RheoSense™

Simply Precise™

VROC®

Technology

PHARMACEUTICAL APPLICATIONS

AGENDA

1. About RheoSense
2. What is VROC® Technology?
3. Products
4. Applications
5. Results
6. Summary
7. Q&A

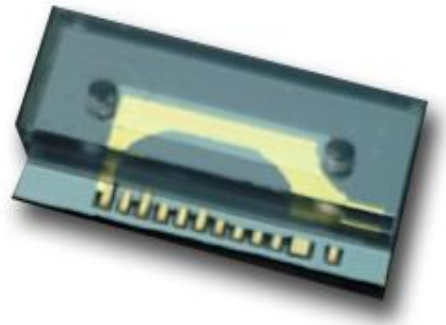
What is RheoSense?

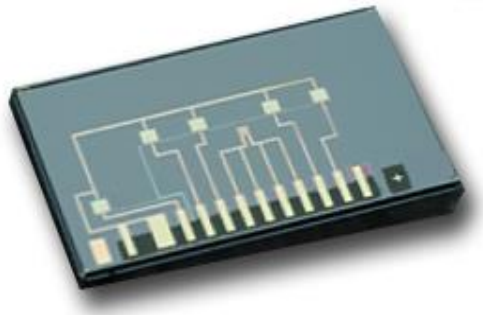
- Headquartered in Silicon Valley
- Founded in 2001
- Patented technology
- Fortune 500 client base
- Market leader in biotechnology, pharmaceutical, and the emerging protein therapeutics applications
- Opening East Coast office 2014

Our Technology

VROC®: The Viscometer/Rheometer-on-a-Chip

- Measures Absolute Viscosity
- Smallest Sample Volume (> 50 µL)
- Exceptional ease-of-use and Accuracy
- Highest Shear Rate Viscosity Measurement
- Widest Dynamic Range in Shear Rates
- Small Footprint
- Fast, Reliable Results

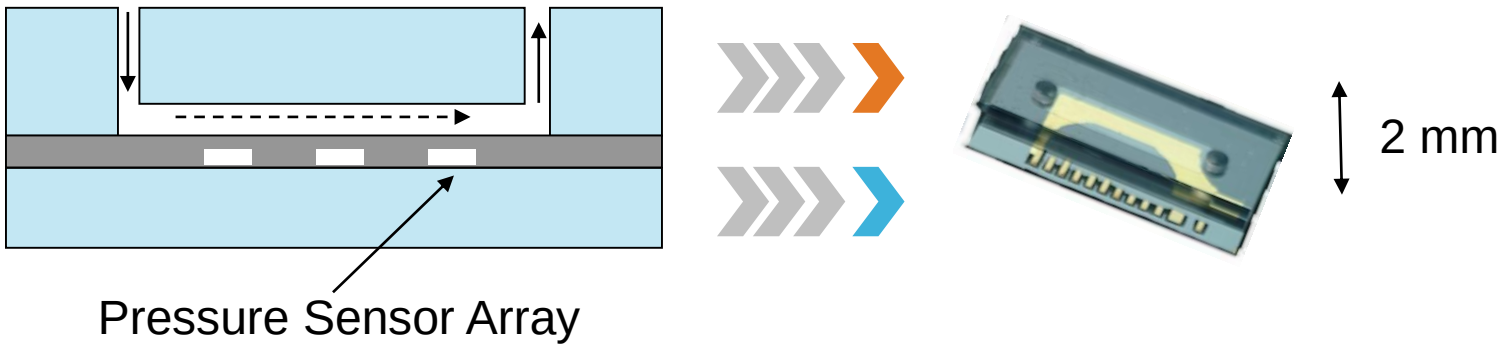




How it Works

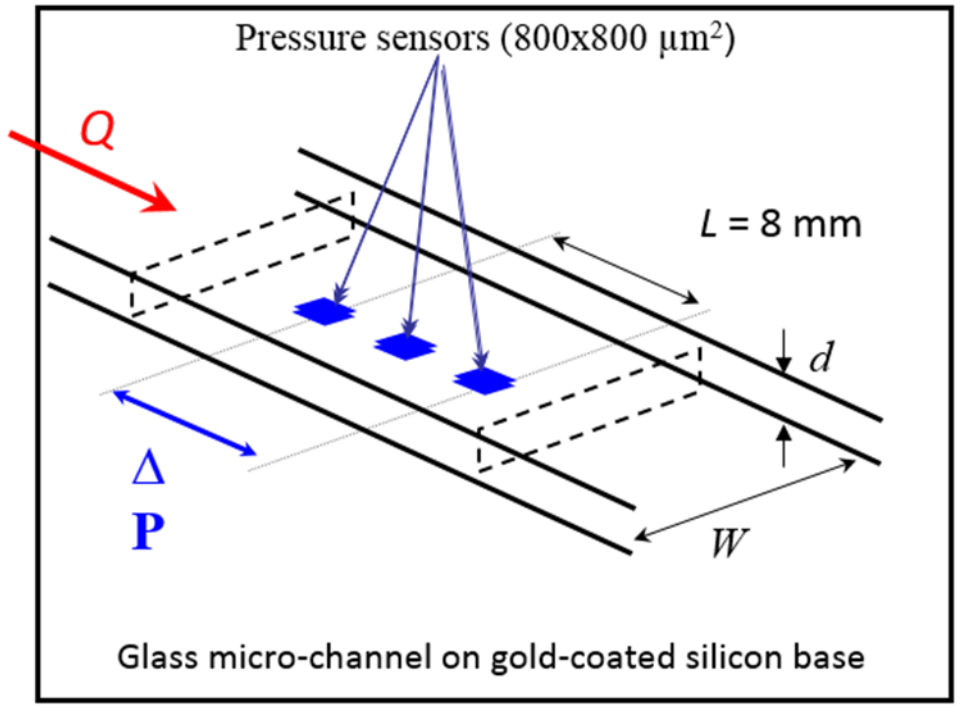
VROC is a hybrid of microfluidic and MEMS (Micro-Electro-Mechanical Systems) technologies:

