



## STUDY OF ANTIHYPERLIPIDEMIC EFFECT ON THE JUICE OF THE FRESH FRUITS OF *LAGENARIA SICERARIA*

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Received: 22 Sep 2010, Revised and Accepted: 24 Oct 2010

### ABSTRACT

The effect of juice of the fresh fruits of *Lagenaria siceraria* on the blood cholesterol level of atherogenic diet rats was evaluated. Twelve adult male Wistar rats divided into 3 groups of 4 animals each were used and were treated orally with normal saline (Control group), atherogenic diet (Positive control) and 200mg/kg of AP seed juice extract (treatment) over a 4-week period. The present study was undertaken to assess body weight, total cholesterol (TC), triglyceride (TG), low density lipoprotein (LDL), high density lipoprotein (HDL) and very low density lipoprotein (VLDL). They were significantly lower in the juice extract treated groups compared to the control group. Phytochemical screening showed that the juice extract contained cardiac glycosides, tannins, saponins, alkaloids and flavonoids. The study showed that juice of the fresh fruits of *Lagenaria siceraria* have the potential to cause a blood cholesterol lowering effect. The serum biochemistry changes may suggest that the juice extract has a tonic effect on the kidneys and the liver and these organs play central role in drug metabolism. Absence of significant lesion in the kidney, liver and testes may indicate that the plant is safe for medicinal use.

**Keywords:** *Lagenaria siceraria*, Atherogenic diet.

### INTRODUCTION

Hyperlipidemia is a condition which characterized by abnormal elevation of lipid such as (triglyceride and cholesterol) and lipoproteins such as (LDL, VLDL) levels in the blood<sup>1</sup>. Increased blood pressure is a common disorder which results in a greatly increased probability of coronary thrombosis, strokes and renal failure<sup>2</sup> which is than supported by an abundance of congruent result from genetic, epidemiological, experimental animal studies and clinical trials that the presence of high plasma lipid cholesterol increases the incidence of coronary heart diseases<sup>3,4</sup>. *Lagenaria* is a genus of gourd bearing vines from the family *Cucurbitaceae*, also known as the "Squash" family. It contains at least seven species, one of which is known as the Calabash (*Lagenaria siceraria*). Its species fruit can either be harvested young and used as a vegetable or harvested mature, dried, and used as a bottle or utensil<sup>5</sup>. Ethnomedically, it has been found to be effective in the patients who suffered from blood pressure<sup>6</sup>. The juice of fresh fruits of *Lagenaria siceraria* was evaluated for pharmacological effects in animal models. The study demonstrated the blood cholesterol lowering effect of *Lagenaria siceraria*. Studies have reported that presence of cardiac glycosides in *Lagenaria siceraria*.<sup>7</sup> Hence assess the phytochemical constituents of this plant, evaluated the effect on the lowering the blood cholesterol level.

### EXPERIMENTAL METHODS

#### Collection and authentication of plant material

The fruit was obtained from the vegetable market of dehradun. They were authenticated at the Taxonomist of Department of Botany, Forest Research Institute, Dehardun, where the voucher specimen was deposited at the herbarium.

#### Sample preparation

The fresh fruit was taken and was peeled off. It was than cut into the pieces and pressed into juicer to obtained fresh juice. The juice was kept under low temperature in refrigerator only for 24 hrs. It was than discarded due to bitter taste which means presence of toxicity.

#### Phytochemical screening

Standard Phytochemical methods were used to test for the presence of saponins, alkaloids, phyosterols, tannins, anthraquinones, cardiac glycosides, cyanogenetic glycosides and flavonoids<sup>8, 9, 10, 11, 12</sup>

### Animal study

The animals used in this study were 12 male Wistar rats weighing between 200-250gms. They were maintained in the laboratory, School of Life Sciences, University of Hertfordshire, England. They were kept in rat cages and fed commercial rat feeds and allowed free access to clean fresh water in bottles *ad libitum*. The twelve animals were divided into 3 groups of 4 animals per group. While group A rats served as normal control, groups B served as positive control and group C animal was administered the fresh juice 5ml once daily and distilled water was administered orally to the animals once daily for 28 days using stomach cannula.

#### Technique for obtaining serum samples

Blood samples were collected by cervical decapitation from diethyl ether anaesthetized rats into clean non-heparinized bottles and allowed to clot. The serum was separated from the clot and centrifuged according to groups into clean bottles for biochemical analysis.

#### Determination of serum biochemical parameters

Serum total protein was measured using Lowry *et al.*, method<sup>13</sup>, total cholesterol and triglyceride was estimated by the method of CHOD-PAP and high density lipoprotein by the method of GPO-PAP. Low density and very low density cholesterol were calculated by using Friedwald formula.

#### Statistical analysis

Results were subjected to the student's t-test and were considered significant at  $P < 0.05$ <sup>14</sup>.

