



z-Movi®

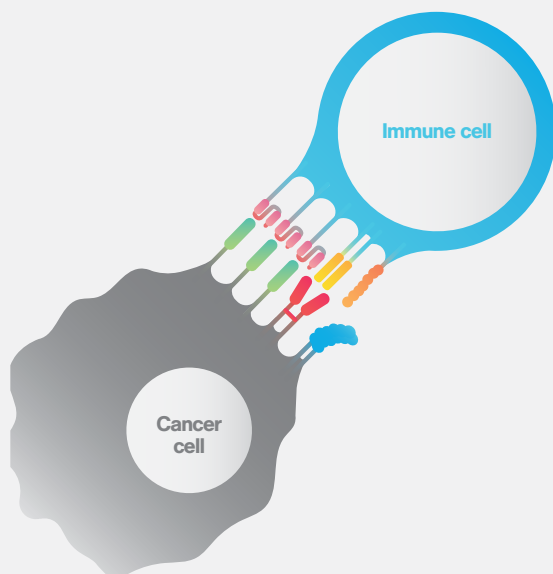
Cell Avidity Analyzer

Identify the most potent effector cell populations to accelerate immunotherapy development

■ Product Brochure

The z-Movi® is a unique instrument that enables you to identify the most potent effector cell populations, by measuring the overall binding strength – or avidity – between effector and target cells. Get avidity data from hundreds of individual cells in a matter of minutes.

The determining event for all downstream function of immune cells



INmune Bio & University College London

"The z-Movi allows us to dissect the temporal nature of the **synaptic formation** between NK cell and tumor cell [...] giving us a perfect tool to measure potency of batches of INKmune NK cells."

- Prof. Mark Lowdell, CSO and Professor of Cell & Tissue Therapy



A crucial biomarker in immuno-oncology

A superior predictor of *in vivo* response

The z-Movi® Cell Avidity Analyzer offers you a fast and simple solution for validating and optimizing immunotherapeutic strategies in a highly predictive and reproducible manner. By measuring cell avidity, the z-Movi provides you with quick and accurate results to characterize optimal immune cells for immunotherapy.

While immuno-oncology has introduced valuable therapeutic options to clinical oncology, the efficacy of adoptive cell therapy is often challenged by hostile microenvironments, relapses, or off-target toxicities. As a result, some cancer patients undergoing immunotherapy either respond partially or not at all, and several patients experience severe adverse effects.

This inconsistent response rate results from the inherently complex immune system and the context-dependent and dynamic nature of cancer cells. Clinical immunotherapeutic strategies like CAR T cell therapies, consequently, are difficult to predict, and the field lacks a proper understanding of what defines a good therapy.

The z-Movi is an easy-to-use benchtop instrument that offers a crucial parameter to the field of immuno-oncology.

Why we study avidity

Binding events between a T cell and its target tumor cell determine the initiation of immunological synapse formation and T cell activation. Cell-cell interactions are, therefore, crucial to consider when trying to comprehend T cell response processes.

While affinity reveals the intermolecular strength between a T cell receptor and a tumor-associated antigen, cell avidity defines the total intercellular binding strength. So, cell avidity refers to the combined strength between multiple parallel interactions, including co-receptor binding, TCR clustering, cell adhesion proteins, and even orientations and valencies.

Compared with affinity, cell avidity provides a more complete and physiologically relevant picture that reflects the bona fide interaction between effector cells and tumor cells. Assessments of these interactions can serve to better predict cellular responses and outcomes during immunotherapy.

The z-Movi fills the scientific gap

A complete solution for avidity studies

The approaches commonly used today to select the best immunotherapeutic effector cell candidates include surface plasmon resonance affinity studies, tetramer staining, and functional assays like cytokine secretion or cell killing analyses. Results from affinity and tetramer assays are inconsistent and do not correlate linearly with immune cell response [1,2]. Functional assays, such as IFN- γ secretion and cell killing, are more predictive of effector cell response *in vivo* but are time-consuming and can be inconsistent between experiments or assays.

Now, with the z-Movi Cell Avidity Analyzer, researchers can investigate cell–cell interactions that correspond to immune cell response. The new technology provides you with **predictive**, **reproducible**, and **fast** high-throughput results at the single-cell level without compromising cell viability. All within a compact little box that is easy to use.



Predictive

Instant binding avidity results that correlate with immune-cell function.



Reproducible

Biophysical measurements that serve as your gateway to reliable results.



Fast

Generate a complete data set within an hour.



Explore the highlights of the z-Movi's product features

High throughput

Analyze hundreds of immune cells in parallel.

Single-cell resolution

Measure the binding strength of each effector–target pair individually.

Compact

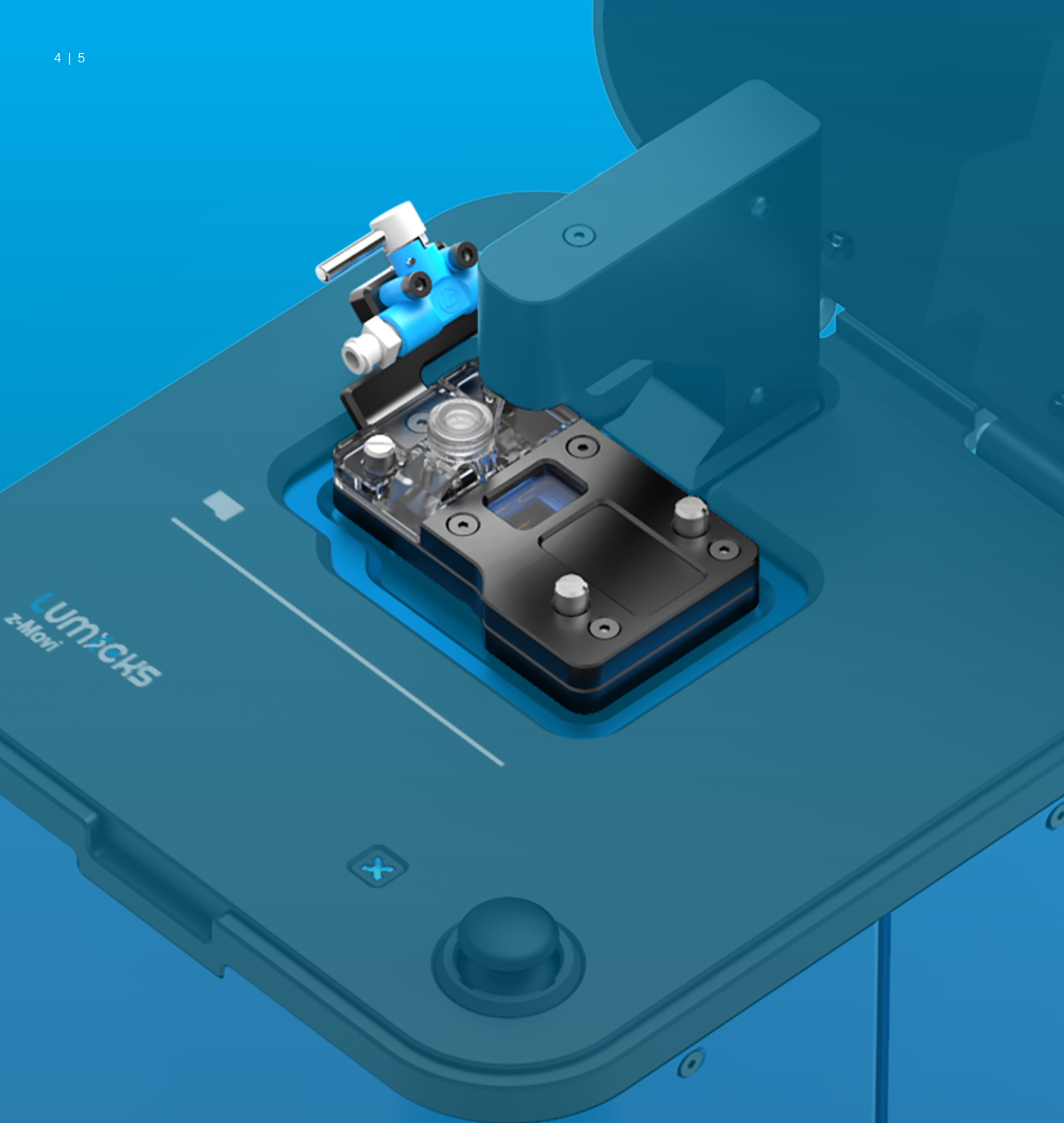
The size of a small centrifuge that easily fits in the flow hood for sterile and safe handling.

