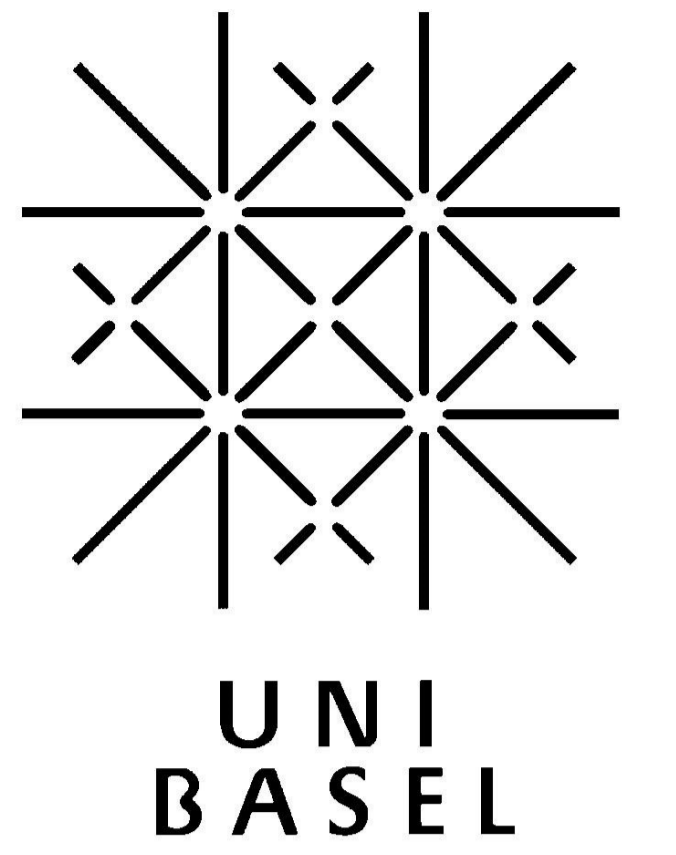


Software design and device development for cantilever array deflection read-out in the FlexAFM head

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1. Motivation

The development is based on the commercially available single-cantilever FlexAFM head from NanoSurf. Our idea is to extend the concept using a cantilever array composed of eight cantilevers. The cantilevers' length is 500 μm , the width is 100 μm and the thickness is 1 μm . The pitch is 250 μm . The cantilever array is housed in a sealed liquid cell. The aim is to detect biomarkers in liquid. Hence the cantilevers have to be coated with corresponding probe molecules to detect the target molecules. Adsorption of biomarkers generates on the functionalized cantilever side a surface stress, which causes its bending (see figure 1.4). In this project we present the development of hardware and a deflection data acquisition software for data visualization and analysis.

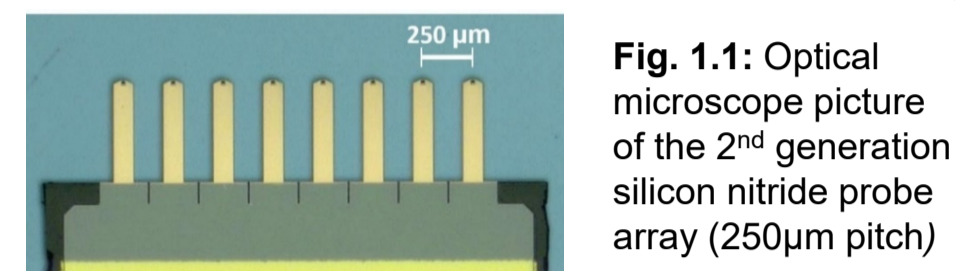


Fig. 1.1: Optical microscope picture of the 2nd generation silicon nitride probe array (250 μm pitch)

