

SFM Series.

Stopped-flow accessories





2,3 and 4 SYRINGE STOPPED-FLOW

A unique design for single, double and triple mixing applications





Configure the **SFM** to meet your specific needs

The SFM family of stopped-flow instruments is renowned for its versatility.

This modular range of instruments is easily adaptable and expandable to a vast array of rapid mixing techniques and applications.

Any SFM can be switched from optical stoppedflow, to chemical quench-flow, to freeze quench, to automatic titration capabilities, to cryo operation, to a beam-line, all in minutes, using off the shelf options.

All of these accessories are user changeable, enabling you toadapt the SFM-2000/3000/4000 to your labs needs without buying a completely new system.

Titration accessory

A programmable titrator exploits high-precision stepping motors for ultra-accurate micro-volume delivery. Volumes **as low as 2 µl can be injected** using a 1.9 ml syringe. The titrator accessory is designed to accept standard 1 cm x 1 cm cells, and includes a micro magnetic stirrer. A 5.5 mm diameter port is available for external devices (pH or nitric oxide electrodes, temperature probes)The titration head is connected to the SFM circulating bath circuit, but an optional Peltier element is available for independent control of cuvette temperature.

Titration sequence design

The titrator is fully automated thanks to Bio-Kine software with its user-defined concentration steps. Automatic increment functions are also available, including a variable function to change the increment during titration. **Bio-Kine automatically tracks solution volumes, and alerts the user to out of range conditions**. When the acquisition is made using a BioLogic spectrometer, a series of titration sequences is performed automatically. External detection devices can be synchronized by adjusting the steps and acquisition duration.

Data collection

The user can choose between single wavelength titrations and spectral titrations. When the titration experiment is carreid out at a single wavelength using a BioLogic spectrometer, the Bio-Kine software automatically builds the titration curve at each step by plotting signal versus concentration.

Depending on the spectrometer model, spectra can also be recorded at each titration step and data displayed and analyzed as a 3D file.

Titration accessory
Cryo-stopped-flow -90°C
Quench-flow upgrade
Freeze quench
EPR stopped-flow
Optical delay line
Optical quench head
mT-jump, rapid mixing temperature jump
MCS-200, conductimetric detector
FT-IR stopped-flow
Umbilical link
Neutron head
X-Ray accessory
Miscellaneous

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Applications include: - denaturation studies, - binding reaction, - pH denaturation, - dissociation constants.





Specifications

Minimum injection volume	2 μl (1.9 ml syringe) 10 μl (10 ml syringe)
Injection ports	3
Detection windows	3
External probes port	1 (Ø 5.5 mm)
Temperature control	yes (-20°C to +85°C)

Compatible with

all SFM models

Titration accessory

Catalog n^e 045-01

CS-90°C, cryo-stopped-flow

Liquid N₂ cryo-stopped-flow

The cryo option extends the range of BioLogic stopped-flow instruments to -90°C and allows transient state kinetics experiments to be performed in single mixing mode.

Temperature control

CS -90°C includes a cryo-bath and heater. The cooling is performed by circulating liquid N_2 in a coil immersed in cryo-solvent. Temperature dependence studies can be carried out from -90°C to +20°C without any reconfiguration. A temperature probe in contact with the cuvette gives a precise temperature reading of the reaction.

Mixer and observation cell assembly

The umbilical connector connects the SFM to the mixing chamber. The chamber, 200 μ l reactant transfer lines, and the last mixing stage are immersed in the cryo-solvent. The CS-90°C is equipped with FFKMO-rings for full solvent compatibility. Anaerobic operation is standard.

Detection is made using fiber optic cables, and the CS-90°C is compatible with all MOS models. A diode array (MOS-DA) is the most popular spectrometer for the CS-90°C. Absorbance spectra can be collected at the fastest rate available (400 μ s per spectrum) with the diode array to trap the structure of reaction intermediates invisible at ambient temperature.

CS -90°C includes:

- stopped-flow adaptor, umbilical link,
- mixing compartment including 1 cm light path cuvette,
- 2 fiber optics (2 m long) with SMA connectors,
- FFKM O-rings for full solvent compatibility,
- temperature probe,
- heater for temperature regulation,
- 22 liters cuve and coil for liquid N, circulation.

