

SPARK OPTIMAS

Clarity Control Module

ENG

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To facilitate the orientation in the **Spark Optimas** manual and **Clarity** chromatography station, different fonts are used throughout the manual. Meanings of these fonts are:

Open File (italics) describes the commands and names of fields in **Clarity**, parameters that can be entered into them or a window or dialog name.

WORK1 (capitals) indicates the name of the file and/or directory.

ACTIVE (capital italics) marks the state of the station or its part.

Chromatogram (blue underlined) marks clickable links referring to related chapters.

The bold text is sometimes also used for important parts of the text and the name of the **Clarity** station. Moreover, some sections are written in format other than normal text. These sections are formatted as follows:

Note:	Notifies the reader of relevant information.
Caution:	Warns the user of possibly dangerous or very important information.

Marks the problem statement or trouble question.

Description: Presents more detailed information on the problem, describes its causes, etc.

Solution: Marks the response to the question, presents a procedure how to remove it.

1 Spark Holland Optimas control module

This manual describes the setting of the **Spark Holland Optimas** autosampler. The control module enables direct control of the instrument over serial line.



Fig. 1: Spark Holland Optimas

Direct control means that the autosampler can be completely controlled from the **Clarity** environment. Instrument method controlling the sample preparation conditions will be saved in the measured chromatograms.

Note: It is recommended to check the user manual of the autosampler for its operating principles and restrictions.

2 Requirements

- Clarity Installation with AS Control module (p/n A26).
- Serial straight DB9F-DB9M cable (p/n SK02).
- *Note:* Cables are not part of **Clarity** Control Module. It is strongly recommended to order required cables together with the Control Module.
 - Free serial COM port in the PC.
- *Note:* Modern computers usually have only 1 (if any) serial (COM) port installed. To use more devices requiring the port, the **MultiCOM** adapter (p/n MC01) is available.
- *Caution:* Communication is only possible if the Spark Optimas instrument has communication (serial) port. Typically Spark Optimas samplers are supplied without a communication port. Samplers supplied to Knauer company typically have the communication port.

3 Installation Procedure

3.1 Hardware - Wiring

Commands for the autosampler are communicated with **Clarity** through the communication cable given by the communication board installed in the **Spark Optimas** autosampler. All cables that can be used (described in the chapter **"Requirements"** on pg. **2**.) are common standard and can be acquired either from **DataApex** company or from local computer stores.

3.1.1 Connections of the autosampler and chromatographic system

The connection of the whole chromatography set is dependent on the many factors, such as control modules available for each particular part of the set. The common options for the **Spark Optimas** autosampler will be either all modules controlled, or some of them not controlled. The typical wirings are shown on schemes below:

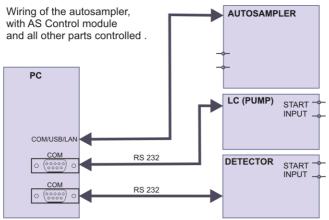


Fig. 2: Wiring of the autosampler - all parts of the set controlled

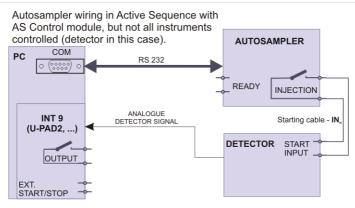


Fig. 3: Wiring of the autosampler - some parts of the set not controlled

Note: Other typical connections of a set with Autosampler can be found in the **Getting Started** manual (chapter **Device Setup and Wiring**).

3.2 Spark Optimas setup - communication

The **Spark Optimas** autosampler can be controlled from **Clarity** via serial line.

In addition, the *ID* of the **Spark Optimas** autosampler must be the same that will be later set in **Clarity**. The ID is hard-set to the value of 60 for **Spark Optimas** autosamplers.

3.2.1 Digital Inputs and outputs

The digital inputs and outputs of the **Spark Optimas** autosampler are both present on the back panel of the instrument and simulated over the communication line. In usual cases, the outputs do not have to be connected by wire with the autosampler as they are communicated to **Clarity** digitally.

