

LF402 Metabolic – Metabolism Detector

General Description

The LF402 Metabolic is a research instrument for the assessment of tissue metabolism by using the uv-induced intrinsic fluorescence pattern. The tool for the scientist is a fibre-optic probe. It enables superficial and minimal invasive experiments on living tissues.

Main technical characteristics are a pulsed uv-laser, a sophisticated dual-channel time-gated signal detection and the optimised method of data analysis.

The uv-induced fluorescence is related to the intra-cellular NADH concentration which in turn is dependent on the metabolic activity in the area of observation. This NADH dependent signal is referenced with a second signal largely not correlated with NADH. In this way a meaningful indicator of the tissue's redox potential is generated that is quite robust against varying tissue optical properties or artefacts.

Intended Use

Medical research work in conjunction with the following set-ups :

- Animal models
- Organ models
- Organ preparations
- Tissue preparations
- Tissue cultures
- Cell cultures



Figure 1: Hamster model

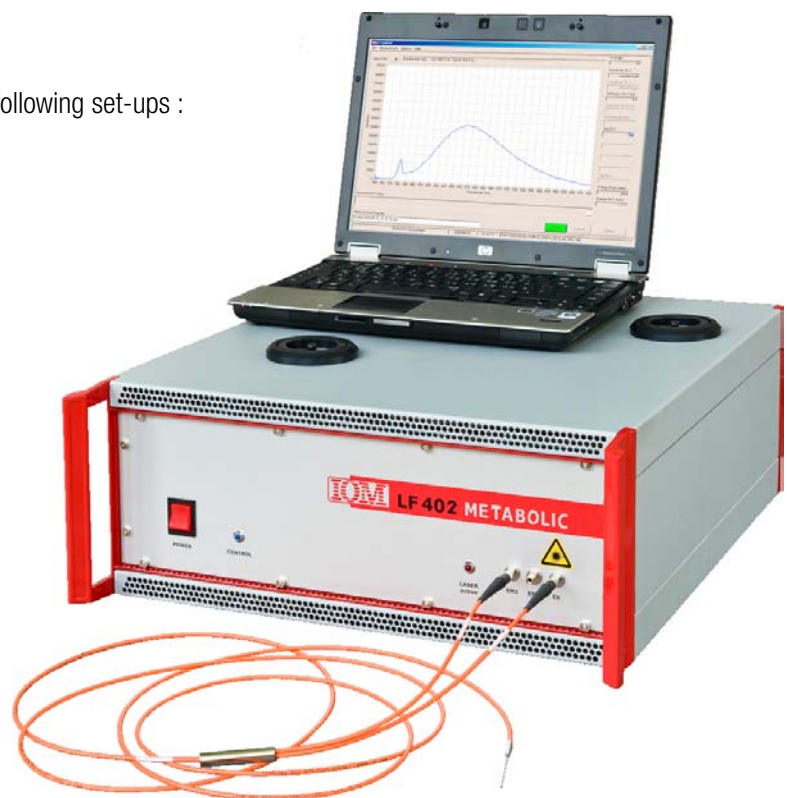


Figure 2: LF402 Metabolic

The LF402 Metabolic is not a medical device!

LF402 Metabolic – Metabolism Detector

Technical Specifications

Light source:	Nitrogen laser (337 nm, 30 Hz)
Detector:	Photomultiplier tube
Detection channels:	2
Detection wavelengths:	NADH channel: 460 nm Flavin channel: 530 nm
Signal detection:	in 2-ns-gate
Fiber-optic probe:	3.15 m length with stainless steel probe tip

Fibre-Optic Probes

The standard fibre-optic probe has 7 x 200 µm fibres assembled in a 1-mm stainless steel cannula. The probe tip is optically polished.

