AFS™ G2

Parallel Single-Molecule Force Spectroscopy

AFS I Product Brochure

Acoustic Force Spectroscopy (AFS™) is a new single-molecule and single-cell manipulation method capable of applying acoustic forces on hundreds of biomolecules in parallel for precise experimentation with high throughput.

LUMICKS
Capture Molecular Interactions

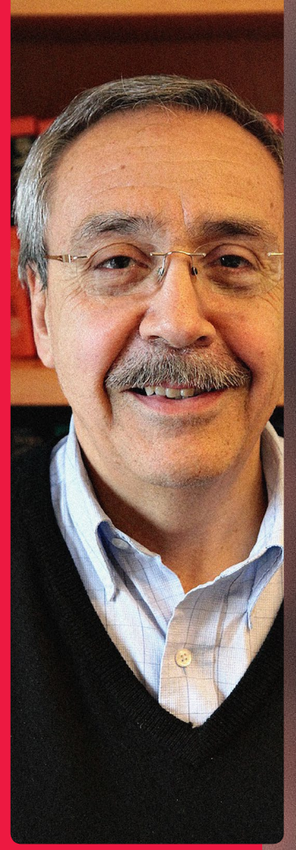
C-Trap | m-Trap | **AFS** | z-Movi

The key to unlocking dynamic single-molecule analysis.

Our dream is to unlock dynamic single-molecule analysis. Understanding the root cause of diseases at the molecular level is one of the greatest scientific challenges of today. Expanding the knowledge of biological processes that are at the basis of disease is key for prevention and the development of future cures. We aim to create the best possible tools for researchers to perform high-quality, high-throughput single-molecule experiments in the most accessible manner.

With this in mind, we bring to you the a new generation of the Acoustic Force Spectroscopy platform. We are always looking for ways to advance the technology and to bring new features that enable breakthrough discoveries.

The new **AFS™ G2** brings novel features and improved performance, such as the new automated XY stage, the drift correction option, integrated temperature control, and an individual electronic fingerprint per chip. These developments increase the data quality and improve the ease-of-use for highly-multiplexed force spectroscopy measurements.



Prof. Carlos Bustamante

HHMI investigator and professor at the University of California, Berkeley

On the importance of single-molecule research.

Adapted from Bustamante, Carlos. "Optical Tweezers: Single Molecule Manipulation in Biochemistry" Lecture, iBiology, September 2010.

*"Single-molecule experiments allow you to follow the dynamics of the reactions; not just the average behavior of molecules, but how individual molecules behave over time, providing **incredible insights** into the function of individual biomolecules."*

Discover the power of sound

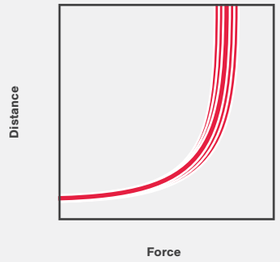
Deciphering complex molecular interactions requires many independent single-molecule measurements in order to distinguish heterogeneous behavior and rare events from intrinsic stochasticity caused by thermal fluctuations. As for cells, many measurements are required to map cellular heterogeneity within a population. With LUMICKS' groundbreaking AFS technology, scientists are now able to probe thousands of individual particles in parallel (such as DNA, RNA, proteins and living cells), allowing statistical analysis of the mechanical properties of biological systems based on a single experiment. The essence of the AFS technology lies in a glass microfluidic chip with a piezo element which generate resonant acoustic waves (ultrasound), used to exert forces on micrometer sized particles with a different density than the surrounding medium.

With this revolutionary method for manipulating large numbers of biomolecules and cells scientists now have the potential to understand life to the smallest detail, which is critical for life science research and drug development. These highly parallel single-molecule and single-cell measurements have proven to be research game changers. And LUMICKS makes this technology available as ready-to-use instrumentation to allow you to focus on their research and make the next wave of scientific discoveries.

Want to learn more about LUMICKS? Visit www.lumicks.com for more information!

Force-distance measurements

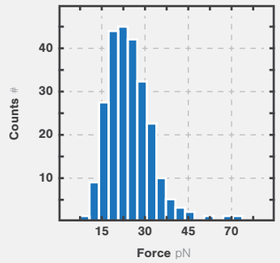
Examples of typical data output using the AFS technology include force-distance curves of multiple single molecules, such as DNA, under the influence of different proteins and binding partners or buffers.



▶ Multiple DNA molecules are tethered between a bead and a glass surface. The molecules are stretched by pulling the beads away from surface while the position of the beads is measured. This makes it possible to derive the force-distance curves of multiple molecules in parallel.

Bond rupture measurements

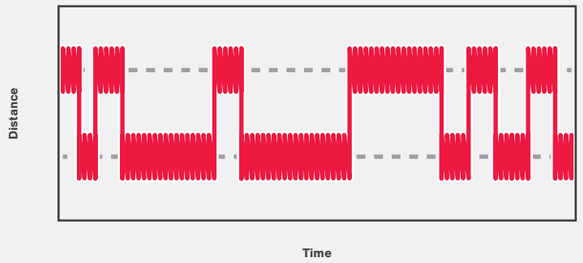
The broad force range (fN to hundreds of pNs) together with the extensive loading rate (fN/s to nN/s) capabilities of the AFS makes it possible to perform massively parallel experiments involving bond rupture. Moreover, controlling the temperature enables experimentation under biologically relevant circumstances.



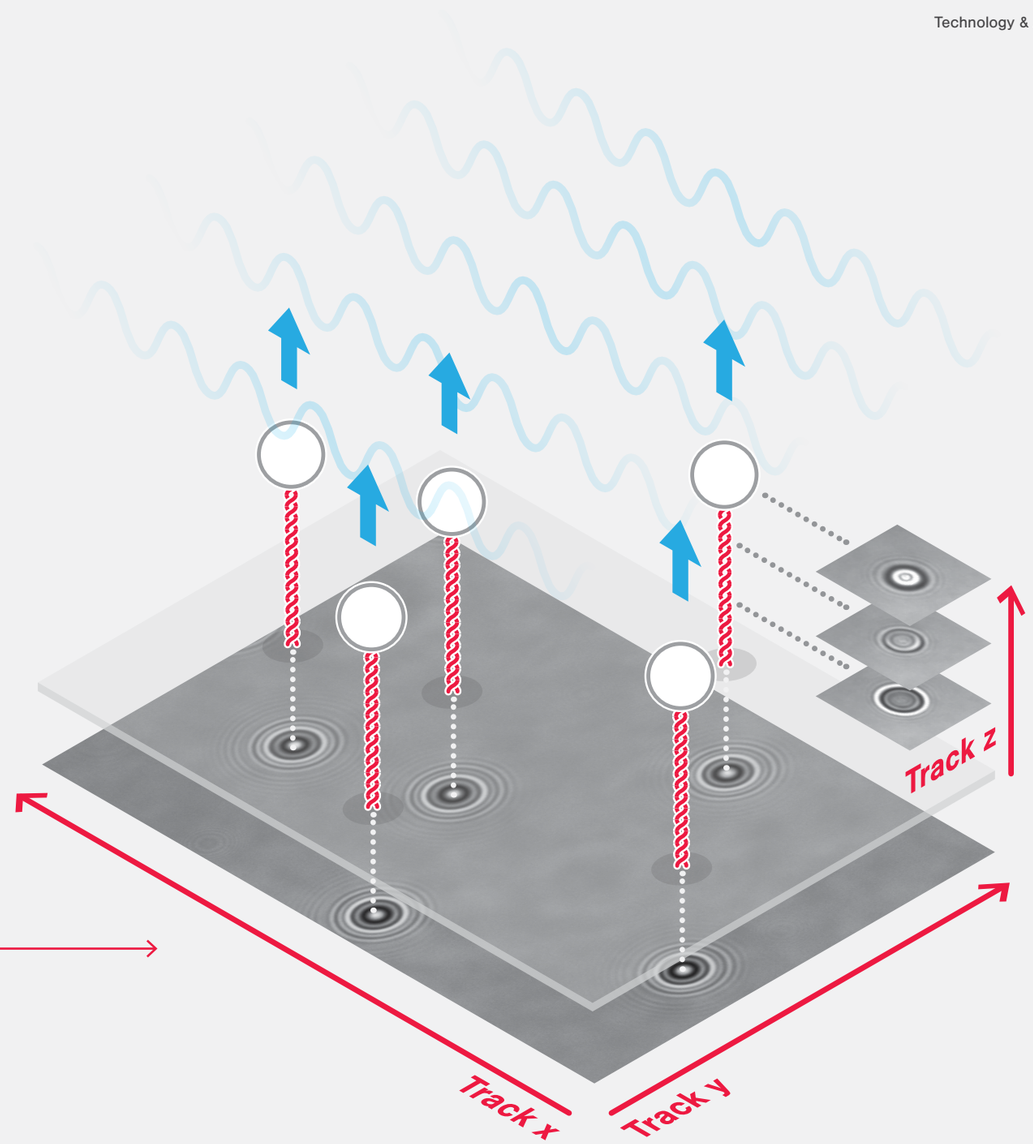
▶ Multiple antibody labeled beads are interacting with antigens connected to the glass surface. By pushing the beads away from the glass surface with an increasing force while measuring the position of the beads, it is possible to count how many antibody-antigen interactions break at which rupture force.