However, when other instrumentation needs to be acknowledged of the analysis start etc. by wire, the I/O connector on the back panel of the autosampler may be used. The mapping of the pins on the connector is as follows:

Pin	Function
1	Output - Common
2	Output - Normally Open
3	Input 1
4	Input 2
5, 7, 8, 9	GND
6	Output - Normally Closed

Tab. 1: I/O connector pins mapping

3.3 Clarity Configuration

System Configuration			— 🗆 X
Setup Control N	Iodules	5 ber of Instruments:	
Name Use	d I	🕜 Instrument 1 🖨 Instrument 2 🖨 Instrume	nt 3 🔘 Instrument 4
GC Detector Balance Valve Fraction Collector Capillary Electrophoresis		Name Instrument 1 Instrument Type LC Name From Sampler 1 Knauer 0	E
L 🚘 Auxiliary	< <	Detector Thermostat Valve Fraction Collector Data Input: & Outputs Ext. Start Dig. Input: Ready Dig. Output:	ce Number ⁽⁸⁾
		Miscellaneous Settings	
		Units Setup	Method Options
Add Remove About	Setup	ОК	Cancel Help
Available Control Modules			– 🗆 X
	Installed Only Filter: All	v optimas 2	0
Name	Status Vendor Com	nent Mo	dule Info
Knauer Optimas	installed Knauer		
GC GC Detector Detector Detector Detector Thermostat Valve Valve Higher Fraction Collector Copillary Electrophores Copillary Electrophores	is		
Add Cancel			Help



- Start the **Clarity** station by clicking on the A icon on the desktop.
- Invoke the *System Configuration* dialog accessible from the *Clarity* window using the *System Configuration...* command.
- Press the *Add* button ① (see **Fig. 4** on pg. **7**.) to invoke the *Available Control Modules* dialog.
- You can specify the searching filter 2 to simplify the finding of the driver.
- Select the **Optimas** sampler and press the *Add* 3 button.

The Spark Optimas Setup dialog will appear.

Serial Port	COM1	~
ID	60	AutoDetect
Status	Unknown	
Sampler Name	Sampler 1	
	Sequence Mode	
	Digital Input Names	Change

Fig. 5: Spark Optimas Setup

• Select the correct *Serial Port* and fill in the correct *ID* for the autosampler. Then press the *Autodetect* button. If the communication is correct, the *Connected* inscription along with the firmware version and serial number of the **Optimas** autosampler will be shown in the *Status* row. The *ID* must match the device identifier set previously in the autosampler setup, as described in the chapter "Spark Optimas setup - communication".

Note: The <u>Spark Optimas Setup</u> dialog is more closely described in the chapter **"Spark Optimas Setup"** on pg. **22**.

The **Optimas** autosampler item will appear in the *Setup Control Modules* list of the *System Configuration* dialog.

- Drag and drop the **Optimas** icon from the Setup Control Modules ④ list on the left side of the System Configuration dialog to the desired Instrument ⑤ tab on the right side ⑥ (or use the -> button ⑦ to do so).
- Set the *Ext. Start Dig. Input* and *Ready Dig. Output* numbers ⁽³⁾ for your acquisition card according to the wires being used for synchronization. If you wish to synchronize the **Clarity** start with the autosampler over serial line, you can set the **Optimas** in the *Ext. Start Dig. Input* drop-down menu, using the *1* as a input *Number*. In such case it is necessary to change the behavior on the *Method Setup Measurement* tab later from *Down* to *Up*, else the start signal will be delayed by pulse length (approximately 2 seconds).

4 Using the control module

New <u>Method Setup - AS</u> tab appears in the *Method Setup* dialog, enabling the setting of the **Spark Optimas** autosampler control method.

4.1 Method Setup - AS

The *Method Setup - AS* dialog consists of six sub-tabs assigned for the various parts of the **Spark Optimas** autosampler method. These sub- tabs are<u>Injection,Inputs & Outputs,Mix,System,Spec. Vials</u> and<u>Tray</u>. Additional buttons allow to display the <u>Hardware Configuration</u> dialog of the **Spark Optimas** autosampler or to read the instrument method from the **Spark Optimas** autosampler. The method is sent to the autosampler every time the *Send Method* or *OK* button is pressed. Other actions in different windows may also cause the sending of the instrument method to the controlled devices including the **Spark Optimas** autosampler - most notable cases being pressing the *Send Method* button in the *Single Run* dialog or starting a new injection from the *Sequence* window (each injection is preceded with sending the instrument method).