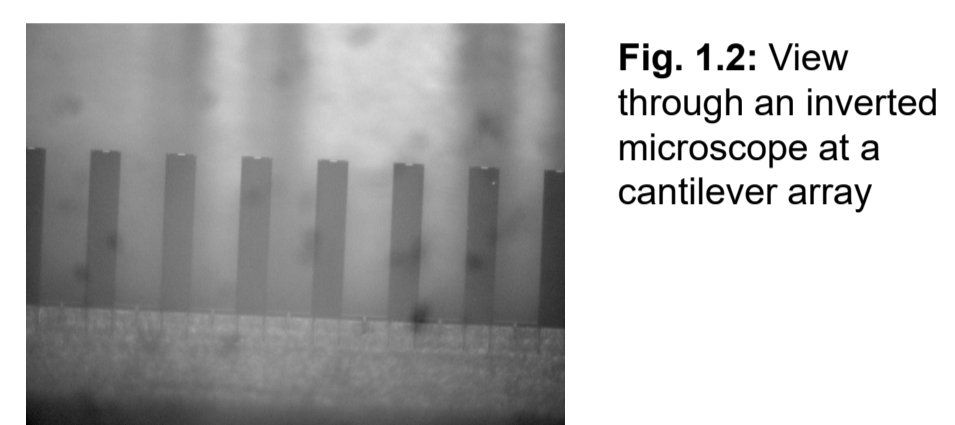


Fig. 1.2: View through an inverted microscope at a cantilever array

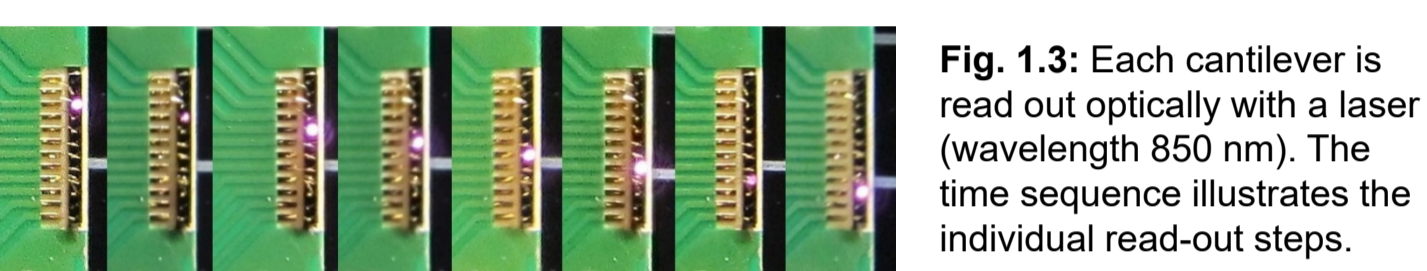


Fig. 1.3: Each cantilever is read out optically with a laser (wavelength 850 nm). The time sequence illustrates the individual read-out steps.

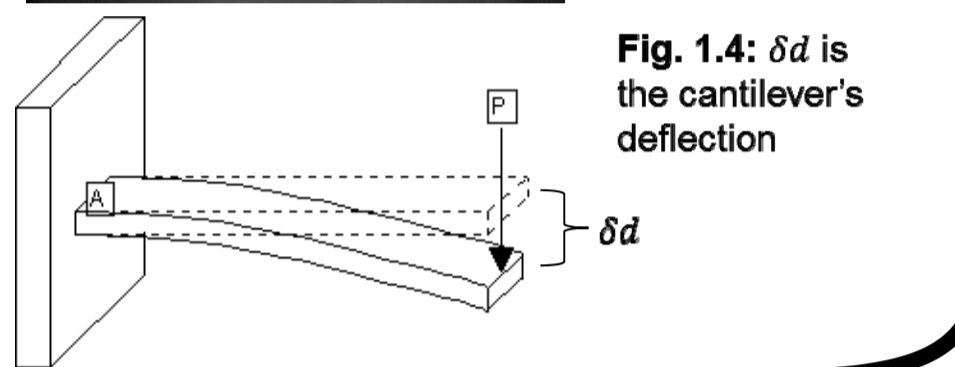


Fig. 1.4: δd is the cantilever's deflection

2. The FlexAFM device

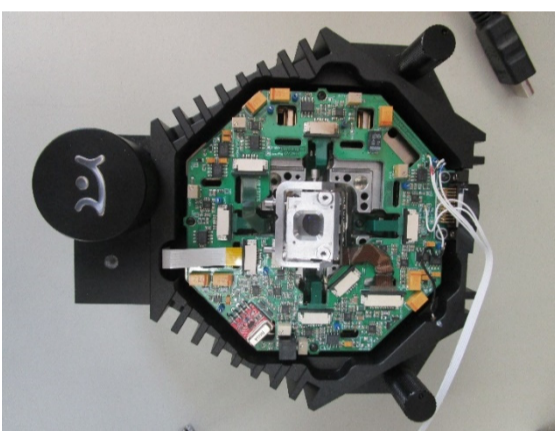


Fig. 2.1: Top view of the FlexAFM device. In the center we placed the holder for the vertical cavity surface emitting lasers (VCSELS). The laser beams for cantilever deflection read-out pass through the central hole.



Fig. 2.4: Top view of the FlexAFM head with mounted VCSELS' holder and VCSELS.

