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## Evaluation of Occurrence of Renal Dysfunction in Goats Using Some Biochemical Markers.

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## SUMMARY

This cross-sectional study evaluated the demographical distribution and occurrence of kidney dysfunction in goats in Umuahia, Abia state, using known biochemical markers. A total of 145 goats presented for slaughter at the Ahiaeke abattoir were evaluated. Prior to slaughter, sex, age, breed and health status of the goats were determined. Blood was collected from the jugular vein at slaughter for haematology and serum biochemical analyses. Urine was collected from the bladder for urinalysis using test strips. Standard procedures were adapted for all the analyses performed. Results showed that 56.6 % of the goats were females, 89 % were adults, 92.4 % were Red Sokoto breed and 95.2 % were apparently healthy. Out of 145 goats sampled, 3 (2.1 %) were diagnosed with kidney dysfunction based on positive serum creatinine and urine protein levels, while 10 (6.9 %) had positive serum urea values. There was significant association (P and < 0.05) between the health status of goats and all the biochemical parameters measured to ascertain renal dysfunction. No such associations exist between sex, breed and the biochemical markers measured. We conclude that kidney dysfunction exists in Nigerian goats, and health status is a determinant of renal dysfunction. **Keywords** : renal biomarkers, health status, sex, breed, age, goat

## **INTRODUCTION**

The domestic goat (*Capra aegagrus hircus*) is a member of the animal family Bovidae and the subfamily Caprinae, meaning it is closely related to the sheep, with 300 distinct breeds (Ezewudo *et al.*, 2017). Goats are one of the oldest domesticated species of animal, and have been used for milk, meat, fur and skins across much of the world (Wilson, 1987). They play an important role in both health and economy as they are not affected by any religious and cultural taboo and so, are reared and consumed in areas where beef,

pork and any other livestock are forbidden (Adeyeye and Adewumi, 2015).

The kidney is one of the vital organs in the mammalian body and is responsible for intricate aspects of homeostasis: regulation of water and electrolytes homeostasis, product excretion, acid-base waste homeostasis, regulation of arterial blood synthesis pressure, of vitamin D. gluconeogenesis, and hormone secretion (Ihedioha et al., 2019).

Renal dysfunction is routinely diagnosed using biomarkers. A biomarker is measured and evaluated as an indicator of normal biological, pathologic processes, or pharmacologic responses to a therapeutic intervention (Ramachandran, 2006) and they play a role in accurate diagnosis, prognosis and treatment of diseases (Gowda etal., 2010).Creatinine, urea, uric acid and electrolytes are used for routine diagnosis of renal dysfunction, and more recently, cystatin and  $\beta$ -Trace protein (Gowda etal., 2010) as well as Kidney Injury Molecule-1, interleukin-18, Nacetyl- $\beta$ -D-glucosaminidase, and L-type fatty acid-binding protein (L-FABP) (Andreucci et al., 2016). Creatinine is produced as the result of normal muscle metabolism. Phosphocreatine, an energystoring molecule in muscle, undergoes spontaneous, non-enzymatic cyclization to form creatine and inorganic phosphorous. Creatine then decomposes to creatinine. In health, production and excretion of creatinine are fairly constant in an animal, resulting individual in low variation (Zuo et al., 2008), it isless influenced by extra-renal factors like hydration and so, is a more accurate biomarker. Urea is produced by the breakdown of proteins and is excreted in the urine and because its level is affected by circadian rhythm, diet, liver function, hydration, and intestinal absorption, it is less accurate as a biomarker (Rosner and Bolton, 2005). Persistent proteinuria that is, presence of protein in the urine, in the absence of lower urinary tract disease or reproductive tract disease, is usually an indication of renal damage or dysfunction (Harley and Langston, 2012).

Renal diseases are important clinical problems and are causes of illness and death in many animals' species, including goats (Mahouz *et al.*, 2015). Although, there is a global increase in kidney damage in man (Lucykx, 2018), reports of kidney damage in animals are scant. However, studies on the prevalence of renal disease in goats currently available in literature were in other climes and mostly abattoir studies using gross lesions (Ansari-Lari, 2007; Dutta *et al.*, 2016). In Nigeria,

however, kidney disease has been reported in cattle (Ihedioha *et al.*, 2019) and there are hitherto, no reports of kidney disease in Nigerian goats known to us. Also, kidney studies using biomarkers for renal disease to detect renal dysfunction in goats are not readily available in literature. This study provides these data.