Want to learn more about the z-Movi?
Visit www.lumicks.com/cell for more information.



Read more

[1] Siebener et al., (2018) Cell; [2] Zhang et al., (2016), Science Translational Medicine



A unique **breakthrough** technology

The working principle behind the newest benchtop instrument with lab-on-a-chip.

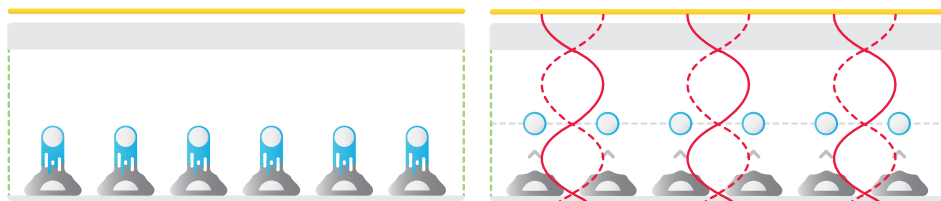
It all starts with the acoustics

At the heart of the z-Movi technology, inside the microfluidic chip, you will find the piezo element that generates resonant acoustic waves. These ultrasound waves are the foundation of cell avidity analyses, as they can pull cells vertically toward the first acoustic node where the force is zero.

You can easily control the magnitude of the force by regulating the voltage.

In a typical avidity experiment, you first culture your target cells on the surface of the z-Movi flow channel. Once the target cells have settled, you can introduce the effector cell population that you want to study, for example, CAR T cells. After a brief incubation period, apply an acoustic force ramp and measure the interaction strength as the effector cells are pulled towards the acoustic node.

This seamless approach to separate interacting cells enables you to directly measure, record, and compare cell avidities between populations of effector cells.



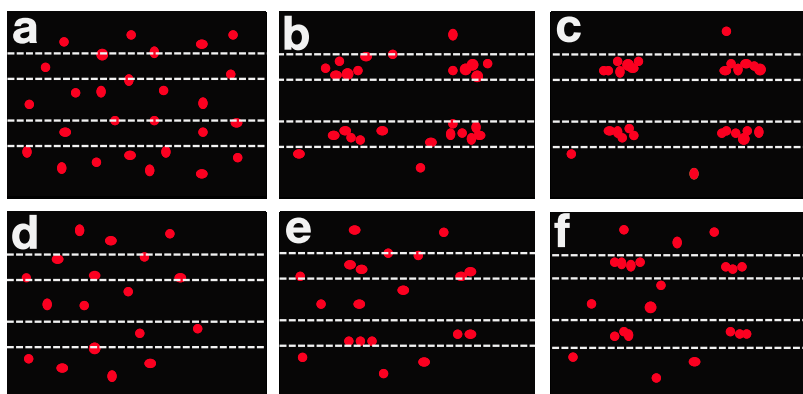
1 Schematic figure showing the principles of the z-Movi. After the effector cells (red) have settled on the target monolayer (gray), an acoustic standing wave is applied to pull the immune cells towards the nodal plane (horizontal dashed line).

And it ends with meaningful, predictive results

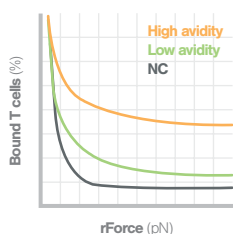
You can follow the detaching effect of the acoustic waves on fluorescently labeled effector cells from the top view of the flow channel. Effector cells will detach from the monolayer in a force-dependent manner and accumulate at the acoustic node. Meanwhile, the z-Movi records the corresponding binding strengths between each effector cell and its target cell.

The figure on the right shows the force-induced detachment of fluorescent effector cells establishing low avidity and high avidity interactions, respectively. Low avidity effector cells will readily release from the target cell monolayer (**a-c**) and gather at the acoustic nodes (**c**), while high avidity effector cells require larger forces to release from the target cells (**d-f**).

Different types of representations can be prepared to compare the avidity between different effector cell populations, as seen below. The avidity score describes the relative differences between populations of effector cells.

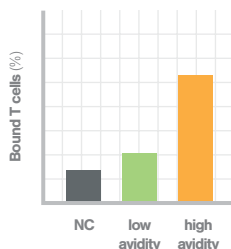


$$\text{Avidity score} = \frac{\text{Average detaching force of effector cells}}{\text{Average detaching force of the negative control}}$$



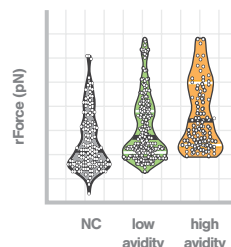
Avidity curves

Percentage of bound T cells as a function of force (NC depicts the negative effector control). rForce is relative force as calibrated with 10 μm polystyrene beads.



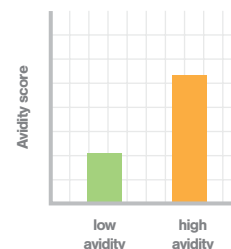
Bar graphs

The percentage of bound T cells upon the minimal NC-detaching force.



Violin dot plots

The force required to detach effector cells at single-cell resolution.



Avidity scores

Factor change in detaching-forces scaled to NC.