Site requirement:

- cryo-solvent (oil, isopropanol, ethanol, etc.),
- pressurized liquid nitrogen tank,
- tubing to connect cryo-stopped-flow to $\mathrm{N_2}$ tank.



Cryo-accessory for external cryostat

BioLogic also offers a cryo-accessory compatible with the CC-905 cryostat from Huber GmbH. The mixing assembly and umbilical are similar to CS-90°C.

A stopped-flow is attached directly on the cryostat. The temperature of the bath is set from the manual control unit and is stable to within 0.1° C.

Customization of the cryo-accessory is also possible for coupling with other cryostat models (minimum bath aperture should be 270 x 150 mm and minimal depth 150 mm), contact BioLogic for customized solutions.

Cryo-accessory includes:

- stopped-flow adaptor,
- umbilical link,
- mixing compartment including 1 cm light path cuvette,
- 2 fiber optics (2 m long) with SMA connectors,
- FFKM O-rings for full solvent compatibility,
- temperature probe.





Specifications

Temperature range	from -90°C to +20°C
Dead time	2 ms
Mixing ratio	1:1 to 1:5
T control of umbilical link	yes
Single mixing applications	

Compatible with



Double mixing cryo-stopped-flow

Double mixing cryo-stopped-flow is possible with the SFM-4000. The solutions flow to the cryo-bath using HPLC tubing. Two mixers are immersed and a 100 μ l delay line is built-in. Users can customize the delay line volume if required. Ageing times as low as 15 ms can be reached before proceeding to a second mixing.

Double mixing is available with liquid N_2 and cryostat cooling methods. The SFM-4000 can also be used for single mixing applications. The delivered items are similar to single mixing set-ups except that one delay line and second built-in mixer are included.



Specifications

Temperature range	from -90°C to +20°C
Dead time	2 ms
Mixing ratio	1:1 to 1:5
T control of umbilical link	Yes
Single mixing applications	

Compatible with



Cryo-accessory for external cryostat

Specifications

-	
Temperature range	from -90°C to +20°C
Dead time	3 ms
Mixing ratio	1:1 to 1:5
T control of umbilical link	no
Single and double mixing applications	

Compatible with



Double mixing cryo-stopped-flow Double mixing CS-90°C

Double mixing cryo-accessory for external cryostat 05

Catalog n°
053-11/29D
053-11/31D

Cataloa nº

053-11/31

Quench-flow upgrade

SFM-3000 and SFM-4000 systems can be quickly converted to high-performance quench-flow systems by exchanging the stopped-flow observation head for the quench-flow accessory. Ageing times are varied by selecting delay lines. Mixing is carried out in continuous flow mode, interrupted flow mode, or in BioLogic's unique pulse mode (single mixing applications only).

BioLogic's stepping-motor-technology controls the flow rate precisely via Bio-Kine software. The independent-stepping-motors eliminate all the limitations of a single-drive mechanism, and mixing ratios can be changed freely, just as in stopped-flow observation mode, while consuming only 50 µl of reagent.



The SFM-4000/Q is the only system available with 4 independent syringes and 3 mixers. It is a unique system for triple mixing applications, such as De/H exchange experiments or radioactive

labeling. The user controls two ageing times from the software and series of "MIX - AGE - MIX - AGE - MIX" experiments can be done rapidly.A long list of papers citing our instruments in this field of application is available on the BioLogic website.

Minimum ageing time	2 ms
Ageing method	set of delay lines
Temperature range	0°C to +85°C
Sample consumption (depending	50 µl
on mixing ratio and delay line	
used)	

Compatible with SFM-3000/4000, SFM-300/400

Quench Flow upgrade	Catalog n°
QF upgrade for SFM-3000	054-11/41
QF upgrade for SFM-4000	054-11/42



Freeze quench

The freeze quench technique uses extreme cold to stop the reaction. Freeze guench is used to study full kinetics or to trap reaction intermediates whose presence has been determined by stopped-flow experiments. Freeze guench techniques are extensively used to study metallic reaction centers in metalloproteins and metalloenzymes. The reactants are mixed and aged for a user-defined time period inside the calibrated ageing loops before ejection for freezing. A built-in ejection nozzle at the exit of each ageing loop sprays the aged solution. Mixing is fully controlled via the Bio-kine software. Freezing is performed by ejection into a -130°C isopentane bath cooled down by liquid N₂ circulation. The mixing process is independent from the freezing process so coupling to cold surface freezers is possible. Sample consumption depends largely on the amount of material necessary for off-line analysis. For example, to obtain around 1 cm of crystals in an EPR tube, the user needs around 200 μ l of solution delivered to the ejection nozzle. Ageing times, including flying time and freezing time, range from 9 ms to several minutes.