- MEMS Sensors – Silicon (Si) Pressure Sensor Array
- Microfluidics – Precision Glass Micro-Channel



The VROC® Principle

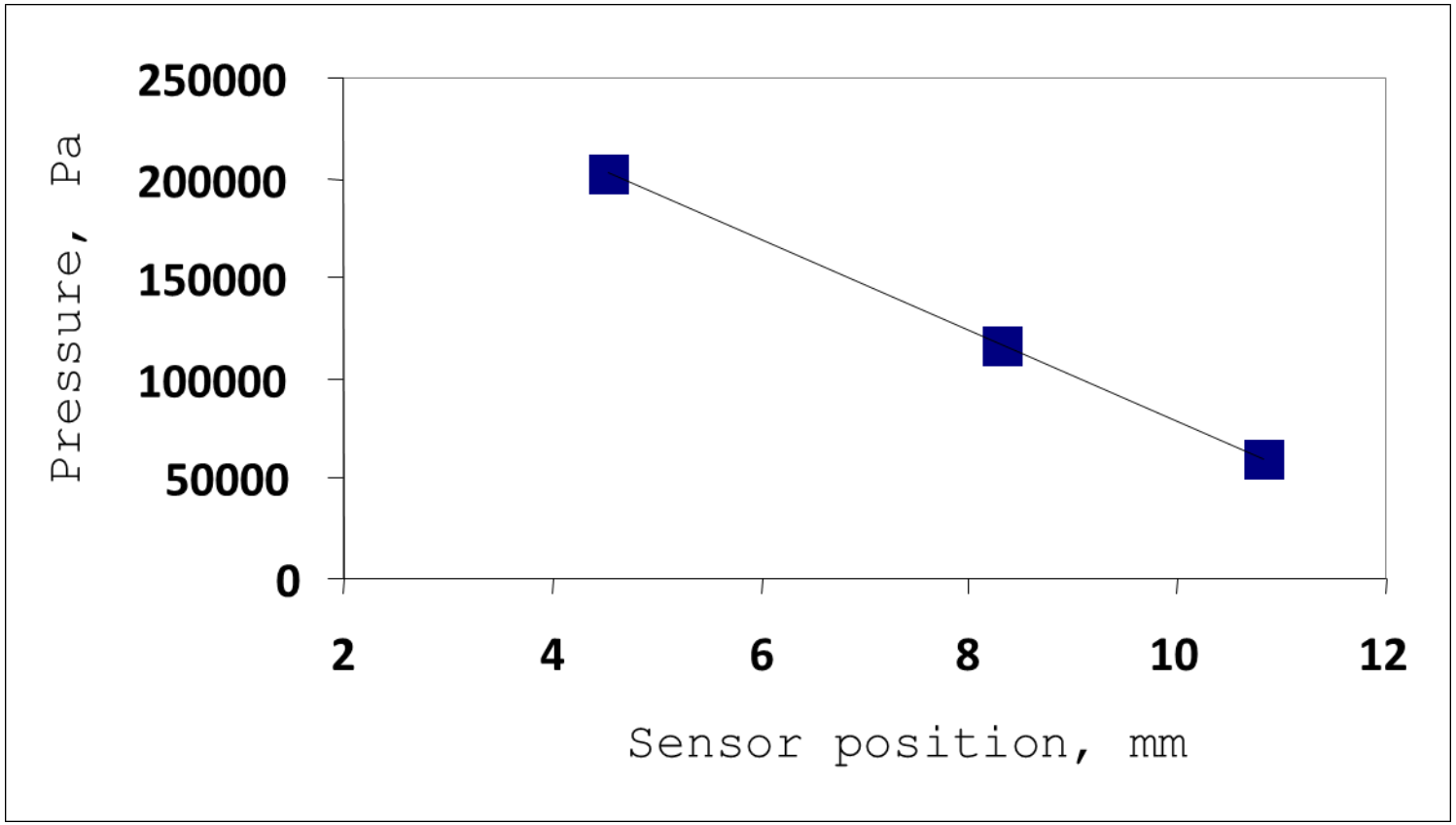
Derivative of Hagen-Poiseuille



Pressure Drop ~ Shear Stress I Flow Rate ~ Shear Rate

- Measure the pressure drop as a test liquid flows through a flow channel
- Pressure is measured at positions of increasing distance from the inlet
- The slope of the straight line in the plot of the pressure vs. sensor position is proportional to the viscosity.