Applications in **immuno-oncology**

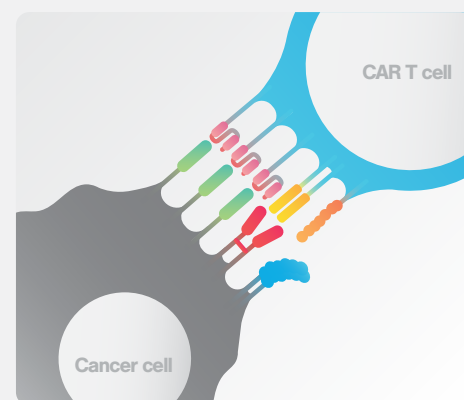
Learn how the z-Movi Cell Avidity Analyzer offers predictive, reproducible, and fast results to advance immunotherapy.

Page 7-8

Chimeric antigen receptor (CAR) engineered T cells

► A powerful therapy based on genetically engineered T cells expressing artificial receptors that specifically recognize antigens on the surface of a tumor cell. Identify the Goldilocks ("just-right") avidity CAR within a panel, and fine-tune the binding strength to avoid on-target/off-tumor binding.

- Experiment: **Identify the Goldilocks chimeric antigen receptor**
- Experiment: **Rank affinity-tuned CAR T cells**

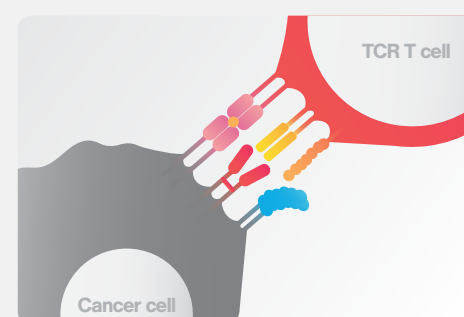


Page 9

T cell receptor (TCR) engineered T cells

► T cells with modified TCRs are optimized to recognize antigens presented by cancer cells through a major histocompatibility complex (MHC). Reveal the TCR-modified T cells with optimal avidity to obtain the best T cell response.

- Experiment: **Improve the functional correlation of TCR T cells by replacing affinity with avidity**

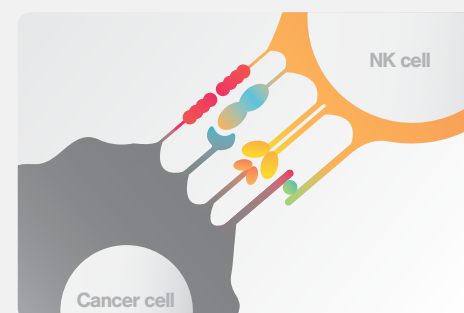


Page 10

Natural killer (NK) cells

► Immunotherapy with NK cells is an efficient allogeneic strategy in immuno-oncology. Compare the avidity of different donor NK cells and correlate the outcome to the respective functionalities, such as a cell killing. This strategy can be extended to CAR-NK cells as well.

- Experiment: **Use avidity to find the best NK donor with the optimal functional response**

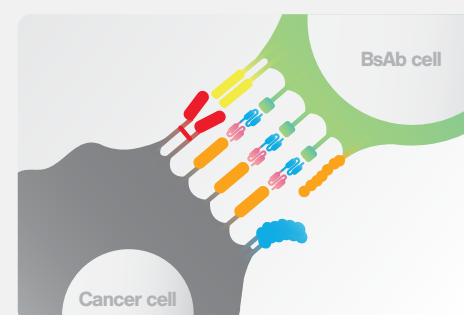


Page 10

Cell engagers

► Cell engagers, such as bispecific monoclonal antibodies, are designed to simultaneously bind a tumor antigen and a T cell. Test the binding avidity associated with different BsAb and compare them to find the most desirable response.

- Experiment: **Evaluate the avidity of T cells engaged by bispecific antibodies**





Identify the Goldilocks chimeric antigen receptor

Dr. Steven Albelda's team tested the predictiveness of CAR T cell functionality on target cells with different levels of antigen expression.

- Aim:** To identify affinity-tuned CAR T cells that bind high antigen-expressing targets without causing on-target/off-tumor toxicities on low antigen-expressing cells.
- Immune cells:** Experimental: low functioning CAR T cells (CAR^{low}) and high functioning CAR T cells (CAR^{high}) based on functional assays. Negative control (NC): non-transduced T cells.
- Target cells:** Low antigen-expressing (antigen^{low}), high antigen expressing (antigen^{high}), or non-expressing adherent cells (confirmed by flow cytometry).
- Comparison assay:** Cell killing.

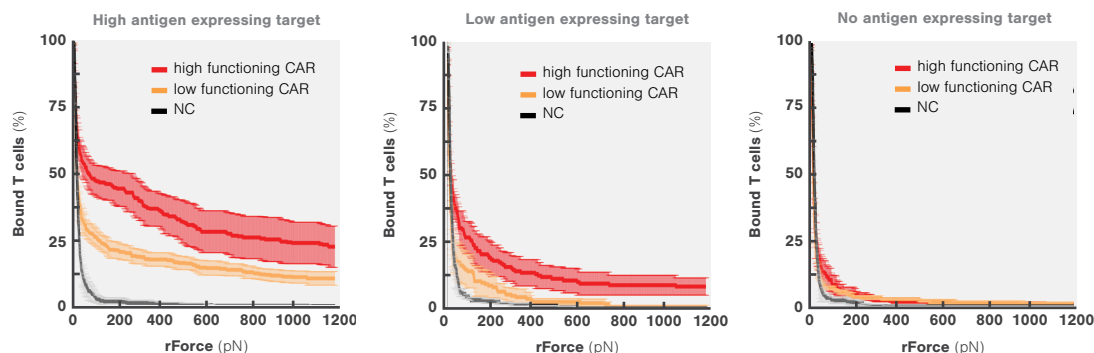


High functioning T cells may bind and kill healthy cells

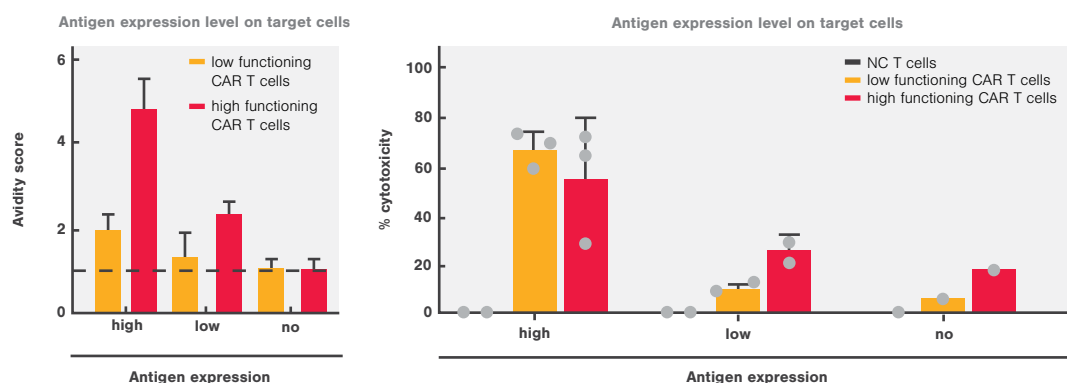
Antigen^{high} target cells induced higher avidity than antigen^{low} cells, regardless of CAR functionality (Figure 1). As expected, target cells without antigen expression established minimal binding independent of CAR functionality.