To read the **Spark Optimas** method from the autosampler it is necessary to use the *From AS* button available from all sub-tabs of the *Method Setup - AS* dialog. If the injection method is already established in the sampler, it is advisable to download it to **Clarity** using the *From AS* button and save it as a **Clarity** method.

4.1.1 Injection

Method Setup Default2 (MODIFIED)			×
New Open Save Save as Report setup Audit trail Send method by e-mail			
Select Sampler 1 🗸 🗸 Enabled			
Knauer Optimas Sampler Method AS Status			
Injection Inputs & Outputs Mix System Spec. Vials Tray			
Use Timebase Methods Use Mix Method Use Mix Method Use Injection Mode Analysis Time [min.] Flush Volume [j.L] Tubing Volume [j.L] Partial Loopfill 0 30 15 Use Wash Method Wash Between Times Between Injections 1			
Use Preparative Mode			
AS Status Demo Mode: Not Ready (Method has not been sent) From AS			
Event Table AS Measurement Integration Calculation Advanced			
GK Cancel	3 5	end Meth	od .:

Fig. 6: Method Setup - AS - Injection

This is the main tab defining the AS control method. It defines which parts of the **Spark Optimas** method will be performed and set some other parameters.

Use Timebase Methods

Sets whether the sampler method part used for outputting signals after injection will be used. This part of the method is then set on the <u>Method Setup - AS - Inputs &</u> <u>Outputs</u> tab.

Use Mix Method

Sets whether the sample pre-preparation part of the injection method will be used. If checked, the method defined on the Method Setup - AS - Mix tab will be performed.

Use Injection Method

This part of method contains the information on the injection routine, analysis time and flush volume.

Injection Mode

Allows to set the injection mode by selecting one of the possibilities:

Partial Loopfill - in this mode, only a part of the sample loop volume will be transferred into the column.

Full Loop - in this mode, the full sample loop will be transferred to the column. Only the value set in the *Loop Volume* field of the<u>Method</u> <u>Setup - AS - System</u> dialog is allowed for setting in the *Inj. Vol.* column in the **Sequence Table** in the *Sequence* window.

 μ *l Pick up* - defined volume of the sample will be injected into the column, preceded and followed by the transport liquid.

Analysis Time [min.]

Defines the time between switching the injection valve to the *INJECT* position and the start of next sample preparation. This parameter is critical for setting the analyses performed in the *Sequence Mode* without the synchronization wire.

Caution: While using the synchronization wire for the **Spark Optimas** autosampler, it is necessary to set the analysis time to *0*.

Flush Volume [µl]

Defines the volume of the sample that will be used for flushing the sample loop and tubing before the sample is aspired. Possible values range from 0 to 9999 μ l, default value is 45 μ l. This option is only available for the *Partial Loopfill* and *Full Loop* injection modes.

Caution: Setting the *Flush Volume* parameter to less than twice the volume of the needle and tubing will cause worse performance.

TubingVolume [µl]

Defines the volume of the sampler needle, including the tubing between the needle and the valve. Possible values range from 0 to 999 μ l, default value is 15 μ l.

Use Wash Method

Defines the type of the washing method performed between injections or vials. The wash method itself is only defined on the Method Setup - AS - Wash tab.

Wash Between

Defines the time when the wash should be performed. Possible values are:

Between Series - the wash will be performed after each finished sequence. The option is only available if the *Sequence Mode* option is checked in the Spark Optimas Setup dialog.

Between Vials - the wash will be performed after each sample measured. The option is only available if the *Sequence Mode* option is checked in the <u>Spark Optimas Setup</u> dialog.

Between Injections - the wash will be performed after each injection.

Use Preparative Mode

Switches the method for the **Spark Optimas** autosampler to the preparative mode. This means that the values for the *Injection Mode* on the *Method Setup - AS - Injection* tab, *Loop Volume* and *Syringe Volume* on the<u>Method Setup - AS - System</u> tab and the *Plate Types* on the<u>Method Setup - AS - Spec. Vials</u> tab are hard-set to those used by the **Spark Optimas** autosampler with Prep mode option installed.