Constant force measurements

Equilibrium dynamics of biomolecular states can be measured by performing constant force measurements on many molecules in parallel.



▶ Multiple proteins are tethered between beads and the glass surface, using DNA handles on each sides. By holding the beads at a constant force, the equilibrium folding and unfolding dynamics are observed.



Multiplexed single-molecule force spectroscopy

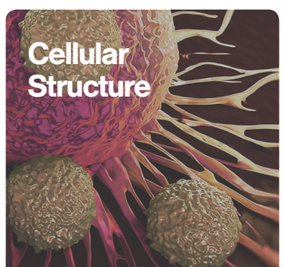
Multiple molecules, such as DNA, are tethered between a bead and the glass surface of the chip. The molecules are stretched by pulling the beads away from the surface towards the acoustic node. By simultaneously measuring the z-positions of the beads using image analysis, the extension and mechanical properties of the molecules or cells can be determined. The importance of measuring many biomolecules in parallel lies in the fact that many independent measurements are often needed to distinguish heterogeneous behavior and rare events from intrinsic stochastic behavior caused by thermal fluctuations.

Applications

Acoustic Force Spectroscopy

The essence of the AFS™ technology lies in a glass microfluidic chip with a piezo element that generates resonant acoustic waves (ultrasound). These resonant acoustic waves are used to exert forces on micrometer-sized particles with a different density than the surrounding medium – e.g. a polystyrene microsphere or a living cell.

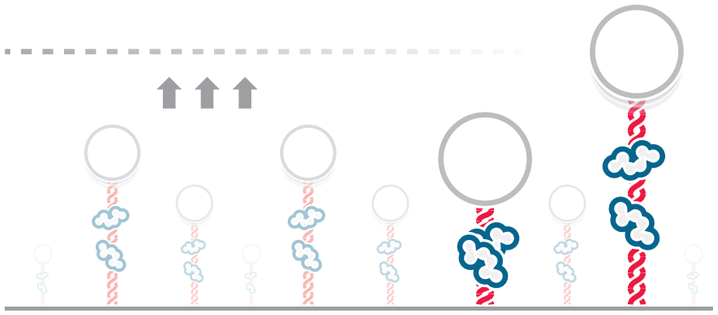
Browse the applications ⟶



DNA/RNA-Protein Interactions

Study molecular mechanisms involved in DNA repair, replication, transcription, translation, and organization with high-throughput.

The highly parallel measurement capability of the AFS makes it very useful for studying the mechanical properties of biomolecules, such as DNA or RNA molecules, in buffer solutions or in the presence of proteins, and generating datasets that have the highest statistical significance. The acoustic forces of the AFS can be used to manipulate, stretch and measure hundreds of DNA or RNA molecules at the same time. The same capabilities can also be extended to the investigation of the DNA-protein complexes involved in repair, replication, transcription, translation, and organization. This allows performing highly-multiplexed single-molecule experiments to investigate the mechanics and protein activity involved in biomolecular processes.

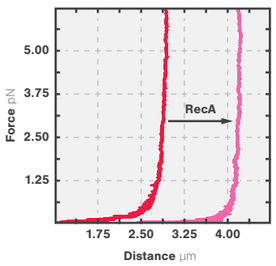


Investigation of the mechanical properties of DNA-protein interactions.

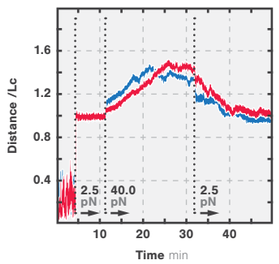
Multiple protein-coated DNA molecules are tethered between a bead and a glass surface. The DNA molecules are stretched by pulling the bead away from the surface while the position of the bead is measured. This makes it possible to obtain the force-distance curve of many protein-coated DNA molecules in parallel.

Read more:

Hill et al.
Biochemical Society Transactions (2017)



1 Force-distance curves of DNA in the absence (left) and presence (right) of RecA. Data courtesy of Prof. Erwin Peterman & Prof. Gijs Wuite at the VU University Amsterdam.



2 Normalized length-time traces of two individual DNA molecules in the presence of RecA shows force dependent binding behavior of the length increasing protein. Data courtesy of Prof. Erwin Peterman & Prof. Gijs Wuite at the VU University Amsterdam.

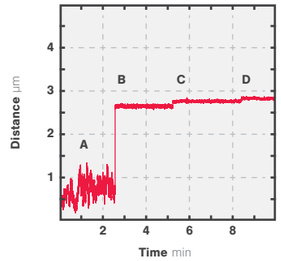
Figure 1 shows the force-extension curves of a DNA molecule (8.3 kbp) measured before (left) and after (right) incubation of 1 µM of RecA, a protein involved in DNA repair. Looking at this graph we can observe that RecA substantially lengthens the DNA.

Figure 2 indicates that AFS can also be used to obtain insights into the dynamics and cooperative behavior of RecA binding the DNA. Normalized length-time traces are shown of two individual DNA molecules in the presence of 0.5 µM RecA. At a force of 40 pN, the DNA length increases to >1.4x the contour length due to RecA binding to the DNA. When the force is set to 2.5 pN again, the length of the DNA decreases due to RecA disassembly. This indicates that the RecA binding strongly depends on tension and is enhanced by increased force. In addition, slightly different behavior of the two molecules can be observed, underlining the importance of obtaining many single-molecule measurements.

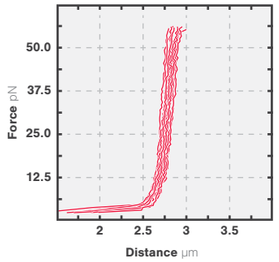
Investigation of the mechanical properties of DNA.