Comparison between CARs revealed that CAR^{low} induced lower avidity than CAR^{high}, independent of the antigen-levels on the target cells. CAR^{high} T cells established relatively stable interactions with antigen^{low}, suggesting that high functioning T cells may bind healthy-like cells.

The *in vitro* cell killing assay outcomes supported the antigen-dependent avidity (Figures 2 and 3). They also demonstrated a significantly higher cytotoxic response induced by CAR^{high} on healthy-like antigen^{low} cells. However, the cell killing assay could not distinguish between CAR^{high} and CAR^{low} on antigen^{high} cells.



1 Avidity curves showing the percentage of bound CAR T cells on target cells with different levels of antigen expression; high (left), low (middle), and no antigen expression (right). CAR^{high} (red) cells require higher forces to detach compared with CAR^{low} (yellow) regardless of antigen levels. (rForce indicates relative force).



2 Avidity scores based on the average detaching forces of the CAR T cell populations relative to the non-transduced T cell control population.

3 Cytotoxic effect of CAR^{high} and CAR^{low} on target cells with different antigen expression levels. High antigen expression levels on the target cells correlate with increased cytotoxicity regardless of CAR functionality compared with low antigen expression.

Can the z-Movi distinguish between T cell specificities to improve CAR T cell therapy?

- Target-cell antigen levels affect both avidity and functionality, indicating that cell avidity is a reliable readout for finding optimized CARs.
- While the cell-killing assay could not distinguish between CAR functionality on antigen^{high} cells, cell avidity measurements were sensitive enough to differentiate them.
- Cell avidity assessment distinguishes "unsafe" CAR functionalities, as observed by the on-target/off-tumor effects of CAR^{high} T cells.

Cell avidity is a valuable strategy to effectively find optimal CAR-antigen combinations with reduced on-target/off-tumor cell binding.



Data courtesy

Dr. Steven Albelda at University of Pennsylvania

Want to learn more about this experiment?

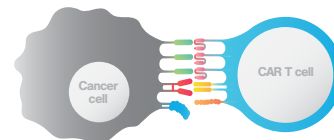
Watch our on-demand webinar at www.lumicks.com/immuno-oncology-webinars



Rank affinity-tuned CAR T cells

Leucid Bio compared cell avidity with cell killing in a panel of CARs to characterize the best functioning CAR T cells.

- Aim:** To screen a panel of affinity-tuned CAR T cells to characterize the most potent candidates.
- Immune cells:** Standard: An analogue of FDA-approved CAR T cells (original CAR).
Experimental: Affinity-tuned CAR T cells (CAR 1-5).
Negative control: non-transduced T cells.
- Target cells:** Tumor cell line.
- Comparison assay:** Long-term cell killing.



A faster way to predict CAR T cell functionality

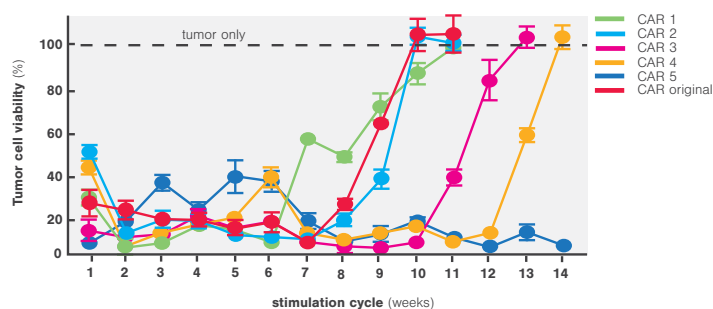
Target cells from a long-term cell viability assay were incubated with CAR T cells, and cell viability was quantified for 14 weeks.

Three of the CAR T cell populations (CAR 3, CAR 4, and CAR 5) caused significant prolonged tumor suppression compared with the rest of the panel, including the CAR original. CAR 5 efficiently suppressed cell growth until the experimental endpoint (**Figure 4**)

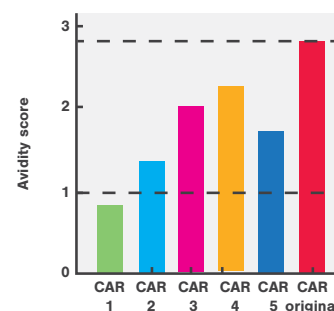
Of note, the late outgrowth of tumors observed for CAR 3 and CAR 4 (11 and 13 weeks, respectively) suggests delayed exhaustion.

The cell avidity data showed similar trends using the same panel of CARs albeit significantly faster – hours instead of weeks. Furthermore, avidity score data illustrated that CAR 3, CAR 4, and CAR 5 outperformed the rest of the CARs, including the original CAR (**Figure 5**).

Overall, the findings provide a starting point to screen and tune the avidity profiles of improved CARs to induce optimal responses with minimal side effects.



4 Long term cell viability assay (14 weeks) showing cell viability of the target tumor cell line upon incubation with specific CAR T cell populations (CAR original and CARs 1-5). Results are normalized to the tumor cells in absence of CAR T cells.



5 Avidity scores demonstrating the average detaching forces of the CAR T cell populations relative to non-transduced T cells.

Leucid Bio

London, United Kingdom

"With a technology like this, we can whittle down candidates rapidly."

- Dr. John Maher, CSO



Can the z-Movi identify affinity-tuned CARs with optimal therapeutic potential?

- Cell avidity analyses are reliable and significantly faster than cell viability assays.
- Cell avidity measurements can identify optimal binding strengths for improved functionalities.
- Intermediate avidity scores correlate with improved CAR T cell functionality – the highest avidity score showed relatively poor cell killing.

The findings provide a starting point to screen and tune the avidity profiles of improved CARs to induce optimal responses with minimal side effects. Studies on larger panels of CARs may ultimately offer quick and reliable avidity scores that discern optimized CARs for targeted cell killing.



Data courtesy

Dr. John Maher at Leucid Bio



Key Publications

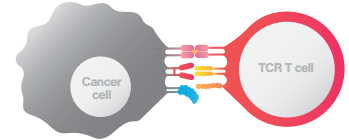
Halim et al. (2022) Front. Immunol.; Katsarou et al. (2021) Sci. Trans. Med.; Fernandez de Larrea et al. (2020) Blood Cancer Discov.



