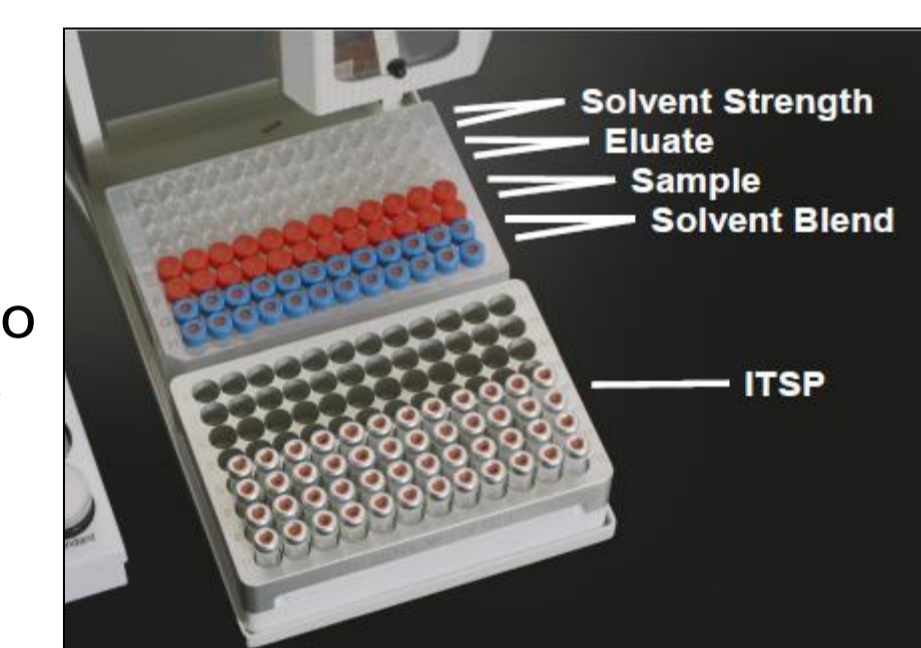


Figure 3: Method Development Tray Setup.

## Example of ITSP application

The kiwi fruit extract contain oils. These interferences are removed by ITSP cleanup (Figure 4).

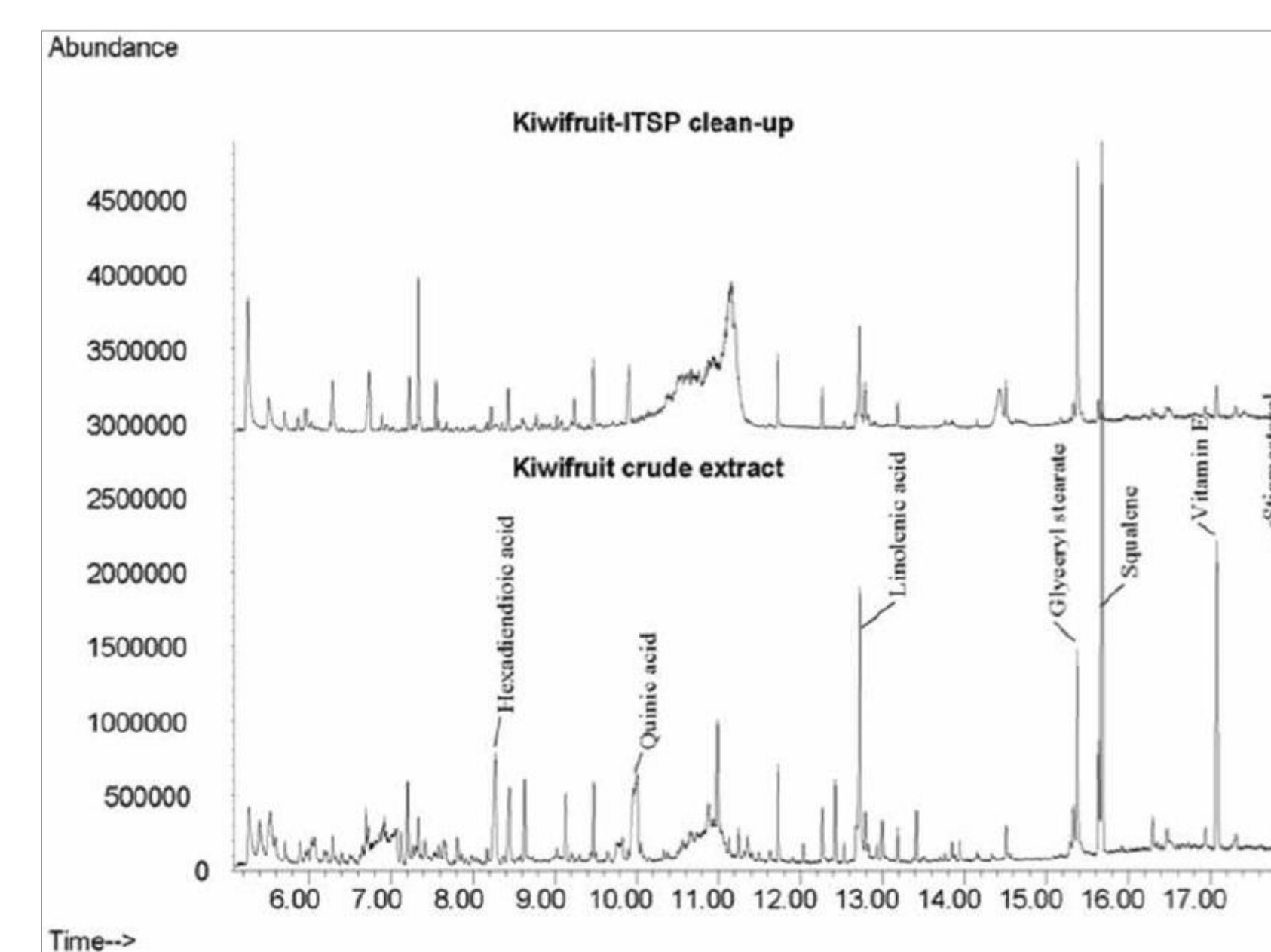


Figure 4: GC-MS full-scan chromatograms showing removal of non-polar matrix by GC-ITSP.

## Final Comments

Combining the sample preparation with the analysis of samples by GC and GC-MS can provide significant reductions (some labs report 90% or more) in solvents, client sample requirements, labor and hazardous waste for sample prep making it a “Green” solution for your lab.

One of the advantages of ITSP SPE is the ability to perform automated sample extraction, cleanup, and extract injection in-line.

ITSP offers an excellent opportunity to improve the performance and throughput of the analytical lab.