## MATERIALS AND METHODS

## Study location.

The study was carried out in the goat abattoir in Ahiaeke, Umuahia North Local Government Area, and Abia State, Nigeria. The coordinates of Ahiaeke abattoir are 5.5120° N, 7.5300° E.

## Study Design

The study was a cross-sectional survey of goats presented for slaughter at the abattoir. Industrial Ahiaeke Market. Umuahia. The study design was a crosssectional survey, and was approved by the Institutional Animal Care and Use Committee (IACUC) of the Faculty of Veterinary Medicine, University of Nigeria Nsukka, Nigeria.

## Animals

The study populations include all goats slaughtered at the Ahiaeke goat abattoir between November 2019 and April 2020. Research visits to the abattoir was done once in a week, and all slaughtered goats during the days of research visit made up the sample population. A total of 145 goats were evaluated. The goats were examined physically before the slaughtering, and data on sex, age, and breed and health status were collected. At slaughter, blood was drawn from the jugular vein and urine samples were collected from the bladder of each of them.

## Sample Collection

Blood for haematology and serum biochemistry analyses were taken immediately to the Veterinary Biochemistry Laboratory, Michael Okpara University of Agriculture, Umudike. Blood samples for haematology and fibrinogen determination were collected in sample bottles containing ethylenediaminetetracetic acid (EDTA) anticoagulant, while those for serum biochemistry were dispensed into a plain glass tubes and allowed to stand for 45 minutes to clot. Clotted blood was separated from serum by centrifugation at 3000 revolutions per minute (rpm) for 10 minutes. Part of the blood collected into EDTA container was centrifuged at 3000 rpm to obtain plasma for fibrinogen determination. Urine for urinalysis was obtained from the bladder at the point of slaughter by cystocentesis (Yam, 1994).

## Laboratory Methods

Serum creatinine levels were assayed using the modified Jaffe method (Blass et al., 1974) using Quimica Clinica Applicada (OCA) Creatinine test kit (QCA, Spain). Serum urea levels were determined by the modified Berthelot-Searcy method (Lamb and Price 2008) using the Dialab Urea (urease/colorimetric) test kit (Dialab, Neudorf, Austria).

Urine protein was assayed using Combostik® 11 urinalysis reagent test strips (DFI Co. Ltd., Korea) based on the tetrabromophenol blue colorimetric method.

## Data Analysis

Data was analysed using SPSS; occurrence was determined using descriptive statistics and presented as percentages. Possible associations between renal impairment and age or sex was analysed using Fisher's exact test, and significance was accepted at p < 0.05. Haematological and serum biochemical profile of goats with kidney dysfunction were compared with that of apparently healthy goats using Student's t test.

## RESULTS

## Demography of sampled goats.

The distribution of sampled goats according to sex, age, breed and health status is presented in Table I. Female goats accounted for 56.6 % of 145 sampled goats, while 89.0 % of the sampled goats were adults. The predominant breed was the Red sokoto (92.4 %) while 95.2 % of goats were apparently healthy (Table I).

Demographic Characteristic	Categories	No. out of the total sample population of 145	Percentages	
Sex	Females	82	56.6%	
	Males	63	43.4%	
Age	Adults	129	89.0%	
-	Young	16	11.0%	
	Mixed	3	2.1%	
Breeds	Red Sokoto	134	92.4%	
	Sahel	8	5.5%	
	Apparently Healthy	138	95.2%	
Health Status	Obviously	7	4.8%	
	Unhealthy			

TABLE I. Demographic characteristics of the sample population of goats evaluated.

## Occurrence of renal dysfunction

Cut-off points for serum creatinine, serum urea and urine protein (Njidda *et al*, 2013); were used to determine the occurrence of renal dysfunction in sampled goats. Goats with serum creatinine higher than 120 mg/kg were classified as having renal dysfunction and they made up 2.1 % of the sampled goats; goats with serum urea greater than 42. 0 mg/kg were positive and made up 6.9 % of the sampled goats while

were positive (Table II).

