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ABSTRACT:

Carica Papaya (CP) plant (paw paw) is largely used for its curative benefit and now being exploited as an anti-fertility agent. The testicular histomorphometric correlation visa versa function is yet to be fully understood. This study aimed at quantifying the effects of aqueous extract of the bark of CP on the testes of adult Sprague–Dawley (S-D) rats. Ninety adult 6-8 weeks old male S-D rats were divided into nine groups [$1DW_{(4wk)}$, $1CP_{50(4wk)}$, $1CP_{100(4wk)}$, $2DW_{(8wk)}$, $2CP_{50(8wk)}$, $2CP_{100(8wk)}$, $3DW_{(16wk)}$, $3CP_{50(16wk)}$ and $3CP_{100(16wk)}$] of 10 rats per group. Rats in groups $1DW_{(4wk)}$, $2DW_{(8wk)}$, and $3DW_{(16wk)}$ served as control and were treated with distilled water (DW) for 4, 8 and 16 weeks respectively. Rats in groups $1CP_{50(4wk)}$, $2CP_{50(8wk)}$ and $2CP_{50(16wk)}$ were fed 50 mg/ml/day CP, while those in groups $1CP_{100(4wk)}$, $2CP_{100(8wk)}$ and $2CP_{100(16wk)}$ were fed 100 mg/ml/day CP. Rats in groups $2CP_{50(16wk)}$ and $2CP_{100(16wk)}$ compared to those in $3DW_{(16wk)}$ were observed for possible reversibility after 8 weeks of withdrawal of the CP extract. Rats were sacrificed after the appropriate duration and testicular histological sections prepared for histometric analysis. Stereological parameters estimated were; tubular diameter, cross sectional area of seminiferous tubules, volume density, number of profiles per unit area, absolute volume of seminiferous tubules and testicular interstitium, numerical density, length density and star volume of the seminiferous tubules. The result showed dose and duration dependent decrease in mean testicular volume, tubular diameter, cross sectional area and star volume of tubules. A converse increase in the length density, numerical density, number of profiles per unit area and volume density of tubules was also observed. Alteration in the histomorphometric data indicates that the CP bark extract can cause impairment in spermatogenesis.

Key words: *Carica papaya,* Rats, Testes, Histomorphometric, Spermatogenesis

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INTRODUCTION:

The primary intent in biological research whether by default or design, is to compare structure with function. The findings grants explanation to why organizational levels are made the way they are and the reason for their varied functions. Conscious attempts have been made in the past to understanding the 3-dimensional (3-D) morphology of the rats' testis and the organizational arrangements [1]. The findings relating its structure to function have remained vague [1]. A clearer understanding is provided when more accurate cytometric quantification is employed. It counts and measures structures using formidable but simple stereological tools [2].

A great deal of first-order stereological information exists for the numbers, sizes and component densities of the various tissue compartments of the testes under normal and experimental conditions [3-6]. These methods are based on random intersections between a geometric probe and the object of interest. Several studies have estimated length of biological objects, including length of seminiferous tubules and capillaries in cerebral cortex [7, 8]. In many biological applications, however, tissue landmarks are difficult to recognize following randomization around one or more axis as required for vertical uniform random (VUR) slices and isotropic uniform random (IUR) plane sections. A recent method that avoids this caveat uses virtual isotropic planes to probe linear objects on arbitrary thick sections [9].

Hitherto, there has been a paucity of complementary quantitative information concerning the 3-D arrangement of testicular compartments. However, with the advent of quantitative methods for exploring second-order stereology, it is now possible to examine associations in various spatial and directional distributions [10-12]. The stereological tools allow quantitative 3-D estimates to be obtained from 2-D slices histological sections of a tissue sample. In a recent biological application on rat testis the analysis of stereological parameters provided quantitative description of differences in 3-D arrangements of testicular components [13]. While direct observations are made from 2-D sections, 3-D information is obtained through mathematically proven relationships [10].

In this study, design-based stereological methods were used to analyze testicular tissues of Sprague-Dawley (S-D) rats treated with *Carica papaya* (CP) bark extract. The findings are expected to contribute considerably in understanding the functional and pathological morphologies in the testes.