Pressure vs Position



RheoSense Systems

m-VROC ™

Min Sample Volume	50 µl
Shear Rate Range, s^{-1}	.5 ~ 1,400,000
Viscosity Range (cP)	0.2 ~ 100,000
Temperature Range	4 ~ 70 °C
Portable	No
Shear/temp Sweep	Yes

*micro*VISC™

Min Sample Volume	100 µl
Shear Rate Range, s^{-1}	1.7 ~ 5,800
Viscosity Range (cP)	0.2 ~ 20,000
Temperature Range	18 ~ 50 °C
Portable	Yes
Shear/Temp Sweeps	No

e-VROC ™

Min Sample Volume	500 µl
Shear Rate Range, s^{-1}	.5 ~ 1,400,000
Viscosity Range (cP)	1.0 ~ 2,000
Temperature Range	4 ~ 70 °C
Extensional Range	0.1 - 1000 s^{-1}
Shear/temp Sweep	Yes

M-VROC™

M-VROC™ Specifications

Min Sample Volume	50 µl
Shear Rate Range, s^{-1}	0.5 ~ 1,400,000
Viscosity Range, mPa-s (cP)	0.2 ~ 100,000
Temperature Range	4 ~ 70 °C
Accuracy	2% of Reading
Repeatability	0.5% of Full Scale
Temperature Sensor	Built-In
Software	Included
Non-Newtonians?	Yes
Temperature Sweep	Yes
Shear Rate Sweep	Yes

Chip module surface material:

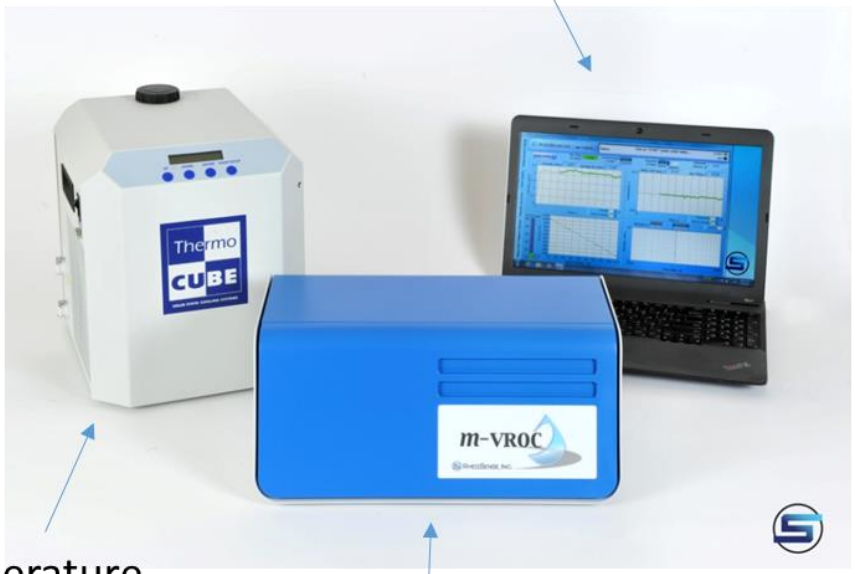
- borosilicate glass, silicon, PTFE, ETFE, PEEK, platinum, Perlast (Kalrez Optional)

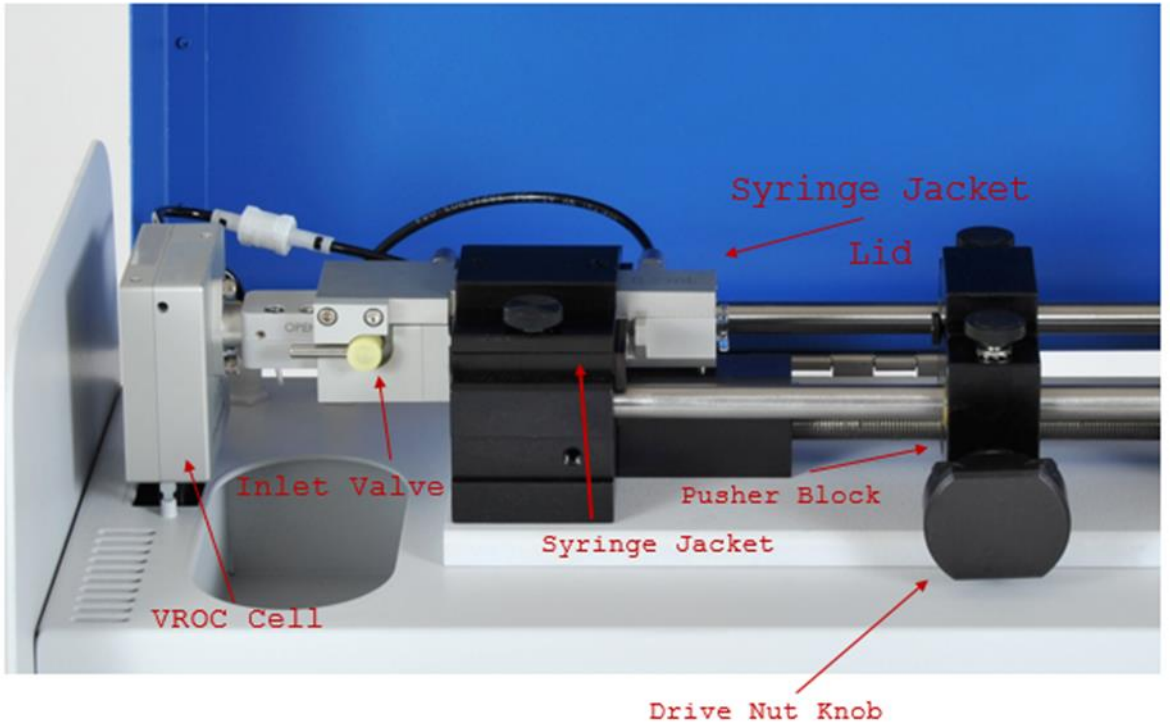
CE certified

Additional Customization (i.e.: 20µl Sample Volume Testing)