### RESULTS AND DISCUSSION

The phytochemical screening of the juice of the fresh fruits of *Lagenaria siceraria* showed the presence of the following: tannins, alkaloids, saponins, flavonoids, and cardiac glycosides. The results however showed that anthraquinone and cyanogenetic glycosides were absent (Table 1). The results thus showed that the juice of the fresh fruits of *Lagenaria siceraria* is rich in cardiac glycosides, alkaloids, saponins, tannins, and flavonoids. Alkaloids usually have marked physiological action on animals. Saponins on the other hand are of great pharmaceutical importance because of their relationship to compounds such as the sex hormones, cortisones, diuretic steroids, vitamin D and cardiac glycosides. Tannins like alkaloids are

substances which show protein precipitation and are related to the physiological effects of herbal medicines. Flavonoid containing plants have influence on arachidonic acid metabolism, thus could have anti-inflammatory, antiallergic, antithrombotic or vasoprotective effects. Cardiac glycosides are steroidal glycosides which exert a slowing and strengthen effect on the failing heart<sup>15</sup>. The implication of all these is that this plant is of great medicinal importance. The presence of cardiac glycosides in this plant has further confirmed its medicinal use as antihyperlipidemic agent. The presence of flavonoids in this plant is therefore evidenced of its anti-inflammatory properties. This therefore supports the earlier work of Olajide *et al*<sup>7</sup> that the plant has anti-inflammatory property. The results of the effect of the juice of the fresh fruits of *Lagenaria siceraria* on the blood cholesterol lowering effect of Wistar rats showed that the juice extract caused significant reduction of the mean body weight when compared with those of the control (Table 2). The study showed that the anti-hypercholesterolemia effect of this juice extract on the mean body weight is comparable to that of juice extract, an adrenergic antagonist, though at a very high dose of the juice extract. The juice extract efficacy in treating hypercholesterolemia as well as most of its toxic effects results from nonselective beta blockade. It decreases blood cholesterol level as a result of decrease in cardiac output<sup>16</sup>. The presence of cardiac glycoside in this plant known to slow and strengthen a failing heart may have accounted for its antihypercholesterolemia effect. Besides, it is known that tannins have diuretic effect hence its presence in this plant may have contributed to the antihypercholesterolemia effect<sup>16</sup>. The results of the safety evaluation of the juice of the fresh fruits of *Lagenaria siceraria* on rats showed that the juice extract caused significant increase in the levels of total protein and globulin (Table 3). It is inferred that alteration in the plasma total protein is most often due to decrease in the quantity of albumin, which may be accompanied by a relative hyperglobulinaemia<sup>17,18</sup>. In this study, all protein experienced an increase, suggesting that there is increased protein synthesis or mobilization. The increase in globulin level may indicate that the plant could have the potential to stimulate immune response by increasing antibody production (immunoglobins)<sup>19</sup>, and this also could be responsible for its use as a medicinal plant. Diseases associated with high TG levels (Diabetes mellitus, obesity, chronic renal disease, primary hyperlipoproteinemia) carry high risk of cardiovascular disorder (CVD)<sup>20</sup>. Hypercholesterolemia is reportedly the major risk factors in life style related diseases such as atherosclerosis and related cardiovascular complications including cerebral paralysis and myocardial infarction<sup>21</sup>.

The results of the safety evaluation of the juice of the fresh fruits of *Lagenaria siceraria* on rats showed that the juice extract caused significant decrease in the levels of serum cholesterol and triglyceride levels. It is shown that atherogenic diet elevates serum triglyceride levels essentially by preventing its uptake and clearance by inhibiting catabolizing enzymes like lipoprotein lipase (LPL) and lecithin cholesterol acetyl transferase (LCAT)<sup>22</sup>.

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**Table 1: Phytochemical Screening of *Lagenaria siceraria***

S.no.	Test	Result
1	For alkaloid	++
2	For cardiac glycoside	+++
3	For anthraquinone	-
4	For cyanogenetic glycoside	-
5	For saponins	+
6	For tannins	+
7	For flavonoids	+
8	For phytosterols	+

+ Mild ++ Average +++ Good — absent

**Table: 2 The Effects Of *Lagenaria siceraria* on body weight of rats**

Groups	Body weight in gm compared with weight first day
Normal Control (Saline)	+11.24
Positive Control (Atherogenic diet)	+ 46.23
<i>Lagenaria siceraria</i> Juice	- 21.72

Values are mean ± SD of six rats from each group

**Table 3: Effects Of The Juice Of *Lagenaria siceraria* On The Serum Chemistry Of Rats**

Parameters	Normal control	Positive control	Fresh juice of <i>lagenaria siceraria</i> (5ml)
Total protein (g/dL)	6.0 ± 0.40	8.2 ± 0.34	7.4 ± 0.26*
Albumin (g/dL)	4.6 ± 0.30	6.3 ± 0.20	5.3 ± 0.42*
Globulin (g/dL)	3.5 ± 0.20	5.7 ± 0.12	4.5 ± 0.35*
Total cholesterol (g/dL)	36.2 ± 0.35	48.4 ± 0.26	40.5 ± 0.31*
Triglycerides (g/dL)	25.1 ± 0.92	37.3 ± 0.45	30.4 ± 0.83*
HDL-c (g/dL)	16.12 ± 0.98	18.64 ± 0.96	17.79 ± 0.45*
LDL-c (g/dL)	28.14 ± 0.12	56.74 ± 1.58	43.31 ± 1.64*
VLDL-c (g/dL)	7.78 ± 1.35	14.87 ± 1.45	11.56 ± 1.46

Values are mean ± SD of six rats from each group; \*  $P < 0.05$  show significant when compare with positive control

#### ACKNOWLEDGEMENT

My regards to Mr. Deepak Nanda, Kapil Kalra, Amandeep Singh and Lokesh tyagi, Faculty, Dev Bhoomi Institute of Pharmacy and Research, for their co-operation to complete the work. I also thanks to Mr. Karan Dhamija, School of Life Sciences, University of Hertfordshire, England, who took interest in our work and carrying out animal experiment and briefing the data.

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