

78 - Analysis of a serum biomarker of exposure to chemical toxicants

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An analytical approach utilizing a sandwich enzyme linked immunosorbent assay (ELISA) was developed to study differences in Clara cell protein (CC16) levels in human serum samples. The National Institute for Occupational Safety and Health is interested in the identification of biomarkers that are related to hazardous occupational chemical exposure. CC16, a secreted protein of human bronchiolar cells, has been observed to decrease in the serum of workers following smoking or silica exposure. This investigation studies differences in CC16 levels in smokers versus non-smokers. Following serum separation from whole blood, standards and serum samples are incubated in microplate wells pre-coated with polyclonal anti-human CC16 antibody. Biotin labeled polyclonal Clara cell antibody is added and incubated with captured CC16. After washing, streptavidin-horseradish peroxidase (HRP) conjugate is added. The remaining conjugate is allowed to react with the substrate solution tetramethylbenzidine (TMB). The absorbance of the resulting yellow product is proportional to the concentration of Clara cell protein. A standard curve is constructed to determine the concentration of CC16 in serum samples. The results are reported as the concentration in ng/ml of CC16. Using this approach the concentration of CC16 in a non-smoker was determined as 12.1 ng/ml. Further analyses of Clara cell protein level in smokers versus non-smokers are compared. The procedure described here can be used to quantitate the level of CC16 biomarker in blood serum samples. This approach is potentially useful for occupational risk assessment for silica exposure, a known chemical hazard in the hydraulic fracturing industry.

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