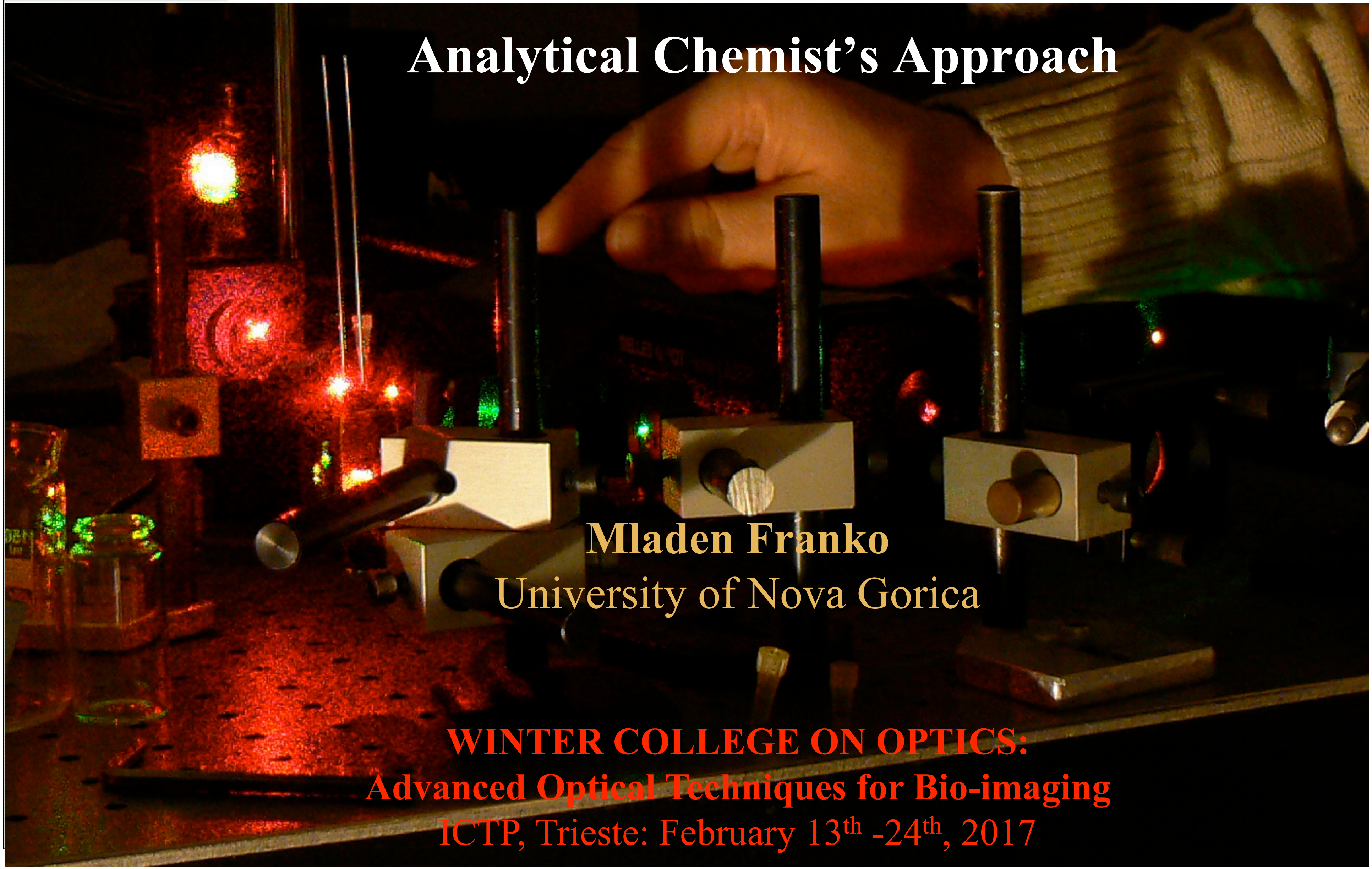
Thermal Lens Spectrometry and Microscopy



Analytical Chemist’s Approach

Mladen Franko
University of Nova Gorica

WINTER COLLEGE ON OPTICS:
Advanced Optical Techniques for Bio-imaging
ICTP, Trieste: February 13th -24th, 2017

Requirements for analytical methods in bio-chemical analysis

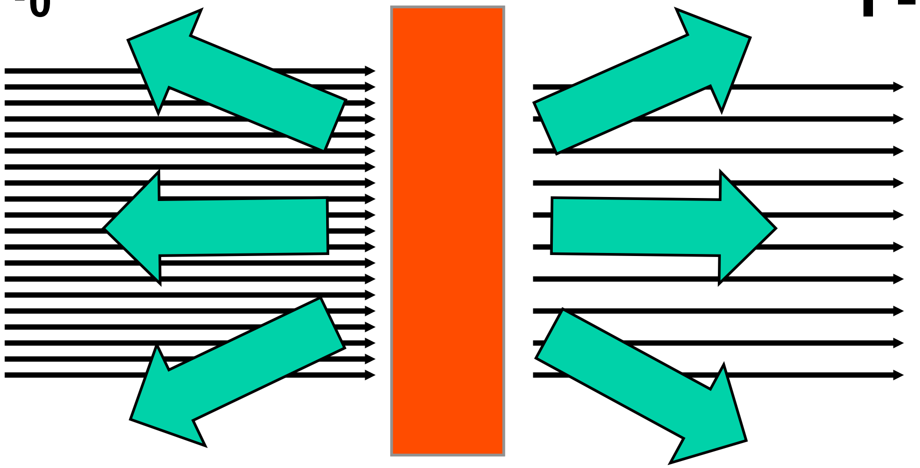
- Sensitivity
- Selectivity
- Cost efficiency
- High sample throughput
  - Conventional (certified, rearguard) analytical methods are labour intensive, time consuming and costly
  - **quick** answers and analysis of **large numbers** of samples (and large number of compounds - screening) for **low cost – vanguard methods** (frequently semi-quantitative or just binary YES/NO response)

Transmission techniques - how to improve sensitivity?

$A = -\log T = -\log(I/I_0)$

for low absorbances:

$A = \varepsilon L C \cong (I_0 - I)/2.303 I_0$



I_0

I - transmission

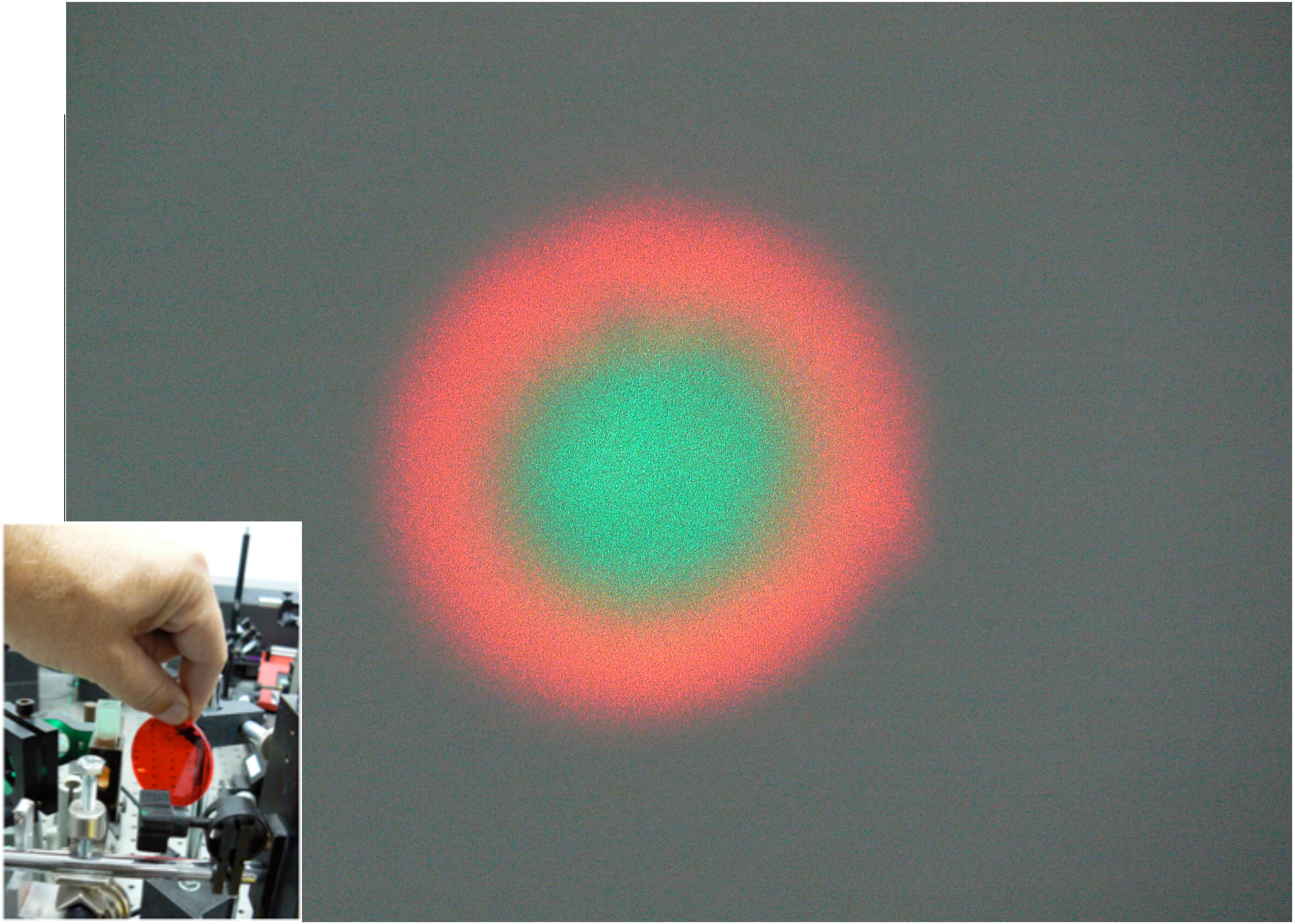
Emission

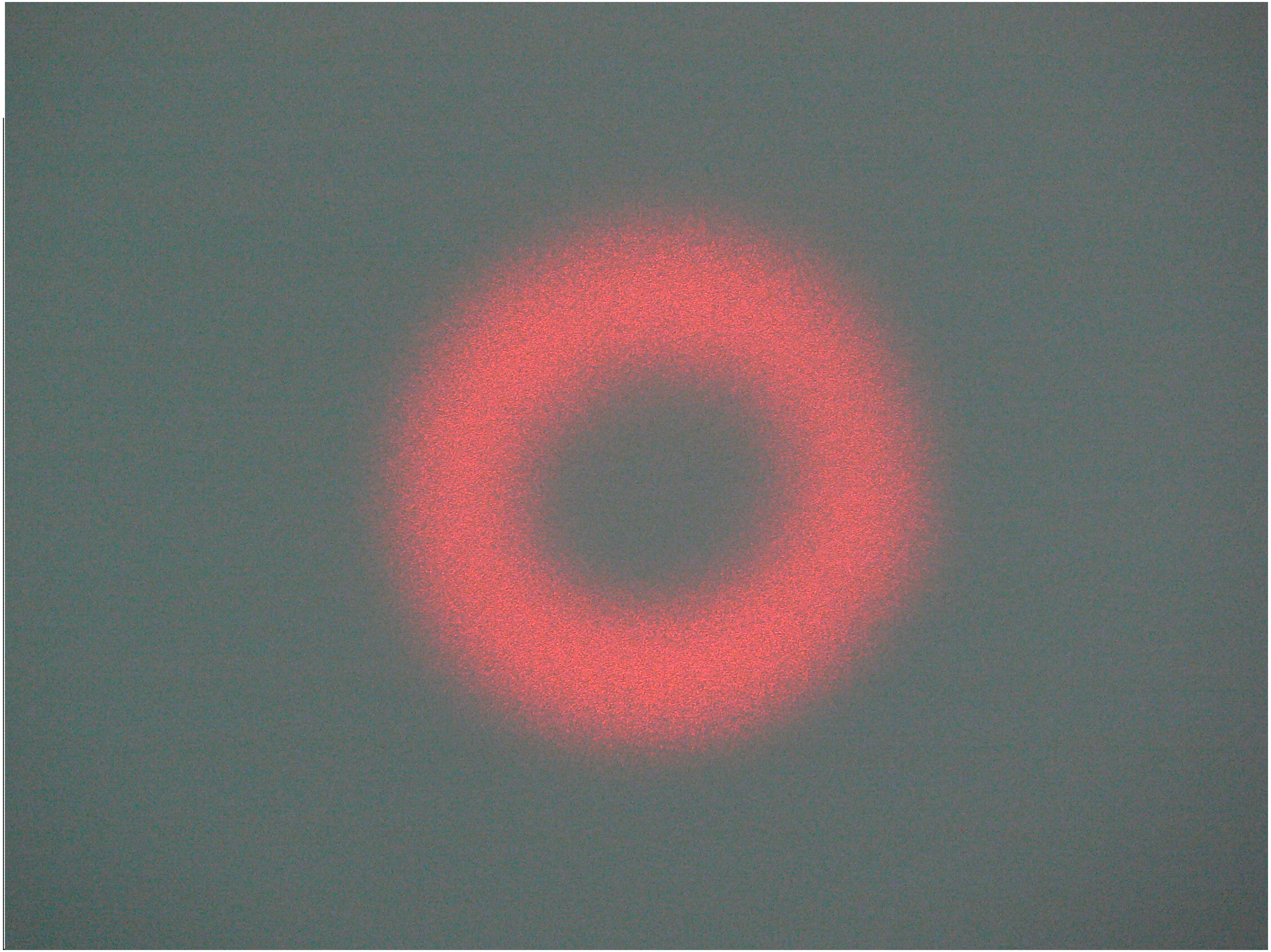
Sample

Radiatioless deexcitation

Basics of thermal lens effect

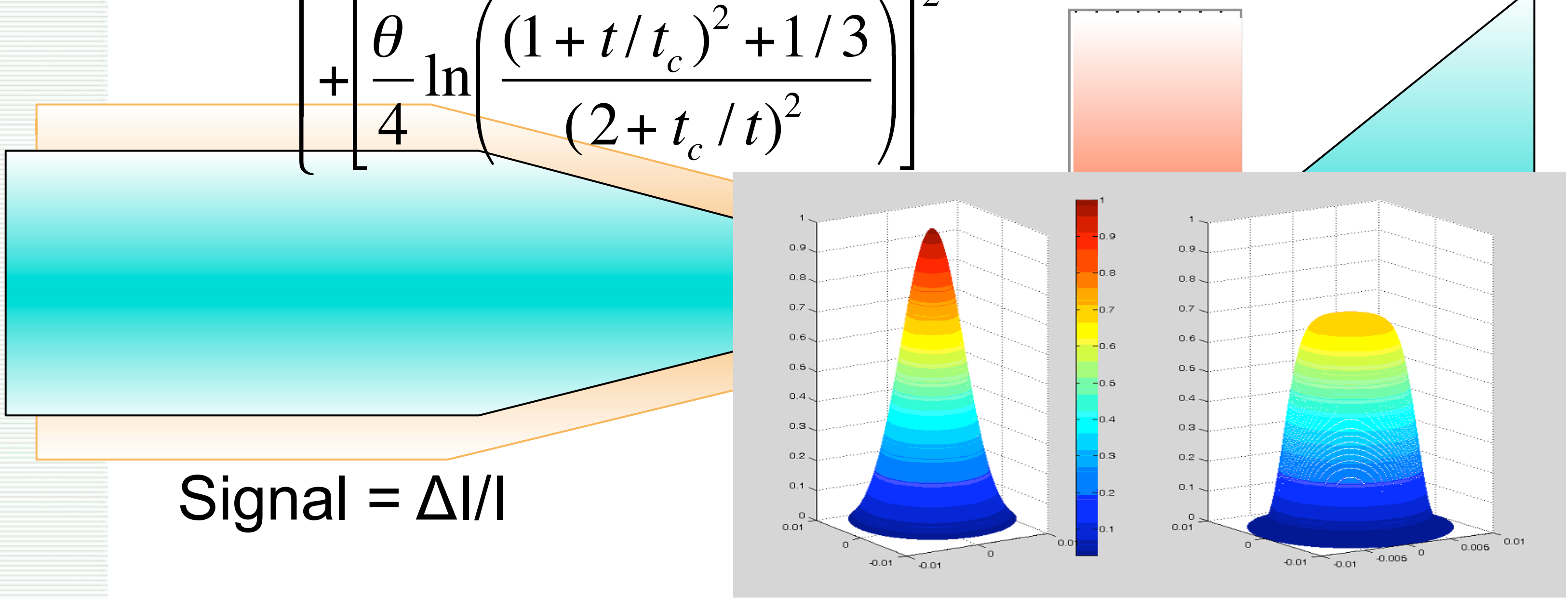
- During non-radiative relaxation of excited species temperature in the sample increases (10^{-4} - 10^{-3} K)
- a temperature gradient is generated with maximum temperature at the axis of the excitation beam
- the resulting refractive index gradient acts as a lens (mostly: $dn/dT < 0$, diverging lens)
- laser beam is defocused (single beam or pump/probe (collinear, crossed beam) configuration)
- beam radius and its intensity at the beam axis changes
- relative change in the beam intensity is proportional to the absorbance of the sample and to the power of the excitation beam.





TLS – how to make good use of something not desired in laser technology?

$$I_{bc}(t) = I_{bc}(0)\left\{ 1-\theta\tan^{-1}\left(\frac{1}{(1+t_c/t)\sqrt{3}}\right) + \left[\frac{\theta}{2}\tan^{-1}\left(\frac{1}{(1+t_c/t)\sqrt{3}}\right)\right]^2 + \left[\frac{\theta}{4}\ln\left(\frac{(1+t/t_c)^2+1/3}{(2+t_c/t)^2}\right)\right]^2 \right\}$$



Signal = ΔI/I

SAMPLE

$$t_c = \frac{a^2\rho C_P}{4k} \qquad \theta = \frac{2.303AP(-dn/dT)}{\lambda k}$$

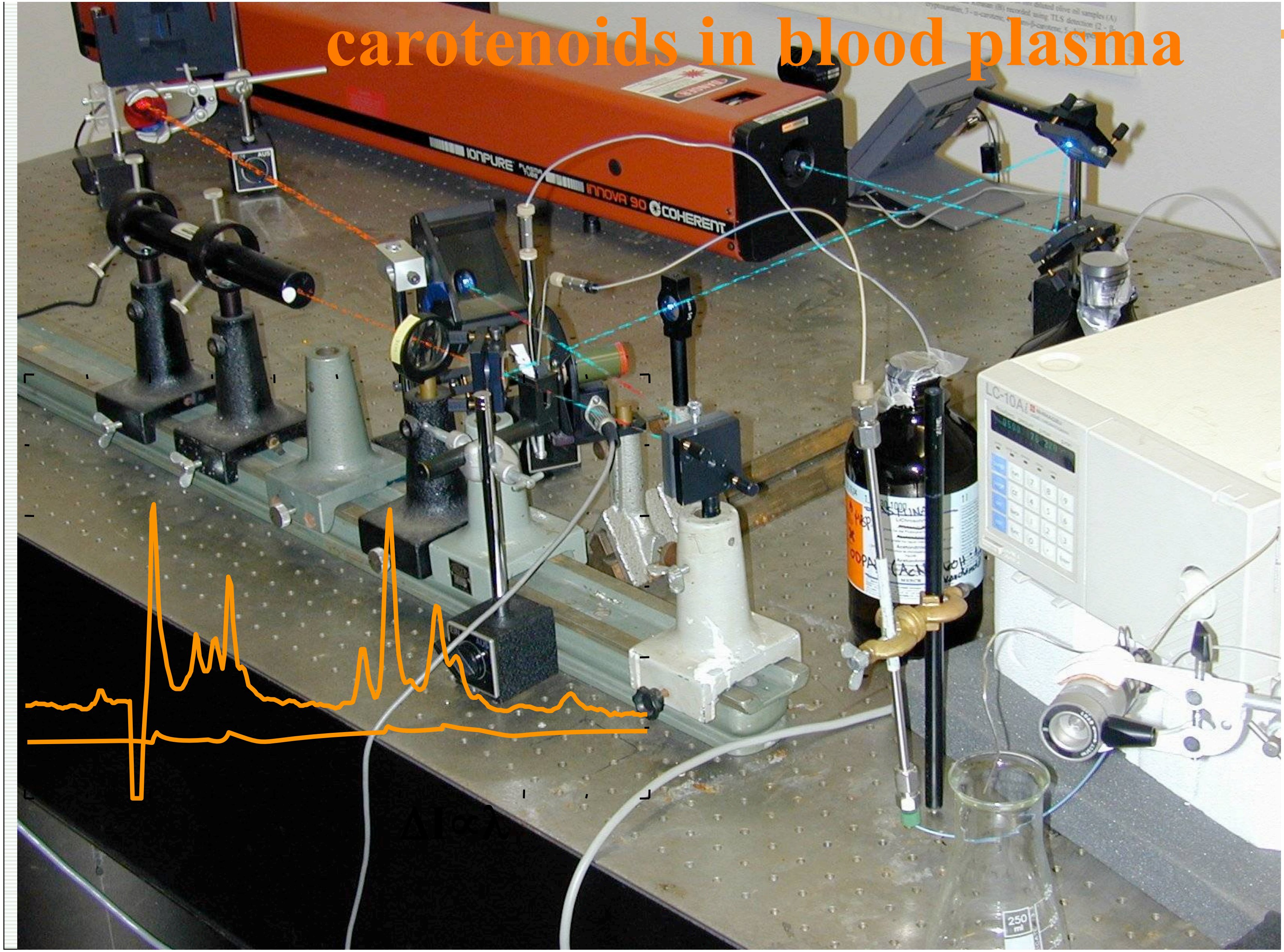
TLS - advantages

- High sensitivity
    - signal proportional to excitation laser power
    - absorbances as low as 10^{-7} can be measured
- Enables On-line detection
    - fast response of TLS signal (on μs to ms time scale)
- Capability of measuring small samples
    - sub-pL volumes can be probed
    - detection in microfluidic systems

TLS – drawbacks and solutions

- Sensitivity still needs improvement
  - Higher laser power? (photo-labile compounds)
  - Modify solvents
- Limited availability of laser sources
  - Coloring reactions, indirect detection
- Poor selectivity
  - Single wavelength measurements
  - Coupling to separation techniques (HPLC, IC, CE)
- Photodegradation
  - Measure in flowing systems

HPLC-TLS degtermination of carotenoids in blood plasma



Nonsteady thermal diffusion

$$\frac{\partial T(r,t)}{\partial t} = D\nabla^2 T(r,t) - v_x \frac{\partial T(r,t)}{\partial x} + \frac{1}{\rho C_p} Q(r,t)$$

- $T(r,t)$….. temperature
- D………thermal diffusivity
- ρ………. Density
- C_p……….heat capacity
- $Q(r,t)$……source term (“heat”)
- v_x………..velocity of the medium in x direction

By solving nonsteady the thermal diffusion equation, changes in refractive index and related TLS signal can be calculated for different beam geometries and excitation regimes (pulsed, cw)

Pulsed and cw excitation with a Gaussian beam

Assuming complete conversion of absorbed energy into heat, source terms are:

- Pulsed:

$$Q(r,t) = \frac{2\alpha E_0}{\pi a^2 t_0} \exp\left[-2(x^2 + y^2)/a^2\right]$$

- cw:

$$Q(r,t) = \frac{2\alpha P_{av}}{\pi a^2} \left\{\exp\left[-2(x^2 + y^2)/a^2\right]\right\} \times (1 + \cos\omega t)$$