Improve the functional correlation of TCR T cells by replacing affinity with avidity

Dr. Nathalie Rufer's lab investigated the relationship between avidity and functionality of effector cells to find a reliable readout of TCR T cell function.

- **Aim:** To screen a panel of affinity-tuned TCR T cells and find the most potent candidates.
- **Immune cell profiles:** Control: One T cell population with wild type NY-ESO-1-specific TCR (WT).
Experimental: Eight T cell populations with affinity-matured TCRs derived from WT.
Immune cells selected for cell avidity assay: WT TCR transgenic T cells (baseline), DM_β (medium affinity), QM_α (high affinity).
- **Target cells:** Na8 melanoma cell line (NY-ESO-1_{neg}; +/- peptide pulsing).
- **Comparison assays:** IFN- γ secretion, Cell killing.



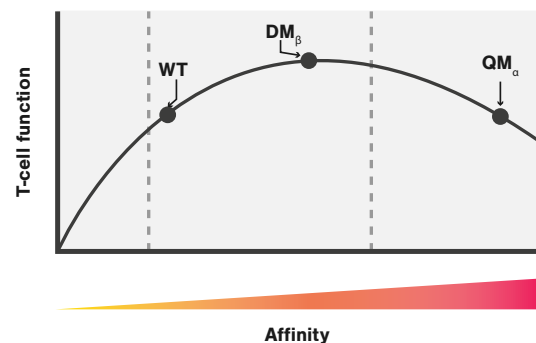
Cell avidity as a predictor of cytokine secretion and cell killing

In general, T cell populations with high antigen-affinity TCRs are functionally superior to those with low antigen-affinity. However, upon breaching an optimal K_D window for TCR affinity, T cell functionality suffers diminishing returns with reduced responses and negative T cell regulation.

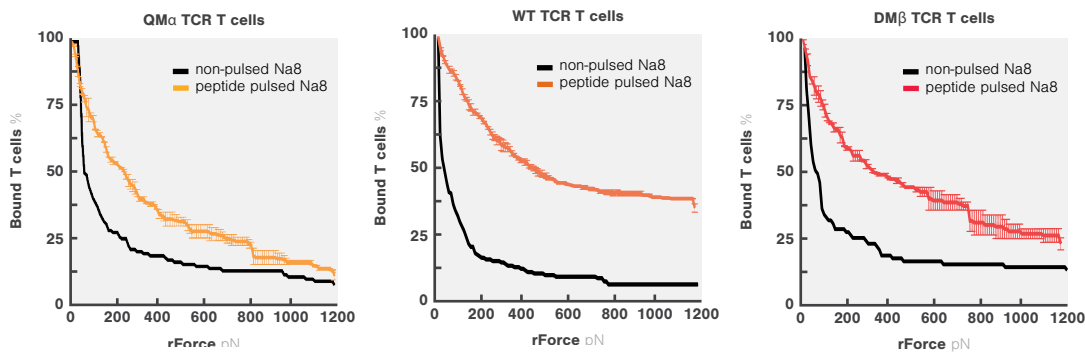
Figure 6 depicts a bell-shaped graph demonstrating increasing T cell function with higher affinities with a subsequent decline among cells with the highest affinities.

Three TCR-transduced T cells were selected for cell avidity measurements from a panel of nine populations based on their affinity levels (WT, DM_β , and QM_α ; see **Figure 6** and **summary**) for cell avidity measurements.

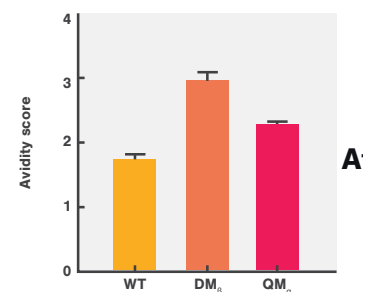
NY-ESO-1 peptide pulsing on QM_α and DM_β resulted in marked avidity enhancement compared with the non-pulsed conditions (avidity scores for pulsed conditions: 2.1 and 3.0, respectively; **Figure 7** and **8**). Of note, the difference between non-pulsed and peptide-pulsed WT cells resulted in a minimal difference (avidity score for pulsed condition: 1.7).



6 Schematic figure showing the relationship between T cell function and affinity of nine different TCR T cell populations. The two dashed lines encompass the cells in the optimal K_D window. WT, DM_β and QM_α were selected for avidity assessments.



7 Avidity curves showing the proportion of bound TCR T cells to non-pulsed or peptide-pulsed Na8 melanoma cells at increasing acoustic forces. Each plot represents measurements performed on WT (left), DM_β (center), or QM_α (right). (rForce indicates relative force).



8 Avidity scores from the three experiments based on the average detaching forces of the TCR T cell populations relative to the non-transduced T cell control population.

Can the z-Movi discriminate between affinity-matured TCR T cells?

- Avidity scores resembled the bell-shaped relationship between T cell function and affinity, explaining the diminishing returns associated with high-affinity TCR T cells.
- The reverse relationship between affinities and avidities (DM_β and QM_α) can explain the reduced functionalities of high affinity TCR T cells.

The outcomes indicate that cell avidity is a better predictor of TCR T cell functionality compared with affinity.



Data courtesy

Dr. Nathalie Rufer at the University of Lausanne



Read more

Hebstein et al. (2013) Front. Immunol.

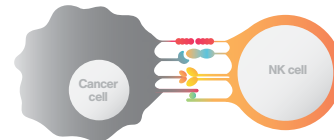


Beyond CAR and TCR T cells

Two additional proof of principle applications showcase the versatility and future of cell avidity analysis.

Use avidity to find the best NK donor with the optimal response

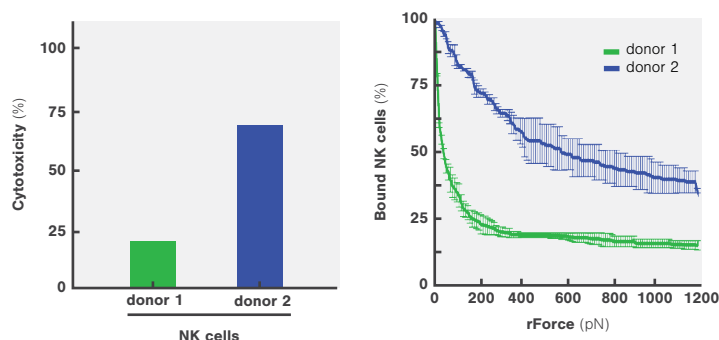
In addition to autologous cellular therapy, allogeneic strategies may facilitate an off-the-shelf solution, including NK cells as a prime example.



Glycostem researchers compared cell avidity data with cell killing by evaluating NK cell functionalities from two different donors (donor 1 and donor 2). The results showed that:

- NK cells from donor 2 induced a 3-fold higher cytotoxicity compared with NK cells from donor 1 (74% and 24%, respectively).
- NK cells from donor 2 required higher forces than those from donor 1 to detach from target cells (**Figure 9**)

The data suggest that cell avidity is a reliable predictor of NK cell function and that cell avidity analyses can be applied to different types of immune cells.