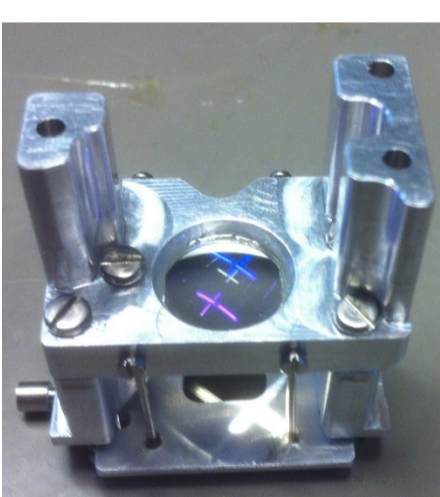


Fig. 2.2: Detailed view of the VCSELS' holder containing a platform which can be moved in three directions for positioning the laser spots close to the free end of the cantilever. In the central axis of the holder, we inserted a lens for focusing the laser beam.

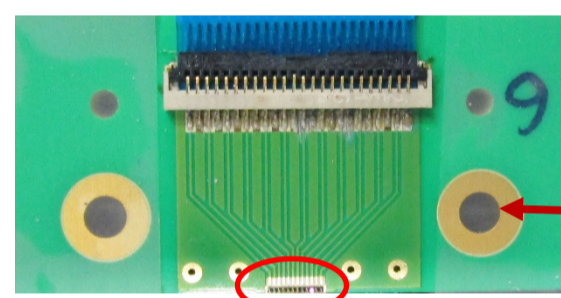


Fig. 2.5: Detailed view of the VCSELS' board. VCSELS are marked by the red ellipse.



Fig. 2.3: The VCSELS' holder is inserted in the FlexAFM head to replace the production series laser unit. The optical path is chosen in such a way that the laser light falls onto the position sensitive detector (PSD).



Fig. 2.6: Cantilever array holder. The gold colored part is the cantilever array (marked by a red circle).

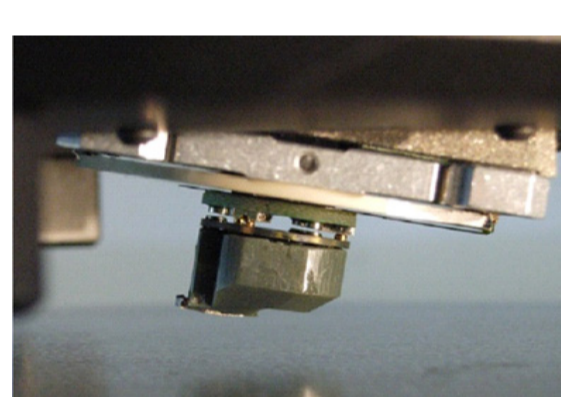
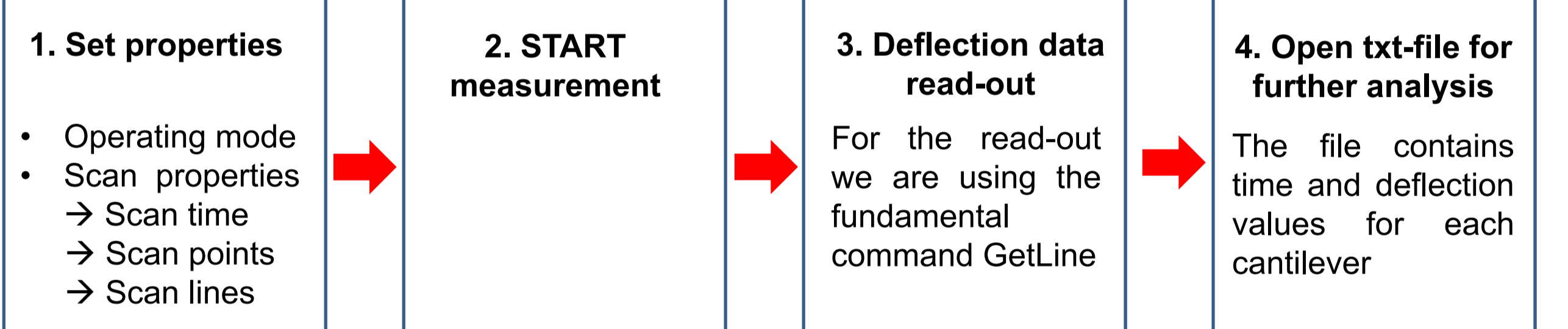