TABLE II. Occurrence of renal dysfunction based on the levels of serum creatinine, serum urea
and urine protein cut off points*.

Parameter	Categories	No. out of the total of 145	Percentages
Serum Creatinine (mg/dl)	$\geq$ 1.20 (positive)	3	2.1%
	< 1.20 (negative)	142	97.9%
Serum Urea (mg/dl)	$\geq$ 42.0 (positive)	10	6.9%
	< 42.0 (negative)	135	93.1%
Urine Protein (mg/dl)	$\geq$ 1000.0 (positive)	3	2.1%
	< 1000.0 (negative)	142	97.9%

\* Njidda et al. 2013

Relationship between sex and occurrence of renal dysfunction.

Based on positive serum creatinine levels, 1.2 % of 82 female goats had renal dysfunction, while 3.2 % of 63 male goats sampled were positive. Based on positive serum urea levels, 3.7 % of female goats sampled had renal dysfunction while 11.1 % of male goats were positive. Based on urine protein, 1.2 % of female goats, and 3.2 % of male goats had renal dysfunction. However, there were no significant correlation between sex and occurrence of renal dysfunction based on serum creatinine, serum urea and urine protein (Table III).

 TABLE III. Contingency table showing the relationship between sex and occurrence of renal dysfunction in the goats sampled.

Based on Ser	um Creat	inine levels:							
	No. w	vith serum	No.	with se	erum T	Total of	f each sex	Percentag	ge of
Sex	creatini	ne $\geq$ 1.20	creat	tinine <	1.20 in	n the	sample	each	sex
	mg/dl (	positive)	mg/c	ll (negativ	e) p	opulati	on	positive	
Females	1		81		8	32		1.2%	
Males	2		61		6	i3		3.2%	
<b>Totals</b>	3		142		1	45			
No significar	t associat	ion between	sex a	nd occurr	ence of	renal d	ysfunction	based on s	serum
creatinine lev	els, $p = 0$ .	58					-		
Based on Serum Urea levels:									
	No. w	vith serum	No.	with se	erum T	Total of	f each sex	Percentag	ge of

	No. with serum	No. with serum	Total of each sex	Percentage of
Sex	urea $\geq 42.0 \text{ mg/dl}$	urea < $42.0 \text{ mg/dl}$	in the sample	each sex
	(positive)	(negative)	population	positive
Females	3	79	82	3.7%
Males	7	56	63	11.1%
<b>Totals</b>	10	135	145	

No significant association between sex and occurrence of renal dysfunction based on serum urea levels, p = 0.103

	No. with urine	No. with urine	Total of each sex	Percentage of
Sex	protein $\geq$ 1000	protein < 1000	in the sample	each sex
	mg/dl (positive)	mg/dl (negative)	population	positive
Females	1	81	82	1.2%
Males	2	61	63	3.2%
Totals	3	142	145	

No significant association between sex and occurrence of renal dysfunction based on urine protein levels, p = 0.58

No significant association (p > 0.05) between sex and occurrence of renal dysfunction in the goats sampled.

## Relationship between breed and occurrence of renal dysfunction

Table IV shows the relationship between breed and occurrence of renal dysfunction in sampled goats. The red sokoto breed accounted for all the positive cases (2.2%) on the basis of serum creatinine; the sahel breed recorded the highest number of positive serum urea cases (12. 5%); while the positive cases based on urine protein was mainly attributable to the red sokoto goats (2.2%). These were however not statistically significant (P > 0.05).

