

## Evaluation of effects of aqueous extract of *Ficus capensis* (thunb.) leaves in adult albino rats

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### Abstract

The study evaluated the effect of aqueous extract of *Ficus capensis* leaves in albino rats. The study specifically investigated the effect of the extract on some liver enzymes (aspartate aminotransferase (AST) and alanine aminotransferase (ALT)) activities, the packed cell volume (PCV), total white blood cell count and differential white blood cell count of the rats. Effect of the extract on serum urea and creatinine levels were also studied. Twenty-five (25) adult male albino rats were used for the study. The extract was prepared and the solution was filtered and fed the rats (25%, 50%, 75% and 100%) of the filtrates. Tap water was fed to the control rats. All the parameters were assessed using standard methods. One way analysis of variance was used to obtain the means from the biochemical result. The means obtained were separated using Duncan Student Multiple Range Test. Results obtained showed that ALT, AST and creatinine levels of the rats that were fed 100% *Ficus capensis* extract were significantly higher than those that were fed diluted *Ficus capensis* extract. Hence, this had adverse effects on the liver cells of the rats. Also result showed that there is improved nutritional status and immunity when properly diluted.

Key words : Effects, aqueous extract, *Ficus capensis*, Albino rats

### INTRODUCTION

Traditional leafy vegetables are integrated in a community's culture for use as food over a long span of time<sup>[1]</sup>. Many of these are home grown or from wild gardens. They are mutually important both in the rural and the urban set ups<sup>[2]</sup>. Traditional green leafy vegetables are highly recommended for regular consumption because of their high nutritional value, diversity to daily food intake, adding flavour and zest to the diet<sup>[3]</sup>. They serve as a source of medicine. They are important in their ecological, agronomic and cultural values<sup>[4]</sup>. They are usually associated with traditional production system, knowledge, and local history of selection and usage. They are consumed daily, particularly in rural communities. Despite their advantages, several studies had established that some vegetable species are potentially toxic to humans and animals. The concentration of these toxic elements is a function of the concentrations in the soil they grow<sup>[5]</sup>. Plant chemical compounds, toxic to humans and livestock are produced as part of the plant's defense mechanism against pest and herbivores or gain advantage over competing plants<sup>[6]</sup>.

Nigeria is endowed with many varieties of green leafy vegetables, most of which are wild forest vegetables. "Okazi". (*Gnetum africanum*) and Uturukpa (*Pterocarpus stanoloides*) are traditional green leafy, wild forest vegetables which had gained a lot of interest from consumers. These vegetables are found in homes in virtually all locations in Nigeria<sup>[5]</sup>.

The use of most of the wildy grown leafy vegetables is not popularized in other surrounding communities apart from the user community. They remain underutilized. This is due to ignorance of the uses as well as diversity in Nigerian languages. It is very difficult for one to recommend some vegetable species due to variation in names.

'Akukoro' leaf (*Ficus capensis*) or bush fig is one of the Nigerian traditional green leafy vegetables consumed in Orba and Ugbaik towns, both in Enugu State. It is utilized in Ayamele

Local Government Area in Anambra State. This plant has great use among the Igede people of Benue State in Nigeria<sup>[7]</sup>. The leaves of *F. capensis* are available throughout the year. Many claims were laid on both the nutritional and medicinal uses of the various parts of this plant. The young leaves are cooked and consumed with soups and porridges as vegetable. The foliage is for cattle, sheep and goats' consumption<sup>[8]</sup>. *Ficus capensis* leaves are used in the treatment of convulsion, stomach ache, respiratory disorders, cough and threatened abortion<sup>[9]</sup>. It is often recommended to Women and children with increased need of nutrients during and after serious ailments or fevers for use. Among the Orba Community in Enugu State, *F. capensis* is known as "blood tonic" (immune booster). The infusions or extracts of the fresh leaves are drunk as quick sources of nutrients and medicine" among the local communities. The dried leaf powder of *F. capensis* is rich sources of tannins, saponins, cardiac glycosides and flavonoids<sup>[7]</sup>. These phytochemicals are precipitate prevention of the activities of free radicals in formation of cancerous cells in the body. Plant poisons are highly active substances that may cause acute effects when ingested in high concentrations and chronic effects when accumulated over time<sup>[10]</sup>. In many cases, consumption of endogenous toxicants in vegetables causes death or prolonged and serious disabilities were reported<sup>[11]</sup>. Moreover, women and children among the vulnerable groups are the major consumers of vegetables because of their increased needs. Consumption of toxic substances in vegetables as one of the only source of their daily nutrient intake would further increase their vulnerability to diseases. Most village dwellers are not aware of the negative effects of the toxicity of plant materials in humans and animals. Based on this, they would continue to use these freely for their detriment or for positive impact.