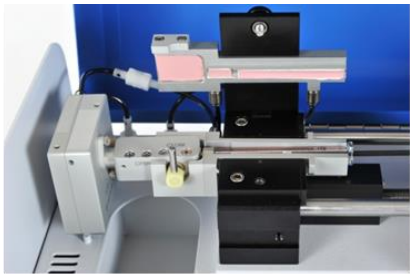
Listed in USP

The System

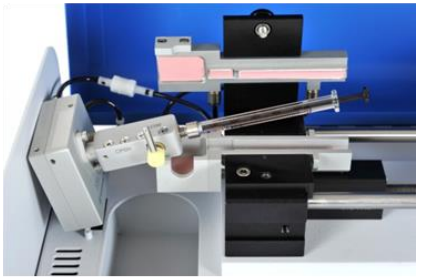

PC Interface
Thermo
CUBE
m-VROC
Temperature
Controller
m-VROC™


Syringe Jacket
Lid
Inlet Valve
Pusher Block
Syringe Jacket
VROC Cell
Drive Nut Knob

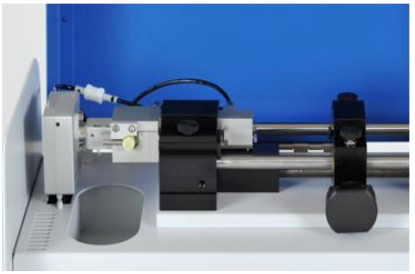
Using the m-VROC™



Step 1: Load the Syringe with your sample



Step 2: Screw the syringe into the chip enclosure and place it inside the thermal jacket



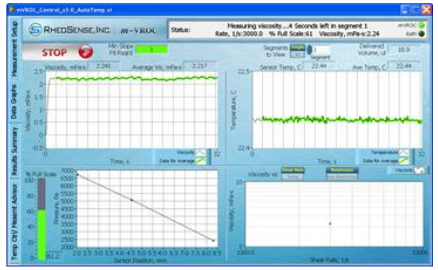
Step 3: Bring down the top of the thermal jacket enclosure and lock it by turning the black thumb screw clockwise. Now, you are ready to measure your sample!

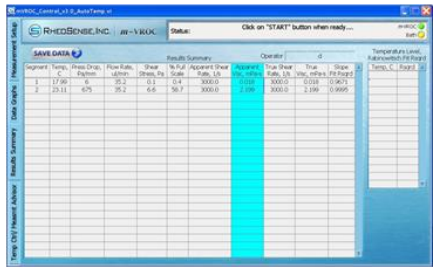
The Chips or Flow Cells

A Cells (A02, A05, A10)	Low Viscosity (0.2 - 200 cP)	* Small Sample Volume * Low Shear Rate
B Cells (B02, B05, B10)	Medium Viscosity (0.2 - 2,000 cP)	* General-Use Viscometer * Sufficient amount of sample required for low viscosity samples
C Cells (C02, C05, C10, C20, C30)	High Viscosity (0.2 - 10,000 cP)	* Low Viscosity Samples: high shear rate, sufficient sample required * High Viscosity Samples
E Cells (E02, E05, E10, E20)	Highest Viscosity (500 cP – 100,000 cP)	* Highest Viscosity Samples * Highest Shear Rates

m-VROC™ is equipped with an interactive measurement advisor in the control program to help determine which VROC® Cell to use for a specific viscosity or shear rate

Intuitive User Interface

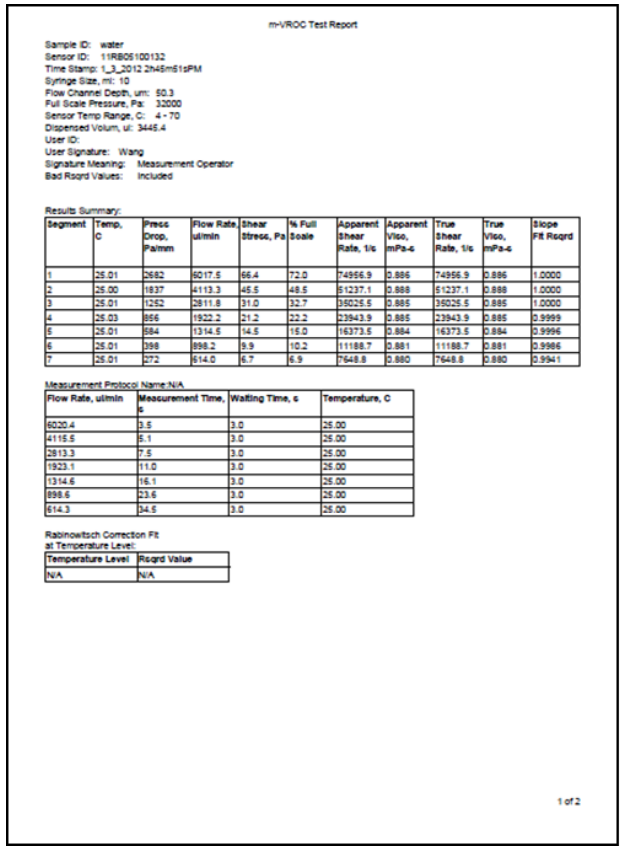




Data Collection

Each test is logged and reported in two formats:

- Pdf. Report of test
- Exl. Spreadsheet with data



m-VROC Test Report

Sample ID: water
Sensor ID: 11RB06100132
Time Stamp: 1_3_2013 2h45m51sPM
Syringe Size, ml: 10
Flow Channel Depth, um: 50.3
Full Scale Pressure, Pa: 32000
Sensor Temp Range, C: 4 - 70
Dispensed Volum, ul: 3446.4
User ID:
User Signature: Wang
Signature Meaning: Measurement Operator
Bad Rsqrd Values: Included

Results Summary:

Segment	Temp, C	Press Drop, Pa/mm	Flow Rate, ul/min	Shear Stress, Pa	% Full Scale	Apparent Shear Rate, 1/s	Apparent Visc, mPa-s	True Shear Rate, 1/s	True Visc, mPa-s	Slope Fit Rsqrd
1	25.01	2682	6017.5	66.4	72.0	74956.9	0.886	74956.9	0.886	1.0000
2	25.00	1837	4113.3	45.5	48.5	51237.1	0.888	51237.1	0.888	1.0000
3	25.01	1252	2811.8	31.0	32.7	35025.5	0.885	35025.5	0.885	1.0000
4	25.03	856	1922.2	21.2	22.2	23943.9	0.885	23943.9	0.885	0.9999
5	25.01	584	1314.5	14.5	15.0	16373.5	0.884	16373.5	0.884	0.9996
6	25.01	398	898.2	9.9	10.2	11188.7	0.881	11188.7	0.881	0.9986
7	25.01	272	614.0	6.7	6.9	7648.8	0.880	7648.8	0.880	0.9941

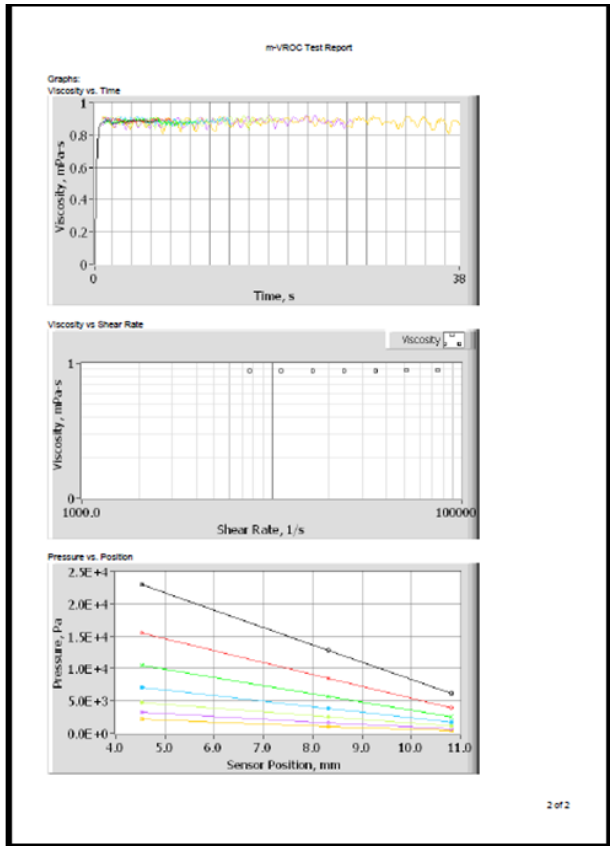
Measurement Protocol Name:N/A

Flow Rate, ul/min	Measurement Time, s	Waiting Time, s	Temperature, C
6020.4	3.5	3.0	25.00
4115.5	5.1	3.0	25.00
2813.3	7.5	3.0	25.00
1923.1	11.0	3.0	25.00
1314.6	16.1	3.0	25.00
898.6	23.6	3.0	25.00
614.3	34.5	3.0	25.00

Rabinowitsch Correction Fit at Temperature Level:

Temperature Level	Rsqrd Value
N/A	N/A

1 of 2

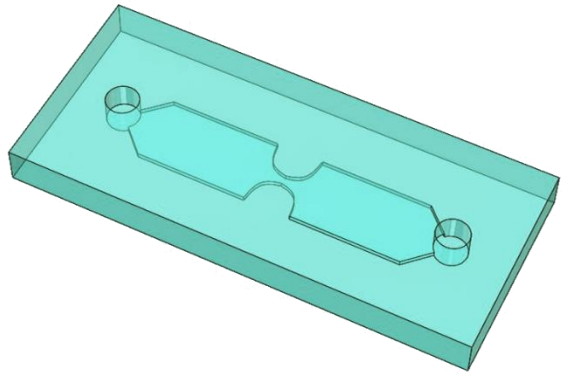


E-VROC™

Measures both the extensional and shear viscosities simultaneously.