Based on Serum (	nction in the goats sa Greatining levels:	inpicat			
<i>Buseu on Serum</i> (		No. with serum	Total of cosh based in	Demonstere	
Durad	No. with serum		Total of each breed in	Percentage o	
Breed	creatinine $\geq 1.20$	creatinine $< 1.20$	the sample population	each breed	
	mg/dl (positive)	mg/dl (negative)		positive	
Mixed	0	3	3	0%	
Red Sokoto	3	131	134	2.2%	
Sahel	0	8	8	0%	
Totals	3	142	145		
No significant a	ssociation between bree	d and occurrence of ren	al dysfunction based on se	rum creatinine	
		levels, p = 0.882			
Based on Serum L	Jrea levels:				
	No. with serum urea	No. with serum urea	Total of each breed in	Percentage of	
Breed	≥ 42.0 mg/dl	< 42.0 mg/dl	the sample population	each breed	
	(positive)	(negative)		positive	
Mixed	0	3	3	0%	
Red Sokoto	9	125	134	6.7%	
Sahel	1	7	8	12.5%	
Totals	10	135	145		
No significant ass	ociation between breed a	and occurrence of renal	dysfunction based on seru	m urea levels, p =	
0		0.733	,	, 1	
Based on Urine Pr	rotein levels:				
	No. with urine	No. with urine	Total of each breed in	Percentage of	
Breed	protein ≥ 1000	protein < 1000	the sample population	each breed	
	mg/dl (positive)	mg/dl (negative)		positive	
Mixed	0	3	3	0%	
Red Sokoto	3	131 134		2.2%	
Sahel	0	8 8		0%	
Totals	3	142	145		
No significant ass	ociation between breed a	and occurrence of renal	dysfunction based on uring	e protein levels, p	
-		= 0.882	-	. ,	

TABLE IV. Contingency table showing the relationship between breed and occurrence of renal dysfunction in the goats sampled.

No significant association (p > 0.05) between breed and occurrence of renal dysfunction in the goats sampled.

# RELATIONSHIP BETWEEN HEALTH STATUS AND OCCURRENCE OF RENAL DYSFUNCTION IN GOAT

Obviously unhealthy goats accounted for 28.6 %, 42.9 % and 28.6 % of sampled goats, on the basis of positive serum creatinine, serum urea and urine protein levels respectively (Table V). These associations were significant (P < 0.05).

## TABLE V. Contingency table showing the relationship between health status and occurrence of renal dysfunction in the goats sampled.

There was a significant association (P < 0.05) between health status and occurrence of renal dysfunction in the goats sampled.

Based on Serum (	Creatinine levels:			
	No. with serum	No. with serum	Total of each health	Percentage of
Health status	creatinine $\geq$ 1.20	creatinine < 1.20	status in the sample	each health
	mg/dl (positive)	mg/dl (negative)	population	status positive
Apparently	1	137	138	0.7%
healthy				
Obviously	2	5	7	28.6%
unhealthy				
Totals	3	142	145	
There was a sign	ificant association betwee		-	nction based on
		n creatinine levels, p = 0	0.006	
Based on Serum l				
	No. with serum urea	No. with serum urea	Total of each health	Percentage of
Health status	≥ 42.0 mg/dl	< 42.0 mg/dl	status in the sample	each health
	(positive)	(negative)	population	status positive
Apparently	7	131	138	5.1%
healthy				
Obviously	3	4	7	42.9%
unhealthy				
Totals	10	135	145	
There was a sign	ificant association betwee			nction based on
		rum urea levels, p = 0.00	07	
Based on Urine P				
	No. with urine	No. with urine	Total of each health	Percentage of
Health status	protein ≥ 1000	protein < 1000	status in the sample	each health
	mg/dl (positive)	mg/dl (negative)	population	status positive
Apparently	1	137	138	0.7%
healthy				
Obviously	2	5	7	28.6%
unhealthy				
Totals	3	142	145	
There was a sig	nificant association b	etween health status	and occurrence of rer	al dysfunction
	based on	urine protein levels, I	P = 0.006	

## DISCUSSION

The results from the demographic study showed that female goats were more predominant than the males in the sampled animals. The predominance of female animals in slaughter houses has been documented by previous studies (Berhanu *et al.*, 2012, Okorie-Kanu *et al.*, 2018, Aba *et al.*, 2020). This trend is an evidence of the primordial nature of livestock production in our environment. In more organized livestock production as seen in developed economies, female animals are rarely slaughtered. It could also be due to a preference by the buyers or cost implication (Aba *et al.*, 2020). However, female goats were more positive for all the markers of kidney function assayed than male goats.

