Tapered optical fibres for local pH detection

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Abstract. This paper deals with preparation of optical probes based on tapered optical fibres and their using for local detection of pH in samples simulating native conditions of plant cells. Optical probes, so-called V-tapers were prepared for this purpose. Fluorescence pH transducer 2′,7′-Bis(2-carboxylethyl)-5(6)-carboxyfluorescein was immobilized onto the end-face of cut V-tapers in a thin xerogel layer. Changes of fluorescence-intensity spectra caused by pH changes were employed for pH measurements. Indium-tin oxide or aluminium coated fibre probes with suitable optical and mechanical properties were successfully tested. It has been confirmed that the selected pH transducer is suitable for pH detection from 5.0 to 7.0. This approach has allowed us to determine extra-cellular pH of in-vitro samples.

1. Introduction
Local detection of intra- and extra-cellular pH has become interesting issue e.g. for botanists, physiologists and also for researchers dealing with optical sensing [1-5]. Tapered optical fibres can be employed as appropriate tools for investigating chemical changes in plant cells, e.g. tobacco leafs with dimensions up to approx. 50x10 µm. We have developed fibre-optic probes based on tapered optical fibres with diameters and properties of the tips that can be used for measurement of intra- or extra-cellular pH of plant cells [6]. These fibre-probes have been intended especially for monitoring of auxins (phytohormones essential for plant growth), which are weak acids, pH 5.0 – 7.0 [7, 8]. The prepared probes with high spatial resolution employ an appropriate optical-chemical transducer for the given pH scale. These probes have suitable optical and mechanical properties making possible the penetration of the tip through surfaces (cell walls and membranes) of plant cells.

2. Tapered optical fibres for pH detection

2.1. Tapered optical fibres
Fibre tapers were manufactured from polymer-clad silica (PCS) fibres (125/200 µm) by using a PC-controlled tapering machine. Tapers in so-called “V-taper” configuration with waist diameters from 60 µm down to 4 µm were prepared. V-tapers were made by fusing two fibre tapers together. The surfaces of the tapers were sputtered with a layer of Indium-tin oxide (ITO) of a thickness ~ 150 nm or aluminium of a thickness ~ 100 nm immediately after the tapering. Optical-chemical pH transducer 2′,7′-Bis(2-carboxylethyl)-5(6)-carboxyfluorescein (BCECF) was immobilized onto the end-face of the taper tip by using a thin xerogel layer prepared from a tetraethoxysilane (TEOS) sol in which the transducer was dissolved [8].
2.2. pH detection

Fluorescence spectroscopy was employed for pH measurements in buffers with a pH range from 5.0 to 7.0 as well as in plant cell cultures simulating native conditions. The BCECF transducer was excited by a laser diode (LD) beam at wavelength of 473 nm (FWHM = 11 nm). Light from LD source was coupled into one PCS pigtail of the V-taper and spectra of the fluorescence intensity were directly collected from second PCS pigtail of V-taper by a spectrometer. Drops of pH buffers (a volume of 1 ml) were used for the calibration of the probes. Maxima of fluorescence responses to pH 5.0 and 6.2 were observed at wavelength of 580 nm. Measured curves of excitation and fluorescence response were completely separated without any overlaps. A shift of fluorescence maximum of about 107 nm from wavelength of excitation maximum was observed. This shift can be attributed to the influence of xerogel matrix encapsulating the transducer. The response to a particular pH value depended on time. This phenomenon can be caused by saturation of the porous xerogel layer.

3. Conclusion

Fluorescence probes based on aluminium or ITO-coated “V-tapers” with immobilized BCECF transducer on the fibre tip have been prepared. The developed probes have suitable optical-chemical and mechanical properties. A novel experimental set-up based on excitation and detection with V-taper probe has successfully been implemented. A shift of fluorescence maximum of 107 nm from wavelength of excitation maximum was observed. This significant shift made possible to detect fluorescence response without optical filtering of the excitation. This approach has allowed us to determine extra-cellular pH of in-vitro samples in a range from pH 5.0 to pH 7.0.

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References
[1] Paciorek T, Zazimalova E, Ruthardt N, Petrasek J, Stierhof Y D, Kleine-Vehn J, Morris D A, Emans N, Jurgens G, Geldner N, Frim J 2005 Nature 435 1251
[2] Rosenzweig Z, Kopelman R 1995 Anal. Chem. 67 2650
[3] Tan W, Shi Z, Kopelmann R 1992 Anal. Chem. 64, 2985
[4] Vo-Dinh T, Kasili P 2005 Anal. Bioanal. Chem. 382 918
[5] Rosenzweig Z, Kopelman R 1996 Anal. Chem. 68 1408
[6] Martan T, Kasik I, Podrazky O, Mrazek J, Pospisilova M, Aubrecht J, Matejec V, Kanka J 2008 Proc. MADICA 6 A53-1
[7] Podrazky O, Mrazek J, Seidl M, Kasik I, Tobiska P, Matejec V, Martan T, Aubrecht J 2007 Proc. SPIE 6585 65850Y.1
[8] Martan T, Podrazky O, Kasik I, Pospisilova M, Matejec V, Kanka J 2008 Proc. Europtrode IX, 276