

## **“CYTOCHECK SPACHIP® pH detection kit” as a new reliable method for measuring intracellular and extracellular pH in cell cultures.**

### **Authors:**

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### **Abstract:**

Therefore, the control of cytosolic pH is pivotal for the cell and the organism itself and, thus, measuring pH changes can be of importance for understanding cell physiology and, on the other hand, for controlling those bio-industrial processes based in cell cultures, as production of antibodies, recombinant proteins, or adeno-viral vectors.

CytoCHECK SPACHIP® pH-single detection technology for simultaneous measurement of cytosolic and extracellular pH in imaging systems and Flow Cytometry (FC). SPACHIPS® are small silicon oxide chips manufactured and functionalized with a fluorescent pH-sensing probe through a combination of surface silanization and microcontact printing techniques.

SPACHIPS® displayed linear response to pH changes between 4.5 and 7.5, with precise reproducibility and coefficient of variation below 10%. When added to cell cultures, an average >25% of the cells internalized a SPACHIP® in the cytosol, where they remain for days without altering cell viability or losing performance. In comparison, probes in solution demonstrated to be toxic in the short or middle term, which impeded to extend the study over time. As a proof-of-concept of our technology in a real FC application, we used CytoCHECK SPACHIP® pH-single detection kit in 293T cells subjected to different uncouplers or inhibitors of oxidative phosphorylation. CytoCHECK SPACHIP® pH-single detection kit detected a significant decrease in cytosolic pH with minor changes in cell medium.

CytoCHECK SPACHIP® pH-single detection technology is a new and reliable tool for performing cell analysis in imaging systems and flow cytometry. Its capability to detect pH changes along time in the cytosol and cell environment simultaneously makes it suitable, not only for basic cell biology and cell physiology studies, but also as quality control in bio-industrial processes based in cell cultures, where controlling extracellular pH affects product yield, as recombinant antibody or AAV production.