

Quantitative imaging of potential protein biomarkers in oral cancer tissues with LA-ICP-MS using bioconjugated gold nanoclusters

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ICP-MS is being used in life sciences for trace element determination, enhancing the understanding of the role of proteins in biological systems. The analytical capabilities of ICP-MS are extended by the use of laser ablation (LA) for direct probing of solid samples. Two of the most interesting features of LA-ICP-MS are its good sensitivity and lateral resolution, allowing the elemental direct mapping of the sample surface. The combination of immunoassays (based on the specific binding of antibodies with their corresponding antigens) with LA-ICP-MS can facilitate multiparametric analyses through proper elemental tagging, allowing the mapping of proteins in biological tissues.

The use of metal nanoclusters (NCs) as elemental tags will provide signal amplification. As compared to polymeric tags, its ratio “number of metal atoms/size” is very high because they do not contain carbon or other nonmetals, resulting in a potential advantageous strategy to determine antigen distributions. In a first approach, AuNCs has been investigated in our experiments. Metallothionein (MT) distribution is known for healthy and cancerous oral tissues, so it was selected as model to validate the proposed methodology.

- Methods

Our synthesized AuNCs have a particle size of 2.7 nm and 580 atoms per NC [1]. We bioconjugated the AuNCs with an Anti-MT antibody and the immunoassay in the tissue section (5 microns thick) was developed. After the immunoassay protocol, detection and imaging was carried out by LA-ICP-MS and by fluorescence (confocal microscope). Studies were performed for tumor and adjacent control regions from the same patient. Calibration for LA-ICP-MS was performed with spiked gelatin standards.

- Results

Once the immunoassay was optimized, the MT distribution was successfully obtained by measuring the Au signal (observed background signal was negligible); the imaging of other metals in the tissues (like zinc and copper) was simultaneously carried out. The image patterns in control tissues and tumor regions were in agreement with conventional immunohistochemistry. The validation of our quantitative methodology was achieved by results comparison with HPLC-ICP-MS.

- Conclusions

A new and reliable analytical procedure with advantageous features for determination of MT in tissues using bioconjugated AuNCs and LA-ICP-MS has been developed. The methodology can be extended for multiplexing using different metal NCs.

- References

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