Most of the goats slaughtered were apparently healthy. This is in contrast with reports from abattoir studies where many animals slaughtered had poor body condition scores (Lamy *et al.*, 2012; Shittu *etal.*, 2014). Animals presented for slaughter in abattoirs are usually stressed probably due to transportation and disease. However, from our study, goats that are apparently healthy were predominant. This could be due to the fact that the slaughter house is located right within the goat market and people purchase these healthy goats and take them directly to the abattoir. Red Sokoto breed of goats were the most predominant. This agrees with previous findings (Shittu et al., 2014; Okorie-Kanu et al., 2018). The predominance of Red sokoto goats over the local breeds could be attributable to the demand for animal protein, hence the procurement of large sized breeds over the small sized local breeds.

There were more positive urea cases than positive creatinine in the goats. Creatinine is more specific for renal dysfunction than urea because the amount of creatinine secreted daily is a function of the muscle mass. It amounts to approximately 2% of the body stores of creatinine phosphate and is roughly 1-2g/day for adult (Njidda et al., 2013). The diagnosis of renal failure is usually suspected when serum creatinine is greater than the upper limit of the "normal" interval. In chronic renal failure and uraemia, an eventual reduction occurs in the excretion of creatinine by both the glomeruli and the tubules (Edmund and David, 2006). However, there are reports that creatinine values may alter as its generation may not be simply a product of muscle mass but influenced by muscle function, muscle composition, activity, diet and health status (Banfi and Del, 2006). The increased tubular secretion of creatinine in some patients with kidney dysfunction could give false negative value (Branten et al., 2005). The Jaffe reaction creatinine assay is affected by substances non-creatinine including glucose, uric acid, ketones, cephalosporins, furosemide. hemoglobin. paraproteins. paraquat, and diquat which may lead to false elevations in serum creatinine values (Stevens et al., 2007). Furthermore, Andreucci (2016) opined that it is both a

achieved

late and indirect marker of kidney injury Consequently, (Andreucci, 2016). creatinine alone is not enough to test for kidney function. Therefore, assay of urine protein was done to determine if goats with positive creatinine also had proteinuria. From the results, goats with positive creatinine also had positive proteinuria; although the normal alkaline pH of goat urine could influence protein reaction and lead to elevated protein levels (Stockham and Scott, 2008). Proteinuria can be physiological (following a protein meal) or pathological as seen in acute renal disease and is a sensitive biomarker for drug induced kidney injury (Griffin et al., 2019). Therefore, simultaneous diagnosis with positive creatinine is highly indicativeof renal dysfunction. Urea, on the other hand, is a metabolite synthesized from ammonia in the liver during protein metabolism and is largely dependent on protein intake (Rosner and Bolton, 2005). is increased in many non-renal It conditions, including dehydration, heart failure, use of drugs like tetracycline and corticosteroids (Luke, 1981). This could explain the occurrence of more positive urea cases, compared to other biomarkers. Therefore, positive urea cases alone are not sufficient to diagnose renal disorders. There were significant associations (P ≤0.05) between health status and occurrence of renal dysfunction. Health status was determined by body condition, and visibly emaciated goats had renal dysfunction, based on creatinine, urea and urine protein levels. Studies have shown that renal diseases rarely present with signs and symptoms at the early stages (Rosner and Bolton, 2005). The implication is that by the time the patient presents with signs, the damage to the kidney would have been severe and the body condition would have

deteriorated. Renal biomarkers therefore.

are very useful in detection of early renal

disease so that effective treatment can be

#### CONCLUSION

It was concluded that the occurrence of renal dysfunction in the Red Sokoto goats sampled in this study is 2.2 % based on serum creatinine and urine protein RI, and the occurrence was significantly associated with the health status of the goats, but not with their age or sex.

A major limitation of this study is the absence of an abattoir exclusively for slaughtering of goats in the study area. This made sample collection very difficult, and is of urgent public health significance as there is absence of proper meat inspection for goat meats slaughtered in the study area. It will also be impossible to carry out retrospective studies due to absence of data on goats slaughtered.

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