On the other hand, lack of authentic information on local foods and vegetables are problems to Dietitians and Nutritionist to offer evidence-based counseling on vegetables consumption. In addition, traditional medicines prepared from medicinal and

food plants are not always safe. *Ficus capensis* leaves contain toxic substances<sup>[12]</sup>. Its root and bark decoctions prepared as herbal remedies had caused death<sup>[12]</sup>. Much more care is advised for use of this plant<sup>[12]</sup>. *Ficus capensis* leaves and their extracts are used freely without fear of toxicity. The lethal dose for the dried leaf extract of *F. capensis* was between 900mg to 1200mg/kg in mice<sup>[13]</sup>. This level may not be attained by consumption of the leaves as vegetable. However, attention is drawn to the hot water extract consumption for a long time. The root and the bark decoction of this plant are toxic. Based on this, proper authentication for safe levels for the leaves extracts is necessary. It is known that plant toxins decrease as it moves from the root via the stem to the leaves. Determination of the optimal safe levels for *F. capensis* extract is important before Dietitians and Nutritionist could support its consumption. This is because over dose of many herbal remedies precipitated various ailments ranging from the liver to the kidney failures or even death. The same caution is imperative for herbs popular among a community's food habit without proper knowledge of its acceptable level for consumption.

The objective of the study was to evaluate the effect of aqueous extract of dried leaves of *F. capensis* in adult albino rats. The study specifically investigated the effect of the extract on some liver enzymes (aspartate aminotransferase (AST) and alanine aminotransferase (ALT)) activities, the packed cell volume (PCV), total white blood cell count and differential white blood cell count of the rats. Effect of the extract on serum urea and creatinine levels were also studied. This study will therefore serve as a baseline study for further research to provide a scientifically evidence based guideline for the use of aqueous extract of dried leaves of *F. capensis*.

## MATERIALS AND METHODS

### Procurement and identification of plant materials

*Ficus capensis* leaves were harvested from a family farm in Imiliki Agu in Udenu Local Government Area of Enugu State, Nigeria. The plant material was authenticated by the staff of Bioresources Development and Conservation Programme (BDGP) Centre, Nsukka, Enugu State, Nigeria.

### Preparation of sample

The leaves were harvested, picked, shade dried at room temperature (25°C) for 14 days and pulverized into coarse texture.

### Extraction of plant material

One hundred and fifty (150g) of shade dried leaf sample was added in 1.5L of boiling water and was allowed to simmer for 5 minutes at 100°C. The solution was filtered with a muslin cloth. The filtrate was analysed as well as the pulverized sample for various components. This extract was used to feed the rats at various levels.

### Animal study

Twenty-five (25) adult male albino rats weighing between (150-250g) were purchased from Department of Veterinary Pathology, University of Nigeria, Nsukka, Nigeria. The rats were housed and maintained in the Laboratory Animal facility of the Department of Home Science, Nutrition and Dietetics, Faculty of Agriculture, University of Nigeria Nsukka. The rats were housed individually in metabolic cages and fed water and rat chow ad libitum for acclimatization to diet and environment for 5 days. These rats were allotted to five groups of five rats each on basis of

body weight. Each group of the rat was fed with different levels of the aqueous extract ad libitum for 14 days.

### Grouping of animals: The rats were grouped into A, B, C, D, and E.

Group A = was fed 100% of the extract.

Group B = was fed 75% of the extract + 25% water ad libitum.

Group C = was fed 50% of the extract + 50% water ad libitum

Group D = was fed 25% extract + 75% water ad libitum

Group E = was fed tap water only as the control group.

### Blood collection

At the end of the 14 days, blood was collected from the retro-bulbar plexus of the medial cantus of the eye of the rats. Serum blood was used for kidney and liver function tests and the other uncoagulated blood was used for PCV, total and the differential white blood cell counts.

Biological determination of alanine amino transferase (ALT) and aspartate amino transferase (AST) were done using colorimetric method. In-vitro determination of GPT/ALT and GOT/AST in serum or plasma using Quimica Clinica Applicada (QCA) test kit, Spain<sup>[14]</sup>. Serum urea determination was conducted using the modified method of Berthelot Searcy for the in-vitro determination of urea in serum using Quimica Clinica Applicada (QCA) Enzymatic urea test kit<sup>[15]</sup>. Serum creatinine was determined using the modified Jaffe method<sup>[16]</sup>. The packed cell volume (PCV) was determined by the micro haematocrit method<sup>[17]</sup>. Haemocytometer method was used to determine the total and the differential white blood cell counts<sup>[18]</sup>.