4.1.2 Inputs & Outputs

New	Open	Save	Save as	Report		C trail	Send method e-mail	i by Help			
lect San	npler		Sar	npler 1		Er Er	abled				
				Knauer Opt	imas Sampler Me	ethod		AS	Status		
Injecti	on Input	ts & Output	ts Mix	System Spec	.Vials Tray						
			Use	Aux 1 On	Aux 1 Off [min.]	Use	Aux 2 On [min.]	Aux 2 Off [min.]			
				0,30	0,50		0,00	0,00			
				0,00	0,00		0,00	0,00			
				0,00	0,00						
					0,00		0,00	0,00			
Statu	ind Time (m 20 Js		de: Not R		as not been sen	t)	0,00		rom AS		
	20 JS	Demo Mo		eady (Method h			0,00		rom AS		

Fig. 7: Method Setup - AS - Inputs & Outputs

Tab defining the behavior of the **Spark Optimas** autosampler while performing the Time Program. The tab governs the assignment of the inputs and output on the connector on the back side of the autosampler and allows to set the timetables for installed optional valves.

End Time

Sets the ending time of the Time Program. Without the value set, no options on this tab can be used or even set. The value of the *End Time* may be greater than the *Run Time* set on the *Method Setup - Measurement* tab to set signals or switch valves after analysis, the difference will be evaluated as a *CONTROL* time.

Aux table

Governs the switching of the digital output between *HIGH* and *LOW* states based on the time. The default state of the output depends on the connection to the Input/Output connector (for more details see the chapter **Spark Optimas setup communication** on pg. **5**). and it can be switched to *HIGH* (*Aux 1/2 On*) and back to *LOW* (*Aux 1/2 Off*) or vice versa up to four times for each output.

4.1.3 Mix

lethod Setup Default2 - #58; 31.03.2023 12:33:56				×
New Open Save Save as Report setup Audit trail Send method by email	? Help			
elect Sampler 1 🗸 🖉 Enabled				
Knauer Optimas Sampler Method	AS Status			
Injection Inputs & Outputs Mix System Spec. Vials Tray				
Action Time [r	nin]			
Wait ~ 1	Add			
1 Add, from Sample to Destination volume 100 [µL]	Insert			
2 Add, from Reagent-A to Destination volume 100 [µL] 3 Mix, times 2, volume 50 [µL]	Change			
4 Wait, time 1,00 [min]	Delete			
5				
	Alexa Terra			
	Move Top			
	Move Up			
	Move Down			
	Move End			
AS Status Demo Mode: Not Ready (Method has not been sent)	From AS			
Event Table AS Measurement Integration Calculation Advanced				
R OK Cancel		2 s	iend Meth	bor

Fig. 8: Method Setup - AS - Mix

Mix part of the sampler method (defined on the *Method Setup - AS - Mix* sub-tab) governs the pre-preparation of sample before the injection. Several operations are possible with vials defined on the <u>Method Setup - AS - Spec. Vials</u> tab. The table fields are context-based, that is their content change according to the action defined in the first column on the actual row.

Note: When any special vials (Reagent, Destination, Transport) should be used in the Mix method, check that there are these vials defined on the<u>Method Setup - AS -</u> <u>Spec. Vials</u> tab. If they are not defined, the default values will be used, which may invalidate given sample or other sample's analysis data.

A new row is added (or actual row is modified) using the *Action* drop-down list. When an action is selected, other parameters of the action emerge and can be modified. Using the *Add* or *Insert* button the action can be added as a new row into the table, either to the end of the list (*Add* button) or above the selected row (*Insert* button). The existing row can be changed by selecting it, changing the necessary parameters and pressing the *Change* button. A row can be deleted using the *Delete* button.

When the row is selected, it can be moved up and down through the list by using the *Move Top*, *Move Up*, *Move Down* and *Move End* buttons.

Note: Up to 15 lines can be programmed in the **Mix Table**.

The items that can be selected for the particular row in a Mix Table are following:

Add

Performs the sequence of steps involving aspiration from the vial (or port) defined in the *From* field and dispension into the vial defined in the *To* field. The volume that should be transported this way is defined in the *Volume* [*µ*] field.

Mix

Mixes the liquid and the Destination vial by aspiring and dispensing the volume defined in the *Volume* [μ I] field. The number of aspirations and dispensions is defined in the *Times* field.

