Abstract

1. ABSTRACT

The Word “chronic kidney disease (CDK)” is define as to include conditions that affect the kidney, with the potential to cause either progressive loss of kidney function or complications resulting from decreased kidney function. Chronic kidney disease was thus defined as the presence of kidney damage or decreased level of kidney function for three months or more, irrespective of diagnosis. Worldwide, increasing numbers of patients are affected by CKD. Metabolic factors have been implicated in the progression of CKD. The progression of established CKD is variable and depends on several risk factors or markers like urolithiasis and nephrotoxicity. The poor diabetes control accelerates the progression of diabetic nephropathy in both type 1 and type 2 diabetes. Obesity or hyperlipidemia is associated with insulin resistance and compensatory hyperinsulinemia, metabolic derangements that may lead to the formation of calcium containing kidney stones. Experimental evidence has also shown a link between hyperlipidemia and the progression of diabetic and non-diabetic nephropathies.

*Hordeum vulgare* Linn. seeds is belonging to family Gramineae or Poaceae. It is locally known as ‘Barley’ or ‘jav’ is distributed throughout India. It is an annual climber with reported to be high medicinal activities. Locally in India, the seeds of *Hordeum vulgare* are being used for the different medicinal purpose. As per the indigenous system of Ayurveda medicine and the Ayurveda literature survey indicated the use of seeds of *Hordeum vulgare* Linn. have been reported to be useful in the treatment of a wide range of kidney disease, including urinary stones, diabetes, hyperlipidemic, carminative and expectorant as well as other complications of kidney. In the light of above facts, the present investigations were examined the use of *Hordeum vulgare* Linn. seeds within the context of the modern scientific framework with the following aim:-

1. Pharmacognostical and phytochemical standardization of Extract of *Hordeum vulgare* seeds.
2. To study the effect Ethanolic extract of *Hordeum vulgare* seeds against kidney stone by different models of urolithiasis.
3. To study the effect of Ethanolic extract of *Hordeum vulgare* seeds on Gentamicin induced nephrotoxicity.
4. To study the effect of Ethanolic extract of *Hordeum vulgare* seeds in STZ-diabetic rats with special reference to diabetic nephropathy.

5. To study the effect of Ethanolic extract of *Hordeum vulgare* seeds on fructose induced hyperlipidemia in rats.

6. To study *in-vitro* antioxidant activity of Ethanolic extract of *Hordeum vulgare* seeds.

The air-dried powdered seeds of *Hordeum vulgare* (2 kg) were extracted with ethanol in soxhlet apparatus for 24 h. The extract was evaporated to dryness under reduced pressure to give solid residues. The residue was stored below 4 °C for subsequent experiments. The yield of the extract was 5.20% w/w. the extract was subjected to preliminary phytochemical analysis for the detection of various class of compounds like alkaloids, triterpinoles, anthraquinones, flavonoids, steroids, amino acids, tannins, and Coumarins using specific reagents. The phytochemical studies revealed that the Ethanolic extract of *H. vulgare* seeds contained alkaloids, flavonoids, saponins, carbohydrates, amino acids, tannins and phenolic compounds.

In present study we observed the effects of Ethanolic extract of *H. vulgare* seeds on risk factors associated with chronic kidney disease (CDK). The ethylene glycol (EG), Glycolic acid (GA) and surgically induced calcium oxalate crystals was the potent urolithiatic agent which induced urolithiasis in animals. The EG was given in the dose of 0.75% w/v in drinking water for 28 days in both preventive and reversal study for induction of urolithiasis. The GA was fed with commercial diet mixed with 3% glycolic acid for 42 days. While, the Calcium oxalate seed (experimentally induced approximately 3 mm diameter in bladder) were inserted in to bladder of animals by the surgical intervention. In GA induced urolithiasis, the test drugs can administered for 28 days and 14 days in preventive study and reversal study respectively. While in GA induced urolithiatic model the drug were given for 42 days. The Due to these urolithiatic agents, there were significantly decreased body weights, water intake, and urine output in urolithiasis induced animals. The Cystone (Himalaya Herbal Healthcare, Bangalore) purchased from the local market was taken as the standard drug. The treatment with ethanolic extract of *H. vulgare* seeds and standard drug Cystone reversed such changes of the body weight, water intake and urine out put in
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urolithiatic animals. There was significant effect of pH was observed i.e. shifted pH to alkaline in treatment group of animals.

Because of the urolithiasis, the urolithiatic control rats significantly increased levels of urolithiatic promoters like calcium, oxalate, inorganic phosphate and uric acid. While there was a decrease the concentration of urolithiatic inhibitors like magnesium and citrate was observed in various biological samples like urine, serum and kidney homogenate. Due to the urolithiasis promoters there was crystal deposition was reflected by the presence of the crystals in the urine which indicate the alteration of kidney function. Damage to the kidney was confirmed from the results of serum creatinine, BUN, uric acid, and creatinin levels. Due to the presence of stone in kidney, the oxidative stress, this was stated by increased malondialdehyde level and decreased superoxide dismutase and catalase levels. The morphological changes, damage to kidney and accumulation of stone were confirmed by histopathological examination. The treatment with Ethanolic extract of *H. vulgare* seeds, significantly improve these alteration in preventive as well as reversal study.

Deposition of calcium oxalate crystals in urinary bladder by surgical method in rats showed increase salt deposition, crystal aggregation within fourteen days. At the end of experimental period, it was confirmed by x-ray analysis. Crystal aggregation also altered physical and biochemical parameters similarly, as observed in ethylene glycol and glycolic acid induced urolithiasis model. Ethanolic extract of *H. vulgare* seeds as well as standard drug cystone treatment significantly prevented alteration in these parameters.