9 Left: Cytotoxic effect of NK cells from donor 1 (green) and donor 2 (blue) on their target cells. Right: Avidity curves showing the proportion of target-bound NK cells, to the same target cells, upon application of a force ramp. (rForce indicates relative force).

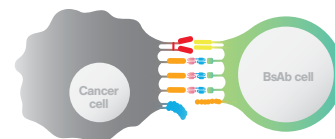


Data courtesy

Glycostem Inc

Evaluate the avidity of T cells engaged by bispecific antibodies

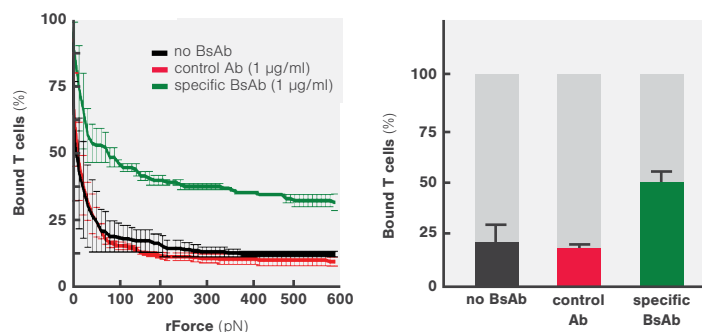
Bispecific antibodies are power tools adopted by immuno-oncology that can enhance the recruitment of T cells to specific cancer cells. The z-Movi expands the set of parameters to select the best antibodies for optimal T cell responses.



In a collaborative study, we compared cell avidities between T cells and target cancer cells in the presence of bispecific antibodies (BsAb) or control conditions (no antibodies or non-specific antibodies).

T cells with BsAb required stronger pulling forces to detach from the tumor cells when compared with the two controls (**Figure 10**).

Future cell avidity analyses of engineered BsAb, will enable researchers to discover and fine-tune optimal antibodies for T cell therapies.



10 Left: Avidity curve representing the average proportion of bound BsAb-engaged T cells upon increasing forces. Right: Bar graph representing percentage of target-bound T cells at the minimal force detaching the negative control (no BsAb) gated from the avidity curve. (rForce indicates relative force).

Perform experiments in hours instead of weeks.
Learn more about the **workflow in page 13**.

Explore the z-Movi

The newest benchtop instrument



Predictive



Reproducible



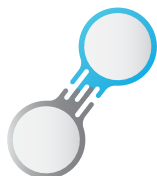
Fast





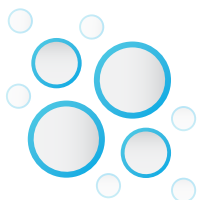
Direct measurement of binding avidity

The z-Movi introduces the robustness of a biophysical-based method to immuno-oncology to uncover the essential properties of effector cells.



Single-cell resolution

Measures the binding strength between every effector cell and its target to ensure that not a single piece of information is lost. Because each cell matters.



High throughput

Analyzes avidities of up to 400 cells in a single run and generates instant statistically relevant data. The fast workflow and easy-to-use z-Movi Chip enable you to measure multiple experimental conditions per day.



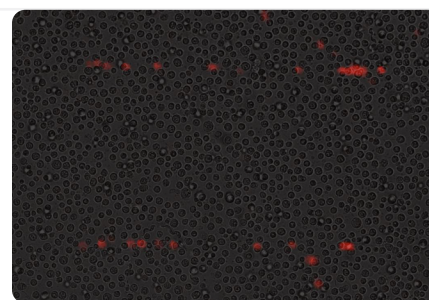
Safe to cells

Offers non-invasive acoustics to measure avidity, leaving your cells healthy and viable.



Fluorescence cell imaging

Distinguishes and follows individual fluorescent effector cells with unparalleled accuracy as they detach from their target cells and gather at the acoustic node.



