

## Detection of Heroin and Its Metabolites in Urine Samples: A Chemiluminescence Approach

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**Abstract :** A sensitive chemiluminescence immunoassay (CIA) for heroin and its major metabolites is reported. The method is based on the competitive reaction of horseradish peroxidase (HRP)-labeled anti-MAM antibody and free drug in spiked urine samples. A hapten-protein conjugate was synthesized by using acidic derivative of monoacetyl morphine (MAM) coupled to carrier protein BSA and was used as an immunogen for the generation of anti-MAM (monoacetyl morphine) antibody. A high titer of antibody (1:64,000) was obtained and the relative affinity constant ( $K_{aff}$ ) of antibody was  $3.1 \times 10^7$  l/mol. Under the optimal conditions, linear range and reactivity for heroin, mono acetyl morphine (MAM), morphine and codeine were 0.08, 0.09, 0.095 and 0.092 ng/mL respectively. The developed chemiluminescence inhibition assay could detect heroin and its metabolites in standard and urine samples up to 0.01 ng/ml.

**Keywords :** heroin, metabolites, chemiluminescence immunoassay, horse radish peroxidase

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