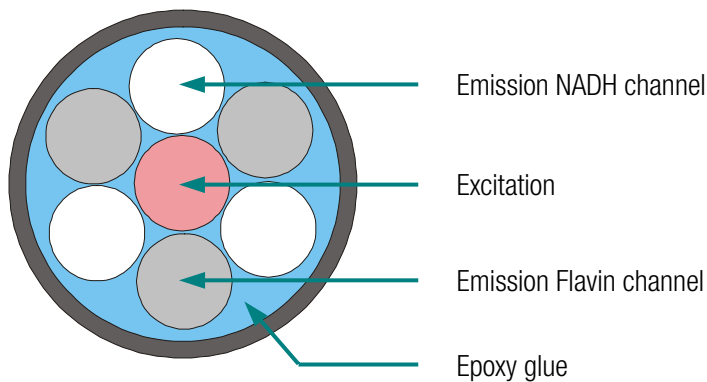


Figure 3: Cross-section of a standard probe

Fibre-optic probes with different geometries of probe tips, with different fibre diameters or which can be sterilised by different methods are available upon request. The following picture shows a miniaturised probe with implantable probe tip:

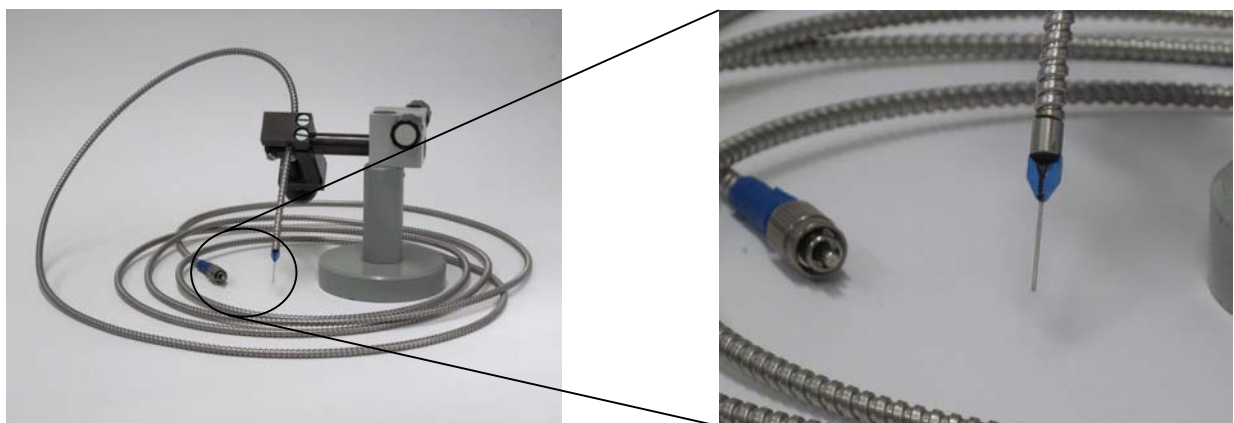
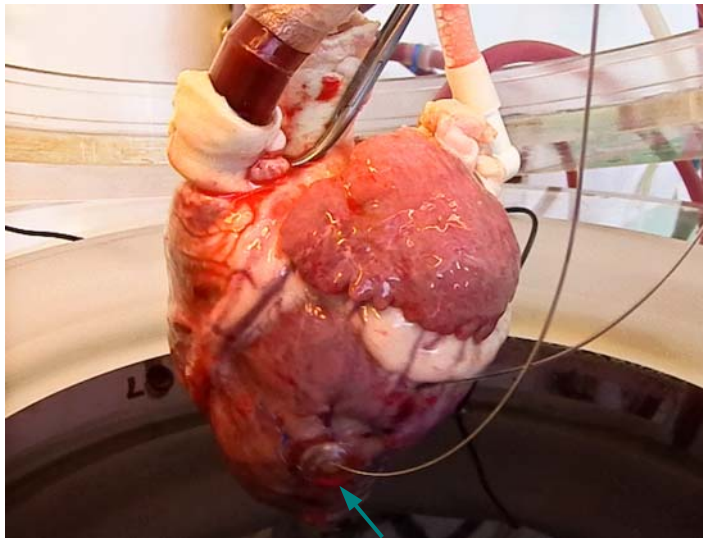


Figure 4: Miniaturised and implantable probe tip

LF402 Metabolic – Metabolism Detector

Application for monitoring of a perfused heart model:

The fluorometric assessment of the redox potential of heart muscle tissue was investigated with pig hearts perfused in a four-chamber working heart model (see [1]). The fibre-optic probe tip was placed on the lateral surface in the region of the left atrium.