MATERIALS AND METHODS:

Ninety adult male S-D rats weighing between 180-250 g were used. They were procured

from a breeding stock, maintained in the Animal house of the College of Medicine, University of Lagos (CMUL) Nigeria. They were allowed to acclimatize for 2 weeks in the animal control room Department of Anatomy CMUL, with an ambient temperature maintained between 26-28ºC. They were permitted free access to water and food *ad libitum.*

Plant Source and Extract Pharmacognosy: The bark of the CP plant was obtained from a forest in Lagos Nigeria in the month of September. It was authenticated in the Department of Botany, University of Lagos, Nigeria where the voucher specimen was deposited (ascension number LUH 2151). Aqueous extraction of the bark was carried out in the Pharmacognosy Department, Faculty of Pharmacy University of Lagos, Nigeria. Briefly, the completely oven-dried (40˚C for 4 days) bark (350g) was crushed and powdered. The powder was placed in distilled water and allowed to boil, simmering for one hour. The water extract was dialyzed and the internal solution lyophilized.

The residue obtained (4.05g, 1.16% yield) were stored at 4°C before use. When required, the residues were dissolved in distilled water and the desired pharmacological concentrations administered based on the animal's individual body weight.

Experimental protocol: The rats were randomized into 9 groups (10 rats per group) identified as: $1DW_{(4WK)}$, $1CP_{50(4WK)}$, $1CP_{100(4WK)}$, $2DW_{(8WK)}$ $2CP_{50(8WK)}$, $2CP_{100(8WK)}$, $3DW_{(16WK)}$, $3CP_{50(16WK)}$ and $3CP_{100(16WK)}$ respectively. The groups consist of 3 treatment intermissions: 4, 8 and 16 weeks. The rats in groups $1DW_{(4WK)}$, $2DW_{(8WK)}$, and $3DW_{(16WK)}$ (control groups) were used to compared events in the other groups.

- Groups: $1DW_{(4WK)}$, $1CP_{50(4WK)}$ and $1CP_{100(4WK)}$ were treated with 5.0 ml distilled water (DW), 50 and 100 mg/ml/kg/day CP bark extracts orally for 4 weeks.
- Groups: $2DW_{(8WK)}$, $2CP_{50(8WK)}$ and $2CP_{100(8WK)}$ were treated with 5.0 ml DW, 50 and 100 mg/ml/kg/day CP bark extracts orally for 8 weeks.
- Groups: $3DW_{(16WK)}$, $2CP_{50(16WK)}$ and $2CP_{100(16WK)}$ were treated with 5.0 ml DW, 50 and 100 mg/ml/kg/day CP bark extracts orally for 8 weeks. The bark extract of CP discontinued and treated subsequently with 5.0 ml DW alone for another 8 weeks.

At the end of the 4th, 8th and 16th week experimental periods, the rats in the respective groups were sacrificed. The experimental intervals were guided by the period taken to complete a spermatogenic cycle in rat [15].

Preparation of tissues for Histological Study: Rats were sacrificed according to the methods described previously by Osinubi *et al* [23]. The rats were made unconscious by cerebral dislocation; this was followed by ventral laparotomy to gain access to the testes via the abdomen; the testes were excised,

weighed and fixed in Bouin's fluid and processed for morphometric studies. After 48 hours the organs were removed from Bouin's fluid and further fixed in fresh Bouin's fluid for another 72 hours [23]. Fixed testes were dehydrated through graded series of ethanol, cleared in chloroform, and infiltrated and embedded in molten paraffin wax. Before embedding, sections were orientated perpendicular to the long axis of the testes and designated as vertical sections. Serial sections obtained from the solid block of tissue attached to a wooden holder that was fixed to a manually operated microtome and sections were cut at 5.0µm. Selected sections were mounted and subsequently stained by Haematoxylin and Eosin staining techniques for light microscopic examination [23].