Multiple DNA molecules are tethered between a bead and a glass surface. The DNA molecules are stretched by using the acoustic force to pull the bead away from the surface while the position of the bead is measured. This makes it possible to obtain characteristic force–distance curves of many DNA molecules in parallel.

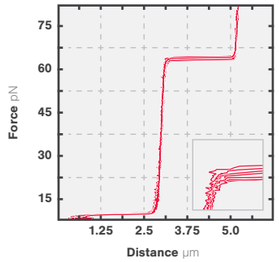
Figure 3 shows the ability of AFS to apply various levels of tension on a single DNA molecule (8.3 kbp) tethered to a polystyrene bead (Ø=4.5 µm) while measuring its length using the AFS tracking software. During the measurement, the acoustic force is increased in four steps: (A) 0 pN, (B) 1.3 pN, (C) 8.1 pN and (D) 15.8 pN. Increasing the force results in an increase of the DNA extension and lowers the thermal fluctuations of the beads, such that a length accuracy of 12.7 nm is achieved (standard deviation of the DNA extension at 60 Hz). **Figure 4** shows how the characteristic force-distance curve can be determined on many DNA molecules in parallel. Each molecule can be (over)stretched several times as observed in **Figure 5**, which shows ten sequential overstretching measurements of the same DNA molecule.



3 Extension of DNA at an acoustic force of (A) 0 pN, (B) 1.3 pN, (C) 8.1 pN and (D) 15.8 pN



4 Characteristic force-distance curves of 20 individual double-stranded DNA molecules in parallel.



5 Ten sequential overstretching measurements of the same DNA molecule

User insights

Prof. Mikhail Khodorkovskii & Dr. Georgii Pobegalov
St. Petersburg Polytechnic University

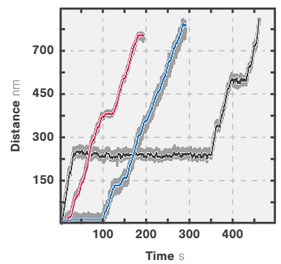
"The AFS enables our group to simultaneously observe hundreds of transcription complexes, offering a considerably higher throughput than other single molecule methods. This is a unique advantage when screening for novel inhibitors."

Multiplexed protein activity investigation involved in DNA processing.

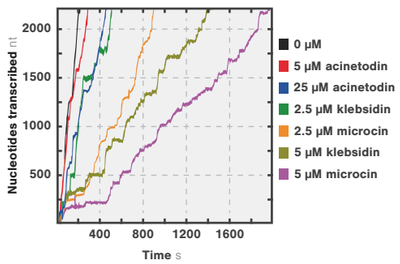
Beads can be tethered to a glass surface via a DNA-protein complex. Measuring the Z-position of the bead, while keeping the force constant, enables the study of the enzymatic activity of the protein as the z-position of the bead changes proportional to the position of the protein on the DNA.

Measurement data from a transcription activity experiment involving DNA molecules with stalled E.coli RNA polymerase (RNAP) complexes are shown in Figure 6. Once transcription is initiated, the activity of many individual E.coli RNAP molecules can be precisely monitored in parallel by measuring the RNAP position in real-time. The graph shows the typical complex nature of the protein activity; stochastically occurring enzymatic activity is frequently interrupted by pausing events of different nature.

Next, the effect of two lasso peptides, acinetodin and klebsidin, is investigated with respect to transcription elongation generated by RNA polymerase. The detection of transcription elongation was determined by the presence of varying concentrations of acinetodin, klebsidin and microcin J25; the last peptide being a known transcription inhibitor. The kinetic analysis shows that, just as microsin J25, both acinetodin and klebsidin inhibit transcript elongation by E. coli RNA polymerase, with the inhibitor activity of klebsidin being comparable to that of microcin J25 and more active than the activity of acinetodin (Figure 7).



6 Activity bursts of multiple RNA polymerase (RNAP) proteins. Courtesy of Anatoly Arseniev, Georgii Pobegalov & Mikhail Khodorkovskii at Peter the Great St. Petersburg Polytechnic University, Russia. Read more: ACS Chem. Biol., 2017, 12 (3), pp 814–824



7 Inhibition of RNAP in a single-molecule AFS experiment. Representative elongation profiles for individual RNAPs for various concentrations of microcin J25, klebsidin, and acinetodin and in the absence of inhibitors plotted as nucleotides transcribed vs time. Data were filtered by a 0.5 Hz low pass filter. Reprinted with permission from ASC Chem. Bio., 2017, 12 (3), pp 814-824. Copyright 2018 American Chemical Society.

Read more:

Metelev et al.
ACS Chemical Biology (2017)

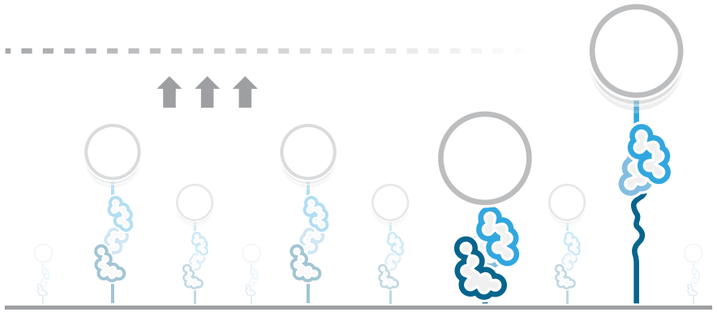
Multi-domain Protein Unfolding & Protein Function

Unravel protein (un)folding pathways and energy landscapes.

AFS lets you measure the folding and unfolding mechanisms of hundreds of proteins in parallel at the single-protein level. Investigating the dynamic mechanism of single proteins folding in detail under different biological circumstances, for example in the presence of specific ligands, represents a valuable method to extract a detailed picture of the (un)folding pathways and energy landscape of individual proteins with statistical significance.

Moreover, with the AFS it is possible to characterize the affinity between antibodies and antigens at the single-molecule level in a highly multiplexed way. Antibodies and antigens interact with each other to protect the body from unwanted complex molecules, such as pathogens. Quantifying at what force these bonds rupture provides valuable information about the affinity between these molecules.

By measuring many rupture events in parallel, the AFS platform is able to provide hundreds of independent measurements necessary to distinguish heterogeneous behavior and rare events from intrinsic stochastic behavior, and gather large datasets with statistical relevance within a single experiment.



Multi-domain protein unfolding.

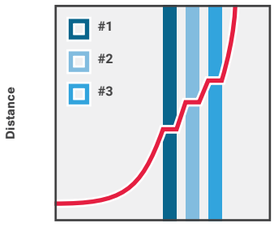
Multiple proteins are tethered between a bead and a glass surface using DNA handles on each side. By pulling the bead towards the acoustic node, the force on the protein is increased causing the different protein domains to unfold. By measuring the Z-position of the bead, the extension – and thus the unfolding pathway – of the protein can be determined. Alternatively, the bead can be held at constant force such that the equilibrium folding and unfolding dynamics can be observed.

The illustrated force-extension curve of a single protein in **Figure 1** shows that the length of the protein increases as the force is increased. Multiple protein domains unfold, causing a sudden increase of the distance between the glass surface and the bead. In this example, the protein unfolds in three steps, corresponding to three different protein domains. **Figure 2** shows an illustration of a typical protein unfolding experiment performed at constant force. Equilibrium dynamics show the transition between different intermediate states. AFS is ideal to measure equilibrium dynamics because of the intrinsic force clamp that is created when the piezo is driven with constant voltage. Additionally, AFS allows measuring many molecules in parallel, which boosts the experimental data throughput.

