



**Spark Holland introduces
INTEGRITY™**

Resetting the standard in
LC autosampling

BETTER **SAMPLE** CARE



Sample Barcode Identification



Injection Performance Monitoring

Resetting the standard in LC autosampling

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Resetting the standard in LC autosampling

State-of-the-art injection technology is important, naturally. And rapid sample turn-around time is very helpful, of course. Large sample capacity is very convenient, no doubt. But this all becomes irrelevant if you have only the slightest doubts about the integrity of the samples that you injected! Yet, while injection technology and capacity have seen great advances over the last 30 years, measures to maintain and control sample integrity have had little attention from instrument manufacturers. Until now.

The new INTEGRITY™ autosampler from Spark sets a new standard in sample care.



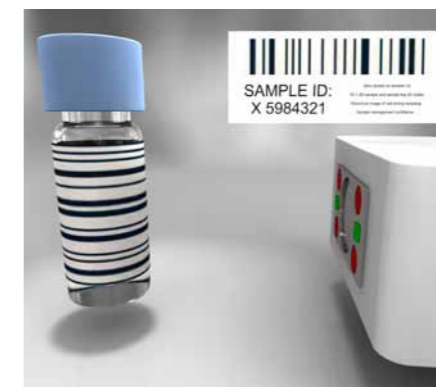
- Sample and tray bar code reading
- < 20 sec injection cycle
- True 4 °C sample cooling of all samples
- Injection performance monitoring
- Vial bottom detection – inject 1 µL out of 1 µL
- Advanced wash capabilities to eliminate carryover
- Dual independent concentric needle concept
- Optimized for UHPLC
- Advanced reagent addition and derivatization
- Integrated on-line sample prep capabilities
- Up to 4 well plates or 216 standard sample vials
- Flexible workstation for many liquid handling needs of analytical samples

Sample care first

A number of innovative features ensure better and safer control of handling and tracking of samples – ensuring that you feel much more confident about the results.

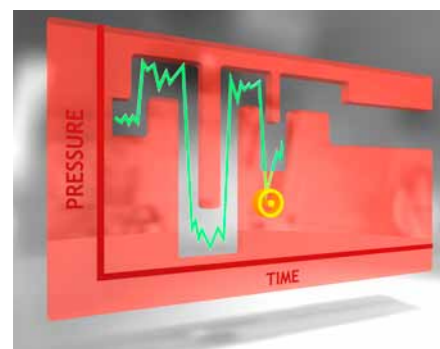
SBIT™ - Zero doubts on sample identity

1D and 2D sample and sample tray identification codes can be read, filed and linked to sample assay results. It is even possible to record an image of the vial during sampling for visual verification of the injection and the sample vial afterwards. Combined with positive feedback motion control for the sample tray positioning, the risk of assigning results to the wrong sample is zero.



IPM™ - Injection performance monitoring

Injecting the proper sample is one thing but how to verify that the injection itself has been performed correctly? The answer is pressure monitoring of the sample flow path during the entire injection cycle! Pressure profiles of injection cycles are recorded for every injection and by comparison with reference profiles, indications for malfunctioning caused by blockage, leakage, air



bubbles, needle damage, valve problems, etc can be easily recognized in case of suspected erroneous results. The injection pressure profile (“pneumogram”) can be linked to assay reports for easy verification afterwards. Obviously, preventing malfunction is better than correcting the consequences. Therefore INTEGRITY™

also checks for needle obstruction and missing vials before injecting a sample. Plus, the injection pressure monitor prevents formation of air bubbles by adjusting the aspiration speed of the dispenser in case of viscous samples or partly blocked needle. Improper injections have become virtually impossible, but if it happens – you will know it!

STC™ - Accurate sample temperature control - low and high!

Many biological samples require cooled storage during processing to avoid deterioration. Therefore, many autosamplers offer cooling, but very few autosamplers offer adequate cooling across the entire

sample space. INTEGRITY™ not only offers cooling, but also offers sample temperature control from 4°C up to 40°C for the entire sample tray and sample processing space. Actual readout of tray temperature shows when the autosampler is ready to accept new samples. Actual tray temperature can also be linked to sample assay results to verify correct sample temperature afterwards. And, in addition, the cooling device has been designed to reduce condensation in the sample area to a minimum. Elevated sample temperature also improves sample solubility and thus helps to prevent precipitation of poorly soluble analytes and adsorption of analyte to vial walls.



solvent pump provides wash solvents to the needle pair, and a smart combination of air pressure and solvent streams ensures thorough cleaning of the needle pair, inside and outside, in seconds. And also provided – jet-stream drying of the needles before entering the next sample! Multiple solvents can be selected by the wash pump for maximum clean-up ensuring zero carryover for the stickiest samples. Because the wash pump is much faster than the syringe-dispenser used for the injection, needle rinsing is not only better, but also much

faster. INTEGRITY™ can do an entire injection-wash cycle in less than 20 seconds!

