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Simultaneous monitoring of multiple human CVD protein biomarkers in circulation by quantitative multiplex immunoassay

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Pardiovascular diseases (CVD) are one of the major causes of death worldwide. Researchers of CVD are increasingly required to ✓ provide highly validated reproducible data which will ultimately translate to the clinic. CVD includes a wide range of pathological phenomena, multiplexed biomarkers analysis therefore is crucial. MILLIPLEX® Human CVD Magnetic Bead Panels 1-6 present 62 significant protein biomarkers that cover all aspects of CVD biology. Newly developed panels 5 and 6 simultaneously measure 14 and 7 biomarkers respectively with superior sensitivity (MinDC<50 pg/mL for 17 analytes), specificity (no detectible cross reactivity) and reproducibility (<10% interCV). Here, we present data using panels 5 and 6. Human serum/plasma samples of patients with acute ischemic heart attack (n=10), stable stage heart failure (n=18) and control serum/plasma from patients with no known CVD risk (n=28) were assayed. Under acute ischemic heart attack, the proteins ADM, CHGA, HSP60, IGF1R, LDLR, LEPR, PTGDS, sST2 and SYND4 showed the most notable elevations over control values. Particularly, IGF1R levels increased over 30-fold and sST2 levels increased ~20 fold. In samples of stable stage heart failure, CHGA, HSP60, IGF1R, LEPR and PTGDS level increased moderately compared to controls. IGF1R showed the greatest increase of ~8 fold. Interestingly, CHGA and PLA2G7 levels were much lower in stable stage samples compared with acute stage samples. Elevations of CD14, Endostatin, LRG1, GRN precursor, and MCAM were observed in both acute ischemic and chronic heart failure samples, while DPP4 and ucMGP levels decreased in both sets of samples. Monitoring of these novel biomarkers provides researchers with a more comprehensive view of CVD pathogenesis, particularly for coronary artery calcification, cardiac hypertrophy, myocardial fibrosis and endothelial inflammation, etc. The assay presented in this study is easy to use, robust, sensitive, and amenable to high throughput and are thus well suited for CVD risk factor discovery towards clinical relevance.

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