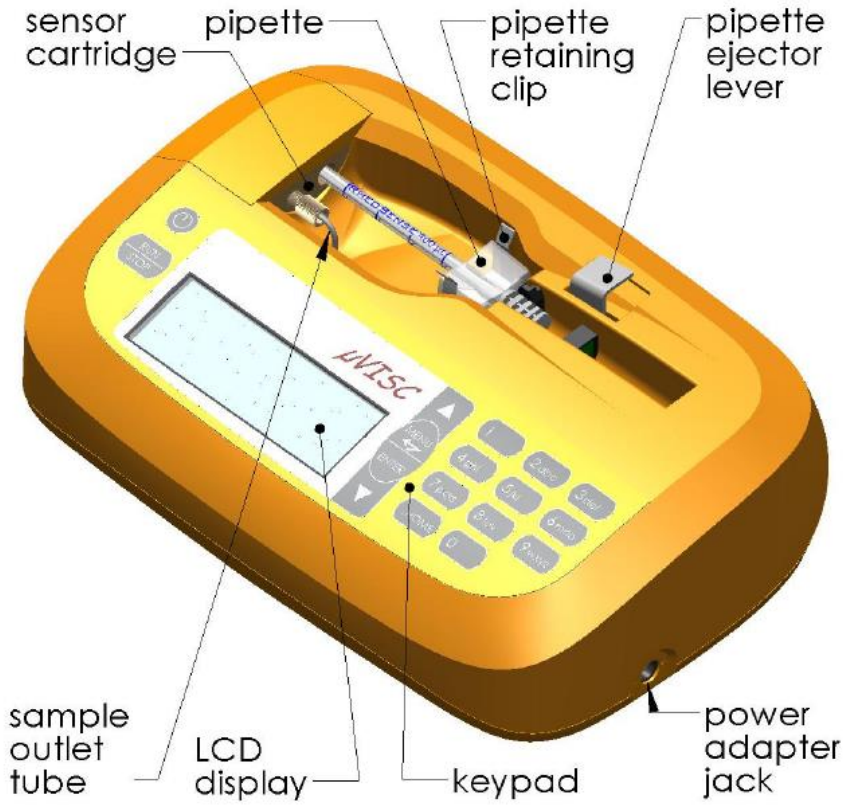
- Hyperbolic contraction/expansion zone in the middle of the channel
- Four monolithically integrated MEMS pressure sensors (two in the upstream and two in the downstream of the contraction/expansion zone)
- A liquid entering the channel first experiences shear flow in the straight channel and then experiences a uniform extension in the contraction zone

e-VROC™ allows the measurement of extensional viscosity at high extensional rates



*micro*VISC™ & *micro*VISC TC

microVISC™ System Overview



Min Sample Volume	100 µl
Shear Rate Range, s^{-1}	1.7 ~ 5,800
Viscosity Range, mPa-s (cP)	0.2 ~ 20,000
Temperature Range	18 ~ 50 °C
Portable	Yes
Accuracy	2% of Reading
Repeatability	0.5% of Full Scale
Temperature Sensor	Built-In
Software	Optional
Non-Newtonians?	Yes
Temperature Accuracy	0.15

Results are 60 Seconds Away…

Step 1: Load the sample

Step 2: Insert the Pipette

Step 3: Press Run

microVISC TC Module

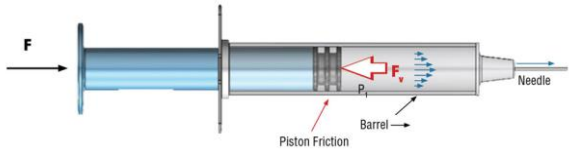
TC Module integrates directly with the *micro*VISC and provides precise temperature control

- Range: 18°C to 50°C
- Stability: +/- 0.07°C
- Control: Peltier heating/cooling
- Temp on/off timer function

Common Bio-Pharma Applications

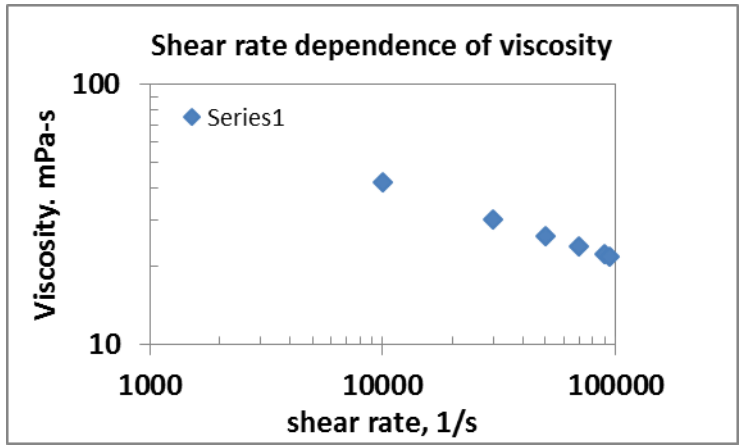
- Protein, RNA &Antibody Therapeutics
- Protein Formulation and Stability
- Accurate Particle Sizing (for DLS)
- Injectability
- Manufacturability

Injectability



- Therapeutic proteins behave like Newtonian, but also exhibit non-Newtonian behavior
- Accurate injection force depends on accurate viscosity
- High shear rates are necessary and difficult to achieve with conventional viscometers and rheometers:

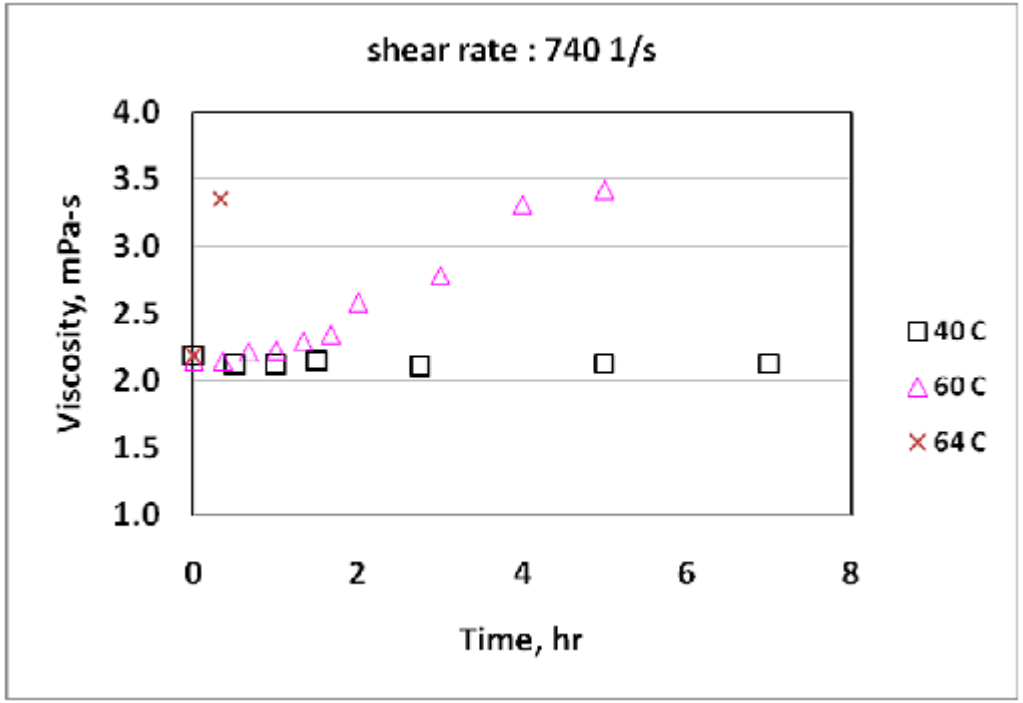
Injection rate, ml/s	shear rate, s^{-1}	
	26 gage	27 gage
0.0625	51,008	102,246
0.1	81,612	163,594
0.2	163,225	327,188



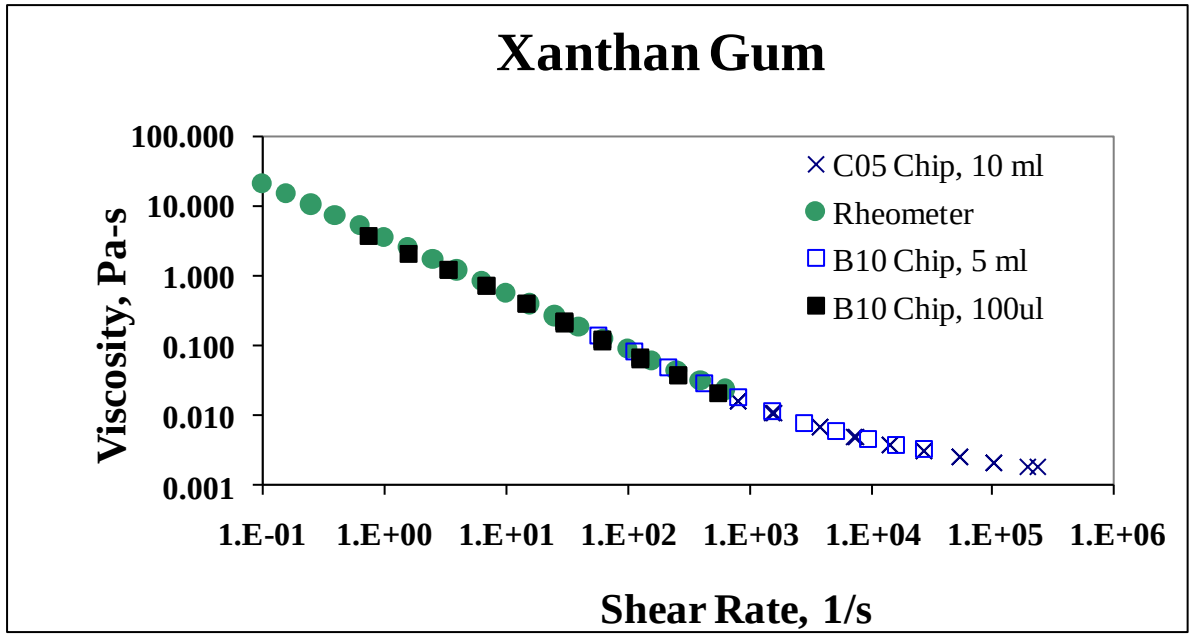
- Viscosity depends on shear rate
- Viscosity at 10,000 s^{-1} is 2X larger than viscosity at 100,000 s^{-1}
- Estimation based on viscosity at ≤10,000 s^{-1} leads to incorrectly assuming a need for a much higher injection force

Formulation and Stability

- Shelf life and efficacy depend on stability
- Most viscometers/rheometers only capable of measuring low concentrations
- Efficacy often requires heavy loading of proteins
- Common methods of determining unfolding (DLS) require diluted concentrations
- Proteins denaturation depends on temperature & time. Denaturation can be immediately or lag depending on temp.
- Aggregated proteins show shear thinning behavior
- VROC can detect even small increases in viscosity due to denaturation
- Viscosity is a bulk property that can be used to detect size changes without dilution