Micro injection volumes and micro sample waste

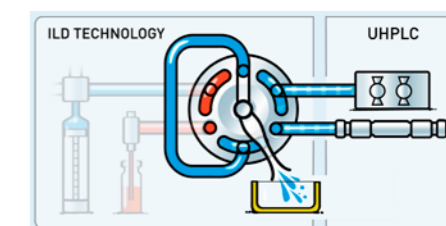
Using a micro syringe for accurate aspiration of small sample volumes, even nanoliter volumes of samples can be injected with high precision and accuracy. The microliter-pick-up mode reduces sample loss to zero and vial bottom detection enables INTEGRITY™ to position the sample needle at just a few tenths of a mm above the vial bottom to make sure you get all your precious sample injected reliably, independent of size variations in vials. Injection cycle time is not compromised by the reduced syringe volume because of the separate wash pump taking care of wash solvent delivery!

ILD™ technology for accurate UHPLC injection

A special injection valve makes INTEGRITY™ ready for injections into UHPLC systems up to 120 MPa

(18,000 PSI). Intermediate Loop Decompression (ILD™) is a new innovative feature on our injection valve for UHPLC. The proprietary design (*) comprises a unique stator-rotor combination with an extra radial relief slit. This configuration enables loop decompression prior to sample aspiration. Ultra-fast valve switching further reduces pressure shocks to a minimum. The result is highly accurate sample injection down to 1 µL and much longer column life time. Combined with the entire range of sample care and sample prep features, INTEGRITY™ adds unsurpassed performance to your UHPLC assays.

(*) Patent pending



Proven injection technology perfected

Our robust concept of closed line sampling using pressure assisted sample aspiration (PASA™) directly from vial to valve, has proven its reliability in more than 30,000 autosamplers. Simplicity of the concept, the absence of a needle port and a syringe that is never in contact with the sample are the most prominent features contributing to its success. With special attention to handling small samples, carryover and compatibility with UHPLC, INTEGRITY™ now brings this concept to a new level of perfection!

PASA™ - Familiar injection modes with higher performance level

Our well known partial loop fill, full loop fill and microliter-pick-up modes are again available on INTEGRITY™. Plus, by reducing the volumes of needle and tubing, adding pressure feedback and refining the needle control, these modes now perform better than ever. Independent motion control for sample needle, piercing needle and vials stripper provides maximum freedom for sampling, needle wash

and vial types. Rigid seals can be pierced without risk of damaging or blocking the sample needle, while the sample needle can be optimized for volume and inertness. PEEK, steel, or even fused silica tubing can be selected to serve as sample needle for optimal sample compatibility. A snap-in needle mount allows needle exchange in seconds. A built-in air pump provides air via the piercing needle, enabling the PASA™ injection concept for bubble free aspiration and assisting needle

wash for the concentric needle pair. Pressure monitoring, plus feedback control during the injection, enable self-adjustment of dispenser speed and valve switching delay times in case of variation in sample viscosity, needle restriction, air bubbles, etc.

Separate needle wash pump for better needle wash and faster injection cycles

A completely new concept for needle wash has been designed for INTEGRITY™. A separate built-in

Integrated sample prep features

Spark has always been leading in innovative integration of sample prep functionality into front-end sample handling systems. We introduced such capabilities as pre-column derivatization, column switching and on-line SPE as integrated functions of autosamplers and front-end systems. No doubt, INTEGRITY™ will strengthen our reputation further!

HoMix™ – Sample homogenizer/reagent mixer

HoMix™ is a separate device for mixing sample with reagents or for sample homogenizing prior to injection and is a unique innovation for HPLC autosamplers. Three separate

mini pumps can provide different reagents from external reservoirs to special self-cleaning overflow wells on the sample tray holder. Sample and reagent volumes are picked up and dispensed into destination vials using the sample needle and syringe

dispenser. Destination vials are then picked up from the tray by HoMix™ and their contents mixed by a very efficient, tumbling-like movement. After placing it back into the tray, the mixture is injected using any of the three available injection modes.



