The colours obtained by application of different reagents at different wavelengths of radiation were recorded (Ali 2003).

14.2.3 Preparation of Plant Extracts

The powdered leaves and stem materials were packed in small thimbles and separately extracted with organic solvents, such as petroleum ether, ethyl acetate and methanol in the increasing order of their polarity using a Soxhlet apparatus. Before extraction with the next solvent, the thimbles were air-dried each time. Finally, the material was macerated using hot water with constant stirring for 24 h and the water extract was also filtered using Whatman No. 1 filter paper. The different solvent extracts were concentrated by rotary vacuum evaporator and then air-dried.

14.2.4 Qualitative Phytochemical Screening

The leaf and stem extracts of *P. hexapetalum* were analyzed for the presence of major phytochemicals such as alkaloids, saponins, phenolic compounds, tannins, flavonoids, glycosides, flavanol glycosides, cardiac glycosides, phytosterols, fixed oils, fats, gums and mucilages according to standard methods.

14.2.4.1 Alkaloids

Hager's test

About 50 mg of solvent-free extract was stirred with 5 mL of dilute hydrochloric acid and filtered, and 2 mL of Hager's reagent (the saturated aqueous solution of picric acid) was added to the filtrate. A prominent yellow precipitate indicated the test as positive (Wagner et al. 1996).

14.2.4.2 Saponins

Frothing test

An extract of 50 mg was diluted with distilled water and made up to 20 mL. The suspension was shaken in a graduated cylinder for 15 min and a 2 cm layer of foam indicated the presence of saponins (Kokate 1999).

14.2.4.3 Phenolic Compounds

Ferric chloride test

About 50 mg of the extract was dissolved in 5 mL of distilled water. To this, a few drops of 5% of a neutral ferric chloride solution was added. Phenolic compounds were indicated by the presence of a dark green colour (Mace 1963).

14.2.4.4 Tannins

Potassium hydroxide test

The extract (0.5 g) was added into 10 mL of freshly prepared 10% potassium hydroxide (KOH) in a beaker and shaken to dissolve. A dirty precipitate indicated the presence of tannin (Williamson et al. 1996).