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Determination of the Feasibility of Automating 25% Sulfosalicylic Acid (SSA) Method for the Quantitation of Urine Protein in Mindray BS 200 Analyzer

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Introduction - Proteinuria is an important diagnostic marker for the early detection of kidney disease. The 25% Sulfosalicylic acid (SSA) manual turbidimetric assay is a nationally patented cost-effective method to quantitate urine protein in the lower range. The present study was conducted to investigate the feasibility of automation of the 25% SSA method to widen its application as a detection tool for proteinuria.

Materials and Methods - Preliminary volume-optimization experiments were performed using a manual spectrophotometer, and the 25% SSA method was subsequently automated on the Mindray BS-200 analyzer. Sample and reagent volumes, primary wavelength, reaction and incubation times for the BS-200 were optimized through several trials. A calibration curve was generated using optimized parameters and the analytical range of the automated method was determined. The method was validated against the gold standard Pyrogallol Red (PGR) method. Difference of median between the 25% SSA and PGR method were analyzed using the Wilcoxon signed rank analysis and the degree of association was analyzed using Spearman's correlation.

Results - The calibration curve at 405 nm showed a polynomial association between absorbance and concentration. The analytical range of the automated method was identified as 20 – 75 mg/dL. Spearman correlation showed a good correlation between the two methods ($p<0.001$, rho=0.933 for N=9). Wilcoxon signed rank analysis showed no significant difference between medians obtained by the two methods ($Z=-0.889$, $P>0.05$). However, poorly controlled sedimentation, which increased with concentration, influenced the readings.

Discussion - It was observed that there is a strong, positive correlation between 25% SSA and PGR methods, with a higher degree of precision of the SSA test. Effects of sedimentation during the assay need to be controlled for further optimization.