Heated
Capillary



Integrated
Stream
Switch



Double
Independent
Concentric
Needles



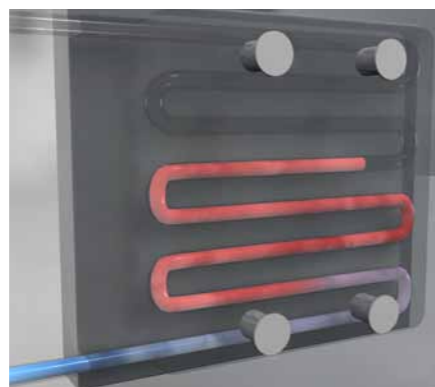
Mixing variables such as tumbling speed and duration can be programmed. Compared to the syringe aspirate/dispense actions offered by other autosamplers, mixing is much better and faster! Typical applications of this unique feature are internal standard addition, dilution, pre-column derivatization, homogenizing etc. Even in-vial liquid-liquid micro extraction is feasible.

HotCap™ - Individual heating of samples and reaction mixtures
Cooling helps to maintain sample integrity, but when deliberate changes of samples are required prior to injection, heat is our best

help. HotCap™ is a piece of capillary tubing that can be heated up to 100°C. When used as sample loop or other position in the sampling line, samples or sample/reagent mixtures can be heated before injection. Typical applications include pre-column derivatization, protein denaturing to reduce protein-analyte binding, disruption of cells, etc. HotCap™ heats up in seconds, has actual temperature read-out and a hardwired temperature limiter for maximum safety.

Controlled elevated temperature for sample trays

The entire sample tray of



INTEGRITY™ can be thermostatted between 4 and 40°C to facilitate such applications as stability testing or to study degradation under physiological conditions at 37°C.

ISS - Integrated stream switching for on-line sample prep

Our favorite way of sample prep is becoming more and more popular! Spark pioneered on-line sample prep (solid phase extraction, column switching) and introduced on-line sample prep as an integrated option on an autosampler over ten years ago. Today, on-line sample prep using high pressure stream switching is considered the best approach to obtain high assay quality and maximum automation. In addition, on-line sample prep using valve switching provides a closed sample processing system – another plus for sample integrity. INTEGRITY™ can be equipped with two extra (ISS) valves, in addition to the standard injection valve.

INTEGRITY^{PLUS}: extended sample capacity

A special version of INTEGRITY™, the INTEGRITY^{PLUS} doubles the sample capacity. INTEGRITY^{PLUS} accommodates 4 well plates or 216 standard vials. Special trays for micro-vials increase sample capacity even further! INTEGRITY^{PLUS} also allows use of well plates in one tray set and (micro) vials in the other, or vice versa for sample reformatting, dilution, etc. Rapid tray positioning will keep your method fast, even when alternating between different trays of vials and wells. Plus... all trays are temperature controlled between 4°C and 40°C!

Dependable OEM partner

Designing instruments for OEM business has become second nature to us. Few instrument companies are so dedicated to customization of hard- and software according to their customers' needs. We aim to be your partner all along the way – from designing the product

to creating and integrating its driver in your software package. We have over 30 years experience in development and production of HPLC instruments and have delivered over 30,000 autosamplers. Comforting numbers if you demand a dependable partner in HPLC instrumentation.

Connecting INTEGRITY™ to the world

For comprehensive control of the extensive capabilities of INTEGRITY™, Spark developed a control software package. The software allows maximum use of the entire INTEGRITY™ functionality in any order of events, to create even the most exotic methods. Methods are linked to samples in a run table for automated processing of sample batches. Both software I/O and hard-wired I/O enable communication with other software and hardware to create full PC control for your entire analytical system.

Drivers for third party software packages

Using the Spark communication protocol, INTEGRITY™ control can be integrated into most third party

software packages. Spark has extensive experience with integrating instrument control into the software packages of our OEM customers, and has assisted in the creation of

a number of drivers for our instruments in commercially available software products for system control and data acquisition. Please consult us for your particular application!



