Chapter 6

Summary and conclusions

Urinary tract and kidney stones ailments have affected human beings since antiquity. Ancient Vedic literature describes stones as Ashmari. Stones are solid concretions or calculi (crystal aggregations) formed in the kidneys from dissolved urinary minerals when urine becomes supersaturated. The occurrence of these stones has been increasing in rural and urban societies [131]. A large population of India suffers from urinary tract and kidney stones, formed due to the deposition of calcium, phosphate and oxalate. The chemicals start accumulating over the nucleus, which ultimately takes the shape of a stone.

Kidney stones modify the victim’s behavior with great fear of intense pain and threat of kidney failure. In modern medicine no satisfactory effective therapy is still available to dissolve or to prevent stones. Acetazolamide are available against uric acid and cystine stone, although several drugs / therapies such as thiazides, cellulose phosphate, magnesium oxide and pyridoxine etc. have been tried. On the other hand, traditional system of Indian medicine “Ayurveda” recommends several medicinal plants for the treatment of Urolithiasis.

Urinary stones contain both crystalloid and colloid components. The crys-
talloid components are mainly calcium oxalate, calcium phosphate, calcium carbonate, magnesium-ammonium phosphate, uric acid and cysteine. Since ancient times, a variety of herbal preparations have been successfully used in renal lithiasis therapy [132, 133, 134]. During recent decades, studies of the antilithiatic effects of herbal extracts have been reported, but in the majority of these reports, the effects did not seem to be mediated by urinary biochemical changes [45, 135]. In the present study antilithiatic properties of two plants, *Tamarindus indica* and *Terminalia arjuna* have been assessed using *in vitro* and *in vivo* experimental models.

1. Effect of both the plants was first studied on calcium phosphate mineralization and on growth of calcium oxalate crystals *in vitro* by using different assay systems. It was found that both *Tamarindus indica* and *Terminalia arjuna* are effective not only in inhibiting initial mineral phase formation, growth and demineralization of the preformed mineral phase of CaP, but also in inhibiting CaOx crystal growth *in vitro*. Crude aqueous extract of *Tamarindus indica* and *Terminalia arjuna* were then subjected to ultrafiltration and two fractions having molecular weight less than 10 kDa and greater than 10 kDa were obtained. Greater than 10 kDa fraction showed greater inhibition and thus protein purification was carried out with greater than 10 kDa fraction.

2. *In vivo* efficacy of *Tamarindus indica* was tested by using aqueous extract on experimentally induced hyperoxaluric rat models. Creatinine clearance, urinary injury marker enzymes and content of serum urea and creatinine levels were restored in animals treated with 5% and 10% aqueous extract of *Tamarindus indica* separately. Histopathological studies further showed marked decrease in crystallization when kidney tissue was seen under light microscope. Consumption of *Tamarindus indica* might be the reason of
low occurrence rate of kidney stones in the areas where its consumption is high.

3. Protein purification of *Tamarindus indica* done by anion exchange chromatography and molecular sieve chromatography carried out with greater than 10 kDa fraction revealed that it has an anionic protein (MW \(\sim 97\) kDa) which is an effective inhibitor of calcium phosphate and calcium oxalate crystals.

4. *In vivo* efficacy of *Terminalia arjuna* was checked by treating hyperoxic-aluric rats with saponin rich n-butanol fraction obtained after bioactivity guided successive solvent extraction from the bark of *Terminalia arjuna*. Parameters studied on serum and urine revealed that saponin rich n-butanol fraction of *Terminalia arjuna*, reduced oxalate mediated manifestations in rat kidney. Decrease mineral deposits were observed in kidney tissue when seen under light microscope. In addition to that, less haemolysis and protein casts were seen in rats treated with saponin rich n-butanol fraction. Morphology was also restored in rats treated with saponin rich n-butanol fraction of *Terminalia arjuna*.

5. Protein purification of greater than 10 kDa fraction of crude aqueous extract of *Terminalia arjuna* was done and an anionic inhibitory protein (MW \(\sim 14\) kDa) was isolated which is capable of inhibiting kidney stone formation.

Hence, both *Tamarindus indica* and *Terminalia arjuna* were found to be efficient in the management of kidney stones not only *in vitro* but *in vivo* too.