Wait

Causes the sampler to wait for the specified time interval. The desired interval is entered into the *Time [min.]* field in minutes.

4.1.4 System

Method Setup Default2 - #58; 31.03.2023 12:33:56			×
New Open Save Save as Report setup Audt trail Send method by Help			
Select Sampler 1 V CEnabled			
Knauer Optimas Sampler Method AS Status			
Injection Inputs & Outputs Mix System Spec. Vials Tray			
Loop Volume [µL] Syringe Volume [µL] Syringe Speed			
10000 2500 V Normal V			
☑ Tray Cooling [°C] (active only if available)			
10			
Reset Outputs			
Headspace Pressure			
AS Status Demo Mode: Not Ready (Method has not been sent) From AS			
Event Table AS Measurement Integration Calculation Advanced			
R OK Cancel	E3 5	end Metho	d

Fig. 9: Method Setup - AS - System

Tab defining various aspects of the Spark Optimas operation.

Loop Volume [µl]

Defines the value of the sampling loop volume used for the injection (in μ I). Possible values range between 0.1 and 1000 μ I. When using *Full Loop Injection Mode* from the <u>Method Setup - AS - Injection</u> tab, only the *Loop Volume* value is allowed in the **Sequence Table** in the *Sequence* window.

Syringe Speed

Sets the syringe speed from the set of predefined values (Low, Normal, High).

Syringe Volume [µl]

Defines the volume of the syringe installed in the autosampler. The value is dependent on the fact whether the *Preparative Mode* is switched on or off, varying between 500 μ l when not using the *Preparative Mode* and 2500 μ l in the *Preparative Mode*.

Tray Cooling [°C]

Allows to enable the tray cooling function and set the temperature for the cooling (in the range 4 - 40 °C when cooler/heater is installed, 4 - 22 °C when only cooler is installed). This option is grayed-out if the **Spark Optimas** sampler doesn't have the function built-in.

Air Segment

Sets whether the air segment should be used during the injection. The air segment normally separates the sample from the transport liquid.

Reset Outputs

While checked, forces the digital outputs to be reset after the sequence has ended.

Headspace Pressure

When checked, the sampler uses the pressure to facilitate the transfer of the sample into the sample loop. The pressure will only be used when the vials are airtight.

Alarm Buzzer

Sets the sound signalization on. See **Optimas** manual for more details.

Skip Missing Vials

Defines the behavior of the autosampler when missing vial is detected. While unchecked, the autosampler will issue an error message and stop the processing of the sequence, but when checked, no error message is issued and the sequence behaves according to the settings in the *When vial is missing* field.

4.1.5 Spec. Vials

Method Setup Default2 - #58; 31.03.2023 12:33:56			×				
New Open Save Save as Proport setup Audit trail Send method by Help							
Select Sampler 1 Sampler 1							
Knauer Optimas Sampler Method AS Status							
Injection Inputs & Outputs Mix System Spec. Vials Tray							
Type of (relative range to default start vial = 1) Plate 24 vials Vials Standard Reagent Vials Reagent Vials Reagent Vials OFF B OFF B OFF B OFF C OFF C OFF C OFF C OFF C OFF C							
AS Status Demo Mode: Not Ready (Method has not been sent) From AS							
Event Table AS Measurement Integration Calculation Advanced	51 :	Send Meth	nod				

Fig. 10: Method Setup - AS - Spec. Vials

Governs the types of trays (or well-plates) used and allocation of special purpose vials (destination, transport and reagent vials) to particular vial well positions.

Plate Type

Defines the plate type inserted into the tray position. The visualization of the tray is displayed on the Method Setup - AS - Tray tab.

First Destination Vial

Defines the position of destination vial(s) used in the *Mix* method. The most common usage of the destination vials is with the *Relative* checkbox checked, as it gives each vial in the sequence it's own destination vial.

Note: When using destination vials and mix method in general, do not use more than one injection per vial.

Reagent Vials

Defines the position of Reagent vial(s) used in the *Mix* method when the *84+3* tray is used. Than two reagent vials can be programmed and they can only be mapped to the large volume vials (85-87).