Stereological measurements: The dissected testes were separated out from the adherent tissue and weighed (TW) to the nearest mg on an electronic balance [5, 6]. The testicular volume (TV) estimated by water displacement method [5, 6]. Seven sections per testis were selected by systematic sampling method that ensured fair distribution between the polar and equatorial region of each testis [1, 2]. This allowed unbiased numerical estimation of morphometric parameters. The diameter of seminiferous tubules (D) with profiles that were round or nearly round were measured for each animal and the mean determined by taking average of two diameters at right angles, D_1 and D_2. They were taken only when $D_1/D_2 \geq 0.85$ this is to abolish different degrees of profile irregularities or tissue contraction. The cross sectional areas (A_C) of the seminiferous tubules were determined from the formula $A_C = D^2 \times \pi/4$; i.e. multiplying the mean of the squared diameters of the seminiferous tubules (D^2) by a constant ($\pi/4$) where π is equivalent to 3.142. The number of profiles of seminiferous tubules per unit area (NA) was determined using the unbiased counting frame proposed by Gundersen [14]. Using the frame, we counted all profiles completely inside and any part inside the frame provided they do not touch or intercept the forbidden line (full-drawn line) or exclusion edges or their extension. Point counting methods were utilized to obtain the volume density (V_V), which is the volume of the seminiferous tubules/unit area of testis [15]. The number of profiles *per* unit volume (N_V) was determined by using the modified Floderus equation: $N_V = N_A / (D+T)$ [16]. Where N_A is the number of profiles per unit area and D is the diameter and T is the average thickness of the section. The length density (L_V) was determined using the equation $2xN_A$ [16]. The star volume (V*) was calculated from the equation $Vv = \pi/3 \times \text{mean } lo^3$ [17, 18]. The star volume of the seminiferous tubules provides a direct and unbiased estimate of volume which has a strict mathematical definition i.e. the volume of all parts of a 3-dimension space

which are visible on every direction from a given point within it.

Statistical analysis: Data were expressed as mean ± standard deviation (SD). Analyses of variance (ANOVA) with Scheffer's post-hoc test were used to analyze the significance of difference and a probability of $p < 0.05$ was considered significant. This was done using the SPSS software package.

RESULTS:

Histological analysis: The administration of CP bark extract to S-D rats for 4 and 8 weeks showed significant alteration in the histology of the testis (Figures 1 and 2). The seminiferous tubules of control rats contained germ cells up to the level of spermatozoa (Figure 1a, 2a, and 3a). In the samples from rats treated with low dose for 4 weeks, the seminiferous tubules showed focal areas with marked hypospermiation and coagulative necrosis of the seminiferous tubules (Figure 1b) while the high dose showed an extensive necrosis of the seminiferous tubules and damage to the germ cells (Figure 1c). There was also destruction to the basement membrane, focal area of disorganization and sloughing. The low and high doses given for 8 weeks (Figure 2b and 2c) showed a more extensive damage with the nuclei of the cells not seen and very scanty Leydig cells. The reversibility after a period (8 weeks) of discontinuation of extract treatment showed some degree of recovery compared to their control counterpart (Figure 3).

Morphometric correlation: The diameters of the seminiferous tubules revealed differences between controls and CP treated rats. Some tubules presented a distorted shape which made it difficult to take the measurement. Consequently only circular profiles were considered in each group, as suggested by Cruz-Orive and Gual-Arnau [19]. The possibility that some tissue shrinkage occurred as a result of the fixation cannot be discounted. However all tissues were treated under the same condition of fixation, embedding and sectioning. It is believed therefore that the differences were most likely due to the different treatments used in the study.

Tubular Diameter: There was a dose-dependent and duration-dependent reduction in the tubular diameter of the experimental rats from 215.6 ± 1.66 v. 183.6 ± 8.0 v. 152.9 ± 27.4 for the 4 weeks group and from 198.0 ± 6.50 v. 173.2 ± 9.7 v. 147.3 ± 16.5 for the 8 weeks group compared to the control. The reversal also showed a dose-dependent decrease in the tubular diameter from 188.8 ± 9.4 v. 160.3 ± 9.0 v. 145.9 ± 5.3 compared with the control (Table 1).

