Microfluidic IEF technique for sequential phosphorylation analysis of protein kinases NAKCHUL CHOI, SIMON SONG, HOSEOK CHOI, BUTFAEK LIM, YOUNG-PIL KIM, Hanyang Univ. — Sequential phosphorylation of protein kinases play the important role in signal transduction, protein regulation, and metabolism in living cells. The analysis of these phosphorylation cascades will provide new insights into their physiological functions in many biological functions. Unfortunately, the existing methods are limited to analyze the cascade activity. Therefore, we suggest a microfluidic isoelectric focusing technique (μIEF) for the analysis of the cascade activity. Using the technique, we show that the sequential phosphorylation of a peptide by two different kinases can be successfully detected on a microfluidic chip. In addition, the inhibition assay for kinase activity and the analysis on a real sample have also been conducted. The results indicate that μIEF is an excellent means for studies on phosphorylation cascade activity.