To limit the number of steps needed during the packing process and avoid the risk of warming up the frozen solution, aged solution is ejected directly into a PTFE funnel. The sample holder is attached to the funnel so the sample can be packed easily with the packing rod (included). Holders are available to collect samples for spectroscopic analysis by EPR/ NMR. Mossbauer, and XAFS.

Freeze quench accessory includes:

- dewar and one collection kit,

- set of ageing loops with built-in ejection

- liquid nitrogen and circulation system,

- magnetic rod and magnetic stirrer plate,



- sample holder (EPR tube, NMR tube, etc.), - packing rods.

- umbilical connector, - built-in Berger Ball mixer,

nozzle,

- packing rods.

- isopentane,

Site requirement:



Specifications

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Priming volume of umbilical	200 µl
Ageing method	set of ageing loops
Flying time	< 1 ms
Minimun ageing time in ageing	3 ms
loop	
Freezing time (using isopentane)	4-5 ms
Minimun ageing time (total)	9 ms

Compatible with



Catalog n°

Freeze quench accessory

053-11/10

EPR stopped-flow

Electron Paramagnetic Resonance (EPR) detection with freeze quench and stopped flow has been used for many years to follow kinetics. The modular design of the BioLogic SFM series now allows easy coupling of a stopped-flow mixer with a variety of EPR flow resonators.

The time dependent change of amplitude of an EPR signal in a distinct field value can be followed. This makes stoppedflow a useful technique for measurements of rate constant for formation, decay or conversion of paramagnetic species.

The EPR stopped-flow accessory consists of an umbilical connector especially engineered so that a Berger Ball mixer can be fitted onto one end to minimize the dead volume of the set-up. A flow resonator is attached directly to the mixer exit using an adaptor ring, or optionally, with HPLC type tubing.

The temperature is controlled from the driving syringe to the mixer. The dead time of the EPR stopped-flow set-up depends on the design of the flow resonator. The chosen geometry will limit the flow rate and determine the dead volume.

Data showing 330 µs dead time using two dielectric rings type resonator* have been published.

EPR stopped-flow accessory includes:

- umbilical connector.
- built-in Berger Ball mixer with open exit,
- adaptor for resonator,
- Viton[®] O-rings.
- Not included:
- EPR flow resonator.

* G. Lassmann, P.P. Schmidt, W. Lubitz, Journal of Magnetic Resonance, 172 (2005) 312-323

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Specifications Umbilical volume

Dead time (depending on

resonator geometry)

Compatible with

Built-in mixer

Material

Optical delay line



The optical delay line is used for applications where light is used to generate a reactant or to trigger a sample change before chemical guenching. The sample flows through the cuvette under constant illumination or the flow is stopped and the sample illuminated at a preset time before mixing with a final reactant/quencher. The cuvette access ports allow beams as large as 5 mm in diameter to be used for illumination. The optical delay line is compatible with SFM-3000 and SFM-4000 only. A FC-15 cuvette is delivered in standard but the optical delay line is compatible with all FC type cuvettes.

Optical quench head



The optical guench head is used for applications where intense light (laser, flash lamp, etc.) is used to guench a reaction. A sample can flow through the cuvette under constant illumination, or the flow can be stopped and the sample illuminated at a preset time. Cuvette exposure is optimized for maximum quenching.

The open air design of the head allows the rapid dissipation of heat without compromising the amount of sample exposed to the light source. The optical guench head is compatible with all SFM models. A FC-15 cuvette is delivered as standard, but all FC type cuvettes are compatible with the accessory.