Integrity™ Specifications

General specifications	
Injection concept	Pressure Assisted Sample Aspiration (PASA™) using ~ 7 PSI sample head-space pressure (built-in compressor) to avoid air bubbles in sample lines. Unsealed wells or vials can be used without PASA™
Injection volume	Programmable from 1 – 10000 µL. Actual volume range depends on installed sample loop volume
Injection Performance Monitoring (IPM™)	Pressure sensor in sampling flow path generates pressure-time plot for diagnostic purposes. - 0-9999 mbar absolute pressure read out - sampling frequency: 10ms For OEM purposes: other sampling frequencies are possible, max 30,000 data points
Needle piercing accuracy	± 0.6 mm
Needle wash	Freely programmable volumes and speed for wash of sample needle. 2 solvents for inside surface and 2 for outside surface of needle from external wash solvent bottles. Note: Option to extend inside and outside wash with 2 extra solvents
Dispenser syringe range	25, 50, 100, 250, 1000 and 2500 µL (user exchangeable; screw lock)
Sample capacity	- 2 well plates according SBS dimension standards - 96 well and 384 well format, low- and high-well (2 plates must be of the same type) - 108 2 ml Chromacol vials or 88 Sarstedt microtube vials (vial adapters required) - see INTEGRITY ^{PLUS} for more sample capacity
Maximum vial or plate height	48 mm (including septa or cap-mat)
Mix and dilute	4 reagent positions of 450 µL each Standard 1 reagent pump for replenishment (also used for µL pick-up injections) Option: 3 extra reagent delivery pumps Note: reagent aliquots are dispensed into vials/wells via sample needle and mixed by asp/disp actions or using HOMIX™ (for suitable vials only)
Valve switching time	< 100 msec
Wetted parts in sample flow path	SS316, PTFE, ETFE, VESPEL®, Glass, PPS
Wetted parts in dispenser and wash lines	ETFE, PTFE, PEEK, Kalrez, Glass, PCTFE

Safety and compliancy	
Safety and EMC compatibility	According to EC-directives. cCSAus (CSA – UL) approved
Installation class	II
Pollution degree	2
Quality	ISO 9001 certified
Missing vial/well plate sensor	Stops run when detecting missing well plate. User selectable response to missing vial: skip sample or stop run
Illumination LED	Full color LED with on/off control. Color selectable For OEM: LED control possible through software
Door sensor	Upon opening the front door, sample processing will be stopped and the tray will be moved to front after completing the current sample run

Electrical	
Power requirements	100 – 240 Volt AC +/- 10%. 50/60 Hz
Power consumption	450 VA

Environment	
Sound pressure level	LAeq <70 dB
Working temperature	10 - 40°C
Storage temperature	-25 / + 60°C
Altitude	Max 2000 m
Humidity	20 - 80% RH

Physical	
Max load on top cover	15 kg
Dimensions	(Width x Depth x Height)
- Standard INTEGRITY™ (HPLC/UHPLC/Micro)	330 mm x 540 mm x 480 mm
- Standard INTEGRITY™ + sample cooling	330 mm x 620 mm x 480 mm
- INTEGRITY ^{PLUS}	330 mm x 680 mm x 480 mm
- INTEGRITY ^{PLUS} with sample cooling	330 mm x 760 mm x 480 mm
Weight	
- Standard INTEGRITY™ (HPLC/UHPLC/Micro)	26 kg
- Standard INTEGRITY™ + sample cooling	30 kg
- INTEGRITY ^{PLUS}	31 kg
- INTEGRITY ^{PLUS} with sample cooling	35 kg

Instrument control	
Graphical User Interface	SparkLink instrument control software including Integrity driver
PC interface	2x Sub-D connector with RS232 protocol and Spark Multilink connection Ethernet RJ45 connector USB
Outputs	5 programmable relay outputs; NO and NC connectable 1 programmable TTL output; programmable as: Inject marker, Auxiliary or Alarm
Inputs	4 programmable TTL inputs for OEM applications



Specifications of various INTEGRITY™ versions

	Integrity HPLC	Integrity UHPLC	Integrity UHPLC Micro
Max operating pressure	35 MPa (5000 psi)	126 MPa (18.000 psi)	84 MPa (12.000 psi)
Loop volume	Standard: 50 µL Other loop sizes possible	Standard: 20 µL Other loop sizes possible	10 µL
Injection volume With standard loop volume* *) larger volumes possible with larger sample loop	- Full loop: 50 µL - Partial loop fill: 1 – 25 µL - µL pick-up: max 15 µL	- Full loop: 20 µL - Partial loop fill: 0.5 - 10 µL	0.05 – 1 µL
Injection precision With standard loop volume	Full-loop: < 0.3% RSD Partial loop-fill: < 0.5% RSD* µL -pick-up: < 1.0% RSD* *) injection volume > 5 µL	Full-loop: < 0.3% RSD Partial loop-fill: < 0.5% RSD* µL -pick-up: < 1.0% RSD* *) injection volume > 1 µL	Full-loop: < 0.3% RSD Partial loop-fill: < 0.5% RSD for injection volume > 0.5 µL; < 1% RSD for injection volume > 0.2 µL µL -pick-up: < 1.0% RSD for injection volume > 0.2 µL
Sample needle	SS 0.25 mm i.d. Volume 10 µL (including connecting tubing)	PEEK SIL 0.2 mm i.d. Volume: 7 µL (including connecting tubing)	PEEK SIL 0.15 mm i.d.; volume 3.6 µL
Sample buffer tubing tubing between syringe and injection valve	Tefzel, 1.0 mm i.d. Volume 500 µL	Tefzel, 0.75 mm i.d. Volume 200 µL	PEEK, 0.5 mm i.d.; volume: 100 µL
Injection valve* *) other valves available on special order	SS stator, PEEK rotor seal, 0.4 mm bore, 1/16" connections	UHPLC valve with ILD™ Coated SS stator VESPEL rotor seal 0.25mm bore 1/16" connections	Coated SS stator, VESPEL rotor seal, 0.15 mm bore, 1/32" connections.
Dispenser Syringe volume	250 µL	100 µL	50 µL
Injection cycle time	Typically 30s, including default wash in partial loopfill mode	30 - 60 sec. depending on selected conditions for injection and wash speed	~ 1 minute depending on injection and wash conditions
Carry-over	< 0.005% under specified conditions	< 0.005% under specified conditions	< 0.01% under specified conditions