Transport Vials

Defines the position of vials containing the transport liquid (used with µl Pick up injection mode and together with the 84+3 plate type). The algorithm defining from which vial will the transport liquid be actually aspired is stored in the **Spark Optimas** sampler. If the transport vials are used, they must form an uninterrupted

row beginning in the well number defined in the first field and ending by the vial defined in the second field.

4.1.6 Tray

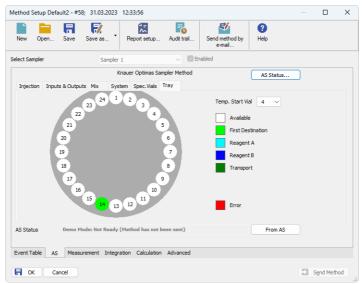


Fig. 11: Method Setup - AS - Tray

Shows the visualization of the **Spark Optimas** autosampler tray(s), along with the mapping of the vials as performed on the**Method Setup - AS - Spec**. Vials tab.

Temp. Start Vial

Allows to set the temporary position of the first sample vial that will be used in the measurement to assess the positions of other special vials. The position of this temporary starting vial will be reset whenever the *Method Setup* dialog is closed.

4.2 Hardware Configuration

Hardware Configura	ition	×
Type of Sampler: Connection: Firmware Revision:	Demo	
Tray cooling:	Available	~
ОК	Cancel	

Fig. 12: Hardware Configuration

The AS Status button in the <u>Method Setup - AS</u> displays the Hardware Configuration dialog. In the full version, this dialog displays autosampler model, automatically detected communication parameters, firmware revision and the presence of optional of the **Spark Optimas** autosampler.

Type of Sampler

Shows the **Spark Optimas** autosampler type as automatically detected by the control module.

Connection

Shows the communication parameters as detected from the sampler.

Firmware Revision

Shows the firmware revision loaded into the Spark Optimas autosampler.

Tray Cooling

Allows to set whether the **Spark Optimas** sampler is equipped with the tray cooling option.

4.3 Spark Optimas Setup

Spark Optimas Setup dialog (accessible through the *System Configuration* dialog) allows to manually set the parameters needed for the communication with the **Spark Optimas** autosampler.

Spark Optimas Setup		×
<u>S</u> erial Port	COM1	~
ID	60	AutoDetect
Status	Unknown	
Sampler Name	Sampler 1	
	Sequence Mode	
	Digital Input Names	Change
ОК	Cancel	Help

Fig. 13: Spark Optimas Setup

Serial Port

Set the COM Port used for the communication between the **Spark Optimas** autosampler and **Clarity**.

ID

Shows the device identifier of the **Spark Optimas** autosampler. The number listed here is hard-set for **Spark Optimas** autosamplers to the value *60*. For more details see the chapter **Spark Optimas setup - communication** on pg. **5**.

Autodetect

When pressed, checks whether there is the **Spark Optimas** autosampler present using the selected *Communication* option, with the given *Serial Port*. The ID is also checked for the successful communication attempt. The result of the autodetection is then displayed in the *Status* row.

Status

Shows the status of the communication with the **Spark Optimas** autosampler after the *Autodetect* button has been used. The displayed information, in case of the successful communication attempt, includes the version of the firmware in the autosampler and the sampler's serial number.

Sampler Name

Allows to set the custom name of the **Spark Optimas** autosampler, which will be then shown in the *Device Monitor* window, in the reports and on other places in **Clarity**.

Sequence Mode

This option governs the mode of the operation with the **Spark Optimas** autosampler control module. While checked, as much of the sequence table as possible is sent to the autosampler in one package (no method sending prior each injection will be performed for the injections package). The sequence should be

written in the manner where following lines are analogical (e.g. same number of injections from each row, but the injection vial is incremented between rows etc.). More information on the *Sequence Mode* synchronization can be found out in the chapter **"How to set freeze synchronization"** on pg. **28**.

While not checked, the operation will be governed by **Clarity** - the method will be sent to the autosampler prior each injection and each injection will thus be considered a single one-injection series by the autosampler. This prevents the use of washing between samples and few other features.

Digital Input Names

Pressing the *Change* button opens the *Digital Input Names* dialog which allows to set the custom name for the virtual **Spark Optimas** digital input. This input allows to start **Clarity** run by the signal of the **Spark Optimas** autosampler simulated over the communication line instead of the usual synchronization wiring, send warnings on autosampler errors or govern auxiliary devices.