EPR stopped-flow adaptor

all SFM models

200 µl

peek

0.33 ms

Berger Ball

Catalog n°

053-11/33

8

Specifications			
Cuvette	FC-15		
Light path	1.5 mm		
Intermixer volume	89 µl		
Dead time (from center of mixer 31 µl to center of cuvette)			
Compatible with all SFM models			

Optical delay line accessory

Specifications				
Cuvette	FC-15			
Light path	1.5 mm			
Illuminated volume	37 µl			
Compatible with				
all SEM models				

all Srivi models

Optical quench accessory

Catalog n° 053-11/11

Catalog n°

053-20

mT-Jump, rapid mixing temperature jump

MCS-200 conductimetric detector

The mT-Jump accessory achieves temperature changes by mixing two solutions of different initial temperatures T1 and T2. The final temperature of the mixture (T3) is calculated from the initial temperatures T1 and T2 and the mixing ratio of the two solutions. Three thermoelectric Peltier elements are used to control the initial temperatures of the two solutions and that of the observation cell after mixing. The user defines the mixing ratio for the reaction and the amplitude of the temperature jump. The temperature of each Peltier element is then automatically adjusted. The mT-Jump system can accommodate all FC-type cuvettes and is compatible with all MOS models.

The amplitude and direction of the jump is entirely controlled by the user (up to $\pm 40^{\circ}$ C jump), offering new domains of investigation, for example, protein refolding studies.

Main applications with millisecond resolution include:

- folding/refolding without chemical denaturant,
- polymerization.



Refolding of horse heart cytochrome c by cold jump from 85°C to 60°C.



Specifications

Mixing ratio	from 1:1 to 1:4
Storage lines	150 µl
Cuvette compability	all FC cuvettes
Dead time	1.5 ms
Communication	USB
Temperature range	from +5°C to +90°C
Maximum temperature jump	±40°C
Precision of T Jump	±0.1°C
T stability (cuvette)	variation < 1% in 30 s
T stability (storage line)	±0.2°C at 80°C
Compatible with	
all SFM models	Cataloa nº
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045-11/4

mT-Jump accessory

Conductivity detection can be an alternative to standard optical detection techniques. Changes in the solution's conductance in the course of a chemical reaction can be induced either by a change in the concentration of positively, or negatively, charged ions or molecules or by a change of their mobility inside the solution.

MCS-200 is based on Fast-Fourier-Transform (FFT) Impedance **Spectroscopy**. The instrument allows recording steady state impedance spectra of the sample in a frequency range of 10 Hz to 200 kHz, and measuring rapid kinetics with a time resolution of 200 µs per data point. Resistance, conductance and specific conductance are measured in real time with a 3 ms dead time.

The conductimetric cuvette is a customized FC-20 cuvette with 1.6 mm diameter gold electrodes (glassy carbon electrodes available on request) built in to the cuvette walls. Simultaneous absorbance measurements are possible. Cyclic voltammetry technique is available for efficient electrode cleaning.

MCS-200 includes:

- dedicated observation head with ground connection,
- conductivity cell .
- control unit equipped with impedance board and low current capabilities,
- hard-stop,
- special Bio-Kine software version
- (for impedance measurements, Nyquist representation included).

Main applications include:

- (in)organic reduction/oxidation,
- proton exchange,
- metal ligand complexation,
- micelles formation from surfactants,
- ion exchange.









Specifications

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Conductivity range	1 to 500 μS.cm ⁻¹
Accuracy	1%
Electrical cell constant	6.4 cm ⁻¹
Light path	2 mm
Fastest acquisition time	200 µs
Minimum dead time	3 ms
Electrode material	gold (Ø 1.6 mm)

Compatible with all SFM models

Catalog n°	
091-64	

MCS-200 d	etector
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FT-IR stopped-flow accesory

Improvements in Fourier Transform Infra-Red (FT-IR) technology and use of step scan acquisition have made the combination of FT-IR with stopped-flow instruments a valuable tool for evaluating reaction rates in the Infra-Red region.

FT-IR spectroscopy provides structural information in terms of bonds and mobility. It can be a very useful accessory to study infrared kinetics in the amide I region of a protein's vibrational spectrum.

The FT-IR accessory is usually attached to stopped-flow using an umbilical connector so only the observation cuvette is fitted into FT-IR sample compartment. The FT-IR flow cell is mounted in a holder compatible with most FT-IR instruments.

