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Evaluation of the Effects of Aqueous Extracts of *Ocimum gratissimum* on Some Kidney Function Parameters in Normal Male Rabbits

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Abstract: The effects of the aqueous leaf-extract of *Ocimum gratissimum* (African basil) on some serum kidney biomarkers of normal rabbits were investigated. The experimental animals were randomly selected into four groups with five animals in each. A group was administered normal saline (control), while the other three groups were administered 75 $mgkg^{-1}$, 150 $mgkg^{-1}$ and 250 $mgkg^{-1}$ of aqueous leaf-extract of *Ocimum gratissimum*. The normal saline and extract were administered orally to the animals twice daily for 21 days. Blood samples were collected from the ear vein of the animals on days 0, 7, 14 and 21 for determination of serum sodium (Na^+), potassium (K^+), chloride (Cl^-), urea and creatinine levels. The results showed significant reduction ($p \square 0.05$) in serum Cl^- (106.00 ± 2.14 $mEqL^{-1}$) in the 250$mgkg^{-1}$ group on day 7. The administration of 75 $mgkg^{-1}$, 150 $mgkg^{-1}$ and 250 $mgkg^{-1}$ of extracts resulted in significant increase ($p ? 0.05$) of 37.00 ± 1.84, 41.00 ± 1.87 and 39.40 ± 2.52 respectively of serum urea on the 7th day. The serum creatinine increased on the 7th day similar to the serum urea but reduced significant ($p \square 0.05$) with the 250$mgkg^{-1}$ on the 21st day. The administration of the extract showed nephrotoxic capacity after seven days, which improved with increased duration of administration.

Key words: Aqueous extract · Kidney · *Ocimum gratissimum* · Rabbits

INTRODUCTION

Plants are rich in bioactive secondary metabolites which vary quantitatively between and within species of plants. Medicinal plants have been an old companion of man providing food, shelter, wealth and have helped in maintaining relatively good health by its preventive and curative potentials when properly utilized [1-9]. The practice is sustained by the ready availability of plants, the insignificant cost of preparation and the new crave to avoid the side effects of chemotherapy [10-14]. To that effect the larger population of Africa and Nigeria, in particular, hinge on their old companion plants for a relatively disease-free state on appropriate use as herbal medicine [11-14].

The people of Southern Nigeria, particularly the south-eastern region are noted for their high consumption of vegetable and spices. The wide variety of vegetables and spices form part of the ingredients of herbal medicine in the region because of their high secondary metabolite constituents [14-18]. *Ocimum gratissimum* Linn which is also called African basil is native to Africa and Asia belonging to the family *lamiaceae*. It is found in the tropical regions of Nigeria and described by different local name, but popularly known as 'Scent Leaf' in most part of the country [19]. Though this plant is mostly used as spice, it is also used widely in folk medicine for management of diabetes (Egesie *et al*., 2006 [6]; as anti-diarrhoea (Ezekwesili *et al*., 2004) [7], mosquitocidal activity (Oparaocha *et al*., 2010) [16], as anti-oxidant (Oboh and Rocha 2008) [15], antifungal activities (Amadi *et al*., 2010) [5] and antibacterial activities (Akujobi *et al*., 2010) [4].

The kidney is important for the regulation of body fluids and electrolytes. Condition when the kidney cannot perform its function is referred to as kidney failure which could be reversible (acute) or irreversible (chronic). The excretion of body fluid with reduced amounts of

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potassium, sodium and water intake restriction, as well as excessive intake of potassium and sodium may result in rare conditions of hyperkalemia and hypernatremia respectively (Kang *et al*., 2002) [8]. The kidney is also an excretory organ of waste substances like creatinine and urea (Pouokam *et al*. 2011) [18]. Creatinine and urea are major catabolic products of carbohydrate and protein metabolism respectively. Creatinine is eliminated from the plasma through glomerular filtration and excreted as a waste product into urine. The reduction in creatinine concentration in the urine is indicative of impaired renal function (Smith and Hampton, 1990) [19].