E_0 ….pulse energy	a…..pump laser beam radius
t_0 …..pulse width	P_{av}…cw laser average power
α…...absorbance (cm^{-1})	ω…..modulation frequency

Thermal lens signal

$$s(t) = \frac{w_2^2(t) - w_2^2(0)}{w_2^2(0)}$$

- $w_2(0)$….radius of an unperturbed probe beam at the detector site
- $w_2(t)$…..time dependent radius of a probe beam perturbed by the thermal lens
- w_0…….radius of the probe beam at its waist

$$w_2^2(t) = w_0^2\left[\left(1 - \frac{z_2}{f(t)}\right)^2 + \frac{1}{z_0^2}\left(z_1 + z_2 - \frac{z_1 z_2}{f(t)}\right)^2\right]$$

Simplifications for usual far field experimental configuration

- $z_2 >> z_1$, $z_2 >> z_0 = \pi w_0^2/\lambda$
- $f(t) >> z_1$, $f(t) >> z_0$
- @ $t = 0$, $f(0) = \infty$
  - λ, w_0 …..probe beam wavelength and waist
  - z_0…..confocal distance
  - z_1….. probe beam waist to sample distance
  - z_2 …..sample-to-detector distance

$$s(t) = -\frac{2z_1}{f(t)}$$

Refractive index change and focal distance of thermal lens

$$n(x,y,t) = n_0 + \left(\frac{\partial n}{\partial T}\right)_{T_A} \times T(x,y,t)$$

- n_0….. unperturbed refractive index at ambient temperature T_A

collinear:

$$\frac{1}{f} = -\frac{\partial n}{\partial T}\,\ell\left(\frac{\partial^2 T}{\partial r^2}\right)$$

transversal:

$$\frac{1}{f} = -\frac{\partial n}{\partial T}\int_{-\infty}^{\infty}\left(\frac{\partial^2 T}{\partial x^2}\right)dy$$

- f…..thermal lens focal length
- ℓ….interaction length

TLS signal for collinear configuration

- Pulsed: ($t_0 \rightarrow 0$)

$$s(t) = -\frac{4AE_0z_1(\partial n/\partial T)}{\pi k a^2 t_c}\frac{1}{(1+2t/t_c)^2}$$

- cw:

$$A = \alpha \ell$$

$$s(t) = -\frac{2APz_1(\partial n/\partial T)}{\pi k a^2}\frac{1}{(1+t_c/2t)}$$

  - t_c….time constant = $a^2\rho c_p/4k=4a^2D$
  - k….thermal conductivity of the sample

TLS signal for transversal configuration

- Pulsed: ($t_0 \rightarrow 0$)

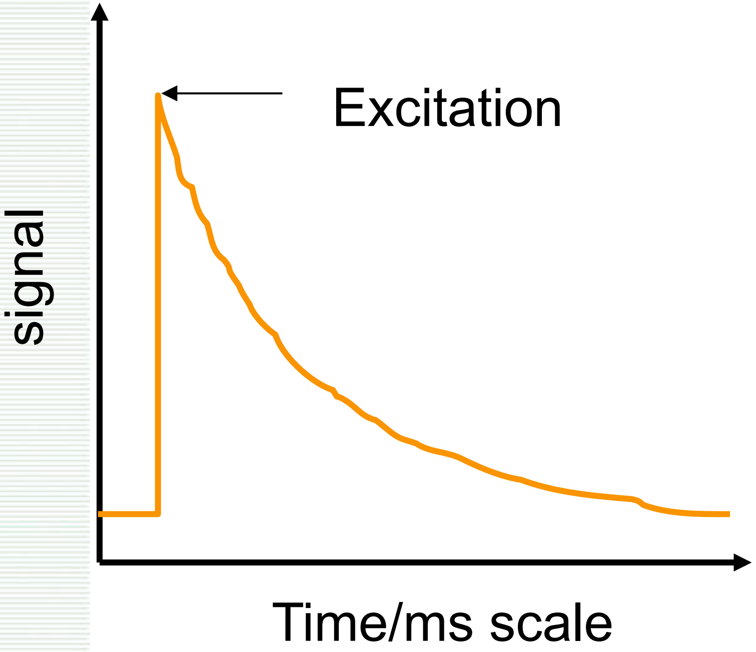
$$s(t) = -\frac{2\alpha E_0 z_1 (\partial n / \partial T)}{\sqrt{2\pi} k a t_c} \frac{1}{(1 + 2t/t_c)^{3/2}}$$

- cw:

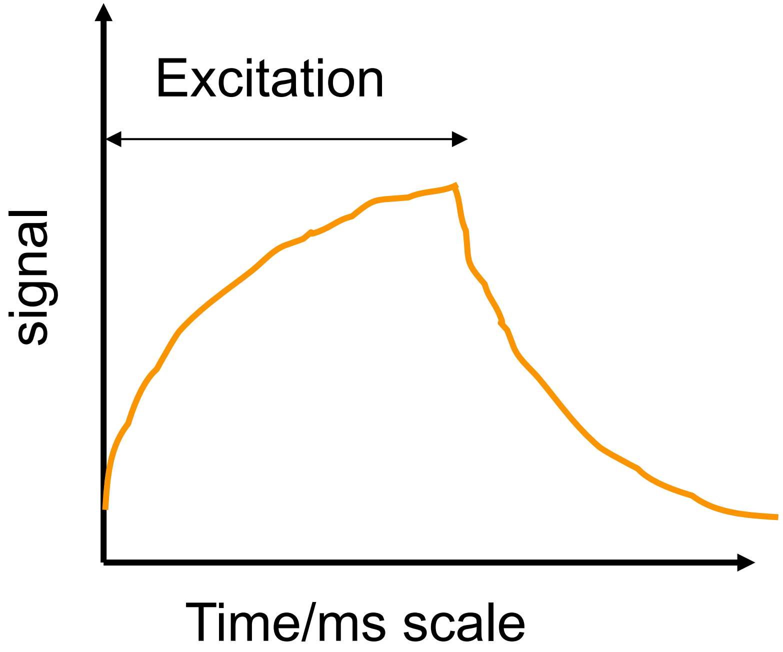
$$s(t) = -\frac{2\alpha P z_1 (\partial n / \partial T)}{\sqrt{2\pi} k a} \frac{1}{(1 + t_c/2t)^{1/2}}$$

TLS signal form

Pulsed



CW



E - Enhancement factor in TLS

$$\frac{\Delta I}{I} = \frac{2.303P\left(-dn/dT\right)A}{\lambda k}\, arctg\left(1/\sqrt{3}\right) = 2.303EA$$

Solvent	$-dn/dT$ (10^{4} K^{-1})	k ($W\,m^{-1}K^{-1}$)	E (10^{-3} W^{-1})
H_2O	0.91	0.607	0.12
CCl_4	5.9	0.103	4.74
acetone	5.42	0.190	2.36

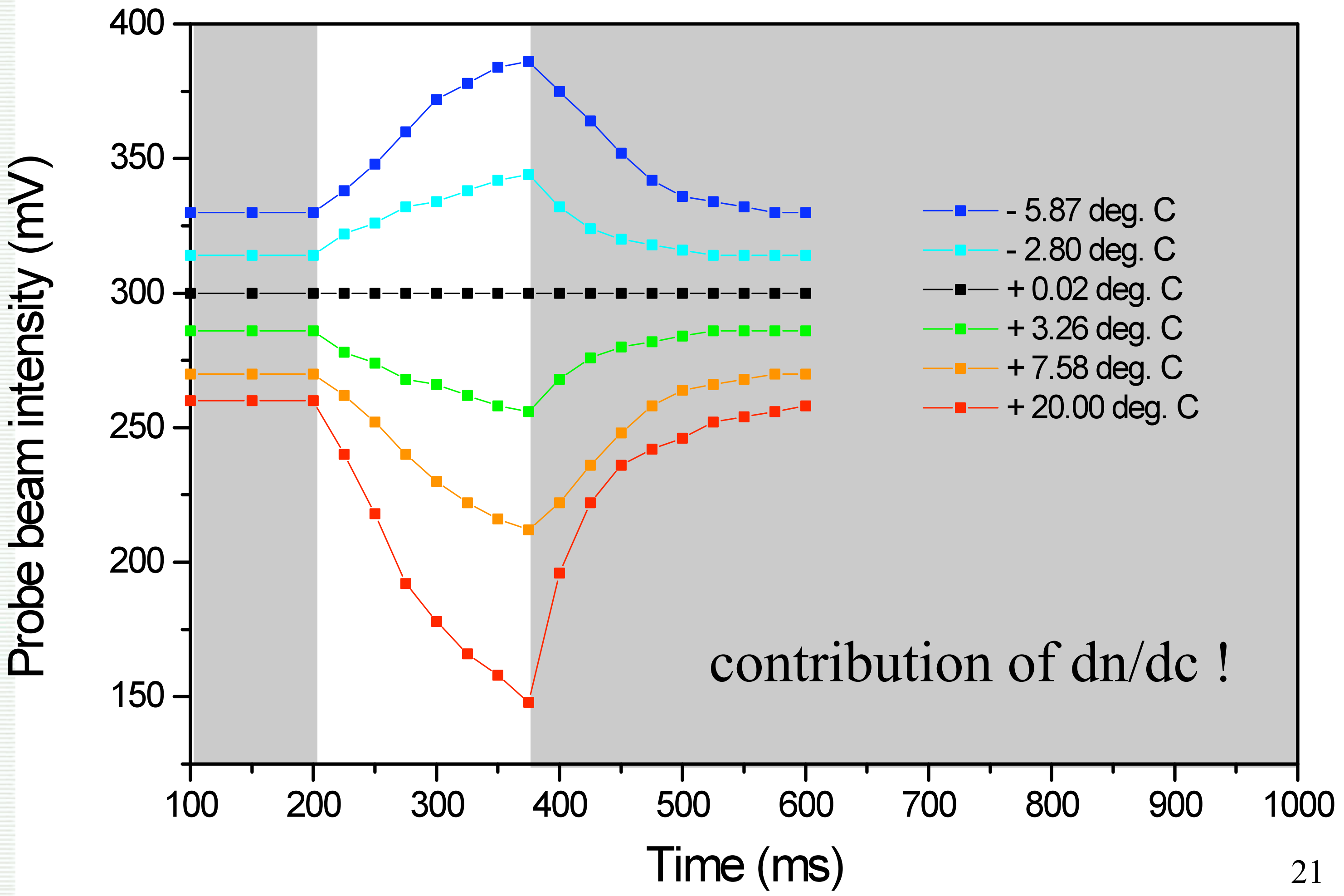
$E = (-dn/dT)/(1.91\ \lambda k)$ is calculated for $\lambda = 632.8$ nm

Thermo-optical properties of solvents for TLS measurements

Solvent	Thermal conductivity, k $mWcm^{-1}K^{-1}$	$10^4(dn/dT)$ K^{-1}	$-\frac{10^4(dn/dT)}{k}$ $cm\ mW^{-1}$
CO_2 (SC)	0.7	-100	143
CCl_4	1.03	-5.9	5.73
Benzene	1.24	-6.4	5.16
C_8MImTf_2N	n.d.	n.d.	4.55
cyclohexane	1.24	-5.4	4.35
$BMImBF_4$	1.78	-7.54	4.24
n-heptane	1.26	-5.0	3.97
$BMImTf_2N$	1.06	-4.0	3.78
dioxane	1.39	-4.6	3.31
$EMImTf_2N$	n.d.	n.d.	2.37
methanol	2.20	-4.7	2.14
water	6.11	-0.8	0.13

