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### ACCEPTED MANUSCRIPT

Ratiometric Quantification of β2-microglobulin Antigen in Human Serum Based on Elemental Labeling Strategy

Gongwei Sun<sup>a</sup>, Yuqing Zhang<sup>a</sup>, Yi Zhang<sup>b</sup>, Zhian Hu<sup>a</sup>, Zhi Xing<sup>a</sup>, Sichun Zhang<sup>a,\*</sup>, Xinrong Zhang<sup>a</sup>

<sup>a</sup>Beijing Key Laboratory for Microanalytical Methods and Instrumentation, Department of Chemistry, Tsinghua University, Beijing 100084, China.

<sup>b</sup>Key Laboratory of Nuclear Medicine, Ministry of Health, Jiangsu Key Laboratory of Molecular Nuclear Medicine, Jiangsu Institute of Nuclear Medicine, Wuxi, Jiangsu 214063, China.

\*Corresponding Author: E-mail: sczhang@mail.tsinghua.edu.cn

#### **ABSTRACT**

Ratiometric quantification for competitive immunoassay based on internal standard element detection utilizing inductively coupled plasma mass spectrometry (ICP-MS) as multiplex readout has been demonstrated. The Beta-2-microglobulin ( $\beta$ 2-MG) associated with clinical diseases was detected by Y-labeled capture antibody used as internal standard probes and Sm-labeled antigen used as report probes via antigen-antibody reaction. The ratiometric quantification was achieved by taking the signal ratio of Sm/Y. The ratiometric method could compensate for the particle loss and suppress the signal fluctuation, which improved the precision of the quantitative result. Under the optimized conditions, the calibration curves for  $\beta$ 2-MG antigen was linear in the range of 0.25-8.0 µg/mL with a detection limit of 0.17 µg/mL ( $3\sigma$ , n = 11). The recoveries of 96.5% - 132% were obtained for serum samples spiked with different concentration standards, and the relative standard deviation (RSD) was less than 10%. The  $\beta$ 2-MG results in serum samples obtained by the proposed method correlated well with those obtained by

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