This work was aimed at evaluating the activities of the aqueous leaf-extract of *Ocimum gratissimum* on some serum kidney biomarkers. The use of the crude extract of *Ocimum gratissimum* is gaining lots of ground in the South-east region of Nigeria but there is dearth of information on the possible toxicity.

MATERIALS AND METHODS

Plant Material: Fresh leaves of *Ocimum gratissimum* were harvested from a local farm in Okada, Ovia North-East L.G.A of Edo State, Nigeria. They were sorted, washed, air-dried at room temperature and milled into powder. The cold extraction was carried out using water as solvents. The extracts were concentrated to about 10% of the original volume using a rotary evaporator (BUCHI, type RE111, Rotavapor).

Experimental Animals: The experimental animals were all male New Zealand rabbits, which weighed between 1.3 – 1.6kg. The animals were obtained from the Central Animal Facility of Igbinedion University, Okada. The animals were housed in well ventilated cages and allowed 12 hours light and dark cycle for the period of experiment. They were given water and food *ad libitum* throughout the duration of the experiment.

Experimental Design: The animals were randomly selected and grouped. There were a total of four groups with five animals per group. One group was administered normal saline, while the other three groups were administered 75mgkg^{-1}, 150mgkg^{-1} and 250mgkg^{-1} body weight of aqueous leaf extract of *Ocimum gratissimum*. The normal saline and extract were administered orally to the animals twice daily for the duration of the experiment (21 days). The normal saline group served as control. The blood samples were collected from the ear vein of the animals before extract administration and every seven days after administration of extract for the 21-day period of experiment. The control group was also treated in the same manner.

Biochemical Assays: Urea, creatinine, sodium ion, potassium ion and chloride ion levels in serum of normal male rabbits were evaluated using assay kits (Randox Laboratories LTD. United Kingdom BT29 4QY).

Statistical Analysis: The results obtained were expressed as mean ± Standard deviation. The comparison of mean values were analysed by SPSS (version 16.0) using paired samples T-Test at 95% level of confidence.

RESULTS AND DISCUSSION

There was no significant (p?0.05) reduction in serum Na^+ levels (Table 1). This was also shown when the percentage changes of all groups were considered after 21 days (Figure 1). There were dose-dependent elevations in serum K^+ levels (Table 2). The same pattern was observed when the percentage changes for the different groups were considered after 21 days. While the control group showed 8% reduction after 21 days, the extract groups of 150mgkg^{-1} and 250mgkg^{-1} had a concentration dependent increase of 28.01% and 53.25% respectively (Figure 1). The serum Cl^- levels of rabbits administered *O. gratissimum* extracts in Table 3 reduced in a concentration dependent pattern. This was most expressed on the 7th day where the increase was significant (p□0.05) with the administration of 250mgkg^{-1}. The pattern was not the same when the percentage changes within the groups were considered (Figure 1). The figure showed a concentration dependent increase of 2.09% and 3.87% with administration of 150mgkg^{-1} and 250mgkg^{-1} respectively after 21 days. The results obtained though not significant (p □ 0.05) were similar to results of Malgwi *et al*., (2014) [10] where oral administration of *Cucurbita pepo Linn* seed extract resulted in significant decrease in serum sodium ion and increase in serum potassium ion levels. This may suggest that increase in concentration of *O. gratissimum* extracts will yield similar results. This pattern of result was observed when comparing the percentage changes between day 0 and day 21 in the different groups but was not obvious when comparing the serum sodium ion and potassium ion levels of the extract groups with the control.