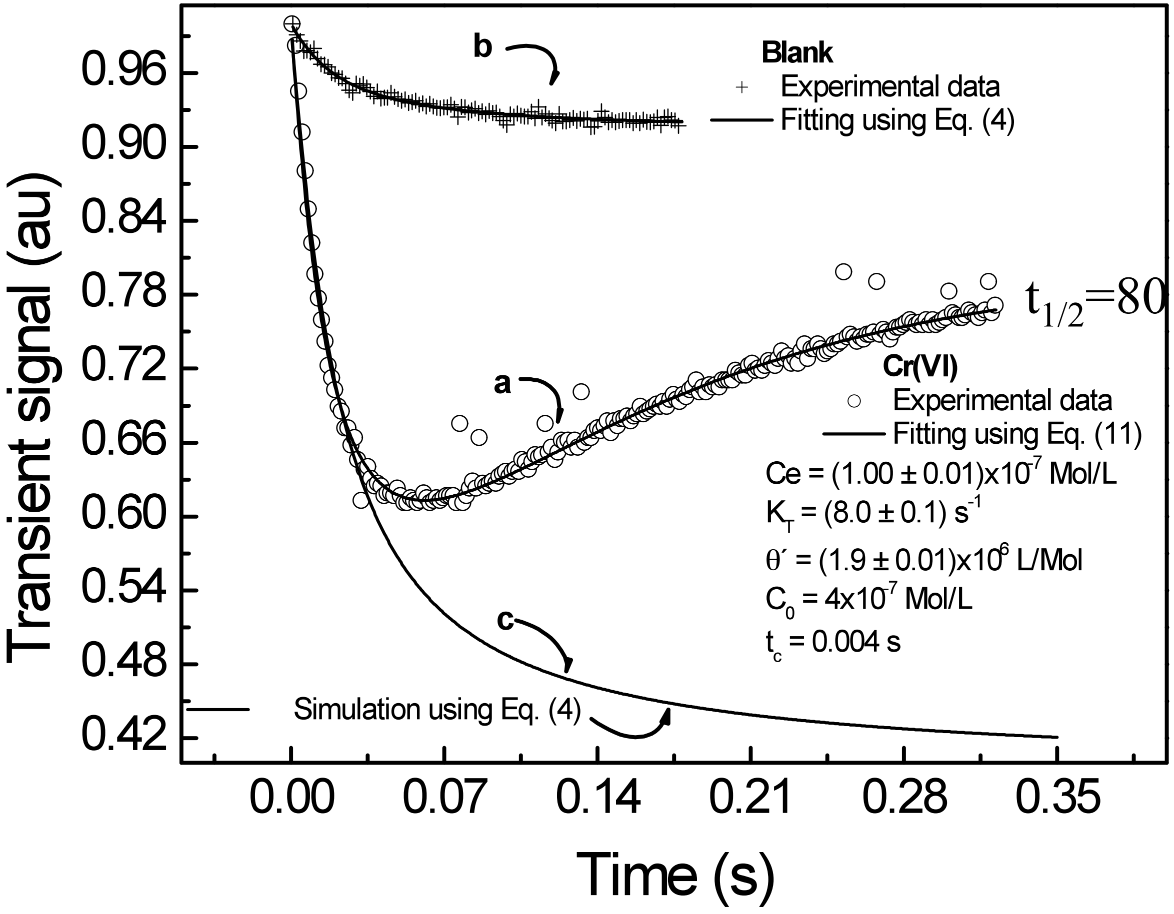
Calc. values (except CO2) taken from Chieu D. Tran and T. A. Van Fleet, Anal. Chem. 60, (1988) 2478

Temperature dependent TLS signal in water



contribution of dn/dc !

The effect of photosensitivity on TLS signal (case of Cr-DPC)

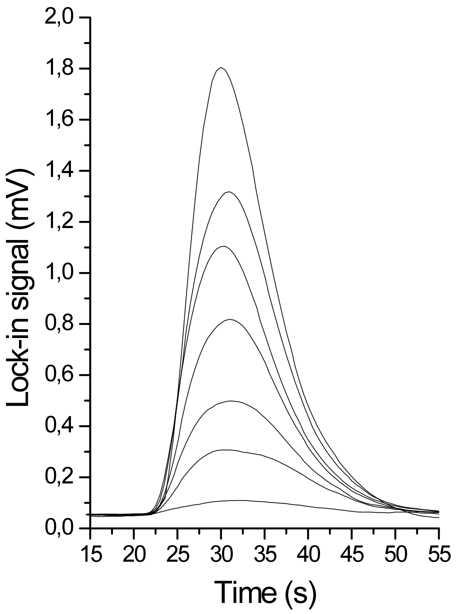


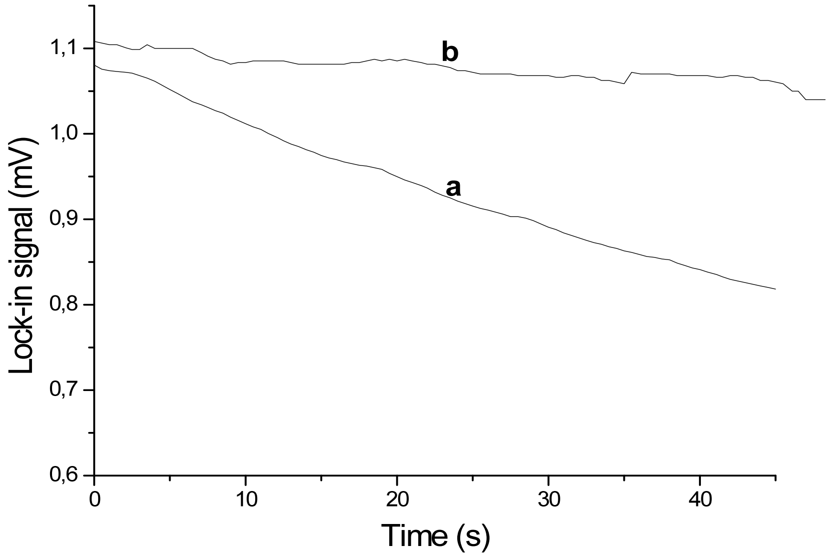
$t_{1/2}$=80 ms

$$I_{bc}(t)=I_{bc}(0)\left[1-\frac{\theta'\left[(C_0-C_e)\exp(-(k_r+k_D)t)+C_e\right]}{2}\tan^{-1}\left(\frac{1}{(1+t_c/t)\sqrt{3}}\right)\right]^2$$

Pedreira et al.: J. Appl. Phys., **100** (2006)
Chem. Phys. Lett. **396(**2004)221

Degradation of photolabile analytes

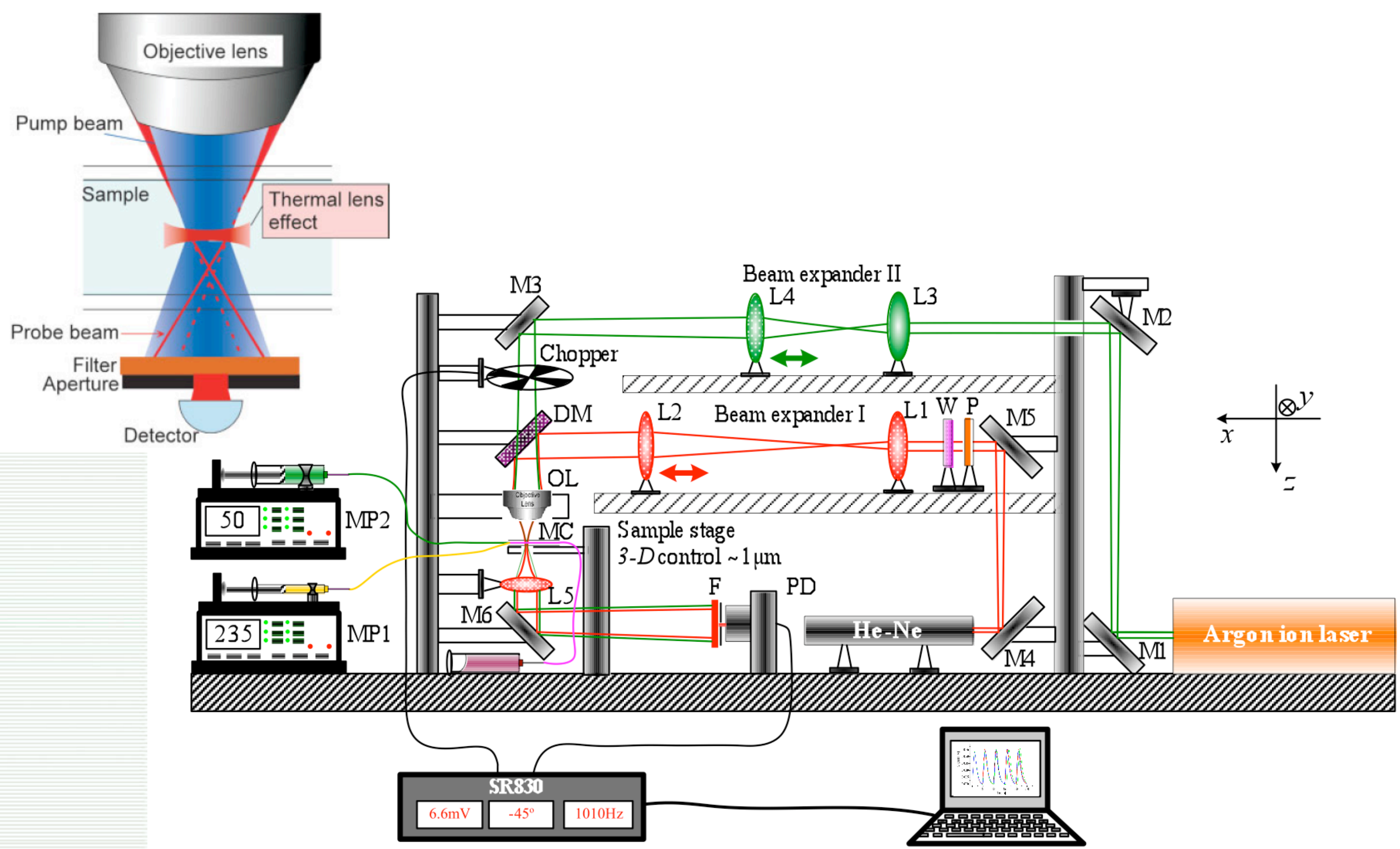




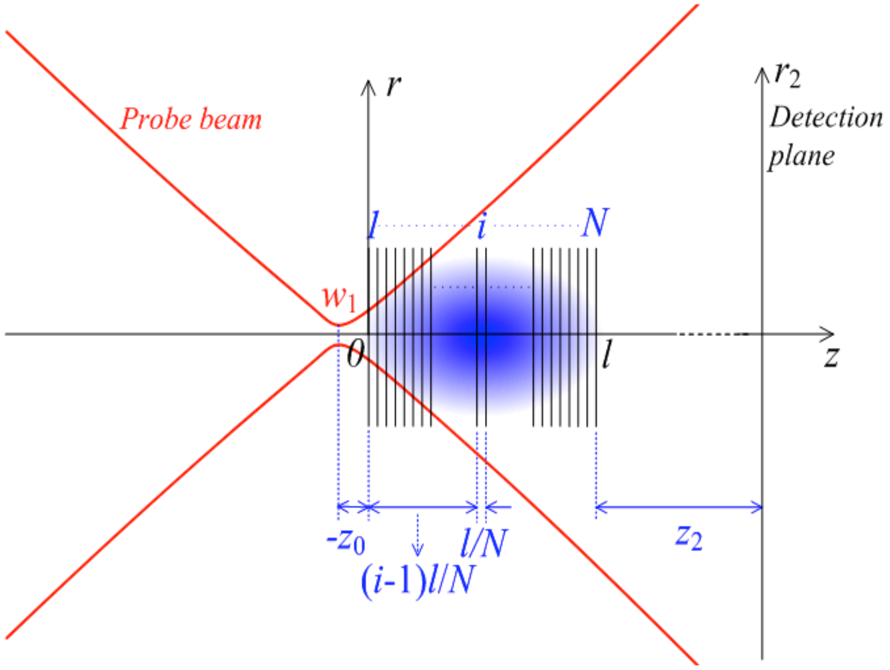
A. Madžgalj, M.L. Baesso, M. Franko, Eur. Phys. J. Special Topics **153** (2008) 503