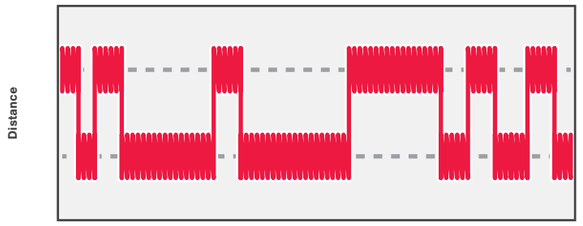
User insights

Prof.
Shishir Chundawat
Rutgers University

"With the AFS we are able to screen hundreds of ligand-protein binding interactions in parallel. This work aims to improve current protein-ligand adsorption models and to develop more efficient protein-based therapeutics as well as novel ligands for diverse applications such as industrial bioseparations."



1 Force-distance curve illustrating multi-domain protein unfolding pathway



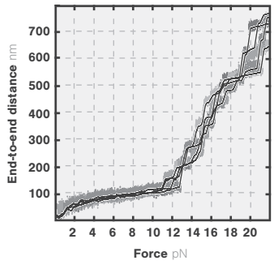
2 Protein unfolding equilibrium dynamics at a constant force

Figure 3 displays multiple force-distance curves obtained by a single Talin protein submitted to alternating stretching and relaxation cycles using AFS. In the extension-force curve shown in **Figure 3a**, consecutive unfolding cycles of an individual Talin protein can be distinguished. By overlapping the unfolding traces of a single protein, individual domain unfolding events can be detected, enabling researchers to obtain valuable information to further understand protein structure and the protein's unfolding free-energy landscape.

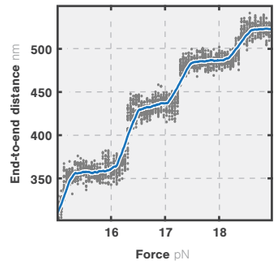
The ultrasounds generated by the AFS instrument do not harm the structural integrity of the tethered biomolecules. Thus, proteins can consecutively unfold and refold for hours in a row, allowing to obtain multiple unfolding and refolding curves using a single protein substrate. This is highly valuable as the information collected from each protein is greatly increased as opposed to other single-molecule force spectroscopy methods which typically, after a limited amount of consecutive stretching and relaxation cycles, damage the protein system due to laser-induced phototoxicity or force-induced disruption, inevitably causing the termination of the measurements.

Figure 3b depicts an enlarged snippet of an individual force-distance curve corresponding to a single pulling cycle. Here, while ramping the force from 15 to 19 pN, we observe a series of four unfolding events–corresponding to four individual protein domains–ranging between 30 and 100 nm. The unfolding events can be clearly distinguished owing to the high-resolution distance measurement capability of the AFS technology.

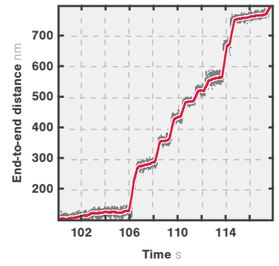
In addition, the time-distance curve of the same molecule as in **Figure 3b** is given in **Figure 3c**, displaying an 18-second range. Generally, with AFS it is possible to measure events ranging from milliseconds to >10 hours, allowing for the kinetic and thermodynamic characterization of the protein dynamics over a wide time regime.



3a Force-distance curves representing multiple stretching cycles of the same individual Talin protein

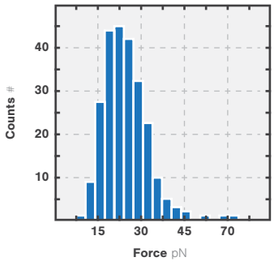


3b Zoom-in of an individual force-distance curve corresponding to a single pulling cycle, covering a force ramp range of 4 pN.



3c Time-distance curve of the same molecule in figure 3b, displaying an 18 seconds range

Sample and data courtesy of NUS, Yan Jie group.



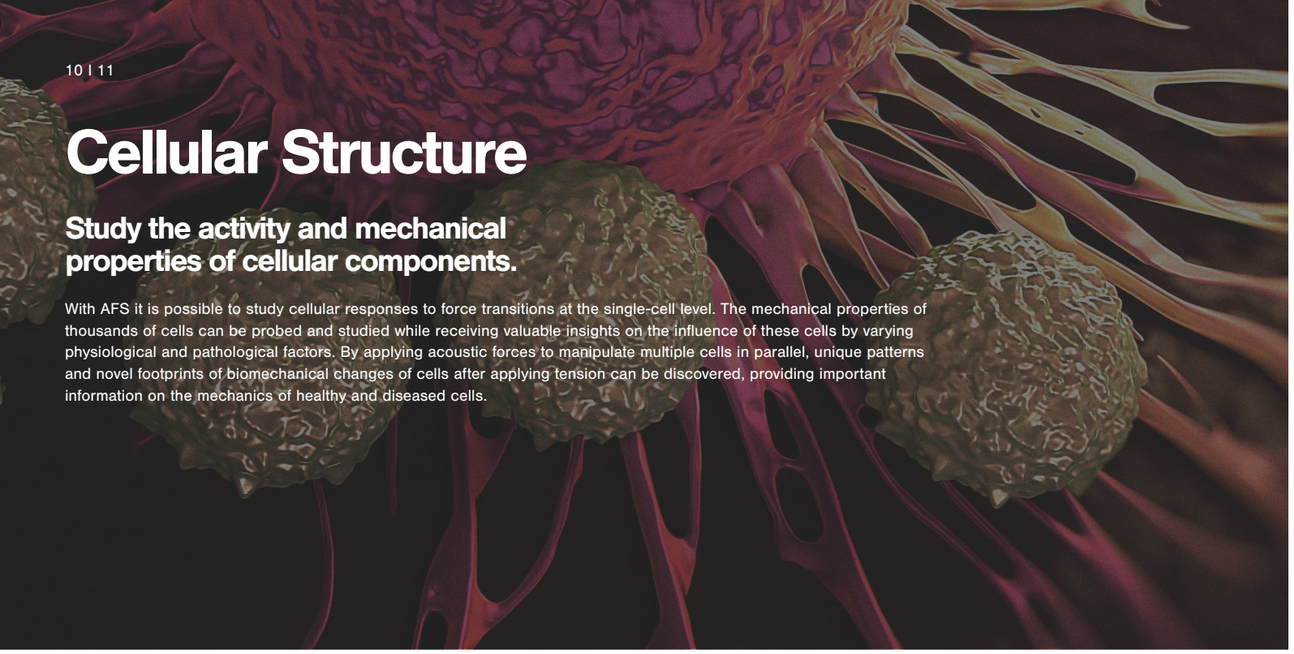
4 Rupture Forces of Dig::anti-Dig bonds.

Data courtesy of Prof. Erwin Peterman & Prof. Gijs Wuite at the VU University Amsterdam.

Antibody-antigen bond rupture measurements.

With the AFS the interactions between multiple antibody labeled beads and antigens connected to a glass surface can be investigated. By pushing the beads away from the glass surface with an increasing force while measuring the position of each individual bead, it is possible to count how many antibody–antigen interactions break and at which rupture force.