Digital Input Na	igital Input Names							
Input no.	Descriptions:							
1	Injection Input							
ОК	Cancel Help							

Fig. 14: Digital Input Names

4.4 Device Monitor



Fig. 15: Spark Optimas Device Monitor

The Device Monitor window for the **Spark Optimas** autosampler enables to control some of the actions of the **Spark Optimas** autosampler.

Initial Wash - Start

When invoked, performs the Initial Wash operation to rinse all tubing.

Reset Error

When invoked, resets the Error state that occurred on the sampler.

AS Status

Opens the <u>Hardware Configuration</u> dialog described in the chapter **"Hardware Configuration"** on pg. **21**.

5 Report Setup

nt 📸 Print to PDF 🐴	Send PDF				
31.03.2023 13:15		Method d:\darity90\DataFile	s\WORK2\Default2.met		Page 1 of 1
		Injection Control - Optim.	as Method Sampler 1		
Firmware Revisio	0 100				
Tray Cooling	: Available				
Inj.Mode	: Partial Loopfill	Prep.Mode	: Yes	Tubing Volume	: 15 µL
Plate	: 24 vials				
Analysis Time	: 0,00 min	Loop Volume		Syringe Volume	: 2500 µL
Flush Volume	: 30 µL	Wash Method			
		Syringe Speed	: Normal		
Tray Cooling	: 10 °C				
Air Segment	: ON	Reset	: OFF	HeadSp.Press.	: OFF
First Dest.	: ON	Relative	: 11		
Reagent A	: OFF				
Reagent B	: OFF				
End Time	: 20.00 min				
			Auxiliary 1 Table		
		Aux 1 On	Aux 1 Off		
		[min.]	[min.]		
		0,30	0,50		
			Auxiliary 2 Table		
		Aux 2 On [min_]	Aux 2 Off		
		[IIIIII]	No step		
		L	ite seek		
		Sampler 1 M	lix Table		
1	Add, from Sample	to Destination volume 100 [µL]			
2		A to Destination volume 100 [µL]			
3	Mix, times 2, volun	ne 50 [µL]			
4	Wait, time 1,00 [m	in]			

Fig. 16: Spark Optimas report preview

All autosampler-specific settings (that means the data from all sub-tabs of the <u>Method Setup - AS</u> tab) are reported as a part of the data displayed by the use of *Injection Control* checkbox of the *Report Setup - Method* dialog.

6 Troubleshooting

When the remedy for some problem cannot be discovered easily, the recording of communication between **Clarity** and the autosampler can significantly help the **DataApex** support to discover the cause of the problem.

The recording can be enabled by adding or amending the COMMDRV.INI file in the **Clarity** installation directory (C:\CLARITY\CFG by default). The file can be edited in any text editor (e.g. Notepad). Following section should be edited or added:

[COM1] echo=on textmode=on filename=SparkOptimas_%D.txt reset=off

- *Note:* Instead of COM1 type the correct port used to communicate with the **Spark Optimas** autosampler. This information is displayed when the *AS Status* button in the <u>Method Setup - AS</u> dialog is invoked or in the <u>Spark Optimas Setup</u> dialog. The correct settings should have the COMx (where x is the number of the COM port) format.
- *Note:* %*D* (or %*d*) in the filename parameter means that the log will be created separately for each day. The *reset=off* parameter disables deleting the content of the log each time the station is started during the same day.

The created *.TXT files will greatly help in diagnosis of unrecognized errors and problems in communication. Note that the file size may be quite significant, so in case the error occurs on a regular basis, it might be better to set the Reset=on, start **Clarity**, invoke the error, close **Clarity** and send the diagnostics file (the file will be once more reset during the next start of **Clarity**).

6.1 Specific Problems

An error message "Cannot establish communication with ..." appears when opening Clarity Instrument.

Solution: Check the power cable (**Spark Optimas** sampler must be switched on), communication cable and communication settings in the <u>Spark Optimas Setup</u> dialog.

An error message "AS Error" appears during the Clarity operation.

Solution: The communication has been interrupted. Check the communication cable as it is most probably disconnected. This message may also occasionally appear after aborting the *ACTIVE* Sequence.

Injection volume set in the Sequence window is not accepted.