The flow cell uses $\text{CaF}_{_2}$ windows and users can freely select light path from 15 μm to 500 μm by changing a spacer between windows.

Dependingonthelightpaththetotalflowratemaybelimitedto1ml/s. The dead time under these conditions is determined by the acquisition speed of the detector.

FT-IR accessory includes:

- FT-IR flow cell holder with built-in Berger Ball mixer,
- set of CaF, windows,
- set of spacers,
- direct exit tube.



Specifications

CaF2
15 μm, 25 μm, 50 μm 100 μm, 200 μm, 500 μm
6 ms

Catalog n°

053-17

Compatible with

all SFM models

FT-IR accessory

Umbilical link

Some stopped-flow applications require special mounting options. This is often the case on a beam line where there is no room to mount the observation head directly on top of the mixer body. The umbilical connection allows the stopped-flow mixer body and the observation head, (with last mixing stage), to be separated by up to 45 cm. With the umbilical connection you can use BioLogic's advanced mixing technology with more devices, and in more applications.

The umbilical connection is compatible with all BioLogic stopped-flow instruments.

For external coupling or difficult to access applications

- Applications where use of the umbilical may be necessary:
- installation on a synchrotron beamline or a neutron scattering line,
- cryo-stopped-flow experiments at sub-zero temperatures,
- high temperature stopped-flow (T \ge 80°C),
- cooling of the observation head and flow lines while the rest of the instrument is maintained at a different temperature (when stock solutions are not your stock) at our simplemental terms
- (when stock solutions are not very stable at experimental temperature), - positioning of the observation head in a spectrometer magnet
- (for this application all parts will be metal free),
- coupling to FT-IR or EPR spectrometer.

Dual Comb spectroscopy

The FT-IR accessory can also be installed in the sample compartment of a dual-comb spectrometer to benefit from the high acquisition speed, the enhanced signal to noise ratio and the wavelength resolution of such technologies. This represents a significant improvement over the acquisition speed limitations of FT-IR instruments.



β-lactoglobulin refolding using FT-IR accessory and a dual comb spectrometer



specifications	S	pe	cifi	ica	tic	ons
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Volume of flow lines ⁽¹⁾	200 µl	
Number of flow lines	2	
Length of umbilical link ⁽¹⁾	45 cm	
Temperature control	Ves	

(1): different length available (multiple of 45 cm)

Compatible with

all SFM models

	U	mk	oilicc	ıl link	acc	essory
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Catalog n° **047-62**

Neutron head

Combining stopped-flow technology with small-angle neutron scattering gives the user valuable information about the early stages of reactions occurring during growth and the formation of micelles or vesicles. It can also be applied to other soft condensed matter studies such as the growth of inorganic particles in an organic matrix or the growth of mesoporous structures.

Small-Angle Neutron Scattering (SANS) is a technique that measures the deviation at small angles (from less than one degree to several degrees), of a neutron beam due to small particles in the sample. These structures could be clusters in alloys, polymers, or biological macromolecules and

their dimensions are usually in the range of 10 to 100 nanometers.

The neutron scattering observation head is designed with a 44° opening for a large solid angle so the detector can get the maximum scattering information. A standard Berger Ball mixer is built into the neutron head. Reactions takes place in a 1 mm light path auartz cuvette.

The stopped-flow instrument can be fitted onto the neutron beamline using the umbilical link to mount the neutron head if needed.While the maximum flow rate is usually limited to 2 ml/s, with the neutron cell, the overall dead time is determined by the acquisition speed of the detector.



X-Ray accessory

Time Resolved Small Angle X-Ray Scattering (TR-SAXS) has become a popular technique to characterize nanostructures.

The X-Ray observation head is engineered with a 60° opening to maximize the solid angle so that both Small Angle (SAXS) and Wide Angle (WAXS) scattering applications can be covered. A standard Berger Ball mixer is built into the X-Ray head. The X-Ray head is available with three sizes of capillary holder (OD 1.1, 1.5 and 2 mm). All holders can be fitted in the same observation head. A special capillary holder with 90° windows is also available for simultaneous optical and X-Ray measurements. Depending on capillary geometry a flow rate up to 7-8 ml/s can be used.

