Bioanalytical determination of vancomycin and ceftriaxone concentration in serum and tissue fluid by HPLC in patients and its application to musculoskeletal infections

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Infection is a significant clinical problem in orthopaedic surgeries and chronic infection can persist intermittently for years. Surgical debridement is the treatment of choice but systemic antibiotics are ineffective in achieving significant concentrations at the site of infection because of local avascularity. Local antibiotic delivery from polymethyl methacrylate (PMMA) cement beads is the current gold standard method of local drug delivery system against which other local drug delivery modalities are compared.

Hence a prospective study was conducted to estimate the concentration of antibiotic in the serum and seroma fluid leached from the antibiotics (Vancomycin and Ceftriaxone) impregnated PMMA beads and the amount of antibiotics absorbed into the systemic circulation estimated by its serum concentration by high performance liquid chromatography (HPLC).

The HPLC method used with C18 (250 × 4.6mm, 5μm) column and analytical guard column 12.5x4.6 mm (5μm), with a gradient elution (1 mL/min) at 40°C column temperature. A mobile phase consisting of a binary mixture of methanol and phosphate buffer adjusted to pH 4 with orthophosphoric acid in a ratio of 30:70. The eluent was monitored at 280 nm. The calibration curve was linear within the range of 2-50 μg/mL ($r^2=0.99$, n=6) for both the drugs. The lowest limit of quantification (LLOQ) was 100 ng/mL for vancomycin and 120 ng/mL for ceftrixone. The retention times of Vancomycin and Ceftriaxone were 2.3 and 3.1 min respectively. Greater than 85% recoveries were obtained for Vancomycin and Ceftriaxone.

The intra and interday relative standard deviation (%RSD) were <5%.

Venous blood will be drawn by venepuncture of upper limb veins and and local seroma fluid from the site of LADS implantation will be aspirated from the previously inserted Ryle's tube at the intervals of 24 hours, 48 hours, 72 hours, 5th day, 7th day and 10th day and marked separately. The collected serum and seroma fluid were labeled "S" and "L" for identification of venous blood and local seroma fluid. Venous blood will be centrifuged at 3500 rpm (rotations per minute) for 5 minutes (Remi centrifugier, India). Both the serum and seroma fluids will be stored at -20 degree centigrade from the time of collection.

Biography
B.M. Gurupadayya has completed his PhD at the age of 34 years from Kuvempu University, Shimoga and post graduation (M.Pharm) from SCS College of Pharmacy, Harapanahalli, Gulbarga University. He is the professor of Pharmaceutical Analysis at JSS College of Pharmacy, Mysore, Karnataka. He has published more than 40 papers in reputed journals.

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