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MIP-based sensing system for early detection of bone loss

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This research proposes a novel real-time, label-free sensing technique for the detection of C-telopeptide of type-I collagen (CTx-I) that can be heelpful in early detection of bone loss. A planar interdigital sensor in conjunction with electrochemical impedance spectroscopy (EIS) was used to study the dielectric properties of the test sample. Molecular imprinted polymer (MIP), including artificial recognition sites for CTx-I molecules was synthesized by precipitation polymerization using CTx-I peptide as a template, ethylene glycol methacrylate as the cross-linker and methacrylic acid as a functional monomer. The sensor was coated using the prepared polymer in order to make the sensor selective for CTx-I molecule. Calibration experiments were performed using different known concentration samples and the reference curve was plotted. Complex non-linear least square (CNLS) curve fitting method was used to estimate electrochemical equivalent circuit parameters. The developed system showed a detection limit of 0.1 ng/ml. Two different unknown samples obtained from sheep blood were measured using the developed sensing system and enzyme-linked immunosorbent assay (ELISA) was used to validate the results.

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Production and characterization of functional recombinant hybrid heteropolymers of camel hepcidin and human ferritin H and L chains

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Hepcidin is a liver-synthesized hormone that plays a central role in the regulation of systemic iron homeostasis. To produce a new tool for its functional properties, the cDNA coding for camel hepcidin-25 was cloned at the 5'end of human FTH sequence into the pASK-IBA43plus vector for expression in *Escherichia coli*. The recombinant fusion hepcidin–ferritin-H subunit was isolated as an insoluble iron-containing protein. Alone it did not refold in a 24-mer ferritin molecule, but it did when renatured together with H- or L-ferritin chains. We obtained stable ferritin shells exposing about four hepcidin peptides per 24-mer shell. The molecules were then reduced and re-oxidized in a controlled manner to allow the formation of the proper hepcidin disulfide bridges. The functionality of the exposed hepcidin was confirmed by its ability to specifically bind the mouse macrophage cell line J774 that express ferroportin and to promote ferroportin degradation. This chimeric protein may be useful for studying the hepcidin–ferroportin interaction in cells and also as drug-delivery agent.

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