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The extraction of peptidome/proteome from *in vitro* to *in vivo*: The *in situ* and timing extraction of cellular peptidome from live cells

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Proteomics reveals the global drafts of proteins in biological systems, with the recent discovery and identification of 92% proteins encoded by human genome. The deeper insights of proteins in living systems call for the complement with dynamic changes in localization, abundance and interactions of proteins. Trials have been made to perform *in situ* and dynamic profiling of proteome/peptidome at subcellular range by the combination of pre-fraction of subcellular components *in-vitro* with isotope labeling at different time points and quantitative proteomics. Other than utilizing *in vitro* protein collection and labeling strategies, proteins obtained from physiological ambient without the chemical derivatizations such as *in situ* extraction with live cells are preferred for proteomic investigation containing high temporal and spatial resolution. To achieve the *in situ* and timing harvest of intracellular proteome and peptidome from a living system, the extraction probes should meet following essential requirements: (1) Biocompatible with live cells, (2) suitable for both general or selective extraction of intracellular proteome/peptidome and compatible to harvest compounds (3) designated intracellular loci and (4) appointed time period. So far, the conventional proteome/peptidome sampling methods including micro extraction, electroporation could hardly satisfy all the above requirements. Nano probes owning high biocompatibility and abilities for subcellular targeting and controllable intracellular transportation, hint promising potentials to extract cellular components from live cells *in situ*. In earlier attempts, Heparin Sulfate Proteoglycans (HSPGs) antibody functionalized magnetic particles were employed to collect targeted endocytotic vesicles for proteomic analysis of HSPGs induced endocytosis, Methotrexate (MTX) linked dendrimers were applied in proteomic discovery of drug substrates from live cells and magnetic carbon nanotubes were used for harvest nucleic acid associated proteins *in situ* coupled with proteomic exploration. However, nanocarriers for dynamic extraction of proteins are not satisfiable yet, since the proteins would be adsorbed by nanocarriers as soon as the cellular internalization. In this paper, a photo-switchable Mesoporous Silica Nanocarrier (MSN with coumarin gates, MSNcg) was constructed as a general nano platform to collect cytosol polypeptides from live HeLa cells for the *in situ* and timing proteomic investigation.

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