Preparation of stock solutions and working solutions

2.2.2.2 Preparation of stock solutions

Preparation of stock solutions of propranolol

PRN (10 mg) was dissolved in 50 ml methanol to obtain a final concentration of 200 µg/ml stock solution of PRN.

Preparation of stock solutions of sildenafil IS

Sildenafil (10 mg) was dissolved in 10 ml acetonitrile to obtain a final concentration of 1000 µg/ml stock solution of sildenafil.

2.2.2.3 Preparation of working solutions

Preparation of working solution of sildenafil (IS)

Sildenafil (250 µl) from stock solution (1000 µg/ml) was diluted to 50 ml of acetonitrile to obtain 5 µg/ml of sildenafil working solution.

Preparation of propranolol serial spiking samples and quality control (QC) samples in serum and buffer.

Calibration curve and QC samples were prepared by taking different volumes (µl) from PRN stock solution (200 µg/ml) as shown in **tables 2.1 and 2.2** to reach 1 ml final volume. Consequently, concentrations of working solutions (µg/ml) were obtained to be used later for serum and Krebs buffer spiking solutions which were prepared by taking 25 µl of each working solution to be spiked in 975 µl of serum or Krebs buffer (final volume 1 ml). Spiked serum and Krebs buffer of PRN serial samples and QC samples were prepared as shown in **table 2.1 and 2.2** respectively.