

بِسْمِ اللَّهِ الرَّحْمَنِ الرَّحِيمِ

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# BIOLOGICAL STUDY FOR SOME DIURETIC PLANTS






# Introduction

- 
- **Diuretics are defined as compounds that increase urine flow.**
  - **Therapeutic use of diuretic compounds is:**
    - **Lower hypertension by decreasing blood volume through the excretion of water in the urine.**
    - **Diuretics are beneficial in many life threatening disease conditions such as:**
      - heart failure, renal insufficiency, hepatic cirrhosis, hypercalcemia, diabetes insipidus, glaucoma, cerebral edema, drug intoxication, hyperaldosteronism and polycystic ovarian syndrome (PCOS).**



# Research Objective

- 
- **A number of diuretics like mannitol, thiazides, furosemide and ethacrynic acid are used in practice. However, there is still need for more effective and less toxic diuretic.**
  - **In this work, we report on evaluation of the diuretic properties attributed to six endemic plant species from Saudi Arabia.**

■ Plant choices were:



*Corchorus olitorius* Linn  
(الملوخيه)



*Indigofera oblongifolia* Forsk  
(دسم، صرح)



*Ocimum basilicum* L.  
(الريحان، الصعتر الهندي)



*Carthamus tinctorius* L.  
(عصفر)



*Lavandula pubescens* Decne  
(خزامى زغبية)



*Alchemilla vulgaris* (hort.)  
(رجل الأسد)

## *Corchorus olitorius* Linn. (Tiliaceae)



- The leaves are used as:  
Demulcent, diuretic, febrifuge and tonic.
- and in the treatment of chronic cystitis, gonorrhoea and dysuria.

## *Indigofera oblongifolia* Forsk (Fabaceae)



- Traditionally used in urinary tract infection, cough and skin infection.



## ***Ocimum basilicum* L. (Lamiaceae)**

- Leaves and flowering parts of *O. basilicum* are traditionally used as antispasmodic, aromatic, carminative, digestive, galactagogue, stomachic, diuretic and tonic agents.



## ***Carthamus tinctorius* L. (Safflower) (Asteraceae) is**

- Traditionally used as purgative
- and in the treatment of menstrual problems, cardiovascular disease, pain and swelling associated with trauma, rheumatism, chronic nephritis and as diuretic.



## *Lavandula pubescens* Decne. (Lamiaceae)

- The plant has been used as an antispasmodic, carminative, diuretic and general tonic.



## Lady's mantle (*Alchemilla vulgaris* hort.) (Rosaceae).

- It has a long history of use, mainly for external treatment for cuts and wounds, and internally for treatment of diarrhea.
- Menstrual problems.
- Diuretic





# Materials and Methods

## ***Extraction***

- The dried ground aerial parts of *I. oblongifolia*, *O. basilicum*, *L. pubescens*, *A. vulgaris*.
- The leaves of *C. olitorius*.
- The flower of *C. tinctorius* (100 g from each plant) were extracted to exhaustion by percolation at room temp. with 90% ethanol and the extract was evaporated in *vacuo* to leave 12.2g, 12.6, 13.3, 11.5, 12.8, 12.9 g of residue.



## ***Pharmacological Evaluation***

### ***Experimental design***

- **5 groups of 6 male Wister albino rats (150-180g/kg, b.w.) were used.**
- **All the animals received normal saline (25mL/kg, b.w) orally, prior to start of the experiment.**
- **Group I which received normal saline was treated as control.**
- **Group II received urea (1g/kg).**
- **Group III received furosemide (Lasix) (5mg/kg).**
- **Group IV and V received the alcoholic extracts at the dose of 200 and 400mg/kg b. w. respectively.**
- **The total urine volume over the period of 5hrs was measured for the all the ethanolic extracts, (200 and 400mg/kg b.w), standard diuretics (urea and furosemide) and control.**
- **Urine volume pH and electrolyte excretion were measured and used to obtain parameters such as activity and diuretic action.**
- **The significance of results was explored using (ANOVA) and student`s *t-test*.**

## Diuretic activity and urinary volume extraction

$$\text{Urinary Excretion} = \frac{\text{Total urinary output}}{\text{Total liquid administered}} \times 100$$

$$\text{Diuretic action} = \frac{\text{Urinary excretion in test group}}{\text{Urinary excretion in control group}} \times 100$$

$$\text{Diuretic activity} = \frac{\text{Diuretic action of drug}}{\text{Diuretic action of urea}} \times 100$$