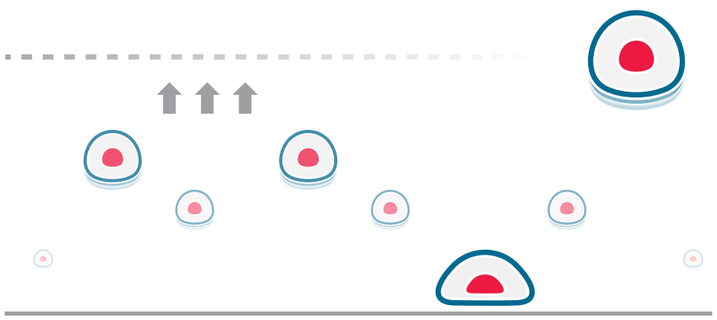
Measurement results of 251 individual Dig::anti-Dig bonds stretched with a loading rate of 2.3 pN/s are shown in **Figure 4**. The rupture force of each bond is recorded and plotted into a rupture force histogram. This histogram was obtained in a single experiment under identical conditions, ensuring experiment reliability and reproducibility.



Cellular Structure

Study the activity and mechanical properties of cellular components.

With AFS it is possible to study cellular responses to force transitions at the single-cell level. The mechanical properties of thousands of cells can be probed and studied while receiving valuable insights on the influence of these cells by varying physiological and pathological factors. By applying acoustic forces to manipulate multiple cells in parallel, unique patterns and novel footprints of biomechanical changes of cells after applying tension can be discovered, providing important information on the mechanics of healthy and diseased cells.

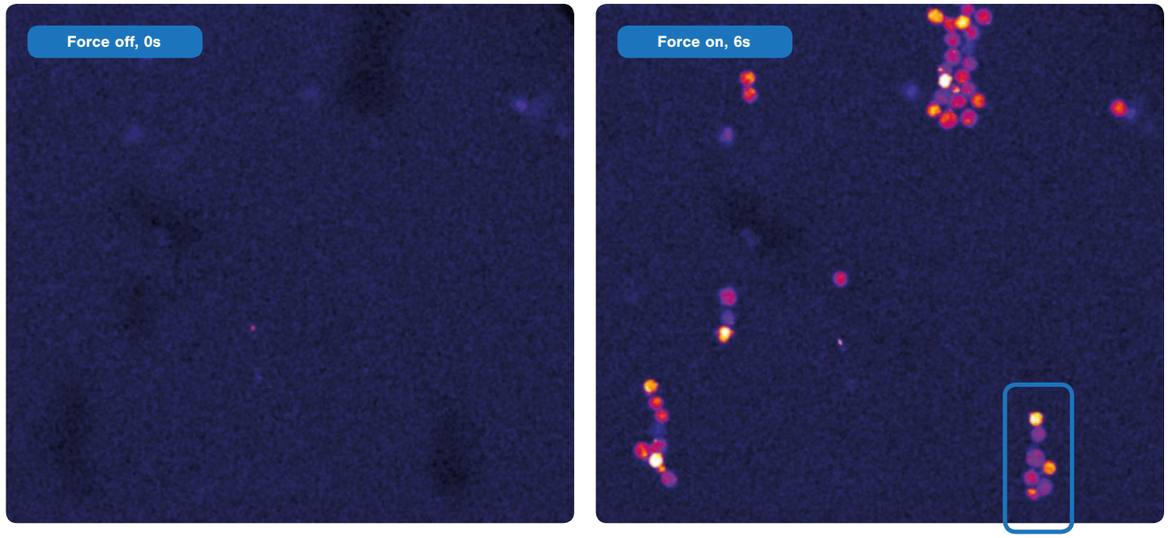


Multiplexed Cell mechanobiology Studies.

With the AFS it is possible to study thousands of cells in parallel by manipulating and imaging them. This application bears great potential for the field of mechano-biology. The illustration above shows multiple cells suspended in a flow-cell. By switching acoustic forces on and off, cell manipulation is possible, as cells cluster in the acoustic node when forces are on and sink to the bottom glass surface when forces are switched off.

In **Figure 1**, forces are applied on neuroblastoma cells (NE-115) using the AFS Module (see page 14) on a commercially available microscope (Leica TCS SP8). The imaging plane of the confocal scanning system is placed congruently with the acoustic node, approximately 40 µm above the bottom surface of the AFS chip. Once the acoustic field is applied, fluorescently labeled cells are rapidly pushed off the surface into the acoustic node and appear in the imaging plane. When the acoustic forces are silenced the cells descend by gravitation, gradually disappearing from the fluorescent image plane.

Alternatively, many cells can be cultured on the surface of the flow-cell and beads can be used to probe each cell with a known force. These types of assays can characterize the mechanical properties of healthy and diseased cells, as well as their force-induced deformations and rheological properties.



a Image captured at 0s

b Image captured at 2s

c Image captured at 4s

d Image captured at 6s

1 Fluorescently labeled cells pushed into the acoustic node where they can be visualized with confocal microscopy.

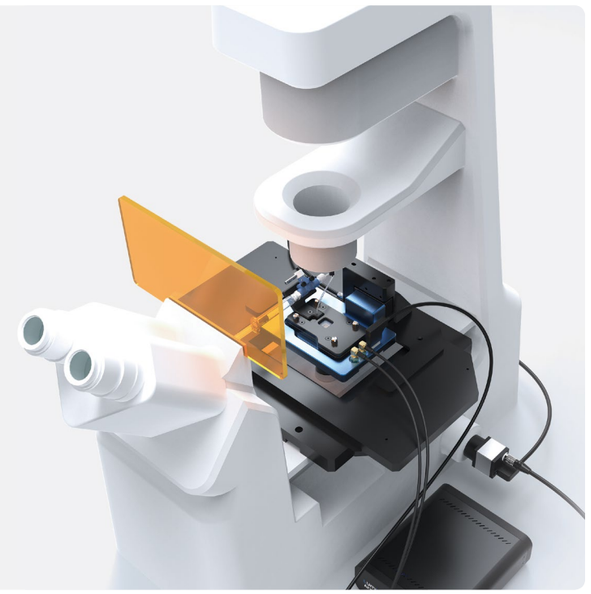
Data courtesy of Kees Janink and Bram van den Broek at Netherlands Cancer Institute.

Cell manipulation and visualization using AFS.

Massively parallel experiments involving cells typically require that dynamic forces can be applied. Furthermore, large force range (fN to hundreds to pN), broad loading rate range (fN/s to nN/s), and temperature control are required to ensure that the experimentation is performed in biologically relevant circumstances. The AFS is available both as a complete integrated stand-alone system as well as an add-on module to a custom or commercial inverted microscope. Relevant specifications for cell manipulation experiments include:

- 3 fN to 400 pN force range
- 10^{-4} to 10^{3} pN/s loading rates
- 3D tracking of up to 5000 cells in parallel
- Temperature control available
- Hours-long static force stability

Learn more about integrating AFS on fluorescence microscopy on **Page 14**



Transparent lid

User interface

Quick access buttons and a joystick allow you to adjust the sample and objective at the system when convenient.

Motorized Stage

The AFS Stand-alone provides automated sample movement in XY with both precision and speed.

Flow cells

Designed for reliability and speed

The smart flow cells contain onboard electronics to interface with the **AFS Stand-alone**, and provide basic onboard fluidics for quick and stable flow, prventing shockwaves, air bubbles and other annoyances. The flow cells can be securely inserted and removed with ease.

x5 Five flow cells are included with each **AFS Stand-alone**, giving you access to parallel sample preparation and rapid execution of multiple experiments.

iD Each flow cell has a unique electronic fingerprint—labeled and recognized by the system—providing instant adjustments and calibration on a chip-to-chip basis.