Cr(VI) (μg/L)	1	3	9	15	20
Mean sig./mV (n = 5)	0.201	0.481	1.227	1.968	2.83
S.D.	0.005	0.005	0.015	0.010	0.014
R.S.D. (%)	2.52	0.95	1.23	0.49	0.48
LOD (μg/L)	0.065				

Adjustable beam size/position TLM

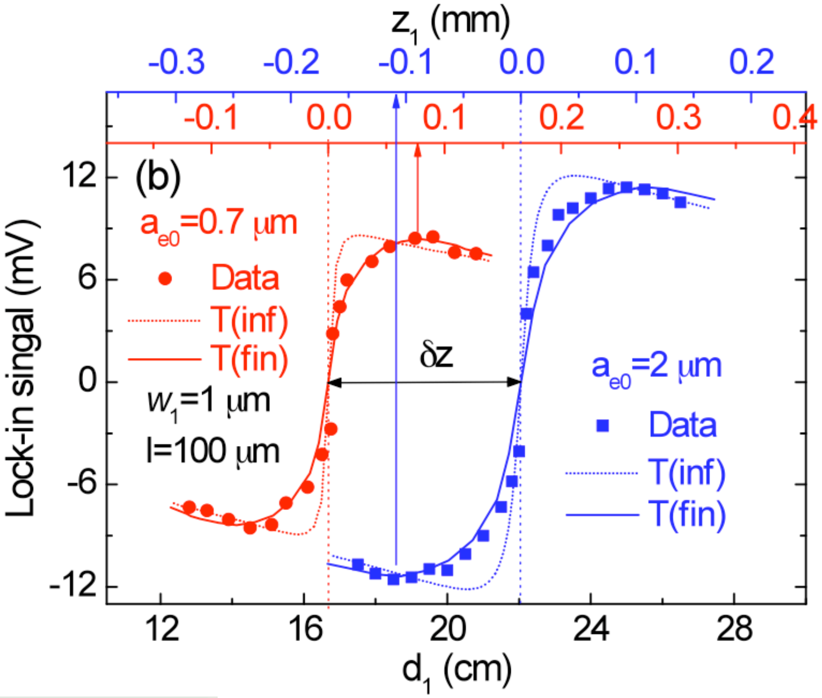


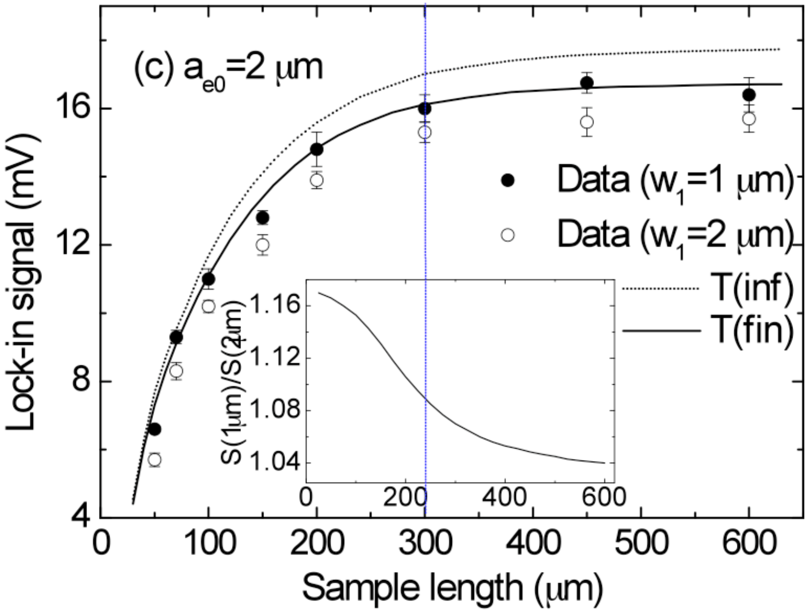
New theoretical model of thermal lens in TLM

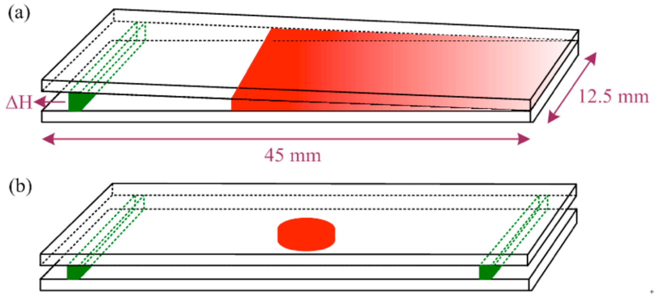


$$S_{tl}=\sum_{i=1}^{N}\frac{\left|E_2(r_2,z_1(i)+z_2,t=n/f+1/(2f))\right|^2-\left|E_2(r_2,z_1(i)+z_2,t=n/f)\right|^2}{\left|E_2(r_2,z_1(i)+z_2,t=0)\right|^2}$$

Effective sample length



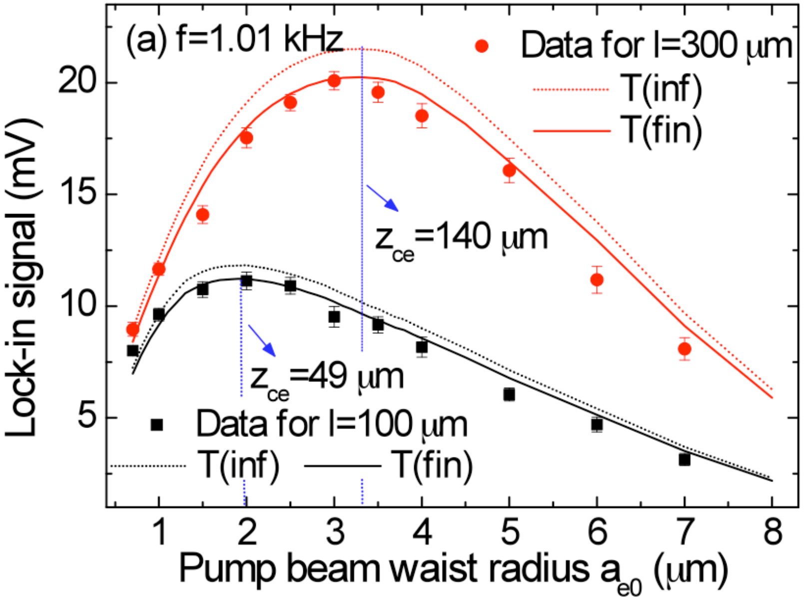


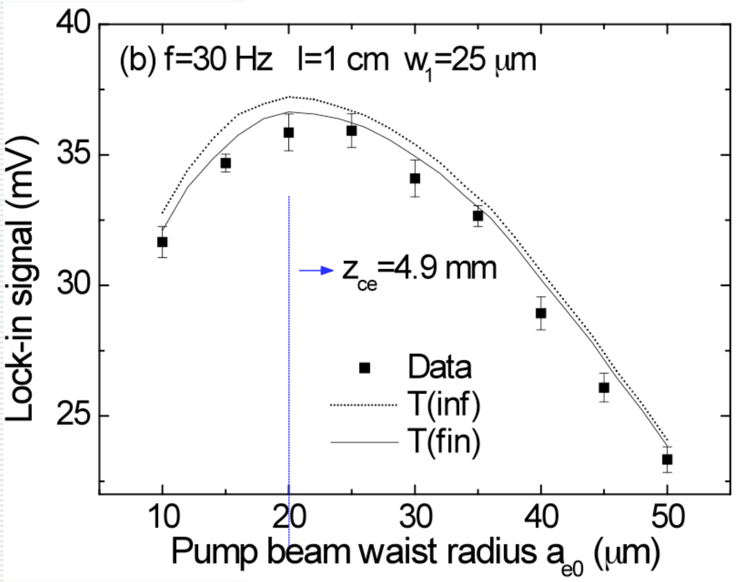


Liu M., Franko M. *Appl. Phys. B,* 2013
DOI 10.1007/s00340-013-5601-4

Optimization of pump beam radius and power density

With 1/10 or 1/20 power density of that at the diffraction limit, 1.36 times higher (l=100μm) or 2.3 times higher (l=300μm) TL signal is obtained at the optimum optical configuration – **important for photosensitive compounds !**





For sample length *l*

Optimal pump beam radius: $$a_{e0} = \sqrt{\frac{\lambda_e l}{4\pi}}$$

Optimal probe beam waist:

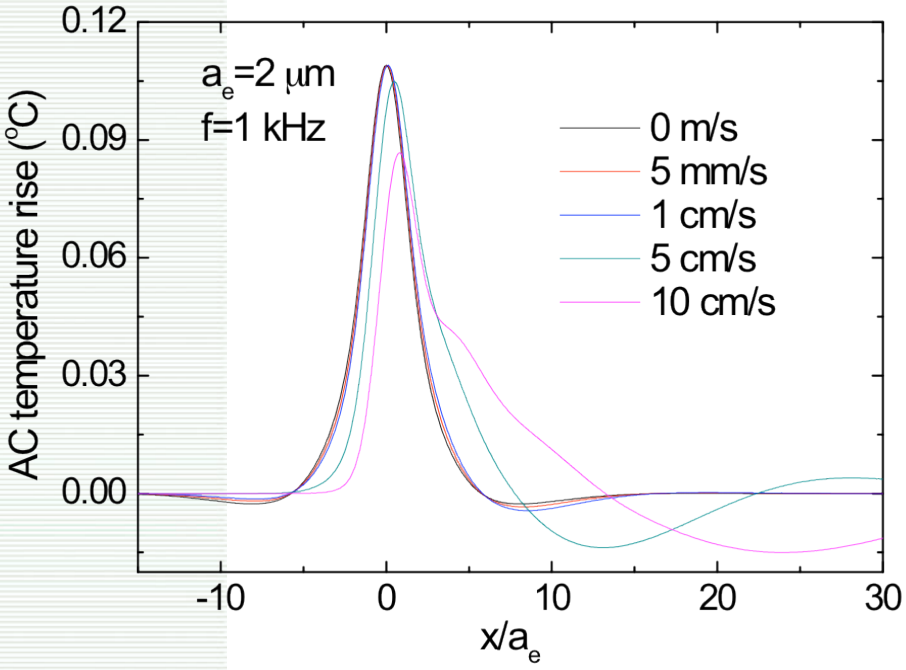
if $l < z_{ce}$, $w_1=0.5a_{e0}\sim a_{e0}$;