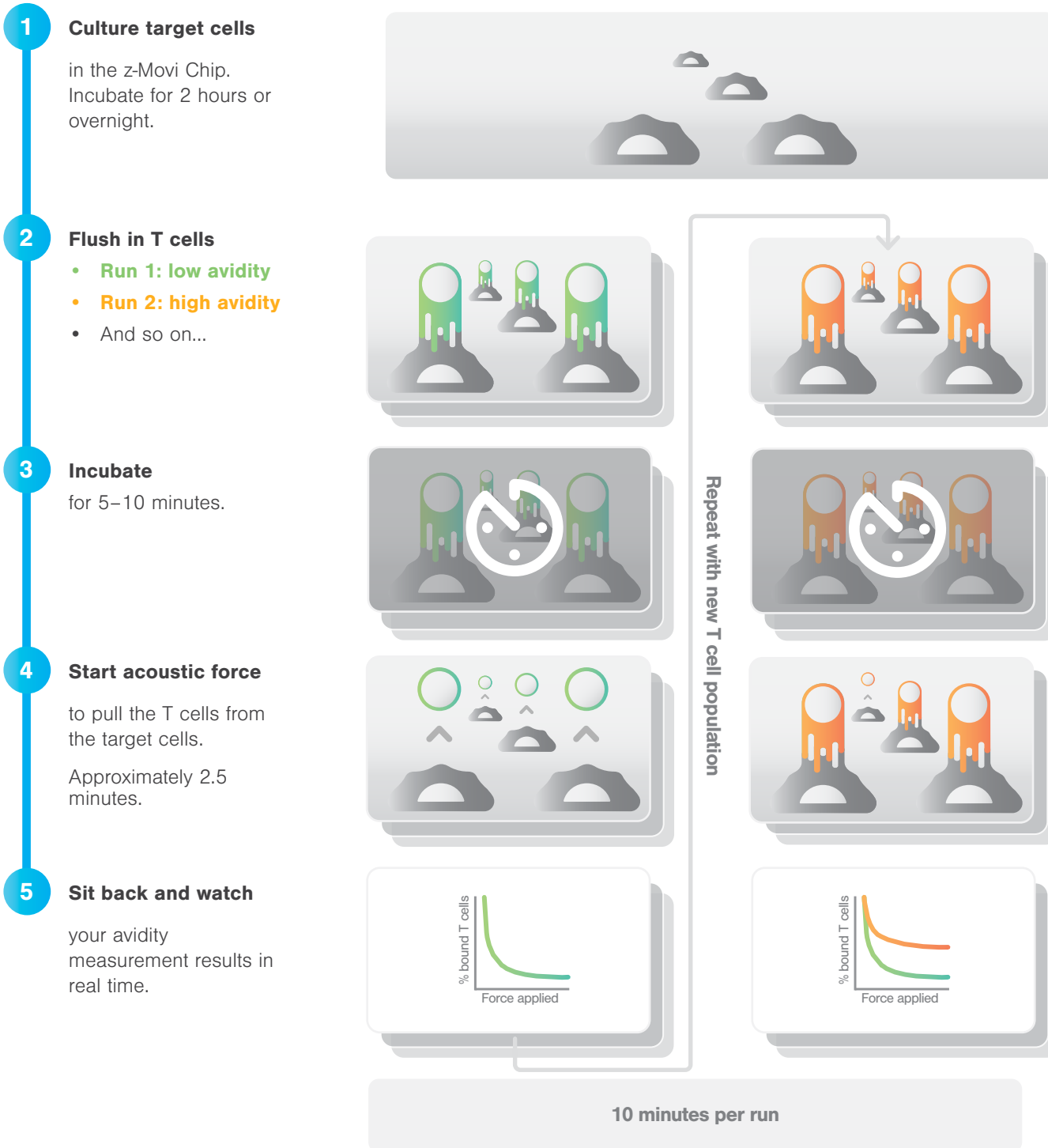
Compact

Small enough to fit inside a flow hood, providing sterile cell handling and protective conditions when working with dangerous reagents.

The z-Movi workflow

Rapid experiments through an optimized workflow

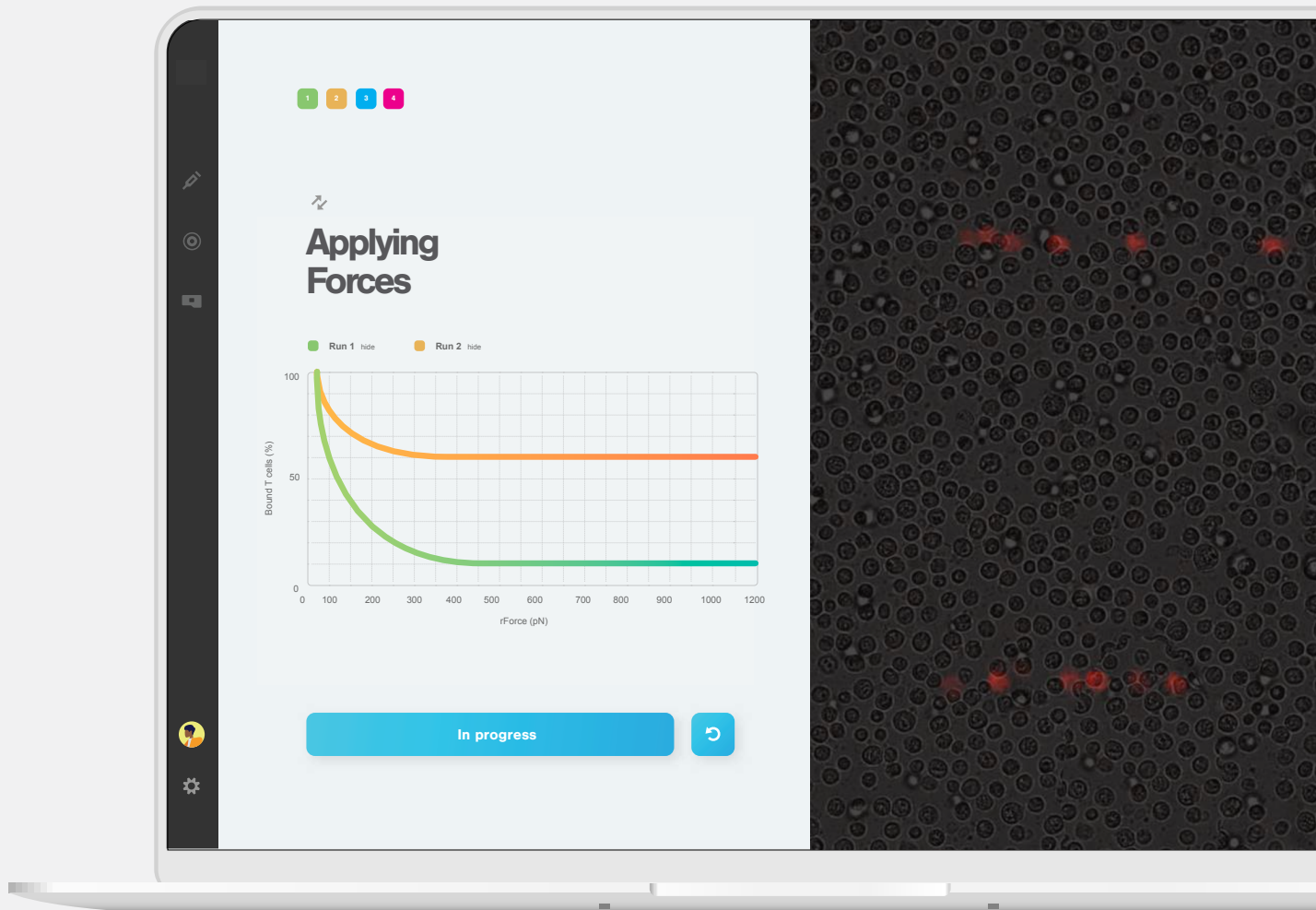
We have optimized the workflow of the z-Movi to facilitate the user experience and provide you with reliable and reproducible results in a matter of minutes. The z-Movi Chip is specialized to simplify cell culturing and maintain your samples in physiological conditions while you perform experiments. Once you have performed one run of analyses, you can simply flush in a new batch of effector cells. Running multiple chips sequentially allows you to measure several experimental conditions per day.



Dive in and discover our software

A user-friendly software that analyzes the avidity of hundreds of cells in parallel and real time

In collaboration with end users, we have developed a fast and intuitive workflow that takes you from defining your parameters to viewing your data in no-time. The powerful tracking algorithms for cell detection and measurements are performed in the background while the software generates your data live on your screen.



The features:



Powerful detection algorithms

Set the parameters and let the Wave software do the rest of the work by automatically detecting and tracking your cells of interest.



Real-time avidity data analysis

Observe your avidity curves being plotted in real time while your effector cells detach from their target cells upon force application.