AFS Stand-alone G2

Statistical measurements, straight out of the box.

The AFS Stand-alone package allows you to perform force spectroscopy measurements straight out of the box. Start your force spectroscopy experiments right away, without the need for additional imaging tools. With a compact inverted microscope, five AFS flow cells, an intuitive image analysis software and a powerful workstation, the **AFS Stand-Alone** is the perfect solution for performing multiplexed single-molecule and single-cell experiments.

What's in the box?

A complete package for multiplex experiments

The AFS Stand-alone features a motorized objective stage with 3 different imaging magnifications, integrated camera, motorized sample stage and a temperature-controlled fluidic solution. Each of these is controlled through our software, which is purposely-designed for highly multiplexed experiments.

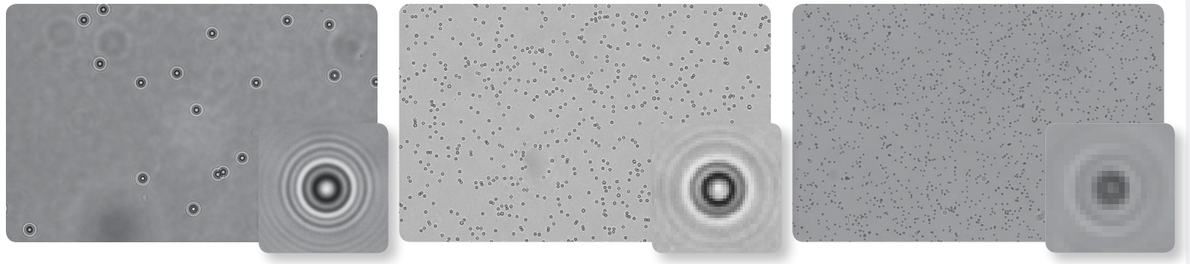
Learn more about the features of our software on page 18!

Reliable and fast workflow

Get started straight away

The AFS Stand-alone does not require extensive knowledge of the acoustics, software, or microscopy in general. The system was designed in such a way to make it easy for anyone to perform experiments from **day 1**.

Continue reading
AFS Stand-alone →



39x Magnification

Field of view: 291 x 183 µm
Bead size = 4.5 µm

This 39x magnification is perfect for use with small particles or if a high tracking accuracy is required.

26x Magnification

Field of view: 437 x 274 µm
Bead size = 4.5 µm

The 26x magnification provides a nice balance between parallelism and tracking precision

13x Magnification

Field of view: 874 x 548 µm
Bead size = 4.5 µm

The 13x magnification is ideal for highly parallel measurements where precision is not crucial.

Precision vs parallelity

Three objectives to match your perfect experiment.

The AFS Stand-alone provides you with access to three levels of magnification, allowing you to weigh precision with parallelity. Objectives can be easily switched in the system.

Integrated Temperature Control

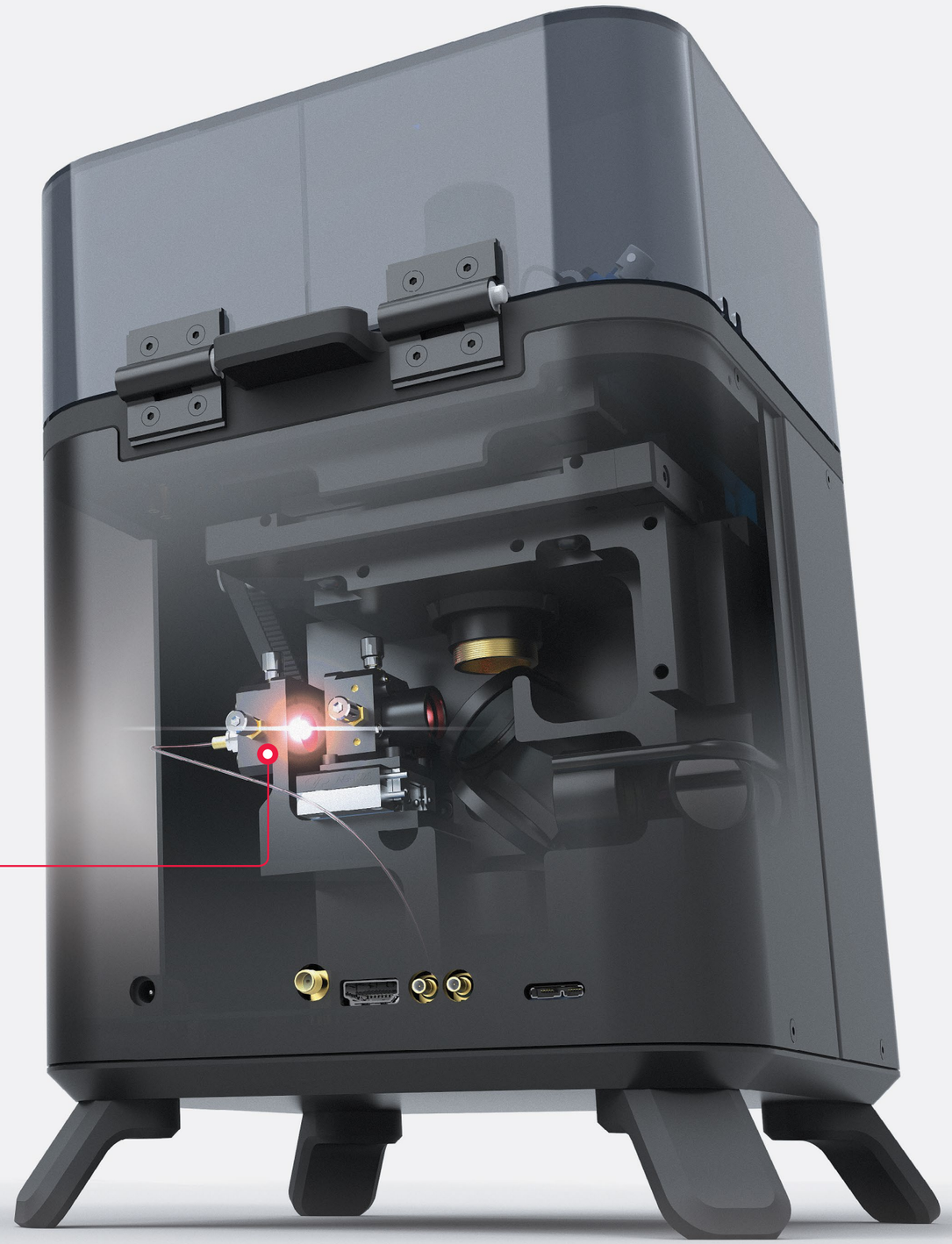
Perform experiments under biologically relevant conditions

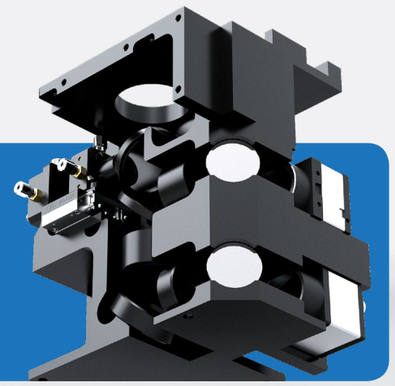
Studying cells and biomolecules often requires precise control over the environment, including thermal circumstances. **AFS flow cells** come with heating ability included, allowing you to perform experiments under physiological conditions and investigate temperature-dependent events. The new integrated temperature control feature allows users to manipulate cells or biomolecules under biologically-relevant condititons and investigate temperature-dependent events in single-molecule experiments. The flow cell applies heat over the full surface with an **absolute accuracy of 0.2°C** up to 40°C and a close loop stability of 0.1°C.