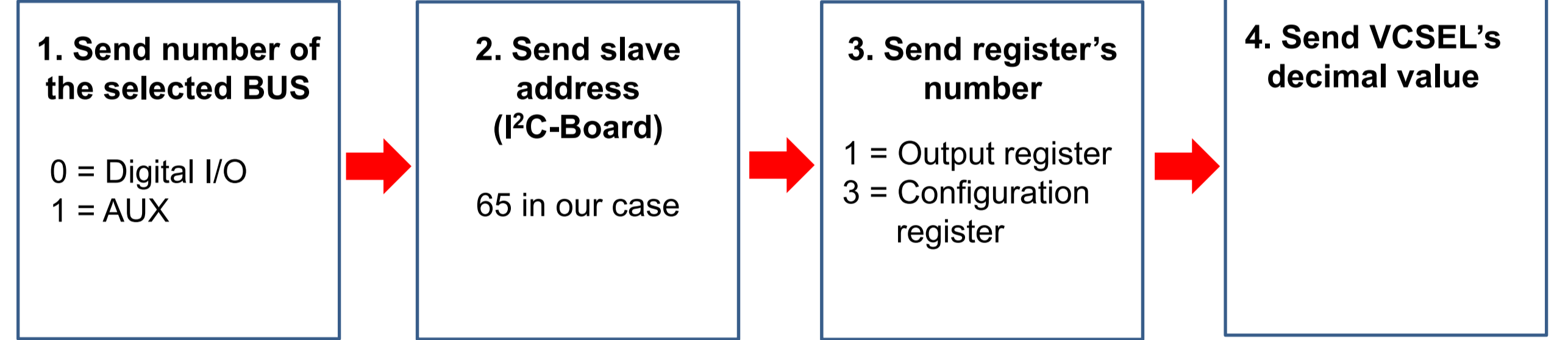
Fig. 2.7: Side view of the cantilever array holder mounted at the bottom of the FlexAFM head. The cantilever array holder is fixed by 3 tiny magnets.

5. Software development

Control of the FlexAFM device is implemented using an object-oriented programming language. It distinguishes different classes: the main class (C3000.Application), online objects and data processing objects. For each object we can set several properties and choose different methods. In our case, we are using online objects, so that the read-out can be simultaneously displayed. The software has been designed based on the following scheme:



During the deflection data read-out our software controls the switching of the VCSELS. This means that the actually measured cantilever has to be illuminated by the corresponding VCSEL. For selection of the VCSEL we use implemented MacroCMDs of the main class «SPMCtrlManager». The following scheme shows the sequence of the bits' transmission:



3. Experimental setup

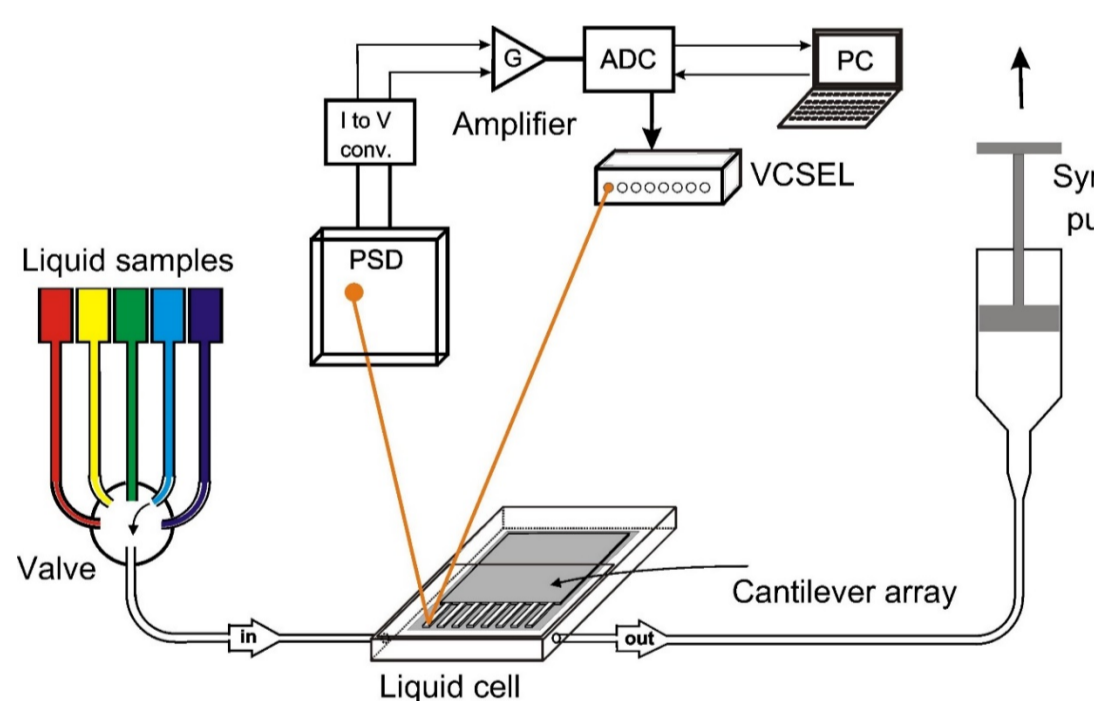


Fig. 3.1: Overview on read-out and liquid-handling system. The FlexAFM head contains the VCSELS, the PSD and the liquid cell with the cantilever array. In addition, we need a syringe pump and a liquid sample selector (multiposition valve device). After selecting a valve position the corresponding liquid is aspirated by the syringe pump and flows through the liquid cell, which contains the cantilever array. The reaction between the biosensing layer on the cantilevers and the target molecules generates a surface stress, which causes their bending. The deflection values are determined using a laser beam with a wavelength of 850nm, which is reflected on the cantilever surface and measured using a position sensitive detector. Data processing and acquisition is performed by specifically created software.

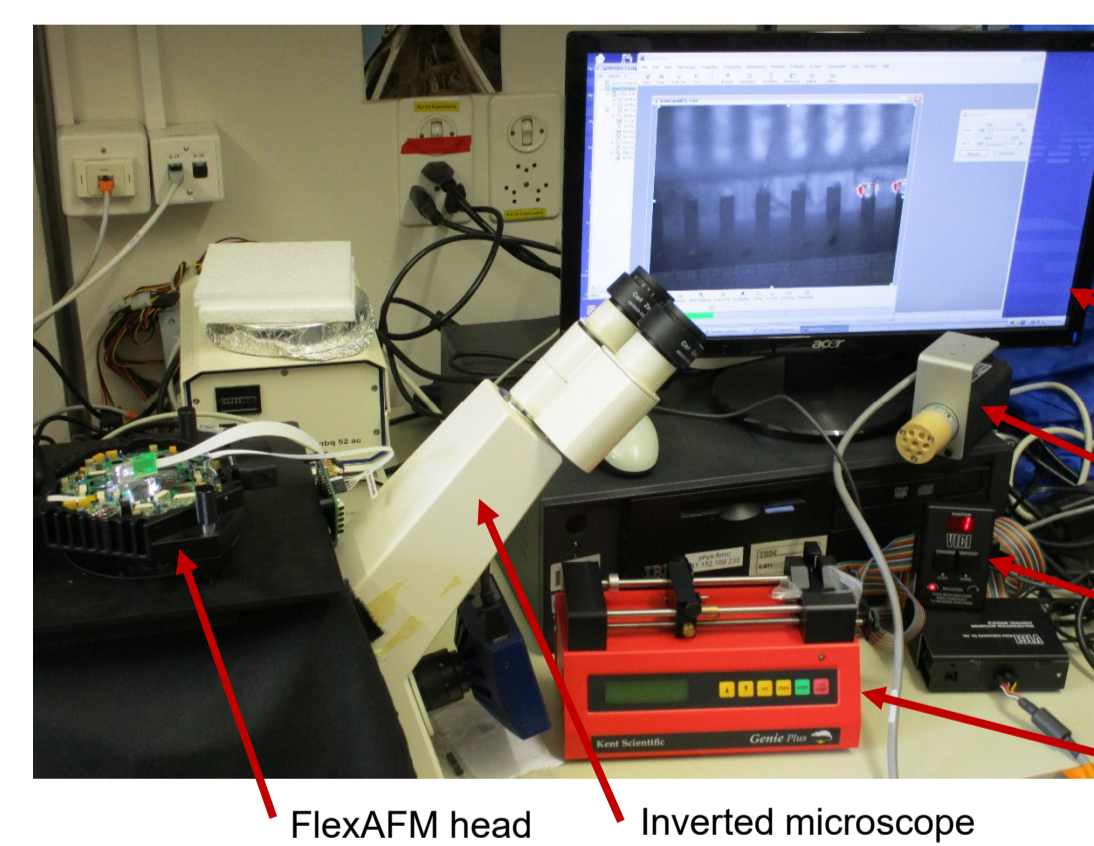


Fig. 3.2: Photo of the experimental setup of our FlexAFM-based device, the Kent Scientific Syringe Pump and the Multiposition valve device.

PC for monitoring the cantilever array and to adjust the position of the laser spots on the cantilevers.
6-position valve device
Valve's indicator
Kent Scientific Syringe Pump

4. I²C-Board

To control the switching of the eight cantilevers we use an I²C (Inter-Integrated Circuit)-Chip. I²C is based on the Master-Slave principle. It has two open drain lines: the **serial clock line** (SCL) and the **serial data line** (SDA), which are pulled up by resistors. The Master generates the clock and the slave responds if it's addressed. To start the bit's transmission we need a START option, this means that SDA has to be low and SCL must be high. Before sending one bit to the addressed slave, it's necessary to set SCL to low and SDA to high. Once SCL is high, sending starts. Each bit's transmission is followed by an ACK-bit (Acknowledgment bit) sent by the slave.