# Results and Discussion

**Table 1: Dose response diuretic activity after 5<sup>th</sup> hour by I.P. administration of the ethanolic extracts of *A. vulgaris* (AV), *I. oblongifolia* (IO), *C. tinctorius* (CT), *C. olitorius* (CO), *O. basilicum* (OB) and *L. pubescens* (LP) in normal rats.**

<b>Groups</b>	<b>Urine Volume (ml)</b>	<b>Urinary Excretion (<math>V_0/V_1</math>)X100</b>	<b>Diuretic Action (<math>UE_t/UE_c</math>)</b>	<b>Diuretic Activity (<math>DA_t/DA_u</math>)</b>	<b>pH</b>
<b>Control ( saline)</b>	<b>0.99±0.15</b>	<b>26.40</b>	<b>-</b>	<b>-</b>	<b>7.63±0.32</b>
<b>Urea (1 g/kg)</b>	<b>1.95±0.33*</b>	<b>52.00</b>	<b>1.96</b>	<b>-</b>	<b>8.55±0.38</b>
<b>Furosemide (5 mg/kg)</b>	<b>3.45±0.34***</b>	<b>92.00</b>	<b>3.77</b>	<b>1.92</b>	<b>9.61±0.19***</b>
<b>AV (200 mg/kg)</b>	<b>1.68±0.17*</b>	<b>45.80</b>	<b>1.69</b>	<b>0.86</b>	<b>8.95±0.30*</b>
<b>AV (400 mg/kg)</b>	<b>2.15±0.26**</b>	<b>57.33</b>	<b>2.17</b>	<b>1.10</b>	<b>9.00±0.28*</b>
<b>IO (200 mg/kg)</b>	<b>2.08±0.27**</b>	<b>55.46</b>	<b>2.10</b>	<b>1.07</b>	<b>8.43±0.32</b>
<b>IO (400 mg/kg)</b>	<b>2.70±0.16***</b>	<b>72.00</b>	<b>2.72</b>	<b>1.38</b>	<b>9.06±0.28**</b>
<b>CT (200 mg/kg)</b>	<b>0.81±0.20</b>	<b>21.60</b>	<b>0.81</b>	<b>0.41</b>	<b>7.63±0.30</b>
<b>CT (400 mg/kg)</b>	<b>0.80±0.06</b>	<b>21.33</b>	<b>0.80</b>	<b>0.40</b>	<b>7.16±0.29</b>
<b>Control ( saline)</b>	<b>1.14± 0.092</b>	<b>30.4</b>	<b>-</b>	<b>-</b>	<b>7.13±0.9</b>
<b>Urea (1 g/kg)</b>	<b>2.57±0.01***</b>	<b>68.53</b>	<b>2.254</b>	<b>-</b>	<b>8.4±0.15***</b>
<b>Furosemide (5 mg/kg)</b>	<b>4.22±0.10***</b>	<b>112.53</b>	<b>3.70</b>	<b>1.64</b>	<b>9.18±0.11***</b>
<b>CO (200 mg/kg)</b>	<b>1.16±0.05</b>	<b>30.93</b>	<b>1.017</b>	<b>0.47</b>	<b>7.31±0.18</b>
<b>CO (400 mg/kg)</b>	<b>1.63±0.07**</b>	<b>43.467</b>	<b>1.429</b>	<b>0.63</b>	<b>8.26±0.10***</b>
<b>OB (200 mg/kg)</b>	<b>2.25±0.09***</b>	<b>60.00</b>	<b>1.973</b>	<b>0.87</b>	<b>8.36±0.21***</b>
<b>OB (400 mg/kg)</b>	<b>2.28±0.10***</b>	<b>60.80</b>	<b>2.00</b>	<b>0.88</b>	<b>8.01±0.24**</b>
<b>LP (200 mg/kg)</b>	<b>1.77±0.10***</b>	<b>47.20</b>	<b>1.552</b>	<b>0.68</b>	<b>8.03±0.20**</b>
<b>LP (400 mg/kg)</b>	<b>2.08±0.10***</b>	<b>55.47</b>	<b>1.824</b>	<b>0.80</b>	<b>8.16±0.15***</b>