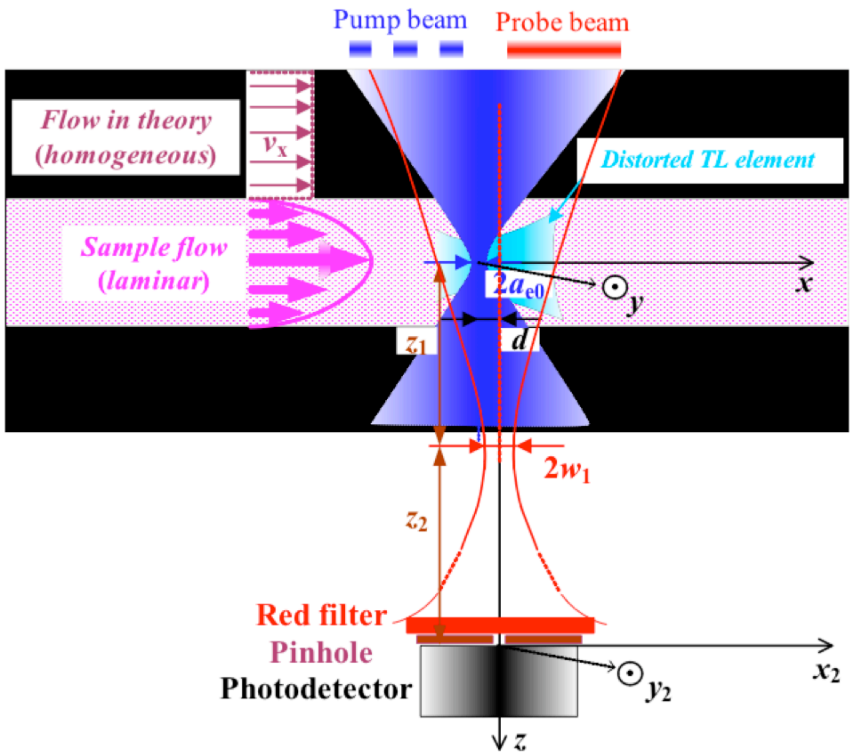
otherwise $w_1 = 0.7a_{e0}\sim a_{e0}$

Liu M., Franko M. *Appl. Phys. B,* 2013
DOI 10.1007/s00340-013-5601-4

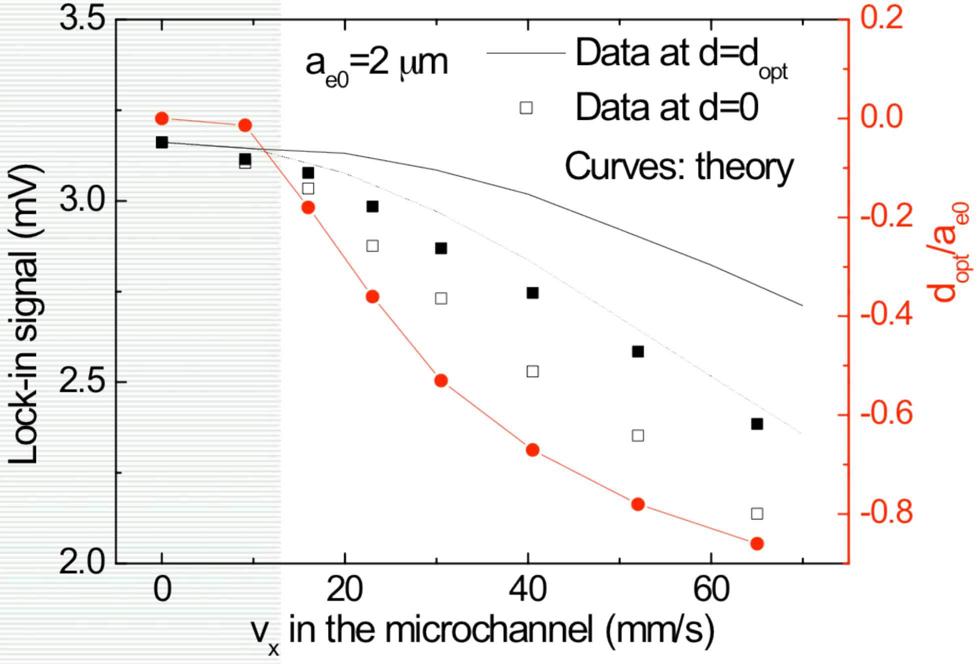
Effect of sample flow on TLM signal

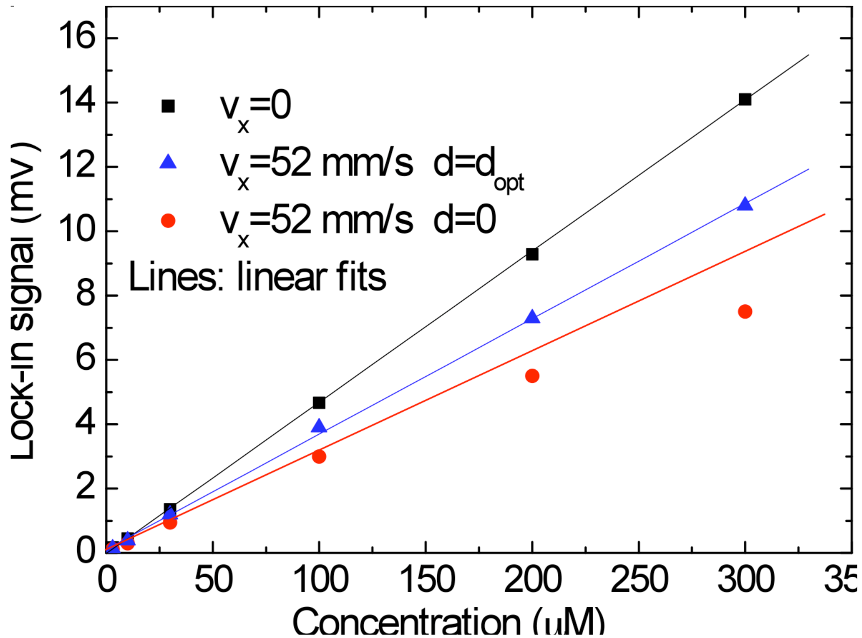
$$\frac{\partial T(r,t)}{\partial t} = D\nabla^2 T(r,t) - v_x \frac{\partial T(r,t)}{\partial x} + \frac{1}{\rho C_p} Q(r,t)$$

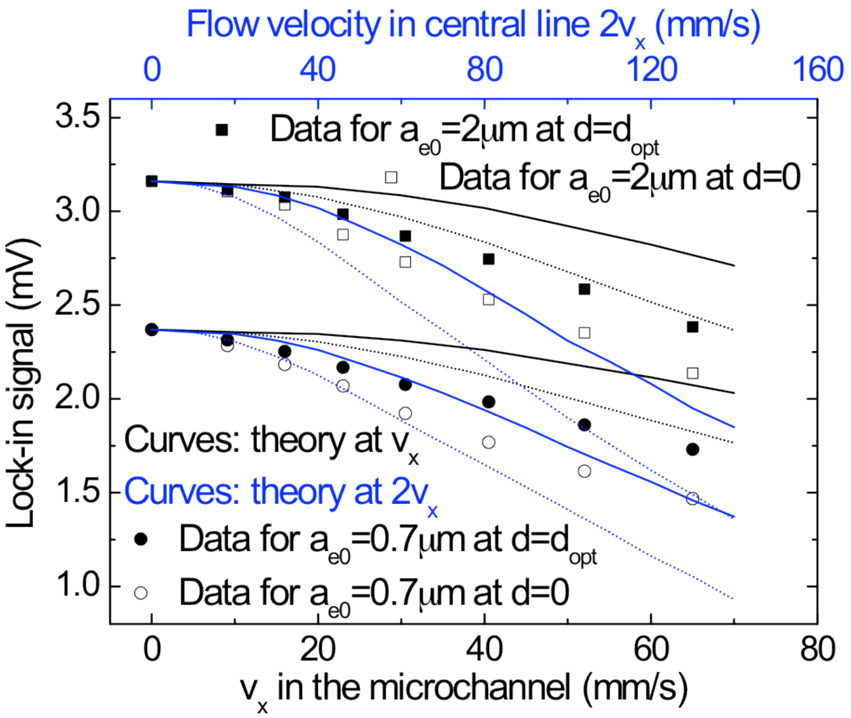




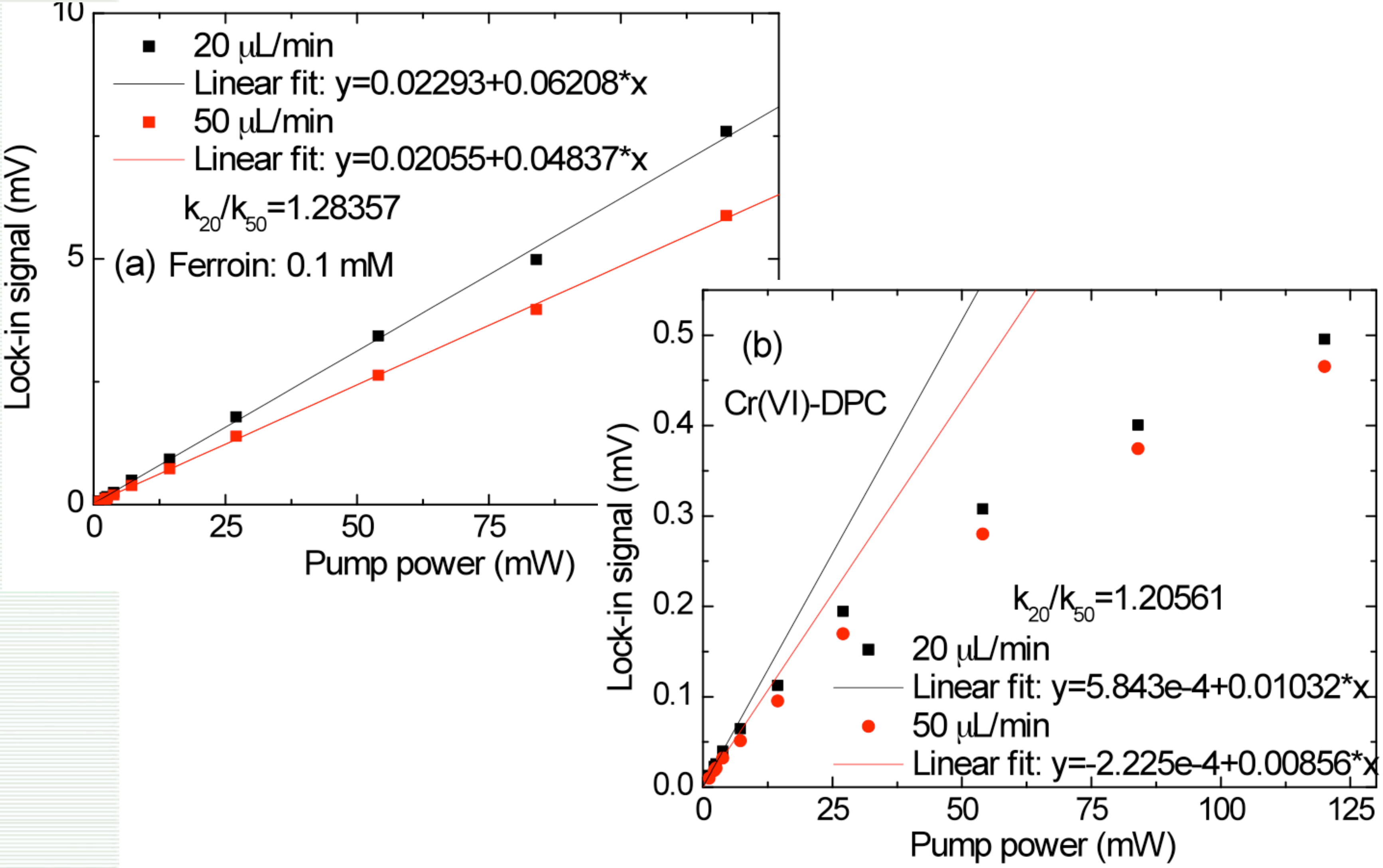
Effects of flow velocity and beam displacement on TLS signal