The Gentamicin is an antibiotic whose clinical use is limited by its nephrotoxicity. Experimental evidence suggested a role of reactive oxygen species in Gentamicin induced nephrotoxicity. The renal damage was induced by Gentamicin at a dose of 100 mg/kg (i.p) for 10 days. The animals were treated with the Ethanolic extract of *H. vulgare* seeds at the different doses consecutively for 10 days. In Gentamicin induced nephrotoxicity, we observed the rise in serum markers like urea, BUN and creatinine and decrease in level of protein in control animals. The extract of *Hordeum vulgare* seeds signicantly reduced the toxicant, elevated levels of above mentioned serum markers and increased in the levels of protein. In gentamycine treated animals, the
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Histopathological investigations revealed the congestion of glomerular, peritubular and blood vessels associated with presence of inflammatory cells in kidney section. The treatment with *Hordeum vulgare* seeds was found to protect the renal micro architecture against Gentamicin induced renal damage. The renal oxidative stress was determined by enzymatic activity of malondialdehyde, superoxide dismutase, reduced glutathione and catalase. The Gentamicin caused significant increased malondialdehyde levels and significantly decreased in superoxide dismutase, reduced glutathione and catalase levels. The Ethanolic extract of *H. vulgare* significantly and dose dependently reduced the malondialdehyde and enhanced superoxide dismutase, reduced glutathione and catalase activities. That indicate that, Ethanolic extract of *H. vulgare* seeds was also induced the protective effect on antioxidant measurement in Gentamicin induced nephrotoxicity.

STZ produced cardinal sign of diabetes mellitus like a significant loss of body weight, polyurea and polydypsia in type I diabetic rats. The diabetes nephropathy was induced by administration of STZ in type I diabetes mellitus and, by STZ and nicotinamide in type II diabetes mellitus. Significant reduction in blood glucose levels were observed in diabetic animals treated with *H. vulgare* seeds. Serum insulin levels were not altered significantly in animals treated with Ethanolic extract of *H. vulgare* seeds. STZ-diabetic rats produced a significant increase urine protein, urea and creatinine level. The Ethanolic extract of *H. vulgare* seeds significantly decreased the levels of urine protein, urea and creatinine dose dependently. The diabetic rats produced significant increased levels of serum triglyceride, cholesterol, LDL-Cholesterol, VLDL-cholesterol and decreased level of HDL-cholesterol. While with the treatment of Ethanolic extract of *H. vulgare* seeds significantly decreased the level of serum triglyceride, cholesterol, LDL-cholesterol, VLDL-cholesterol while, significantly increased the level of HDL-cholesterol. The *H. vulgare* seeds produced significant rise in antioxidant enzymes level in diabetic rats. The Ethanolic extract of *H. vulgare* seeds was showed same effect in STZ-nicotinamide induced type II diabetes. But these effects were less significant as compared to type I diabetes. In both of the model of diabetes there was significant effect was observed on antioxidant enzyme levels.
In fructose induced hyperlipidemia, the animals treated with fructose (10%) in drinking water produced cardinal sign of hyperinsulinemia with increased blood glucose level in animals. The fructose supplementation showed significant increase the level of blood glucose and insulin level in serum. There were significant increase in cholesterol, triglyceride, VLDL-cholesterol and LDL-cholesterol while decreased in HDL-cholesterol as compared to control animals. The Ethanolic extract of *H. vulgare* seeds showed significant effect on decreasing blood glucose level and the insulin level did not affect significantly. Treatment with Ethanolic extract of *H. vulgare* seeds showed significant decrease in cholesterol, triglyceride, VLDL-cholesterol, LDL-cholesterol level and significant increase in HDL-cholesterol levels in a dose dependent manner. The potent antioxidant enzyme activities were observed with the extract treatment.

In conclusion, our data suggests that Ethanolic extract of *H. vulgare* seeds possesses antiurolithiatic activity. The extract also possesses antidiabetic activity and it specifically prevents STZ-induced dyslipidemia and decreased renal functions. The renoprotective potential was further confirmed in Gentamicin induced nephrotoxicity model of rats. Similarly, antihyperlipidemic activity was further confirmed by administration of fructose in drinking water to rats. Antioxidant activity might be responsible for antiurolithiatic, antidiabetic and renoprotective, and antihyperlipidemic activity by *Hordeum vulgare* seeds.
2. INTRODUCTION

The incidence and prevalence of chronic kidney disease (CKD) has increased exponentially in recent years in both developed and developing nations and is consuming a huge proportion of health care finances in developed countries, while contributing significantly to morbidity, mortality and decreased life expectancy in developing ones. This has necessitated renewed interest in global CKD prevention because it is now regarded as a public health threat (Arogundade, 2008). The Work Group defined “chronic kidney disease” to include conditions that affect the kidney, with the potential to cause either progressive loss of kidney function or complications resulting from decreased kidney function. Chronic kidney disease was thus defined as the presence of kidney damage or decreased level of kidney function for three months or more, irrespective of diagnosis (Levey et al., 2005).

The two principal outcomes of CDK are the progressive loss of kidney function over time, and development and progression of cardio vascular disease (CVD). Figure 2.1 shows a conceptual model of the course of chronic kidney disease, which defines stages of CDK, as well as antecedent conditions, outcomes, risk factors for adverse outcomes, and action to improve outcomes. This representation of the course of CDK provides a framework previously lacking for the development of a public health approach to CDK.