
IOCB SERVICE DAYS

Presentation of services by:
Research-Service Groups / Service Groups / Core Facilities

20
23

MicroScale Thermophoresis Research Core Facility

Jaroslav Srp



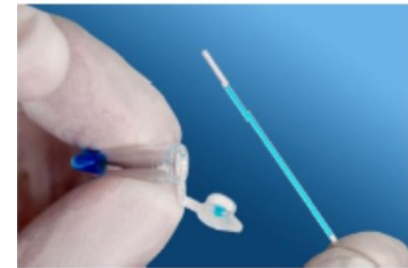
ÚOCHB AV
CR
IOCB PRAGUE



MicroScale Thermophoresis (MST)

What is MST used for?

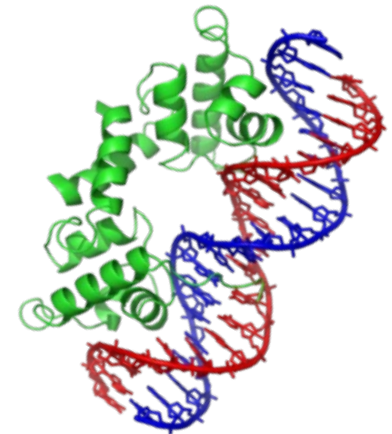
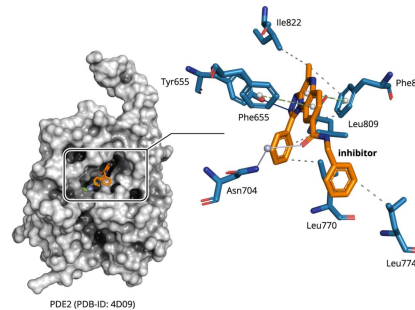
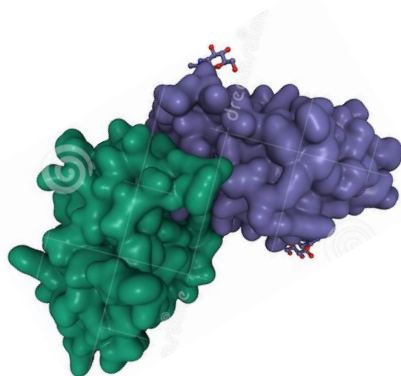
- interaction of macromolecules with ligands
- in solution using capillaries
- you get K_D (dissociation constant)



MicroScale Thermophoresis (MST)

What do you need?

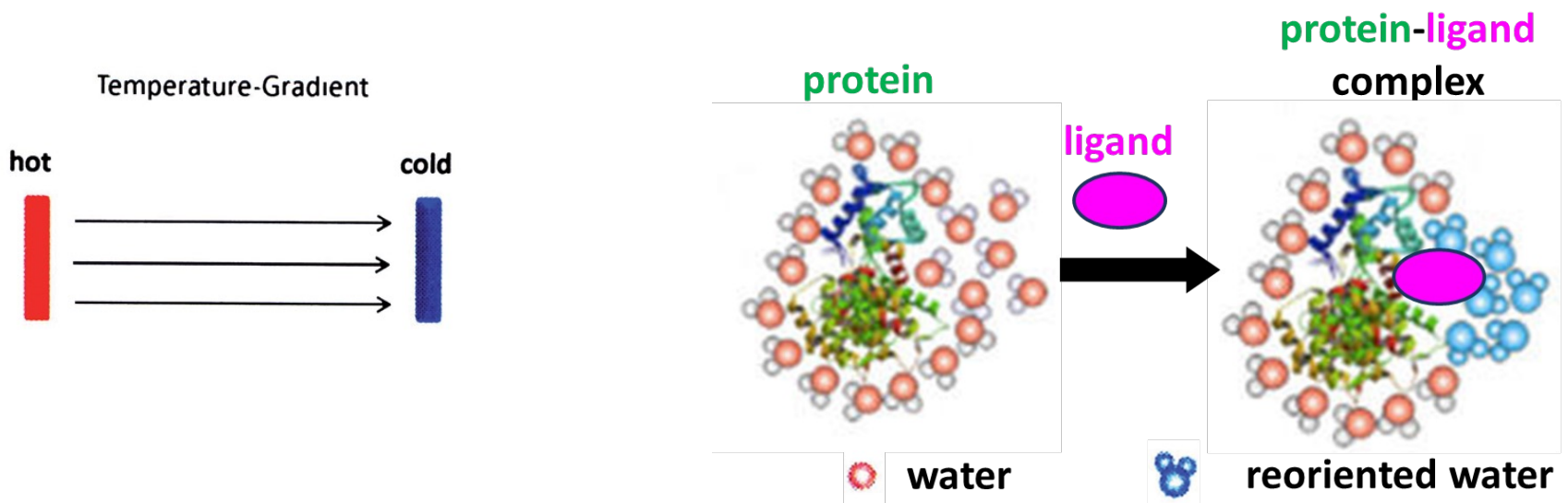
- macromolecule (protein, nucleic acid, ...)
 - < 150 μL of 2-20 μM protein
- ligand for titration
 - almost any type: protein/peptide, DNA/RNA, small molecule
- buffers (additives for optimization)



MicroScale Thermophoresis (MST)

How does the MST work?