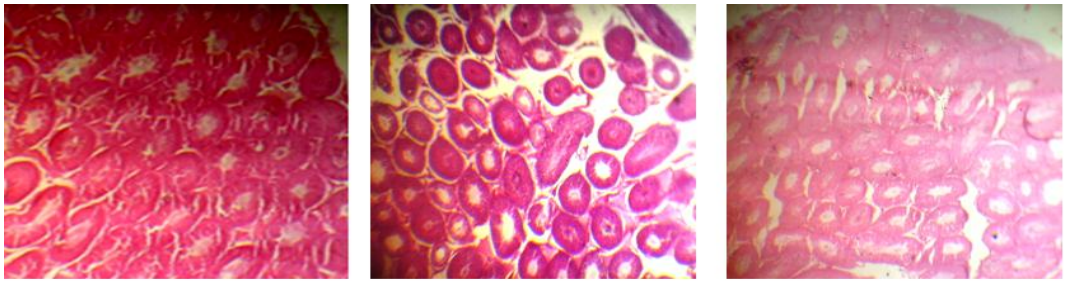

Figure 1: Cross section of testis of (a) control (5 ml of distilled water), (b) Low dose (50 mg/ml/kg/day) and (c) High dose (100 mg/ml/kg/day) at 4 weeks; stained with Haematoxylin and Eosin; Magnification x40

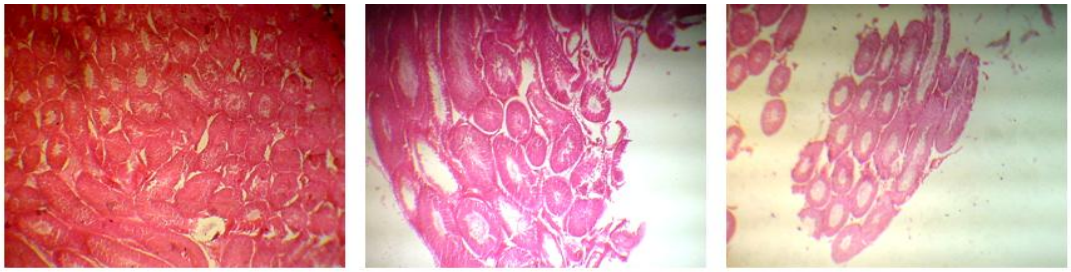

Figure 2: Cross section of testis of (a) control (5 ml of distilled water), (b) Low dose (50 mg/ml/kg/day) and (c) High dose (100 mg/ml/kg/day) at 8 weeks; stained with Haematoxylin and Eosin; Magnification x40

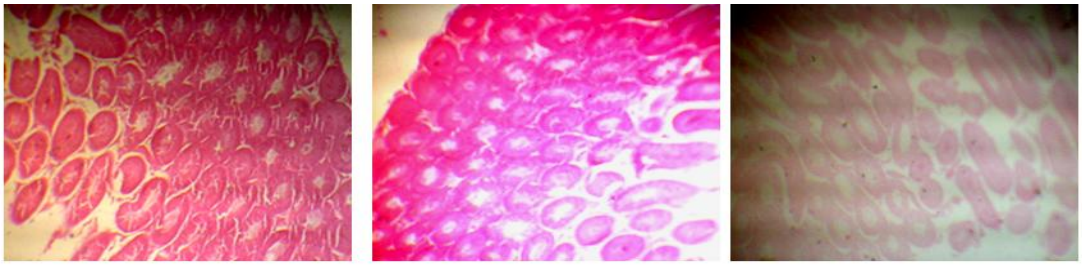

Figure 3: Cross section of testis of (a) control (5 ml of distilled water), (b) Low dose (50 mg/ml/kg/day) and (c) High dose (100 mg/ml/kg/day) at 8 weeks, left for another 8 weeks to assess reversibility; stained with Haematoxylin and Eosin; Magnification x40

Table 1: Effects of the extract of *Carica papaya* bark on the testicular diameter, weight, volume and cross sectional area of the seminiferous tubules of Sprague-Dawley rats for 4 wks, 8 weeks and reversal groups