**Table 1: Dose response diuretic activity after 5<sup>th</sup> hour by I.P. administration of the ethanolic extracts of *A. vulgaris* (AV), *I. oblongifolia* (IO), *C. tinctorius* (CT), *C. olitorius* (CO), *O. basilicum* (OB) and *L. pubescens* (LP) in normal rats**

<b>Groups</b>	<b>Urine Volume (ml)</b>	<b>Urinary Excretion (<math>V_0/V_1</math>)X100</b>	<b>Diuretic Action (<math>UE_t/UE_c</math>)</b>	<b>Diuretic Activity (<math>DA_t/DA_u</math>)</b>	<b>pH</b>
<b>Control ( saline)</b>	<b>0.99±0.15</b>	<b>26.40</b>	<b>-</b>	<b>-</b>	<b>7.63±0.32</b>
<b>Urea (1 g/kg)</b>	<b>1.95±0.33*</b>	<b>52.00</b>	<b>1.96</b>	<b>-</b>	<b>8.55±0.38</b>
<b>Furosemide (5 mg/kg)</b>	<b>3.45±0.34***</b>	<b>92.00</b>	<b>3.77</b>	<b>1.92</b>	<b>9.61±0.19***</b>
<b>AV (200 mg/kg)</b>	<b>1.68±0.17*</b>	<b>45.80</b>	<b>1.69</b>	<b>0.86</b>	<b>8.95±0.30*</b>
<b>AV (400 mg/kg)</b>	<b>2.15±0.26**</b>	<b>57.33</b>	<b>2.17</b>	<b>1.10</b>	<b>9.00±0.28*</b>
<b>IO (200 mg/kg)</b>	<b>2.08±0.27**</b>	<b>55.46</b>	<b>2.10</b>	<b>1.07</b>	<b>8.43±0.32</b>
<b>IO (400 mg/kg)</b>	<b>2.70±0.16***</b>	<b>72.00</b>	<b>2.72</b>	<b>1.38</b>	<b>9.06±0.28**</b>
<b>CT (200 mg/kg)</b>	<b>0.81±0.20</b>	<b>21.60</b>	<b>0.81</b>	<b>0.41</b>	<b>7.63±0.30</b>
<b>CT (400 mg/kg)</b>	<b>0.80±0.06</b>	<b>21.33</b>	<b>0.80</b>	<b>0.40</b>	<b>7.16±0.29</b>

**Table 2: Electrolyte concentration (mMol/L) in urine collected in 5<sup>th</sup> hour by I.P. administration of the ethanolic extracts of *A. vulgaris*, *I. oblongifolia*, *C. tinctorius*, *C. olitorius*, *O. basilicum* and *L. pubescens* in normal rats**

Groups	Electrolytes excretion in mMol/L		
	Na <sup>+</sup>	K <sup>+</sup>	Cl <sup>-</sup>
Control ( saline)	124.46±2.67	64.83±5.94	174.99±31.94
Urea (1 g/kg)	176.19±5.56***	89.33±8.82*	238.18±42.93
Furosemide (5 mg/kg)	195.69±5.79***	111.16±8.58***	277.07±30.59*
AV (200 mg/kg)	191.14±2.46***	75.66±3.98	262.49±41.24
AV (400 mg/kg)	205.48±8.00***	94.66±3.61**	330.54±39.84*
IO (200 mg/kg)	173.21±3.48***	94.33±10.26*	262.49±48.80
IO (400 mg/kg)	180.02±6.73***	96.00±6.81**	369.43±39.84**
CT (200 mg/kg)	124.03±3.86	65.16±4.30	160.41±30.58
CT (400 mg/kg)	124.35±7.06	73.16±6.76	184.71±35.31
Control ( saline)	145.95±1.45	52.5±1.35	170.45±9.00
Urea (1 g/kg)	210.57±13.37***	92.83±2.85***	247.90±12.48***
Furosemide (5 mg/kg)	203.25±1.98***	76.66±2.23***	301.37±17.92***
CO (200 mg/kg)	161.55±8.82	55.5±1.97	204.15±16.84
CO (400 mg/kg)	161.59±3.29**	60.66±1.60**	228.45±17.53*
OB (200 mg/kg)	157.04±2.90**	55.66±2.07	213.87±12.30*
OB (400 mg/kg)	161.74±1.69***	69.16±1.85***	233.32±10.64**
LP (200 mg/kg)	199.81±3.72***	78.83±1.99***	243.05±9.72***
LP (400 mg/kg)	205.26±2.70***	88.00±1.86***	272.21±12.29***