Here, we are using a 4-bit I²C-Bus. It is based on four registers: the input port register, the output register, the polarity inversion register and the configuration register. We send decimal values represented in binary code. In figure 4.2 the sequence is illustrated how the decimal values are sent. Before one LED can be switched on, we need to configure each port as an output.

Our I²C-board also has a current and voltage regulator, which allows to set the maximum values. In this way overheating of the VCSELS can be prevented. Our VCSELS need a typical current of 2mA and a voltage of 1.95V.

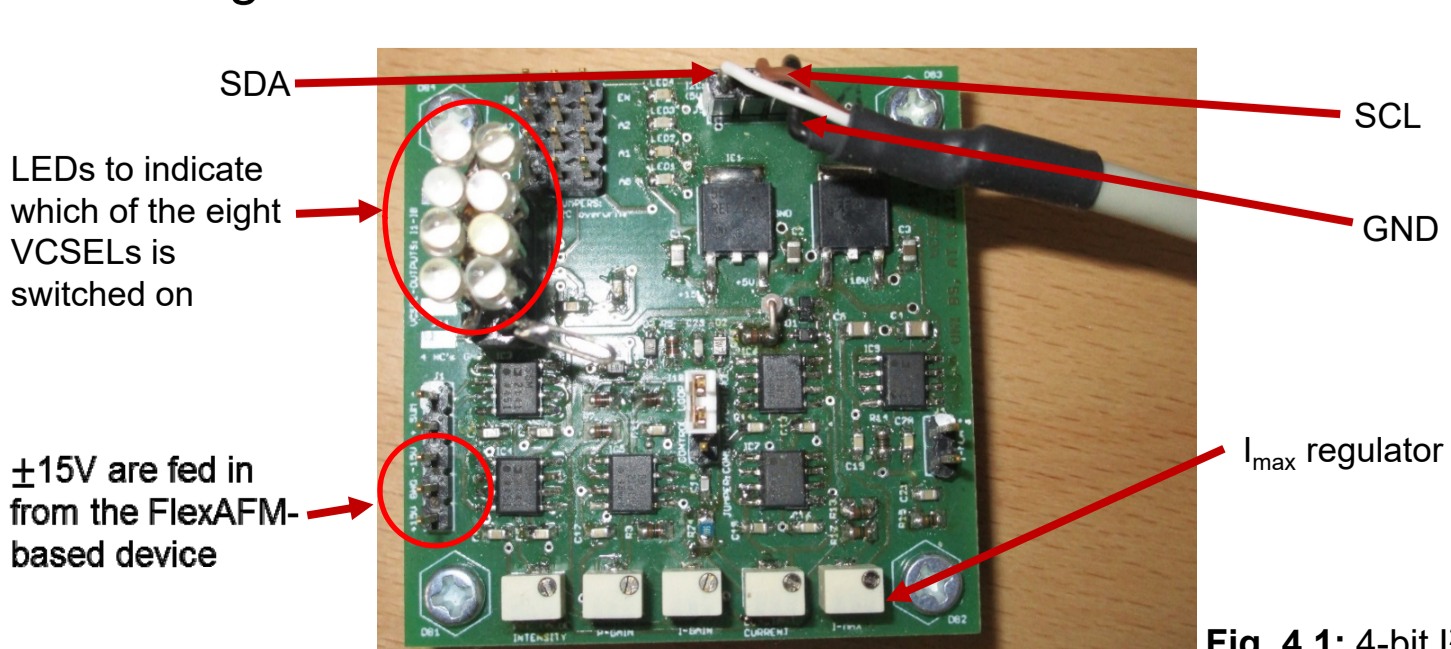


Fig. 4.1: 4-bit I²C-Bus

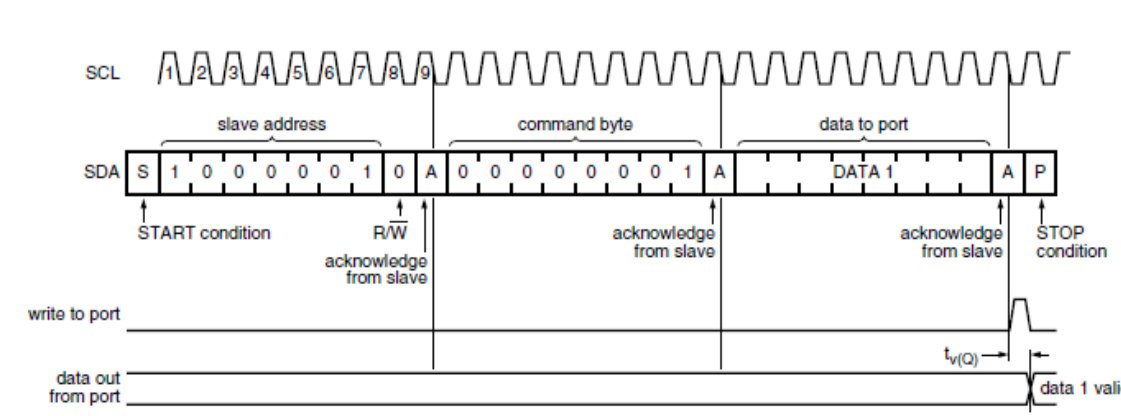


Fig. 4.2: Master sends eight bits and the ninth is the acknowledgment bit (ACK-bit) coming from the slave.