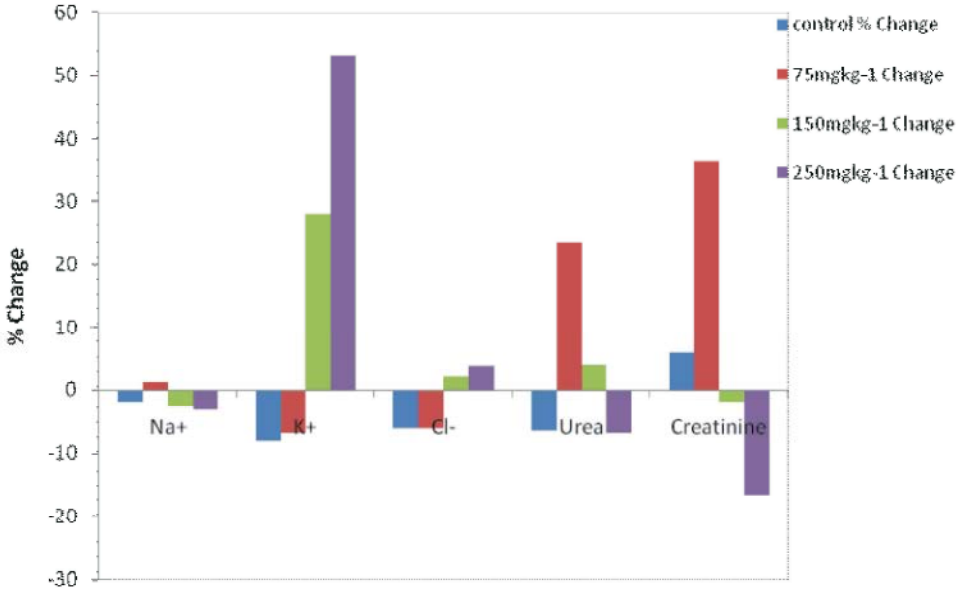

Fig. 1: Percentage changes between day 0 and day 21 in serum kidney markers of rabbits administered aqueous extracts of *O. gratissimum*.

Table 1: The effects of aqueous extracts of *O. gratissimum* on serum Na^+ ($mEqL^{-1}$) of normal rabbits

	Control	75mgkg^{-1}	150mgkg^{-1}	250mgkg^{-1}
Day 0	145.00 ± 2.48	143.00 ± 1.94	143.80 ± 2.09	142.80 ± 1.35
Day 7	144.80 ± 2.14	144.00 ± 1.97	144.80 ± 1.39	145.60 ± 1.29
Day 14	143.80 ± 1.24	141.40 ± 1.33	142.20 ± 1.02	142.80 ± 1.02
Day 21	142.20 ± 1.68	141.20 ± 1.37	140.20± 1.20	138.40 ± 2.07

Values * given as mean ± standard deviation had significant differences, when compared with the control and [a] when compared with day 0. ($p < 0.05$).

Table 2: The effects of aqueous extracts of *O. gratissimum* on serum K^+ ($mEqL^{-1}$) of normal rabbits

	Control	75mgkg^{-1}	150mgkg^{-1}	250mgkg^{-1}
Day 0	5.50 ± 0.44	5.62 ± 0.89	4.70 ± 0.22	4.62 ± 0.57
Day 7	6.04 ± 0.43	6.04 ± 0.47	8.34 ± 0.80[a]	7.14 ± 0.96
Day 14	6.16 ± 0.33	4.60 ± 0.26	6.22 ± 0.66	6.32 ± 0.78
Day 21	5.06 ± 0.25	5.24 ± 0.74	6.02± 0.47	7.08 ± 1.00

Values * given as mean ± standard deviation had significant differences, when compared with the control and [a] when compared with day 0. ($p < 0.05$).

Table 3: The effects of aqueous extracts of *O. gratissimum* on serum Cl^- ($mEqL^{-1}$) of normal rabbits

	Control	75mgkg^{-1}	150mgkg^{-1}	250mgkg^{-1}
Day 0	113.20 ± 1.35	110.00 ± 1.05	105.40 ± 1.85	108.60 ± 1.72
Day 7	116.20 ± 1.57	114.20 ± 1.03	112.80± 1.74	106.00 ± 2.14*
Day 14	110.4± 1.69	108.60 ± 1.22	101.40 ± 2.15	106.00 ± 1.46
Day 21	106.60 ± 2.06	103.40± 1.29	107.60± 2.04	104.40 ± 1.79

Values * given as mean ± standard deviation had significant differences, when compared with the control and [a] when compared with day 0. ($p < 0.05$).