Laser-accurate tracking option coming soon

Drift correction with a variable laser-tracking method

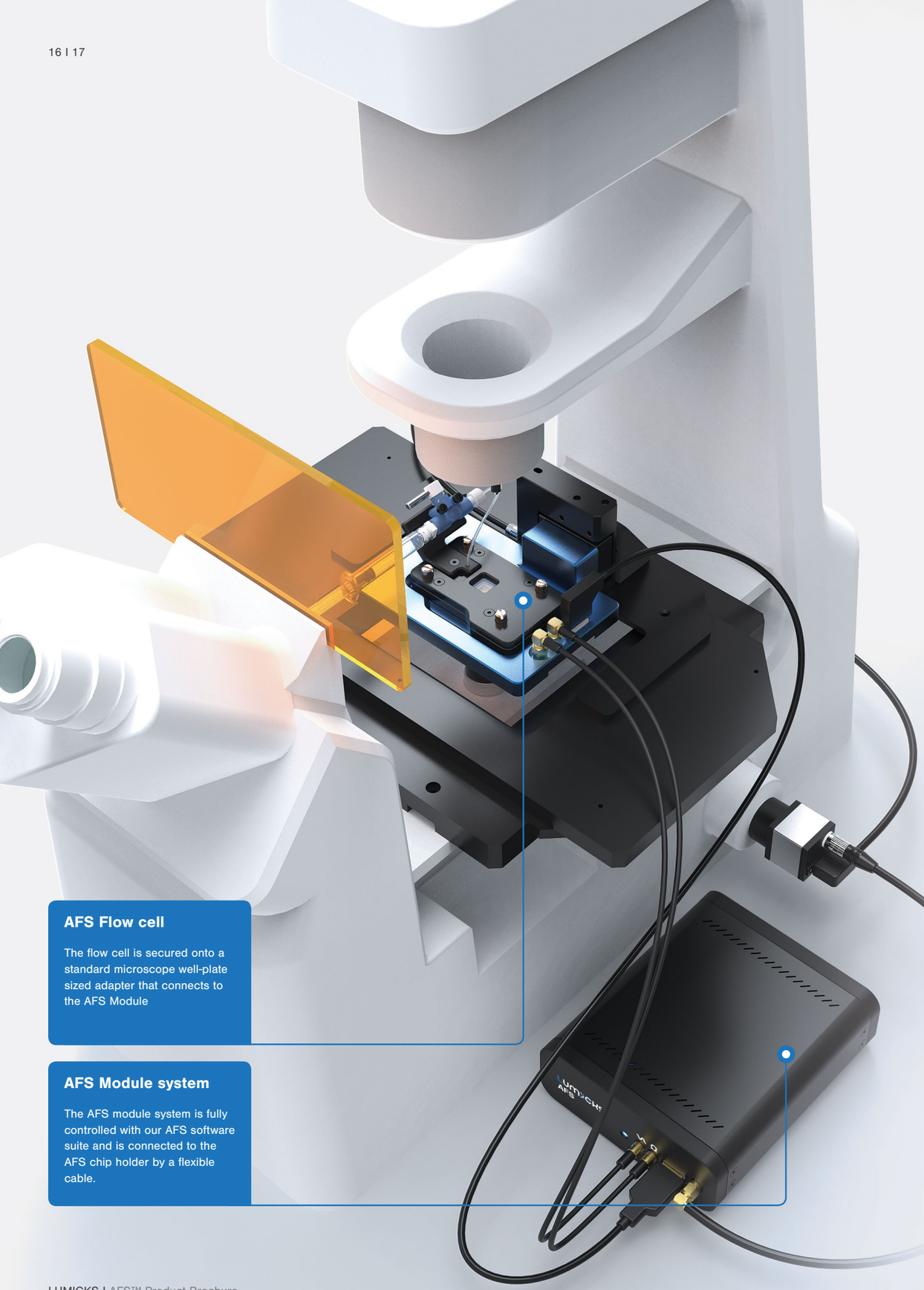
Directly boost your data quality with our new drift correct option. This new features eliminates instrumental drift by measuring sample height with nanometer precision in real-time during experiments. Using a variable laser focus the drift correction is compatible with all magnifications.





Monoblock design

The core of the system reveals our monoblock design approach. By securing all optical components to a single piece of precision-milled aluminum the system benefits from high stability and low drift. The surrounding space fits all of the necessary features and electronics, resulting in a truly compact footprint.



AFS Flow cell

The flow cell is secured onto a standard microscope well-plate sized adapter that connects to the AFS Module

AFS Module system

The AFS module system is fully controlled with our AFS software suite and is connected to the AFS chip holder by a flexible cable.

Integration example

1 XY Sample Stage

An XY sample stage for controlling your sample, which supports the z-stage and AFS flow cell.

2 Motorized Z-Stage

Including a Z-stage enables the full power of our AFS Software package for tracking your sample in Z.

3 LED Illumination

A collimated LED providing illumination for accurate 3D particle tracking can be secured in the condenser mount of commercial microscopes.

4 Camera triggering

Integrating our CMOS camera solution allows for tracking the microspheres in 3 dimensions. Triggering the camera is available for time-crucial experiments.

Flow cells

Designed for reliability and speed

The smart flow cells contain onboard electronics to interface with the **AFS Module**, and provide basic onboard fluidics for quick and stable flow, prventing shockwaves, air bubbles and other annoyances. The flow cells can be securely inserted and removed with ease.

x5 Five flow cells are included with each **Module**, giving you access to parallel sample preparation and rapid execution of multiple experiments.

iD Each flow cell has a unique electronic fingerprint—labeled and recognized by the system—providing instant adjustments and calibration on a chip-to-chip basis.

AFS Module G2

Acoustic Manipulation & Fluorescence Microscopy

Combine parallel acoustic manipulation with your own imaging methods. The **AFS Module** is designed to be combined with inverted (fluorescence) microscopes, both commercial and home-built.

Features & upgrades

A wide range of options

The AFS Module can be configured ranging from a standard system with flow cells, all the way up to a fully integrated solution as shown on the left. This combination of components and software allows you to perform force-spectroscopy measurements concurrent with your inverted (fluorescent) microscope imaging.

View the specsheet on page 21 for a complete overview

Integration options

Solutions for commercial or home-built microscopes

Depending on your microscope set-up, LUMICKS can help you select the best components for the integration of Acoustic Force Spectroscopy into your commercial or home-built microscope. In addition, we have a team of engineers ready to design a solution that fits your system perfectly.

AFS Software

A look into our parallal manipulation and measurement tool.

Bead tracking

Interactive camera view for tracked beads.

xy-Positions

Real-time visualization of the 2D coordinates of your samples.

Z-positions

Image analysis is performed on the beads, giving you a z-position for each of your samples in real time.

high throughput made easy.

A versatile software suite.

AFS systems include a versatile software suite designed for quick and effective parallel experiments and statistical data output. The software provides a variety of features designed for multiplexed manipulation, including complete control over forces, real-time tracking in 3D and quick parallel data output and analysis.