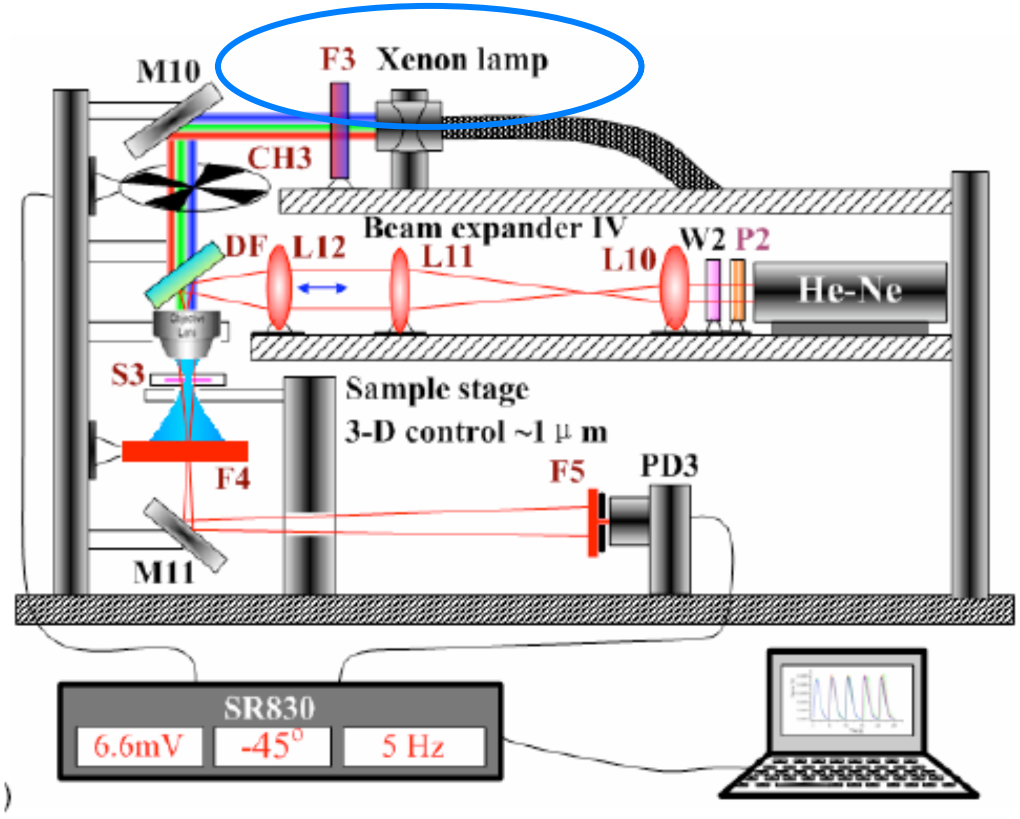




Effect of photodegradation in a microfluidic system



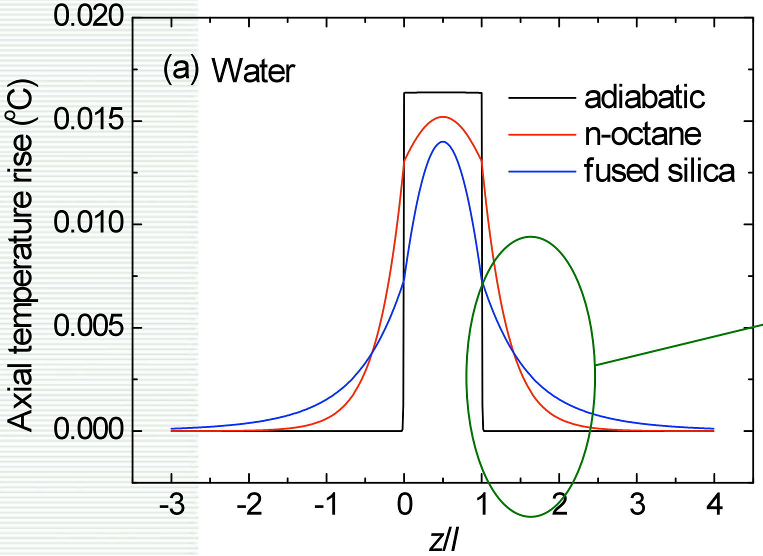
Incoherent light source (ILS)-excited TLM



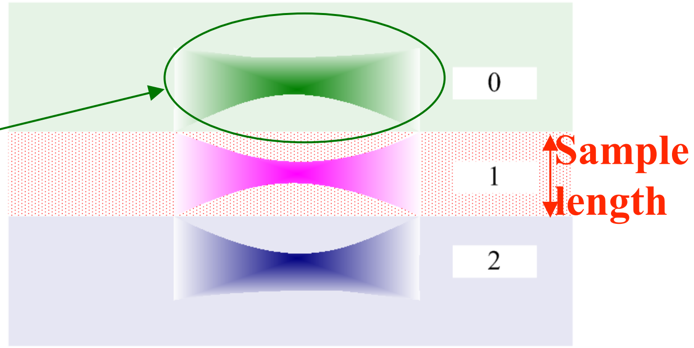
$P_{eq}(514.5\text{ nm})=1.05\text{ mW}$

Liu M., Franko M.: *Appl.Phys.Lett.*, **100**, 2012, 121110.

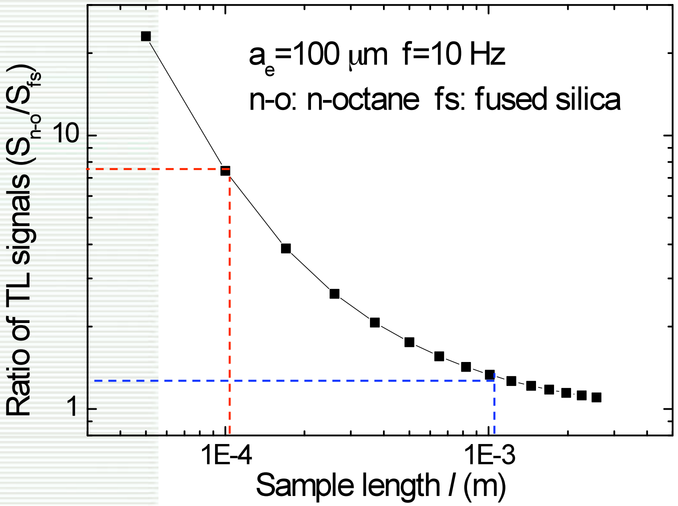
Thermal lens extends beyond the boundaries of microchannel



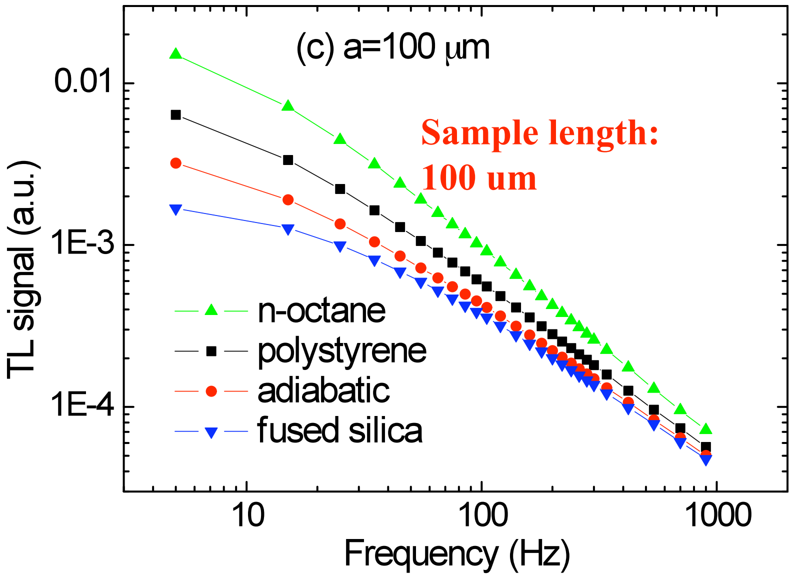
Sensitivity enhancement in ILS-TLM



Sample length



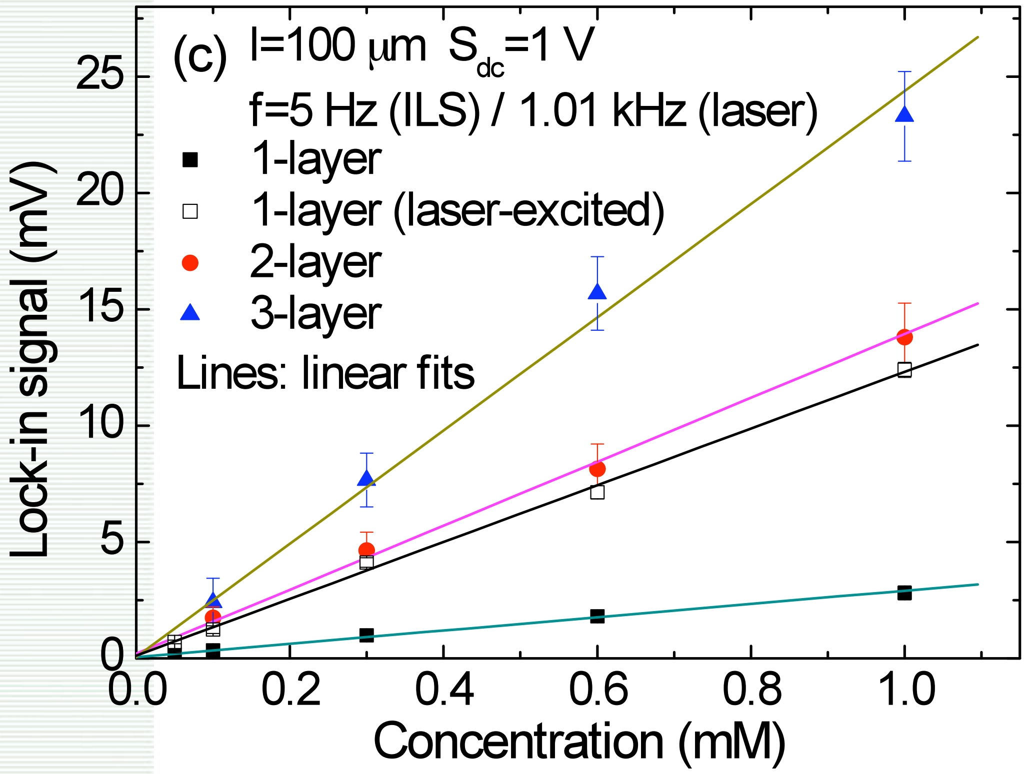
a_e=100 μm f=10 Hz
n-o: n-octane fs: fused silica

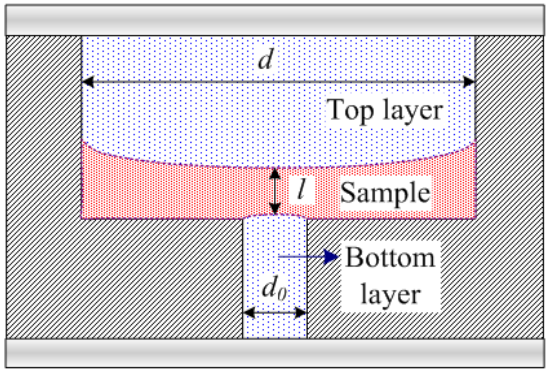


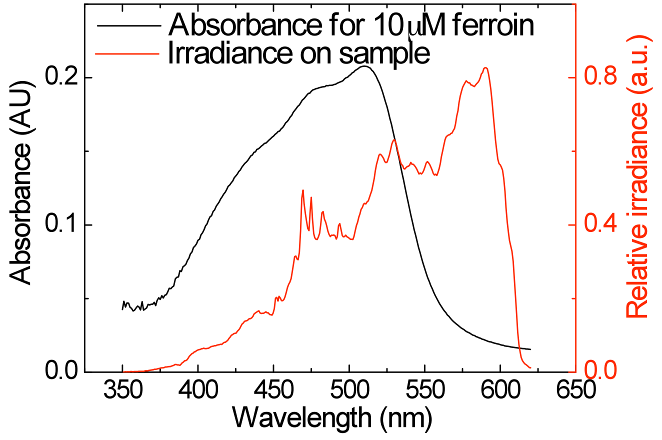
Sample length:
100 um

Sensitivity enhancement in ILS-TLM in layered samples

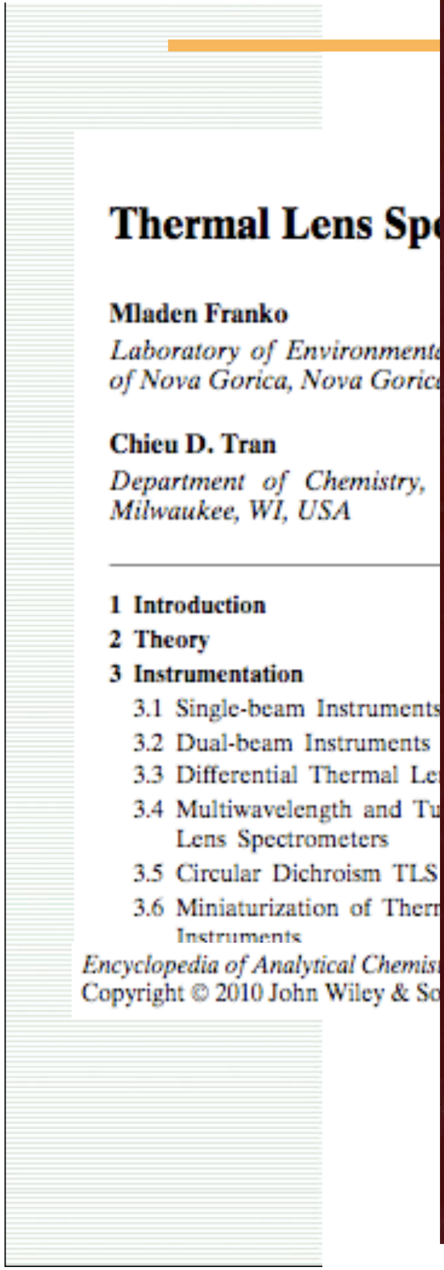
LOD: 5×10^{-7} M at *P*=1.05 mW and *l*=100 µm for ferroin ~ 2.2×10^{-5} AU