Table 4: The effects of aqueous extracts of *O. gratissimum* on serum Urea ($mgdl^{-1}$) of normal rabbits

	Control	75mgkg^{-1}	150mgkg^{-1}	250mgkg^{-1}
Day 0	28.00 ± 2.00	21.20 ± 1.20	25.00 ± 1.76	30.00 ± 1.16
Day 7	28.00 ± 1.22	37.00 ± 1.84*[a]	41.00 ± 1.87*[a]	39.40 ± 2.52*
Day 14	31.00 ± 1.87	26.40 ± 0.60	29.60 ± 0.24	27.60 ± 1.25
Day 21	26.20 ± 1.55	26.20± 1.24	26.00 ± 1.87	28.00 ± 2.00

Values * given as mean ± standard deviation had significant differences, when compared with the control and [a] when compared with day 0. ($p < 0.05$).

Table 5: The effects of aqueous extracts of *O. gratissimum* on Creatinine ($mgdl^{-1}$) of normal rabbits

	Control	$75mgkg^{-1}$	$150mgkg^{-1}$	$250mgkg^{-1}$
Day 0	1.00 ± 0.11	1.54 ± 0.12	1.10 ± 0.11	0.96 ± 0.23
Day 7	1.08 ± 0.03	1.32 ± 0.17*	1.02 ± 0.10	1.02 ± 0.13
Day 14	1.12 ± 0.12	1.06 ± 0.09	0.96 ± 0.04	0.82 ± 0.71
Day 21	1.06 ± 0.05	0.98± 0.04	1.08 ± 0.06	0.80 ± 0.05*

Values * given as mean ± standard deviation had significant differences, when compared with the control and [a] when compared with day 0. ($p < 0.05$).

Table 4 showed the effect of the extracts on the serum urea levels of normal rabbits. Within the 21 days of extract administration, the serum urea levels increased the most on the 7th day. This increase was significant (p□0.05) at all concentrations of the extract administered compared with the control and day 0, but was not concentration dependent. A concentration dependent pattern was revealed when the percentage changes within the groups were considered in Figure 1. The figure showed that increase in extract administration of $75mgkg^{-1}$, $150mgkg^{-1}$ and $250mgkg^{-1}$ resulted in 23.58%, 4.00% increases and 6.67% reduction of serum urea levels respectively. The serum creatinine levels of normal rabbits in Table 5 revealed a concentration dependent reduction with increase in concentration of extracts administered. This was significant (p□0.05) on the 21st day with the $250mgkg^{-1}$ extract group. This pattern was consistent with the percentage changes after 21 days as seen in figure 1. The serum urea level increased significantly on the seventh day of administration of *O. gratissimum* at all concentration but decreased at the end of the 21 days experiment. It is suggestive that the extracts showed nephrotoxicity on the seventh day indicative of impaired kidney function (Afolayan and Yakubu, 2009; Kumar and Padhy, 2011; Ahmed *et al*., 2014) [1, 9, 2]. The serum creatinine increased significantly with the $75mgkg^{-1}$ group and was reduced significantly in the $250mgkg^{-1}$ group. Since crude plant extracts contain myriads of phytochemicals, there is the possibility that this plant extracts contained ingredients which accumulated in the kidney a nd may have led to damage in kidney (Pouokam *et al.,* 2011) [18] after seven days, the accumulation of toxic substance may cause injuries to tubular epithelial cells thereby causing impairment in kidney function (Parke, 1982) [17]. This condition may have been reversed by the stimulation of the xenobiotic enzymes resulting in the metabolism of the toxic ingredients. Also, the presence of polyphenols and flavonoids in *O. gratissimum* extract might be responsible for the antioxidant nephroprotective activities and the reduction of serum urea and creatinine levels (Ajith *et al*., 2007) [3].

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