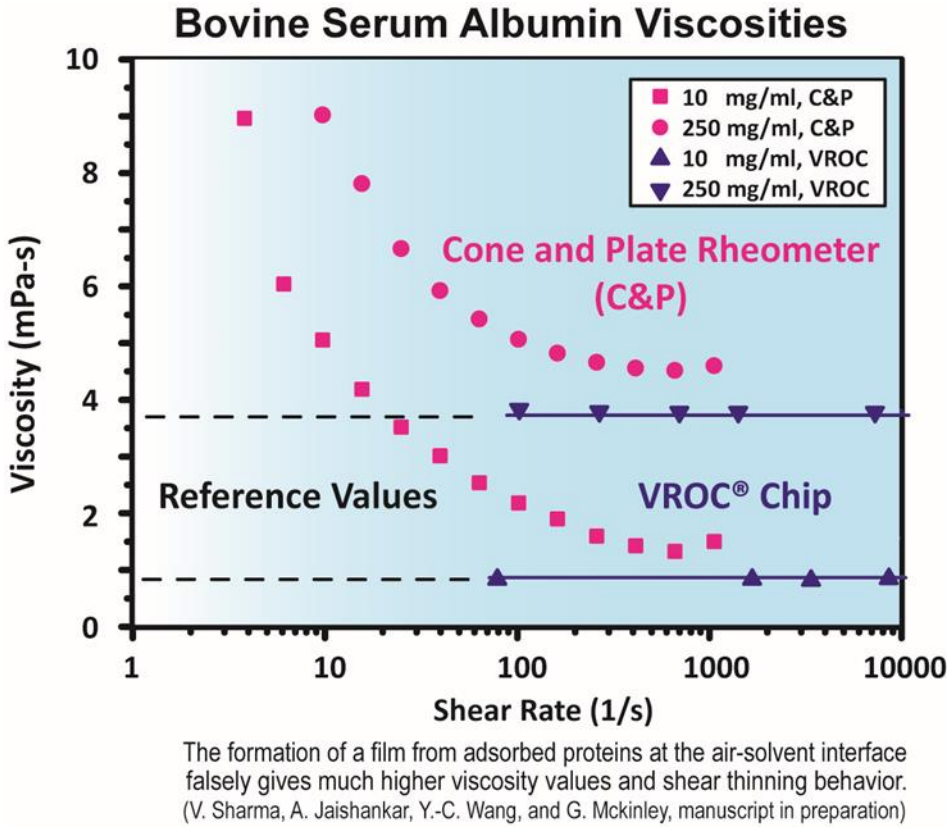


Results- True Shear Viscosity



Customer supplied comparison between *m*-VROC™ and Anton Paar MCR Rheometer
* Notice the shear rate limitation with conventional Rheometers!

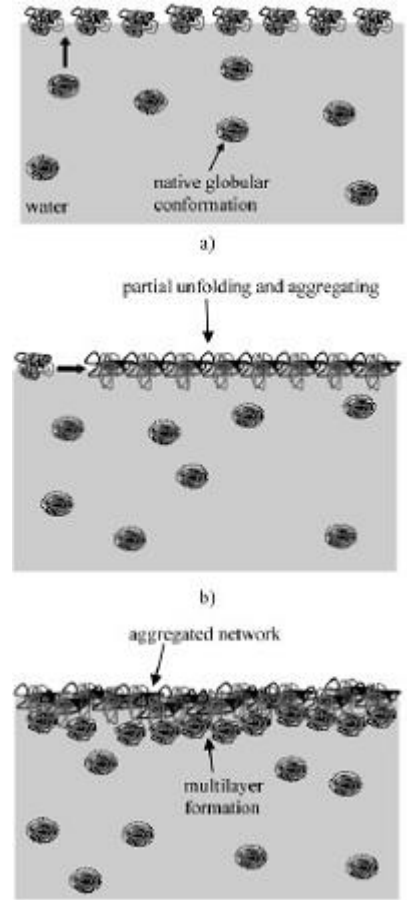
Results- *m*-VROC™ vs. Cone & Plate



The formation of a film from adsorbed proteins at the air-solvent interface falsely gives much higher viscosity values and shear thinning behavior.
(V. Sharma, A. Jaishankar, Y.-C. Wang, and G. Mckinley, manuscript in preparation)

Measuring with cone & plate has two challenges:

- Evaporation
- Irreversible absorption protein molecules at the interface:
  - Proteins migrate to the interface to minimize the interface energy
  - Molecules partially unfold and aggregate
  - Can for gel-like network
- Shows “apparent” shear thinning behavior



Results- True Viscosity

True Viscosity, Not Indexed

- Indexed viscosity instruments will vary machine to machine
- Variation can cause issues if sharing information
  - R&D Formulation to Operations Transfer
  - Multiple Lab Sites
  - Multiple Instruments at One Location
- True viscosity measurement provides consistency

Non-Newtonian Measurement Capabilities

- Don’t have to make assumptions or extrapolations on characteristic over shear rate
- Small sample size helps mitigate cost of additional measurements

The RheoSense Advantage

- Small Sample Volumes (≥ 20 µL)
- Ease of Use (Set-Up, User Friendly Software)
- Rapid Results
- High Precision and Accuracy
- “True” Viscosity, Not Index
- Closed System (No Solvent Evaporation or Contamination)
- Widest Viscosity & Shear Rate Range Capabilities
- Newtonian and non-Newtonian Fluids
- Small footprint
- Extensional and Shear Viscosity

The END…

Thank You!