Liu M., Franko M.: *Adv. Mat. Res.*, **503-504**, 2012, 1480-1483.
Liu M., Korte D., Franko M.: *J. Appl. Phys.*, **111**, 2012, 033109.



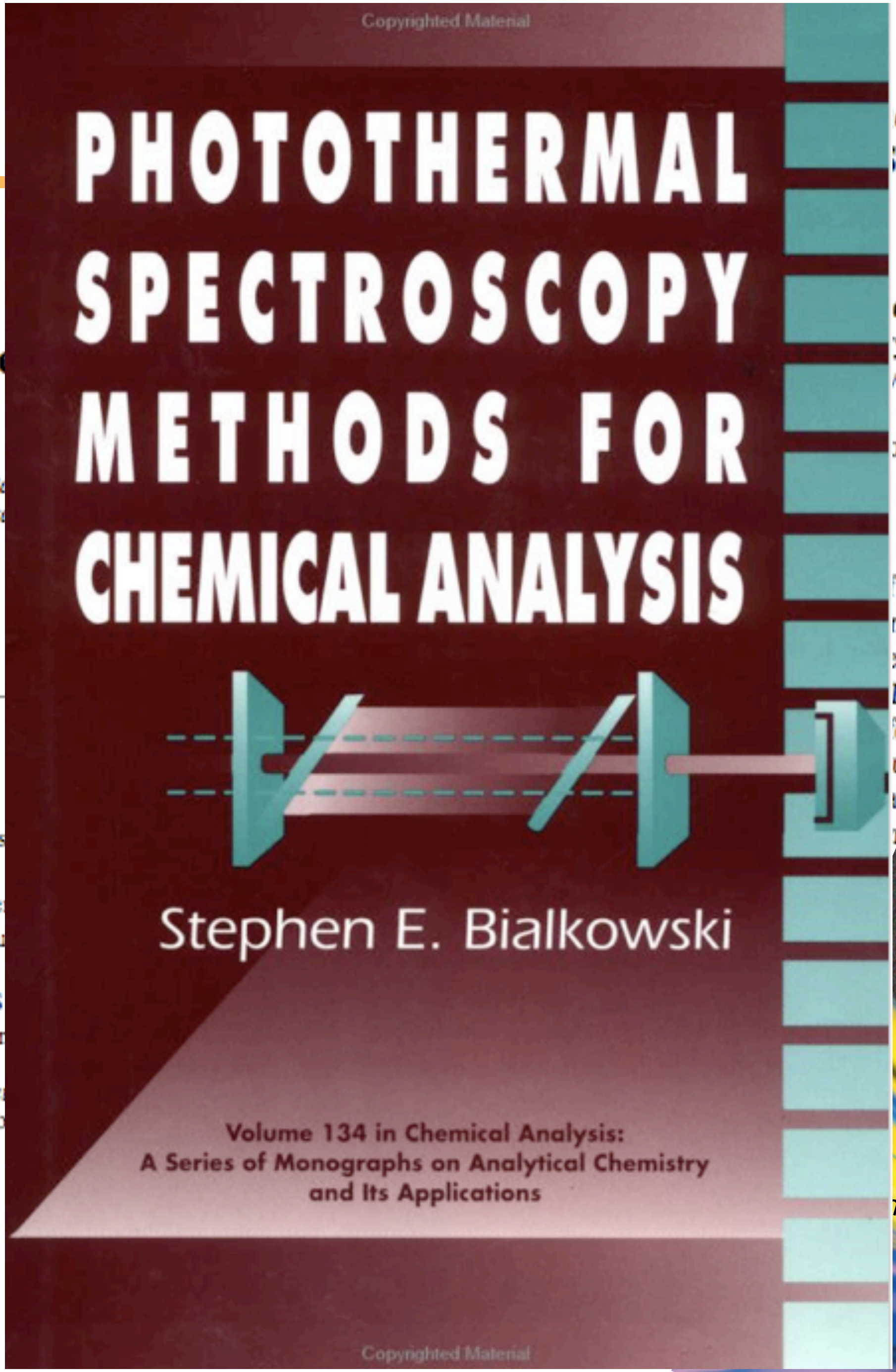
Thermal Lens Sp

Mladen Franko
Laboratory of Environment
of Nova Gorica, Nova Goric

Chieu D. Tran
Department of Chemistry,
Milwaukee, WI, USA

1 Introduction
2 Theory
3 Instrumentation
3.1 Single-beam Instruments
3.2 Dual-beam Instruments
3.3 Differential Thermal Le
3.4 Multiwavelength and Tu
Lens Spectrometers
3.5 Circular Dichroism TLS
3.6 Miniaturization of Ther
Instruments

Encyclopedia of Analytical Chemis
Copyright © 2010 John Wiley & So



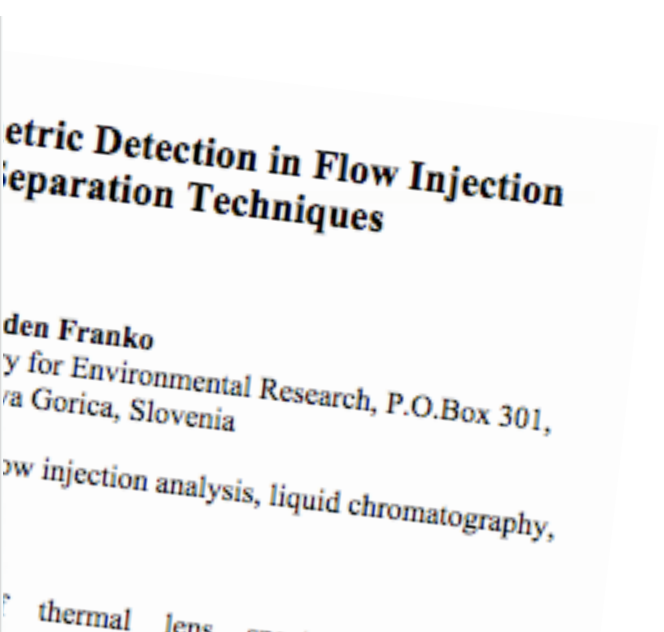
Copyrighted Material

PHOTOTHERMAL SPECTROSCOPY METHODS FOR CHEMICAL ANALYSIS

Stephen E. Bialkowski

Volume 134 in Chemical Analysis:
A Series of Monographs on Analytical Chemistry
and Its Applications

Copyrighted Material

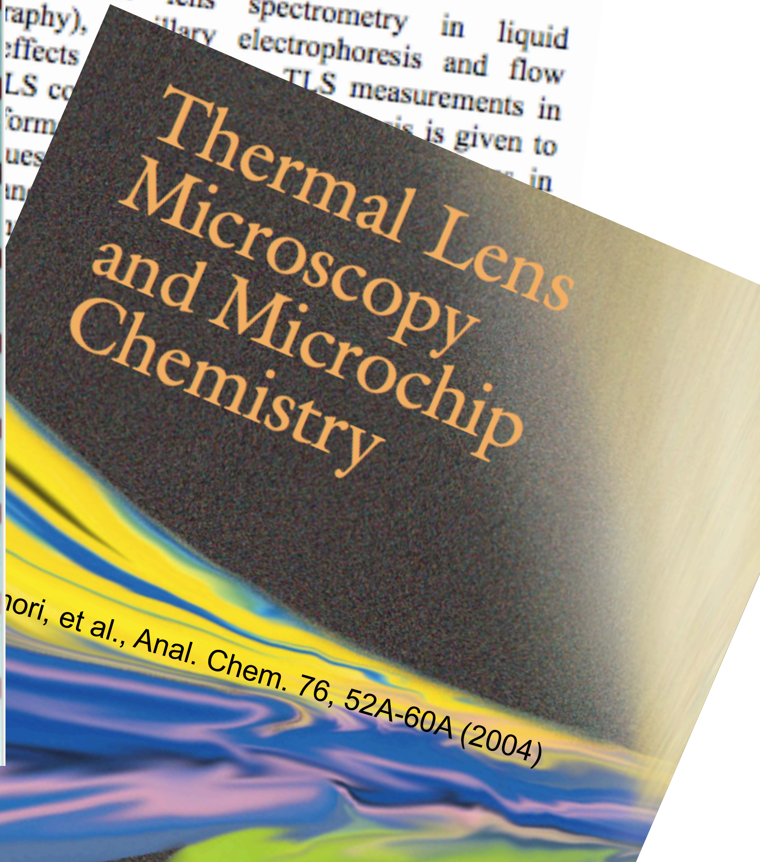


etric Detection in Flow Injection
eparation Techniques

den Franko
y for Environmental Research, P.O.Box 301,
a Gorica, Slovenia

ow injection analysis, liquid chromatography,

thermal lens spectrometry in liquid
raphy), llary electrophoresis and flow
ffects TLS measurements in
LS co is is given to
form
ues



Thermal Lens Microscopy and Microchip Chemistry

ori, et al., Anal. Chem. 76, 52A-60A (2004)

Basic literature on TLS

- S.E. Bialkowski, *Photothermal Spectroscopy Methods for Chemical Analysis*, John Whiley & Sons, Inc., New York, 1996.
- C.D. Tran , M. Franko, "*Thermal Lens Spectroscopy*" in: Encyclopedia of Analytical Chemistry, (Ed. R.A. Meyers), John Wiley: Chichester., 2010. DOI: 9780470027318.
- M. Franko*, "Bioanalytical Applications of TLS"*, in: Thermal wave physics and related photothermal techniques: Basic principles and recent developments, ISBN 978-81-7895-401-1 (Ed. E. Marin), Research Signpost Press, 2009
- M. Franko, Thermal Lens Spectrometric Detection in Flow Injection Analysis and Separation Techniques, *Appl. Spectrosc. Rev*. **43**, 2008, 358-388.

Basic literature on TLS

- Kitamori, T., Tokeshi, M., Hibara, A. and Sato, K., Thermal lens microscopy and microchip chemistry. *Anal. Chem.*, **76**, 2004, 52A-60A.
- M. Franko, C.D. Tran, Analytical Thermal Lens Instrumentation, *Rev. Sci. Instrum*. **67**, 1996, 1-18.
- Liu M., Franko M.: Progress in thermal lens spectrometry and its application in microscale analytical devices, *Crit. Rev. Anal. Chem.* **44**, 2014, 328-353.