6. Results & Conclusion

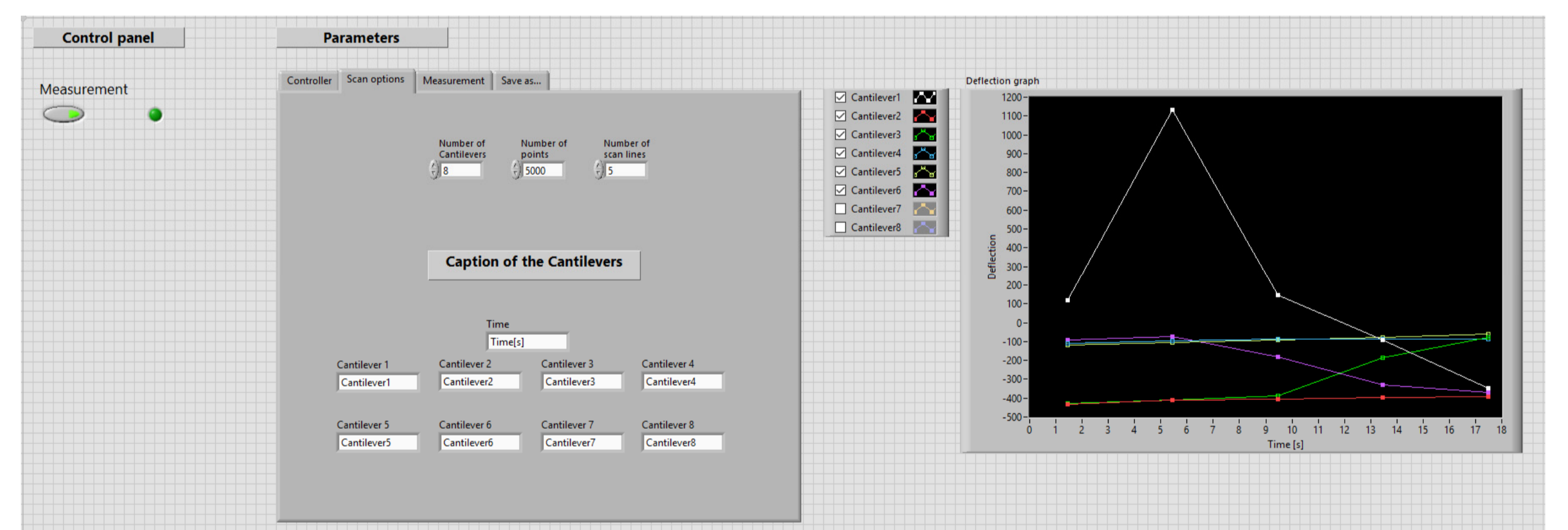


Fig. 6.1: Graphical User Interface. The measurement can be started by pressing the push button in the control panel. To set specific parameters useful for the measurement, we can go to the parameters' panel and there we will find different toolbar pages. Our software allows to label the cantilevers with a description of the coating. In the scan options window we can set the scan lines' number, which corresponds to the number of performed measurements, and the number of scan points measured for each scan line. On the right side we see the shape of the deflection curve. Before or after the measurement we have the possibility to enable or disable specific cantilevers.