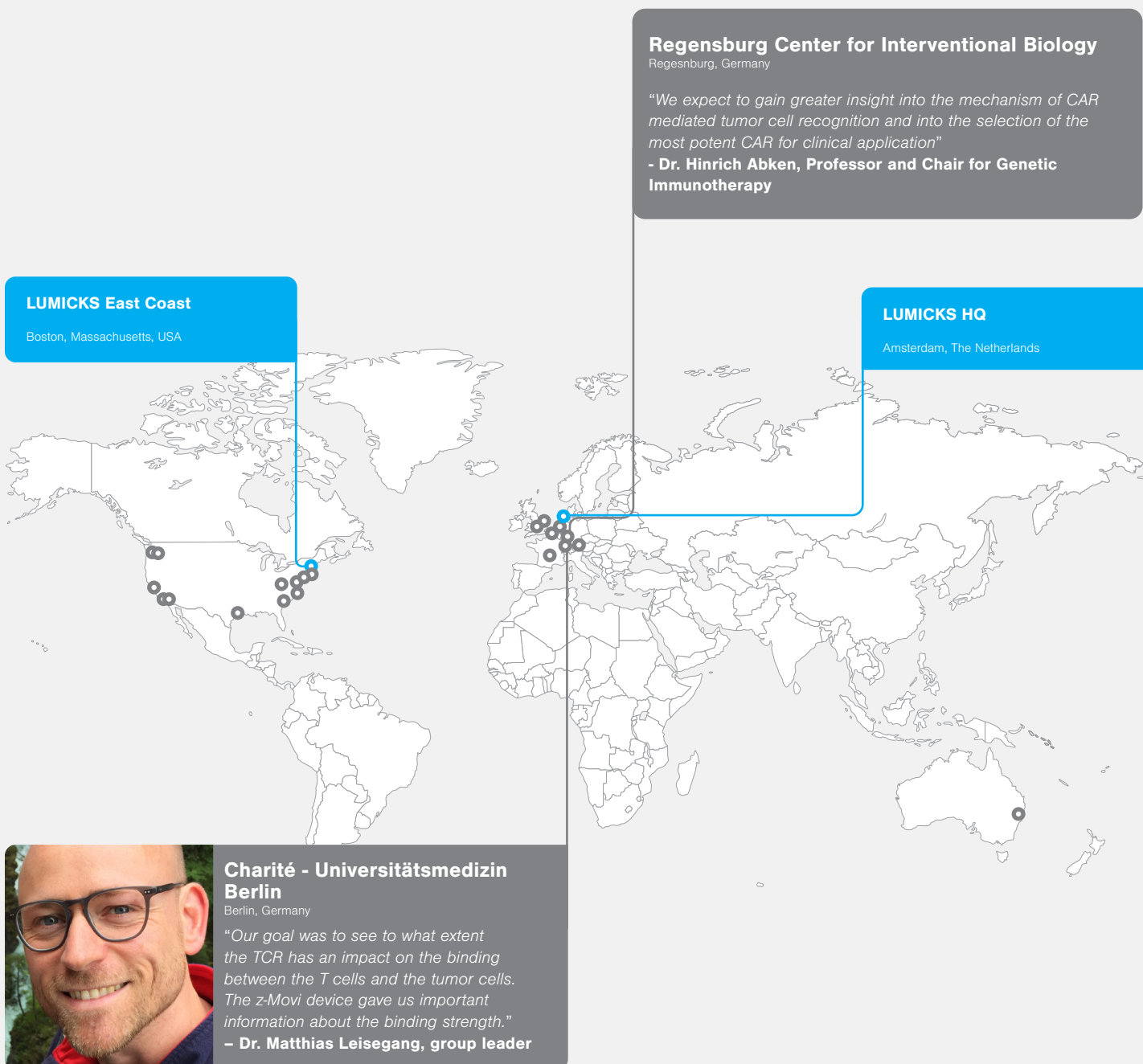
Figure-ready data plotting

Enjoy the benefits of Wave's straightforward post-acquisition data analyses that generate ready-to-present plots immediately after your experiment.



Royalty-free license

The z-Movi Wave software comes with free updates, without license costs, and unlimited support. Analyze your data anywhere and anytime!



A selection of our key collaborators

z-Movi around the world

LUMICKS Support

Our application and customer success scientists constantly travel around the globe to conferences and institutes to perform demonstrations, training, and hands-on workshops. The LUMICKS team is focused to offer the best possible support for your research needs. Are you interested in how the z-Movi works and what it can do for your research? Reach out to us and experience the possibilities yourself!

A history of milestones About **LUMICKS**

2021

Fred Hutch

Oxford University

TU Munich

University of Pittsburgh

- LUMICKS raised \$93 million in series D funding.
- With 128 new hires – including 10 in key senior positions – we more than doubled in company size.
- Continued to empower researchers with more than 40 articles published in high-impact journals using LUMICKS' technology.
- Key sales to Fred Hutchinson Cancer Research Center, Oxford University, Westlake University, Max Planck Institute for Molecular Cell Biology and Genetics, and many more.

2019

Harvard University

Stanford University

Yale University

- Validated the z-Movi for immuno-oncological cell therapies with strategic partnerships
- Built new team for immuno-oncology product line
- Key sales to Harvard University, Stanford University, Yale University, Technical University of Munich, and more
- Grew team to 100 full-time employees
- New Office HQ

2017

Imperial College London

University of Zurich

- Key sales to Imperial College London, University of California, Berkeley, University of Zurich, and more
- Launched the m-Trap® High-Resolution Optical Tweezers
- New office in the US
- 38 full-time employees

2015

- Launch of the C-Trap® Correlated Optical Tweezers – Fluorescence Microscopy
- First sales to BIOCEV, Max Planck Institute of Molecular Cell Biology and Genetics

2020

- Launched the z-Movi® Cell Avidity Analyzer
- New office in China
- Launched new services and initiatives tackling the full DSM experimental workflow from experimental design to data analysis.
- Key sales to LAVA Therapeutics, Leucid Bio, Glycostem, Memorial Sloan Kettering, Imperial College London, and more.



2018

- Key sales to National Institutes of Health, Delft University of Technology, Tsinghua University, Northeastern University, and more
- Established US team in Boston and Bay Area
- Partnership with AstraZeneca
- Developed the z-Movi® Cell Avidity Analyzer prototype

National Institutes of Health

Delft University of Technology

Tsinghua University

Northeastern University

2016

- Introduced into the market Acoustic Force Spectroscopy (AFS®) for single-molecule manipulation
- Key sales to: Rockefeller University, Göttingen University, Shanghai-Tech, and more
- First Nature publication with data captured using the C-Trap®
- 18 full-time employees

Rockefeller University

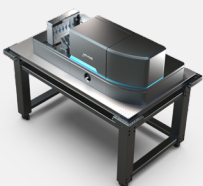
Göttingen University

Shanghai-Tech

2014

- LUMICKS founded

LUMICKS



Specification overview for the z-Movi

Number of cells analyzed	Up to 400 cells*
Maximum force applicable on 10µm polystyrene beads	Up to 1 nanoNewtons
Brightfield/fluorescence illumination	LED (670 nm)/(635 nm)
Objective/Magnification	10x/6.25x
Field of view	1 mm x 2 mm
Fluorescence detection bandwidth	660 - 750 nm
Bright field/fluorescent frame rate	5 Hz
Desktop	Lenovo Thinkstation
Screen	Dell UltraSharp 24"

*depending on experimental conditions, typically 300-400s cells are analyzed

Want to learn more?

We have a team of dedicated scientists that perform z-Movi demos in-house. Get in touch with us to find out more by emailing info@lumicks.com





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