INTEGRITY^{PLUS}

HPLC/UHPLC version with extended sample capacity. Sample capacity extended with 2 SBS well plates. Or 2 trays of 108 2ml vials. Or 2 trays of 88 Sarstedt microtubes. Plate/tray exchange within 3 seconds.

Sample cooling/heating

Temperature range	4-40°C
Temperature accuracy	+/- 2°C
Cooling capacity	Standard Integrity: Maximum ΔT = 21 °C ; INTEGRITY ^{PLUS} : maximum ΔT = 20 °C ΔT = difference between ambient and sample tray temperature Peltier technology

Options

HoMix™; Homogenizer/mixer module	
Vials	Chromacol vials with crimped caps (e.g. 2-CV, 2-CVR, 09-FIV, 1.1-CTVG) Sarstedt screwcap micro tubes (e.g. 72.694.007, 72.703.406, 72.730.406)
Agitation speed	11 speeds selectable
number of agitation cycles	1 - 999

SBI™; Barcode reader	
1D Barcodes for vials or well plates	Code 39, Code 128, Codabar, UPC, EAN, Interleaved 2 of 5, Reduced space symbology, Code 93, Codablock
2D codes for well plates	PDF417, MicroPDF417, Maxicode, Data Matrix, QR code, Aztec, Aztec Mesa, Code 49, UCC Composite
Number of characters	Max. 20
Scan speed	< 0.5s

HotCap™; Heated capillary	
Temperature range	40-100°C Hardwired temperature limiter
Increments	1°C
Accuracy	+/- 2°C
speed	34 s from 25°C to 75°C at 1ml/min

Extended reagent delivery	
3 extra solvent delivery pumps for reagent addition, connected to reagent positions 2, 3 and 4. Replenishment volume of reagent positions on tray is 450 µL.	

Extended wash solvent delivery	
Inside needle wash	2 extra solvents
Outside needle wash	2 extra solvents

ISS 6-port (max 2 per Integrity)	
One extra high pressure 6-port switching valve. Type and make same as injection valve.	
Switching time	< 100 ms

ISS 10-port (max 2 per Integrity)	
One extra high pressure 10-port switching valve	
Switching time	< 100 ms

Ordering information

INTEGRITY™ versions All versions include SparkLink PC control software	
INTEGRITY™ HPLC	SP950.000
INTEGRITY™ UHPLC	SP950.200
INTEGRITY™ UHPLC Cool	SP950.201
INTEGRITY™ Micro UHPLC Cool	SP950.206
INTEGRITY™ UHPLC full option sample cooling, multiple wash solvents, barcode reader, HoMix™ and 4 reagent pumps	SP950.299
INTEGRITY™ UHPLC Plus	SP951.200
INTEGRITY™ UHPLC Plus - Cool	SP951.201

Options (factory installed)	
Extended wash solvent delivery	0950.852
Extended reagent delivery (3 additional reagent pumps)	0950.864
HoMix™ (Homogenizer/mixer module)	0950.860
HotCap™ (heated capillary)	0950.863
SBI (plate/vial barcode scanner) Important note: HoMix module must be installed for reading bar codes on vials	0950.861
ISS 6-port (one additional 6 port switching valve) Note: max 2 additional switching valves can be installed	0950.862
ISS 10-port (one additional 10 port switching valve) Note: max 2 additional switching valves can be installed	0950.865
Prep LC (Serum sample needle, Prep vial adaptor, large-bore injection valve, 2.5 mL syringe and 10 ml sample loop)	0950.866
Bio compatible HPLC (PEEK injection valve and 100uL PEEK sample loop, bio-needle)	0950.867

For more information:

Now interactively explore for yourself all the animated features of the INTEGRITY™ at www.bettersamplecare.com

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