**Table 1: Dose response diuretic activity after 5<sup>th</sup> hour by I.P. administration of the ethanolic extracts of *A. vulgaris*, *I. oblongifolia*, *C. tinctorius*, *C. olitorius*, *O. basilicum* and *L. pubescens* in normal rats.**

<b>Groups</b>	<b>Urine Volume (ml)</b>	<b>Urinary Excretion (<math>V_0/V_1</math>)X100</b>	<b>Diuretic Action (<math>UE_t/UE_c</math>)</b>	<b>Diuretic Activity (<math>DA_t/DA_u</math>)</b>	<b>pH</b>
<b>Control ( saline)</b>	<b>0.99±0.15</b>	<b>26.40</b>	<b>-</b>	<b>-</b>	<b>7.63±0.32</b>
<b>Urea (1 g/kg)</b>	<b>1.95±0.33*</b>	<b>52.00</b>	<b>1.96</b>	<b>-</b>	<b>8.55±0.38</b>
<b>Furosemide (5mg/kg)</b>	<b>3.45±0.34***</b>	<b>92.00</b>	<b>3.77</b>	<b>1.92</b>	<b>9.61±0.19***</b>
<b>AV (200 mg/kg)</b>	<b>1.68±0.17*</b>	<b>45.80</b>	<b>1.69</b>	<b>0.86</b>	<b>8.95±0.30*</b>
<b>AV (400 mg/kg)</b>	<b>2.15±0.26**</b>	<b>57.33</b>	<b>2.17</b>	<b>1.10</b>	<b>9.00±0.28*</b>
<b>IO (200 mg/kg)</b>	<b>2.08±0.27**</b>	<b>55.46</b>	<b>2.10</b>	<b>1.07</b>	<b>8.43±0.32</b>
<b>IO (400 mg/kg)</b>	<b>2.70±0.16***</b>	<b>72.00</b>	<b>2.72</b>	<b>1.38</b>	<b>9.06±0.28**</b>
<b>CT (200 mg/kg)</b>	<b>0.81±0.20</b>	<b>21.60</b>	<b>0.81</b>	<b>0.41</b>	<b>7.63±0.30</b>
<b>CT (400 mg/kg)</b>	<b>0.80±0.06</b>	<b>21.33</b>	<b>0.80</b>	<b>0.40</b>	<b>7.16±0.29</b>
<b>Control ( saline)</b>	<b>1.14± 0.092</b>	<b>30.4</b>	<b>-</b>	<b>-</b>	<b>7.13±0.9</b>
<b>Urea (1 g/kg)</b>	<b>2.57±0.01***</b>	<b>68.53</b>	<b>2.254</b>	<b>-</b>	<b>8.4±0.15***</b>
<b>Furosemide(5 mg/kg)</b>	<b>4.22±0.10***</b>	<b>112.53</b>	<b>3.70</b>	<b>1.64</b>	<b>9.18±0.11***</b>
<b>CO (200 mg/kg)</b>	<b>1.16±0.05</b>	<b>30.93</b>	<b>1.017</b>	<b>0.47</b>	<b>7.31±0.18</b>
<b>CO (400 mg/kg)</b>	<b>1.63±0.07**</b>	<b>43.467</b>	<b>1.429</b>	<b>0.63</b>	<b>8.26±0.10***</b>
<b>OB (200 mg/kg)</b>	<b>2.25±0.09***</b>	<b>60.00</b>	<b>1.973</b>	<b>0.87</b>	<b>8.36±0.21***</b>
<b>OB (400 mg/kg)</b>	<b>2.28±0.10***</b>	<b>60.80</b>	<b>2.00</b>	<b>0.88</b>	<b>8.01±0.24**</b>
<b>LP (200 mg/kg)</b>	<b>1.77±0.10***</b>	<b>47.20</b>	<b>1.552</b>	<b>0.68</b>	<b>8.03±0.20**</b>
<b>LP (400 mg/kg)</b>	<b>2.08±0.10***</b>	<b>55.47</b>	<b>1.824</b>	<b>0.80</b>	<b>8.16±0.15***</b>