### Statistical analysis

One-way analysis of variance (ANOVA) was used to obtain mean treatment of groups of rats and separation of means used Duncan's studentised Multiple Range Test<sup>[19]</sup>. All statistical analysis was done in the computer using the Statistical Package for Social Sciences (SPSS) version 17.

### Ethical Approval

Ethical Clearance for the study was obtained from Faculty of Veterinary Medicine University of Nigeria Nsukka ethics committee.

## RESULTS

Table 1 compared fluid intake, ALT, AST, urea and the creatinine levels for the experimental rats. Group A had the highest ALT, AST and creatinine levels however, their fluid intake was lower relative to those of groups E and D. These rats fluid intake was higher than the other groups whose intake was 31.16ml and 24.62ml respectively. The fluid intake for group E rats was significantly higher ( $P < 0.05$ ) relative to the other groups. However, there was no significant difference ( $P > 0.05$ ) between their ALT levels when compared with those of groups B and C. Group D rats had the least and most stable levels of ALT (18.08IU/L), AST (75.76IU/L) and creatinine (0.57mg/dl) levels when compared with the other groups. There was no significant difference in urea levels for the rats among the groups ( $P > 0.05$ ).

Table 2 shows the effect of *F. capensis* aqueous extract on the PCV and the WBC counts for the rats. The PCV value for the rats ranged from 41 – 46.62%. Rats in group E had the highest PCV value (46.62%) and those in group D had the least (41.38%). The

**Table 1:** Comparison of fluid intake ALT, AST, creatinine and urea levels in rats

Groups	Fluid intake(ml)	ALT (IU/L)	AST (IU/L)	UREA (mg/dl)	CREATININE (mg/dl)
A	21.12±2.21 <sup>a</sup>	30.28±1.54 <sup>a</sup>	85.17±1.70 <sup>a</sup>	2.96±0.31 <sup>b</sup>	0.68±0.34 <sup>a</sup>
B	20.55±1.28 <sup>a</sup>	24.23±1.52 <sup>b</sup>	78.61±1.90 <sup>ab</sup>	3.86±0.39 <sup>ab</sup>	0.62±0.15 <sup>ab</sup>
C	21.72±0.66 <sup>a</sup>	23.45±0.89 <sup>b</sup>	80.36±1.26 <sup>ab</sup>	3.86±0.29 <sup>ab</sup>	0.64±0.12 <sup>ab</sup>
D	24.62±0.47 <sup>a</sup>	18.08±1.67 <sup>c</sup>	75.76±2.03 <sup>b</sup>	4.32±0.29 <sup>a</sup>	0.57±0.04 <sup>b</sup>
E	31.16±1.57 <sup>b</sup>	22.49±1.26 <sup>b</sup>	78.61±1.73 <sup>ab</sup>	3.56±0.51 <sup>ab</sup>	0.64±0.27 <sup>ab</sup>

Means + SEM of 3 determinations. ; Mean values with different superscripts letters are significantly different (P<0.05). A = Rats fed 100% extract; B = Rats fed 75% extract; C = Rats fed 50% extract; D = Rats fed 25% extract; E = Rats fed water only. ALT=Alanine aminotransferase; AST=Aspartate aminotransferase

**Table 2:** Effect of *F. capensis* extract on PCV and WBC count of rats (%).

Treatments	PCV (%)	WBC (%)
A	43.00±2.20 <sup>ab</sup>	13.07±1.35 <sup>a</sup>
B	44.26±1.65 <sup>ab</sup>	15.80±0.60 <sup>a</sup>
C	42.5±1.43 <sup>ab</sup>	15.36±1.98 <sup>a</sup>
D	41.38±1.41 <sup>b</sup>	15.11±0.53 <sup>a</sup>
E	46.62±0.97 <sup>a</sup>	14.47±1.44 <sup>a</sup>

Means of 3 determinations. ± S.E.M; Mean values with different alphabets as superscripts are significantly different (P<0.05). A = Rats fed 100% extract; B = Rats fed 75% extract; C = Rats fed 50% extract; D = Rats fed 25% extract; E = Rats fed water only. PCV=packed cell volume; WBC= White blood cell counts.

PCV value for group E rats was significantly different (P<0.05) from that of group D and similar to those of groups A, B and C. There was no significant difference (P>0.05) in the PCV of rats in groups A, B, C and D. The white blood cell counts were not significant (p>0.05). The total WBC counts for all the rats' individual values for rats in group B had the highest white blood

cell count 15.80%. The rats in group A had the least WBC count (13.07%).