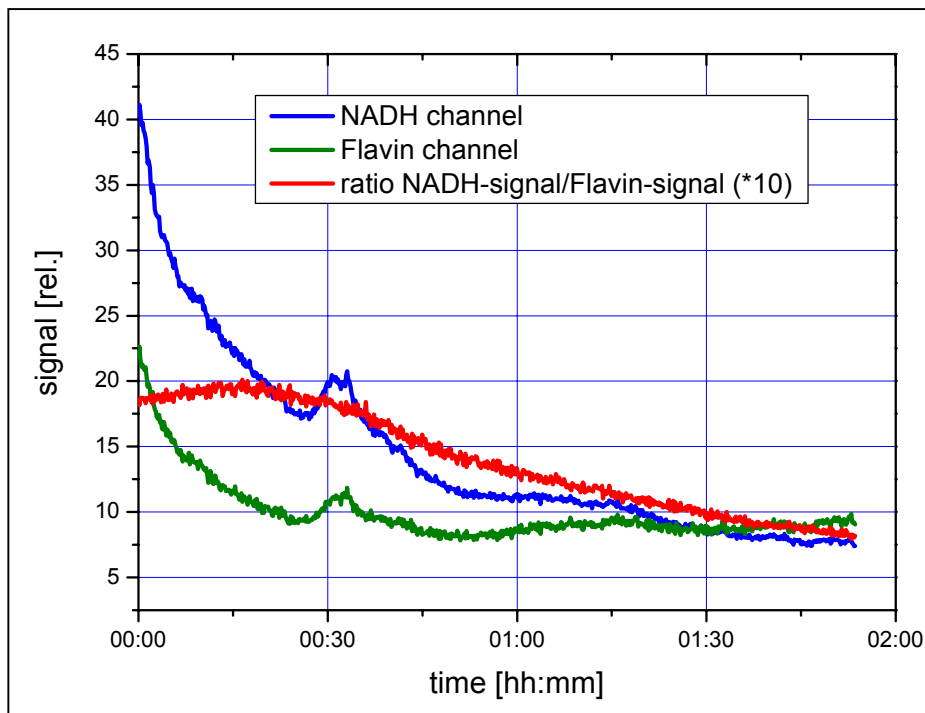


The working phase of the beating hearts lasted several hours. Six organs in total have been monitored during this time.

In intervals of 30 minutes offline parameters like pH, pO_2 , pCO_2 , O_2 saturation and electrolyte concentrations of Na^+ , K^+ and Ca^{2+} were obtained. Perfusate flow through the aorta as well as coronary blood flow were measured using flow meters.

The Troponin-I contents in the blood sample was determined using a commercial immuno assay every 60 minutes.

Figure 4: Perfused pig heart model with attached fibre-optic probe (indicated by arrow)



In steady state phase of the beating heart, after initially being constant, a continuous decrease of the spectroscopically revealed redox state was observed (decrease of the red curve after a stable signal during the first 30 minutes).

This decrease of the redox potential was accompanied by a decreasing heart performance and indications on a changing electrolyte equilibrium (K^+ concentration). At the same time the Troponin I contents in the perfusate was found to grow.