**Table 1: Dose response diuretic activity after 5<sup>th</sup> hour by I.P. administration of the ethanolic extracts of fractions of *A. vulgaris* and *I. oblongifolia* in normal rats**

<b>Groups</b>	<b>Urine Volume (ml)</b>	<b>Urinary Excretion (<math>V_0/V_1</math>)X100</b>	<b>Diuretic Action (<math>UE_t/UE_c</math>)</b>	<b>Diuretic Activity (<math>DA_t/DA_u</math>)</b>	<b>pH</b>
<b>Control( saline)</b>	<b>0.95±0.06</b>	<b>25.33</b>	<b>-</b>	<b>-</b>	<b>6.68±0.22</b>
<b>Urea (1gm/kg)</b>	<b>1.73±0.06***</b>	<b>46.13</b>	<b>1.82</b>	<b>-</b>	<b>7.41±0.16*</b>
<b>Furosemide (5mg/kg)</b>	<b>3.76±0.08***</b>	<b>100.26</b>	<b>3.95</b>	<b>2.17</b>	<b>8.6±0.19***</b>
<b>AV A (100mg/kg)</b>	<b>0.95±0.06*</b>	<b>25.33</b>	<b>1.00</b>	<b>0.55</b>	<b>6.95±0.12</b>
<b>AV A (200mg/kg)</b>	<b>1.08±0.09</b>	<b>28.8</b>	<b>1.14</b>	<b>0.63</b>	<b>6.93±0.15</b>
<b>AV C (100mg/kg)</b>	<b>1.23±0.06*</b>	<b>32.8</b>	<b>1.29</b>	<b>0.71</b>	<b>7.05±0.13</b>
<b>AV C (200mg/kg)</b>	<b>1.36±0.04***</b>	<b>36.26</b>	<b>1.43</b>	<b>0.78</b>	<b>7.1±0.11</b>
<b>AV P (100mg/kg)</b>	<b>1.76±0.08***</b>	<b>46.93</b>	<b>1.85</b>	<b>1.02</b>	<b>7.75±0.13**</b>
<b>AV P (200mg/kg)</b>	<b>2.43±0.13***</b>	<b>64.8</b>	<b>2.56</b>	<b>1.41</b>	<b>7.9±0.13***</b>
<b>IO A (100mg/kg)</b>	<b>1.6±0.08***</b>	<b>42.66</b>	<b>1.68</b>	<b>0.92</b>	<b>7.56±0.21*</b>
<b>IO A (200mg/kg)</b>	<b>1.7±0.10***</b>	<b>45.33</b>	<b>1.79</b>	<b>0.98</b>	<b>8.01±0.13***</b>
<b>IO C (100mg/kg)</b>	<b>0.96±0.06</b>	<b>25.6</b>	<b>1.01</b>	<b>0.55</b>	<b>6.96±0.1</b>
<b>IO C (200mg/kg)</b>	<b>1.21±0.08*</b>	<b>32.26</b>	<b>1.27</b>	<b>0.70</b>	<b>7.51±0.17*</b>

Values are expressed as mean ± SEM (Number of animals, n=6)

$V_0$  = Total Urinary output       $V_1$  = Total fluid input       $UE_t$  = Urinary excretion in test group

$UE_c$  = Urinary excretion in control group       $DA_t$  = Diuretic action of test sample,  $DA_u$  = Diuretic action of urea

\*P<0.05, \*\*P<0.01, \*\*\*P<0.001, student's *t*-test

## Conclusion:

- **The present study justified the use of *A .vulgaris*, *I. oblongifolia* and *L. pubescens* as diuretics.**
- **They were proved to be effective as hypernatramic, hyperchloremic and hyperkalemic diuretic; a fact that correlated well with the traditional use of the plant as a diuretic**
- **The effect may be produced by stimulation of regional blood flow or initial vasodilation, or by producing inhibition of tubular reabsorption of water and anions.**
- **Increased Na<sup>+</sup> and water excretion activity also provide strong basis for the anti-hypertensive action ascribed for the plants under investigation. However, the mechanism of action is still to be explored.**



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