Versatile manipulation

The AFS software provides a variety of ways to exert forces on your assay. Automated and repeatable force profiles—like adjustable constant forces and force ramps—can be created user-specifically.

Real-time tracking

Track hundreds of beads simultaneously in 3D using integrated image analysis. Bead positions are recorded and shown in real-time with nanometer accuracy.

Statistical analysis

Force and position data generated by the software can be exported per bead individually or combined into a single graph with ease, making it an ideal tool for quick statistical data output.

Continuous improvements

At LUMICKS we have a software team working extensively to assure that the AFS software retains versatility for a variety of applications, while also keeping pace with evolving and new use cases. We work with our users to implement features that help them improve their research capabilities, enhancing the possibilities of the system with every release.

Spec Sheet

Unique & enabling features of AFS G2.

LUMICKS is committed to standing by you to ensure your instrument performs to specification throughout its lifetime, provide you with access to our experts for application support and service to facilitate the fastest time to result for your experiments.

Features	AFS Stand-alone	AFS Module	AFS Module + integration kit
XY-Tracking	•		•
Z-Tracking	•		•
Force calibration	•		•
Inverted microscopy compatible	•*	•	•
Applications			
Force-distance curves	•		•
Bond ruptures	•		•
Force application	•	•	•

*If the **AFS Flow cell** is integrated on an inverted (fluorescence) microscope the same functionalities and applications as the AFS Module apply.

AFS Stand-alone G2

Acoustic Forces

Maximum applicable force	>200 pN (Ø4.5 µm silica spheres)
Minium force incremental step	2 fN
Loading rates	10^{-4} to 10^{3} pN/s (limited by viscous drag)
Automated force calibration	Yes (power spectrum analysis)
Static & dynamic force measurement vertical resolution	14 bit

Imaging

Nikon CFI Achromat Air Objective	10x magnification, NA 0.30
Nikon CFI Achromat Air Objective	20x magnification, NA 0.50
Nikon CFI Achromat Air Objective	40x magnification, NA 0.65
Illumination type	685 nm LED

Motorized Objective

Minimal incremental motion (step size)	50 nm
Range	5 mm

Motorized Sample stage

Speed	0.5 mm/s
Travel range	Full piezo coverage
Minimal incremental motion (step size)	0.1 µm

Camera

Type	USB 3.0 CMOS
Resolution	1280 x 1024 px (1 px = 5.3 µm)
Refresh rate	Up to 60 Hz (full fov)

Workstation

Parallel 3D tracking	125 microspheres at 25 Hz (real-time)
Parallel 2D tracking	400 microspheres at 25 Hz (real-time)

Standard tracking

Z-drift correction	Partial (reference bead differential tracking)
Position tracking accuracy (X, Y)	2 nm at 25 Hz (real-time)
Position tracking accuracy (Z)	4 nm at 25 Hz (real-time)

Options

Temperature control

Range	Room temperature up to 40°C
Accuracy	0.2°C
Stability	0.1°C (with cover)

Camera Pro

Type	USB 3.0 CMOS
Resolution	1936 x 1216 px (1 px = 5.9 µm)
Refresh rate	Up to 166 Hz (full fov)

Workstation Pro

Parallel 3D tracking	400 microspheres at 25 Hz (real-time)
Parallel 2D tracking	1500 microspheres at 25 Hz (real-time)

Laser-tracking

Coming soon

AFS Module G2

Acoustic Forces

Maximum applicable force	>200 pN (Ø4.5 µm silica spheres)
Minium force incremental step	2 fN
Loading rates	10^{-4} to 10^{3} pN/s (limited by viscous drag)
Automated force calibration	Yes (power spectrum analysis)
Static & dynamic force measurement vertical resolution	14 bit

Integration

LUMICKS offers a variety of integration options for your commercial or home-built system to enable all the functions of our AFS Stand-alone product. Options include a motorized sample stage, illumination, camera, and a workstation with the full AFS software suite.

Motorized sample for tracking in Z

Minimal incremental motion (step size)	50 nm
Range	5mm

Illumination

Illumination type	685 nm LED

Camera

Type	USB 3.0 CMOS
Resolution	1280 x 1024 px (1 px = 5.3 µm)
Refresh rate	Up to 60 Hz (full fov)

Workstation

Parallel 3D tracking	125 microspheres at 25 Hz (real-time)
Parallel 2D tracking	400 microspheres at 25 Hz (real-time)

Standard tracking

Z-drift correction	Partial (reference bead differential tracking)
Position tracking accuracy (X, Y)	2 nm at 25 Hz (real-time)
Position tracking accuracy (Z)	4 nm at 25 Hz (real-time)

Options

Temperature control

Range	Room temperature up to 40°C
Accuracy	0.2°C
Stability	Depends on room airflow and regulation

Camera Pro

Type	USB 3.0 CMOS
Resolution	1936 x 1216 px (1 px = 5.9 µm)
Refresh rate	Up to 166 Hz (full fov)

Workstation Pro

Parallel 3D tracking	400 microspheres at 25 Hz (real-time)
Parallel 2D tracking	1500 microspheres at 25 Hz (real-time)

LUMICKS HQ
Amsterdam, The Netherlands
AFS, C-Trap, m-Trap

LUMICKS East Coast
Boston, Massachusetts, USA
AFS, C-Trap, m-Trap

Pasteur Institute
Paris, France
AFS

Dr. **Thierry Rose** and colleagues from the CITECH at the Pasteur Institute in Paris use **AFS** to manipulate human lymphocytes in living conditions.

Imperial College London
London, United Kingdom
AFS, C-Trap

In the group of Prof. **David Rueda** at Imperial College London, AFS is used for imaging the interaction and dynamics of various nucleic acid enzymes.

Chinese Academy of Sciences
Beijing, China
AFS

In the lab of Dr. **Peng-Ye Wang**, AFS is used for the study of DNA-protein interactions involved in DNA organization, such as the formation of nucleosomes by DNA.

LUMICKS around the world.

Since our first installation in 2014, our products have found themselves in the hands of many highly-respected research labs and institutes all over the world. Read up on how different labs use our technology below.

Kyushu University
Kyushu, Japan
AFS

Colorado State University
Fort Collins Colorado, US
AFS

Leiden University
Leiden, The Netherlands
AFS

UC Berkeley
Berkeley California, United States
C-Trap ***SR***, u-Flux

Hefei University of Technology
Heifei, China
AFS

Johns Hopkins University
Baltimore Maryland, United States
AFS, C-Trap

University of Münster
Münster, Germany
AFS

University of Zürich
Zürich, Switzerland
C-Trap

Ludwig Maximilian University
Munich, Germany
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CSIC Madrid
Madrid, Spain
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Rockefeller University
New York City, United States
C-Trap

Göttingen University
Göttingen, Germany
C-Trap

Max F. Perutz Laboratories
Vienna, Austria
AFS

National University of Singapore
Republic of Singapore
AFS

ShanghaiTech
Shanghai, China
C-Trap, m-Trap

FOM Institute AMOLF
Amsterdam, The Netherlands
C-Trap

LUMICKS Suppport

Our application scientists constantly travel around the globe to conferences and institutes to perform demonstrations, jobs, training and hands-on workshops. The whole LUMICKS team focuses on offering the best possible support for your research needs. Interested in how AFS works and what it can do for your research? Reach out to us to experience the possibilities yourself!

LUMICKS - Capture Molecular Interactions

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Capture Molecular Interactions