The SFM can be fitted onto the synchrotron beamline or bench-top X-Ray station. If space is not available around the beam line, the X-Ray head can be combined with the umbilical link.

X-Ray head includes:

- observation head with large opening window,
- capillary holder,
- Berger Ball mixer,
- hard-stop (capillaries not supplied).

The Neutron Scattering Qccessory includes:

- observation head with large opening window,
- FFKM O-rings (stopped-flow must also be equipped with FFKM for full solvent compatibility),
- auartz cuvette,
- Berger Ball mixer (hard-stop not included).

Typical applications include:

- soft condensed matter,
- micelles and vesicles.
- protein/protein interactions,
- change of guaternary structure.

S	nec	ific	atic	m
	pec		auc	113

Solid angle aperture	44°C
Light path	1 mm
Temperature control	yes
Spare cuvette part number	OPT3100Q

Compatible with











Specifications

Solid angle aperture	60°C	
Maximum flow rate	8 ml/s	
Temperature control	yes	

Compatible with



V-Day accorren

A-Ruy ucces	SOL Y	Catalog II
X-Rav	for small capillaries, OD 1.1 mm	053-11/18
upgrade	for medium capillaries, OD 1.5 mm	053-11/25
	for large capillaries, OD 2 mm	053-11/36

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Miscellaneous

Cuvette

The SFM observation head was designed so that the cuvette can be changed in seconds without draining the temperature control circuit. The choice of a cuvette is made depending on the detection technique, the signal level needed, and the rate of the reaction to follow. The dead time is inversely proportional to flow rate and therefore cuvette volume. The signal level is proportional to path length. The fastest dead time (0.2 ms) is achievable with the microcuvette accessory (μ FC-08). Longer light paths are usually more adapted for absorbance measurements and shorter light paths for fluorescence.

Cuvette	light path	Catalog n°
FC-08	0,8 mm	054-08
FC-15/7.5	0,75 mm and 1,5 mm	054-15/7.5
FC-15	1,5 mm	054-15
FC-20/10	1 mm and 2 mm	054-20/10
FC-20	2 mm	054-20
TC-100/10F	1 mm and 1 cm	054-60
TC-50/10F	1 mm and 5 mm	054-53
μFC-08	0,8 mm	053-05
µTC-100/10	1 mm and 1 cm	053-05/3
TC-50 cuvette holder		054-62
TC-100 cuvette holder		054-63
FC cuvette holder		054-64

Syringes

All SFM instruments are delivered with 10 ml reservoir syringes as standard. Smaller syringes are available for large mixing ratio applications or titrations. Plungers are also available as spares. Syringes are generally user changeable.

Complete syringues (barrel + plunger)	Catalog n
1.9 ml syringe	950-27/1
6.8 ml syringe	950-27/2
10 ml syringe	950-27/3
3.6 ml syringe	950-27/4
Plungers only	
1.9 ml syringe plunger for SFM-X000	950-27/6
6.8 ml syringe plunger for SFM-X000	950-27/7
10 ml syringe plunger for SFM-X000	950-27/8
3.6 ml syringe plunger for SFM-X000	950-27/9

O-rings

All SFM instruments are delivered with Viton® O-rings as standard. These are suitable for any biochemical application with aqueous samples. For applications requiring the use of organic solvents, the SFM must be equipped with Perfluoroelastomer (FFKM) O-rings. These can be installed at the time of manufacture, or added later.

Standard kit		Catalog n°
Standard Viton® O-ring kit		950-00/20
For full solven	t compatibility	
O-ring kit FFKM	for SFM-2000/S	950-00/30
-	for SFM-3000/S	950-00/31
	for SFM-4000/S	950-00/32
O-ring kit FFKM	for /Q upgrade SFM-3000	950-00/33
	for /Q upgrade SFM-4000	950-00/34

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SFM accessories :

- Temperature jump
- Conductimetric applications
- FT-IR stopped-flow
- Umbilical link
- Neutron head
- X-Ray/beam line studies

- Automatic titration
 Cryogenic applications
 Chemical quench-flow
 Freeze quench
 EPR stopped-flow
- Optical quench
- Optical delay line

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