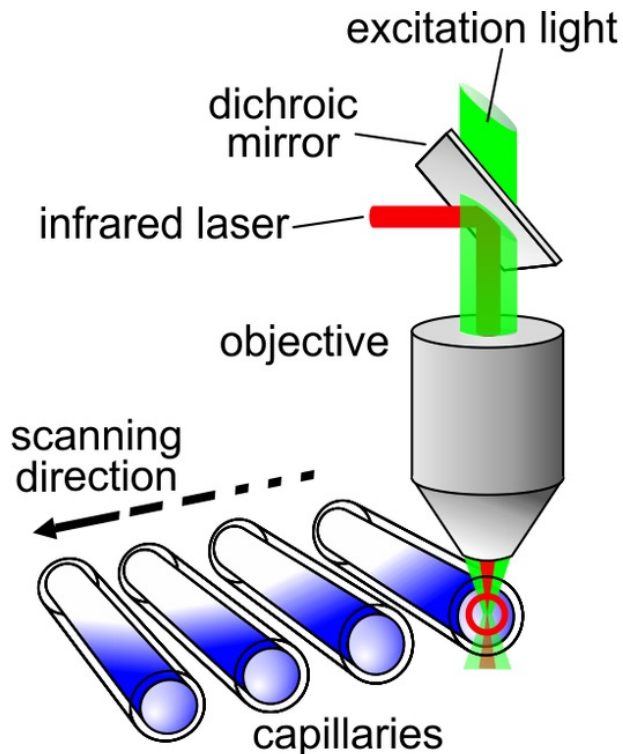
- migration of the molecule in a microscopic temperature gradient
- influenced by charge, size, and hydration shell
(that are changed during complex formation)



MicroScale Thermophoresis (MST)

How does the MST work?

- monitored as fluorescence changes



Infrared (IR) laser generates a microscopic temperature gradient

Detection laser for excitation of fluorescence

MicroScale Thermophoresis (MST)

Our MST instruments



MST NT.115

fluorescently labeled molecules
(labeling kits available)



MST Label Free

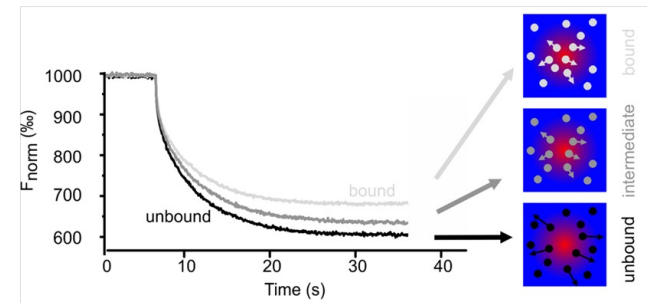
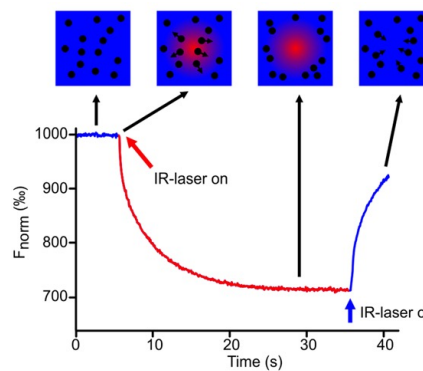
proteins with Trp fluorescence

MicroScale Thermophoresis (MST)

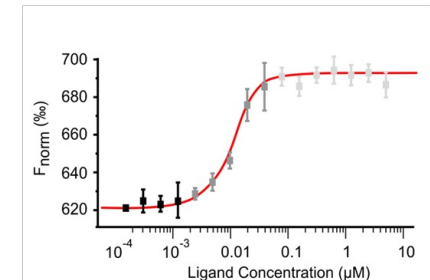
MST measurement

- thermophoresis curve
- 16 parallel measurements with different ligand concentrations
- titration curve

your K_D



your K_D



MicroScale Thermophoresis (MST)

MST measurement

- thermophoresis curve
- 16 parallel measurements with different ligand concentrations
- titration curve

your K_D

Alternative/complementary technologies

- **ITC** (Isothermal Titration Calorimetry)
- **SPR** (Surface Plasmon Resonance)

MicroScale Thermophoresis (MST)

How to start?

- contact me or another trained colleague
srp@uochb.cas.cz; phone: 287; building C, 2nd floor
- application notes on NanoTemper web pages
nanotempertech.com
- online MST training days organized by NanoTemper
in 2023: Apr 25, Jun 27, Oct 24
- expert online support by NanoTemper

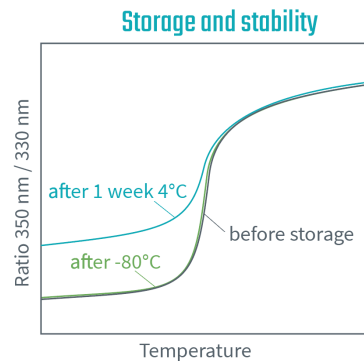
Tycho – thermal stability

What?

- quick detection of protein stability
- quality of recombinant proteins
- storage conditions (buffer, temperature, additives...)

How?

- fluorescence monitoring during thermal denaturation
- > 10 μL of sample
- 10+ $\mu\text{g/mL}$ protein



IOCB SERVICE DAYS

Presentation of services by:
Research-Service Groups / Service Groups / Core Facilities

20
23

MicroScale Thermophoresis Research Core Facility

Jaroslav Srp



ÚOCHB AV
CR
IOCB PRAGUE