Solution: Either you are using the *Full Loop* option and the injection volume doesn't match the one of the installed injection loop, or you are trying to enter the volume that is greater than the half of the installed *Loop Volume* in the *Partial Loopfill* injection mode.

The Spark Optimas autosampler does not start with the injection.

Solution: Check the Freeze input settings on the <u>Method Setup - AS - Inputs & Outputs</u> tab and used wiring. When the Freeze input is used for synchronization, the injection will not start until the respective input is closed (or opened - the logic is set in the ASM).

6.2 How to set freeze synchronization

The **Spark Optimas** autosampler switched to the *Sequence Mode* operation may be synchronized with **Clarity** in two ways - either using the *Analysis Time* parameter from the <u>Method Setup - AS - Injection</u> tab or by using the autosampler's Freeze input.

Either way, for the successful synchronization using the Freeze input, user should do several operations:

• Disable the standard **Clarity** ready-out synchronization. In the **System Configuration** dialog, set the *Number* field for the *Ready Dig. Output* to --.

Data Inputs & Outputs	Device	Device			
Ext. Start Dig. Input:	Knauer Optimas	\sim	1	\sim	
Ready Dig. Output:	Colibrick	\sim		\sim	

Fig. 17: System Configuration - Ready Out settings

- *Note:* This step is probably optional, but cleans the station operation of unnecessary switching of the output at the wrong time.
 - Enter the Method Setup Event Table dialog and insert the following rows there:

N	lew Open		e as	Report setup	Audit		Send method	ev Help			
	open	5476 547	- do	nepoir acrup	Addit		e-mail	by hop			
om	mon for all dete	ctors									
	Name			Input				Output			
		Type	Source	Input	Value	Units	Output Type	Output	Parameter	Store	
	Initial switch	Time Idle >			0,000		Colibrick	Digital Output 1	Low	\checkmark	
	Initial start	Time Idle >			0,400		Colibrick	Digital Output 1	High		
3	Freeze End run	Run Time > Run Time >			1,200		Colibrick	Digital Output 1	Low		
	End run	kun lime >			1,500	min	Colibrick	Digital Output 1	High		
5											
,		_									

Fig. 18	3: Event	Table -	synchronization	settings
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Modify the times entered in the *Value* column to suit your analysis and the items entered in the *Output Type* and *Output* columns to match the digital output used for synchronization. The values should be defined as follows:

- Event called *Initial Switch* should always perform at the *Idle Time* = 0.000 minutes. It serves for initial switching of the output for the first sample in the sequence (or for the first sample in the pack of sequence rows performed as one block) regardless of **Clarity** initial output state settings.
- Event called *Initial Start* should perform at the time reliably sufficient for the preparation of the sample before injection. You have to test that time for your analysis try to run a single-line analysis according to your method and measure the time needed from hitting the Run Sequence button to the injection, plus add a few seconds more. The value can be fine-tuned later, but it is imperative that the sampler will be in the *WAIT FOR INPUT* or *FREEZE ACTIVE* state before the event is triggered. The event is used for starting the first analysis in each block of rows solved together and is performed during the *Idle Time*.
- Event called *Freeze* should be set so that it occurs at the *Run Time* shortly after the internal autosampler analysis time counter ends its method. At that time, a new sample preparation is started and the output has to be switched to block the injection before it occurs. Ideal time of this event would be somewhere between Clarity analysis run time (as defined in the *Method Setup Measurement* tab under the parameter *Run Time*) minus the time defined for the *Initial Start* row, and the Clarity analysis run time (as defined in the *Method Setup Measurement* tab under the parameter *Run Time*).
- Event called *End Run* should be set so that it occurs at the end of the analysis. This time is set in the *Method Setup Measurement* tab under the parameter *Run Time*. It serves for triggering the injections of vials other than the first one in the group of the sequence rows sent together, and is performed in the *Run Time*.
- Ensure that the *End Time* parameter on the <u>Method Setup AS Inputs &</u> <u>Outputs</u> tab is set correctly. It should be set so that it equals **Clarity** analysis run time (as defined in the *Method Setup - Measurement* tab under the parameter *Run Time*) minus the time defined for the *Initial Start* row. At this time, the internal autosampler analysis time will elapse and a new injection preparation will be started.