

cimetidine kindly donated by Jordan Sweden Medical and Sterilization Company (JOSWE, Naur, Jordan).

2.2 Methodology

2.2.1 Chromatographic conditions

The HPLC system FINNIGAN SURVEYOR (Thermo Electron Corporation, San Jose, CA, USA) was used. The detector (UV-VIS Plus Detector), the pump (solvent delivery systems pump) (LC Pump) and the autosampler (Autosampler Plus). Computer system used was Windows XP and the software used was ChromQuest software 4.2.34. HPLC system was set at a wavelength of 214 nm and coupled with a hypersil™ BDS C-18 Column (Thermo Electron Corporation, San Jose, CA, USA); (150 mm x 4.6 mm, 5µm) with a flow rate of 1 ml/min using a 15 µl injection volume and 7 ml/min run time. Sildenafil citrate was used as an internal standard (IS) in this method.

2.2.2 HPLC analysis procedure

2.2.2.1 Preparation of mobile phase

Mobile phase used at a ratio of 15% acetonitrile HPLC gradient, 32.5% methanol HPLC gradient and 52.5% water. Water was prepared by the addition of triethylamine where 1 L of water contains 900 µl of triethylamine then pH was adjusted to 2.75 using phosphoric acid. Water 525 ml was mixed with 150 ml of acetonitrile and 325 ml of methanol to obtain a final volume of 1000 ml.