Table 3 presents the effects of *F. capensis* extracts on differential white blood cell count for the rats. Rats in group A had the highest neutrophils level (29.0%) and those in group B had the least neutrophils level (13.8%). The neutrophils value for rats in group A was significantly different (P<0.05) from those in groups B, C and D. However, the differences were significant (P<0.05) between the neutrophils value for rats in groups A and E. The lymphocyte level for rats in group A was significantly different (P<0.05) from those for the groups B, C and D and comparable to (P>0.05) those in group E. The lymphocyte level for the rats in group B increased to (82.60%) more than that for the other groups. Rats in group A had the least % increase in lymphocytes (68.40%) among the groups.

Monocytes for rats in groups A and B were significantly different (P<0.05) from those in groups C, D and E. There was no marked increase in monocytes for rats in group C. Rats in group A had the highest monocytes level (1.60%) and those in group C had traces of monocytes level (0.00±0.00). There were no significant differences in eosinophils for all the rats. The rats in group E had the highest eosinophils level (2.40%) and those in groups C and D had the least value (1.0%). There was no marked significant difference (P>0.05) basophils levels for rats in all the groups. Rats in group B had the highest basophil 0.02%.

**Table 3:** Effects of *F. capensis* aqueous extracts on differential white blood cell of rats.

Treatment Groups	Neutrophils (%)	Lymphocytes (%)	Monocytes (%)	Eosinophils (%)	Basophils (%)
A	29.00±3.90 <sup>a</sup>	68.40±3.57 <sup>c</sup>	1.60±0.24 <sup>a</sup>	1.60±0.51 <sup>b</sup>	0.00±0.00
B	13.80±0.60 <sup>c</sup>	82.60±0.81 <sup>a</sup>	1.34±0.60 <sup>b</sup>	1.20±0.49 <sup>bc</sup>	0.20±0.20
C	20.80±3.56 <sup>b</sup>	79.00±3.63 <sup>a</sup>	0.00±0.00	1.00±0.55 <sup>c</sup>	0.00±0.00
D	16.80±0.73 <sup>c</sup>	81.80±1.48 <sup>a</sup>	1.00±0.44 <sup>b</sup>	1.00±1.54 <sup>c</sup>	0.00±0.00
E	21.60±3.01 <sup>b</sup>	75.80±3.06 <sup>b</sup>	0.60±0.55 <sup>b</sup>	2.40±0.55 <sup>a</sup>	0.00±0.00

Means of 3 determinations. ± S.E.M; Mean values with different alphabets as superscripts are significantly different (P<0.05). A = Rats fed 100% extract; B = Rats fed 75% extract; C = Rats fed 50% extract; D = Rats fed 25% extract; E = Rats fed water only.



## DISCUSSION

The ALT and AST levels for group A rats fed 100% extract had marked increase relative to the other groups. Tests for ALT are vital to detect liver diseases in dog, cat and primates because this enzyme is in high quantities in liver cells cytoplasm. It increases during cellular degeneration or damage in liver. Serum AST levels are not an organ specific test for liver damage. However, its increased levels were associated with liver diseases in all species<sup>[17, 20-21]</sup>. This slight increase shows that the 100% extract was not safe for consumption even in small concentration. This is true because release of ALT and AST from the cytosol occurs in cellular injury, particularly in membrane damage<sup>[20]</sup>.

Despite the fact that the PCV of this group was higher than that of groups C and Consumption of *F. capensis* extract at 100% level is much more harmful relative to its beneficial effects when consumed for a long time. The increased concentration of neutrophils for rats in group A strongly confirmed tissue damage and inflammation at 100% concentration. It was clear that the much more stability for serum ALT, AST and creatinine at 25% concentration demonstrated that 25% extract has the most membrane stabilizing effects. This appeared to suggest that its consumption for a long time would not have adverse effects. The no increases in both urea and creatinine in all the groups appeared to indicate here was no significant kidney damage. The improved lymphocyte value for the rats meant that the dried leaf extract of *Ficus capensis* could boost the immune system as well as the nutritional status of the consumers. The basophils and eosinophils value for the rat were stable and showed that the plant did not contain harmful substance to cause allergic reactions. Eosinophils and basophils were known to be important during allergic reaction<sup>[21]</sup>.

## CONCLUSION

The extract was good to improve the immune system as well as the nutritional status of the rats. The extract did not impair kidney function however, *Ficus capensis* extract for 100% dose adversely affected the internal liver. When properly diluted *Ficus capensis* dried leaf extract was beneficial to the rat. Further research is required for the work to be replicated and for clinical trials to be conducted.

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