Time[s]	Cantilever1	Cantilever2	Cantilever3	Cantilever4	Cantilever5	Cantilever6	Cantilever7	Cantilever8
1.4370	118.5778	-432.8328	-429.2704	-111.9666	-120.1672	-92.3592	-404.0614	32766.0000
5.4560	1134.3418	-410.8750	-409.7362	-96.1884	-103.5748	-76.0134	-390.4356	32766.0000
9.4620	145.8472	-405.6404	-385.7066	-89.0232	-92.2614	-182.3792	14777.7580	18446.9182
13.4710	-91.5824	-398.1114	-188.0844	-89.4624	-76.8058	-327.8572	29484.2924	8319.3882
17.4790	-346.0460	-391.9394	-77.1438	-85.8280	-60.0414	-371.5038	32766.0000	32766.0000

Fig. 6.2: This is the layout of the corresponding txt-file, which is created during the measurement. The file name contains data and time of the measurement.

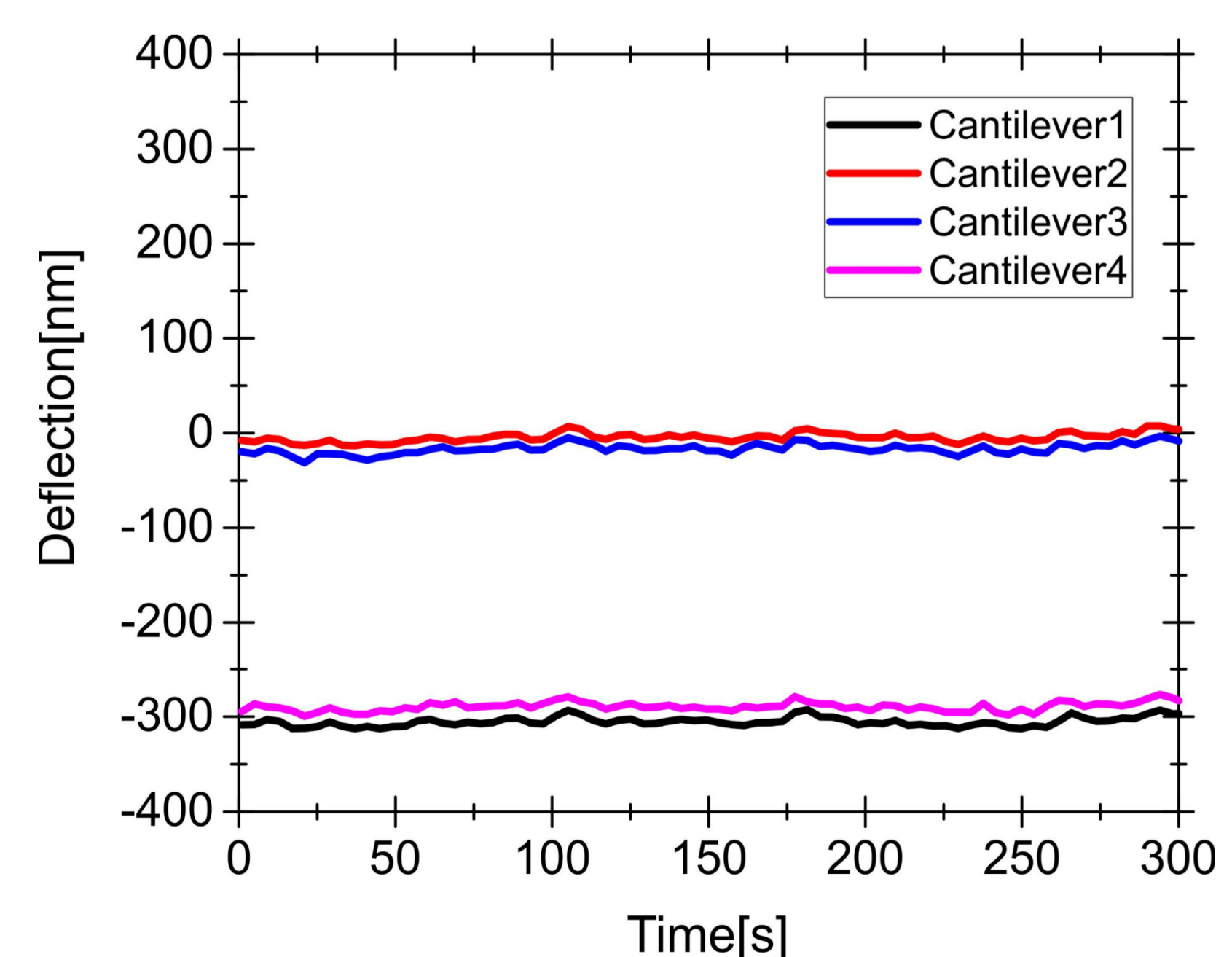


Fig. 6.3: Deflection values of four selected cantilevers. The data have been read out using our software and demonstrate the functionality and stability of the signals.