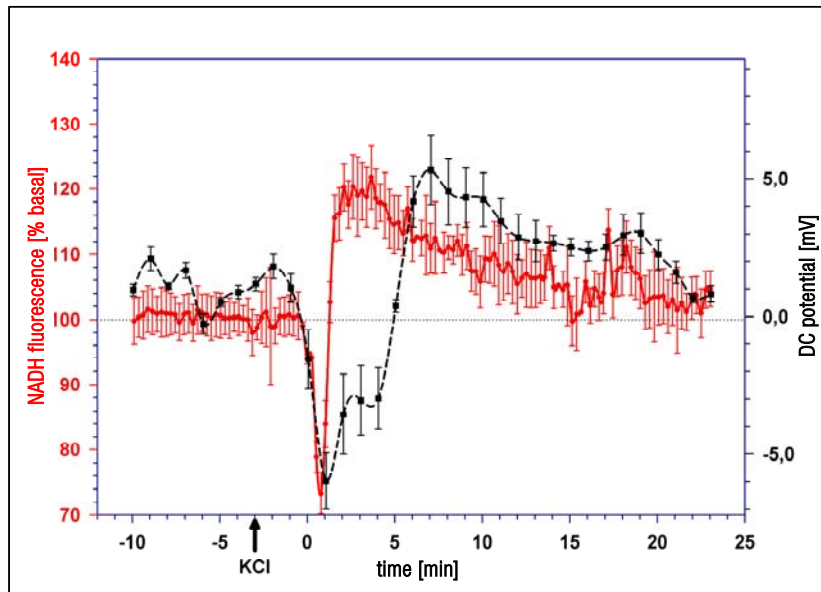
Figure 5: Long-term monitoring of the perfused heart model

By using the multiple linear regression method it was shown that the fluorometrically obtained status of the redox potential of the heart muscle tissue correlated with aorta flow, blood pH and pO_2 , and K^+ concentration.

LF402 Metabolic – Metabolism Detector

Application in Neuroscience:

Aerobic energy metabolism of the brain is closely related to neuronal activity and to the physiological and pathophysiological responses of this organ. The fluorometric assessment of metabolism activity was successfully applied to several different impairments of brain function (e.g. spreading depression, see following graph).



In another work a stimulation of the 5-HT_{1A} receptor in the rat brain was causing a decrease of the neuronal activity in the ventral hippocampus as demonstrated by an increase in NADH fluorescence. [3]

Beside investigations in combination with anesthetized animals in another study the relation between brain metabolism and neuronal activity in freely moving animals exhibiting a movement disorder was characterised [2]. Data clearly confirm that the expression of paroxysmal dystonia in dt^{sz} mutant hamsters is associated with enhanced striatal activity.

Figure 6: Short term response of NADH auto fluorescence (red curve) in relation to the depolarisation curve during a spreading depression measured on the surface of a rat brain

Summary:

The metabolism detector LF402 Metabolic has proven its potential to yield high-quality information on the metabolic activity of different tissue and organ types in living animals and in tissue preparations. Particularly interesting for the scientist is the fibre-optic probe representing the experimental tool. It allows remote investigations on tissue surfaces or with minimal invasiveness due to small diameter probe tips. In contrast to most other sensor systems real online data without sample removal are provided.

Literature (selection):

1. Pfeifer L, Gruenwald I, Welker A, Stahn RM, Stein K, Rex A. Fluorimetric characterisation of metabolic activity of ex vivo perfused pig hearts. *Biomed Tech (Berl)*. 2007;52(2):193-199.
2. Hamann M, Richter A, Fink H, Rex A. Altered nicotinamide adenine dinucleotide (NADH) fluorescence in dt sz mutant hamsters reflects differences in striatal metabolism between severe and mild dystonia. *J Neurosci Res*. 2009 Feb 15; 87(3):776-83.
3. Rex A, Fink H. Effects of 8-OH-DPAT on Hippocampal NADH Fluorescence In Vivo in Anaesthetized Rats. *Journal of Neuroscience Research* 83, 551–556, (2006)

Acknowledgement

We wish to thank Dr. André Rex for providing data and graphs related to the living animal investigations. (Dr. André Rex, Experimental Neurology, Charité Berlin, Germany)