DOSES	TD (µm)	TW (g)	TV (ml)	A_C (x10^3 µm^2)
		4 Weeks duration		
Control	215.0± 1.6[a]	1.20± 0.55	1.0 ± 0.10	36.51± 6.56
Low dose	183.6± 8.0	1.23± 0.67	0.9 ± 0.49	26.47± 4.62
High dose	152.9± 27.4	1.25± 0.40	0.8 ± 0.35	17.04± 4.66
		8 Weeks duration		
Control	198.0± 6.5	2.0± 0.95	1.0 ± 0.10[b]	30.79± 8.34
Low dose	173.2± 9.7	1.9± 0.70	0.7 ± 0.24	23.56± 7.40
High dose	147.0± 16.5	1.8± 0.80	0.7 ± 0.24	15.06± 4.22
		16 Weeks duration (Reversal)		
Control	188.8± 9.4	2.0± 0.65	0.9 ±0.49	20.03± 2.42
Low dose	160.3± 9.0	1.8± 0.50	0.9 ± 0.49	18.42± 1.80
High dose	145.9± 5.3	1.3± 0.45	0.7 ± 0.24b	16.53± 1.20

Control: 5.0 ml of distilled water; Low dose: 50 mg/ml/kg/day of *Carica papaya*; High dose: 100 mg/ml/kg/day of *Carica papaya*; a = mean ± S.D; b = $p < 0.05$; n = 10; TD =Tubular diameter; TW = Testicular weight; TV = Testicular volume; A_C = Cross sectional area.

Testicular Volume: Although there was a dose-dependent reduction in the testicular volume of the experimental rats from 1.0 ± 0.1 v. 0.9 ± 0.49 v. 0.8 ± 0.35 for the 4 weeks group and from 1.0 ± 0.1 v. 0.7 ± 0.24 v. 0.7 ± 0.24 for the 8 weeks group compared with the control; the reductions were not significantly different. For the reversal group, the testicular volume was also not significantly different (Table 1).

Cross sectional area of the seminiferous tubules: There was a dose and duration dependant reduction of the cross sectional area of the seminiferous tubules of the experimental rats from 36.51 ± 6.56 v. 26.47 ± 4.62 v. 17.04 ± 4.66 for the 4 weeks group and from 30.79 ± 8.34 v. 23.56 ± 7.40 v. 15.06 ± 4.22 for the 8 weeks group. There was a decrease from 20.03 ± 2.0 v. 18.42 ± 1.8 v. 16.53 ± 1.2 with the reversal group compared with the control (Table 1).

Number of profiles of seminiferous tubules per unit area of testis: The experimental rats showed an increase in the number of profiles per unit area from 12.1 ± 2.03 v. 13.0 ± 2.63 v. 13.4 ± 3.61 for the 4 weeks group and from 14.4 ± 3.39 v. 15.9 ± 4.38 v. 19.9 ± 6.23 for the 8 weeks group compared with the controls.

The increase was dose and duration dependent. The reversal group also showed an increase from 18.2 ± 3.08 v. 24.9 ± 6.80 v. 30.1 ± 11.8 (Table 2).

Table 2: Effects of the extract of *Carica papaya* bark on the number of profiles per unit area, numerical density, length density and the star volume of the seminiferous tubules of Sprague-Dawley rats for 4 wks, 8wks and reversal groups

DOSES	N/A (x10 µm^{-2})	NV (x10^{-10} µm^{-2})	LV (x10^{-8} µm)	SV (x10^{6} µm)
		4 Weeks duration		
Control	12.1± 2.03	55.0± 9.38	24.2± 2.03	10.50± 0.22b
Low dose	13.0± 2.63	68.9± 10.4	26.0± 2.63	6.48± 0.19
High dose	13.4± 3.61	97.2± 7.57	26.8± 3.61	2.46± 0.17
		8 Weeks duration		
Control	14.4± 3.39[a]	70.9± 8.53	28.8± 3.39	8.13± 0.21
Low dose	15.9± 4.38	89.3± 8.24	31.8± 4.38	5.44± 0.15[b]
High dose	19.9± 6.23	130.7±16.43	39.8± 6.32	3.35± 0.12
		16 Weeks duration (Reversal)		
Control	18.2± 3.08	18.5± 2.05	30.4± 27.0	12.76± 4.30
Low dose	24.9± 6.80	18.5± 1.30	49.8± 8.86	23.48± 4.80
High dose	30.1± 11.8	19.9± 1.20	60.2± 23.2	32.06± 3.50

Control: 5.0 ml of distilled water; Low dose: 50 mg/ml/kg/day of *Carica papaya*; High dose: 100 mg/ml/kg/day of *Carica papaya*; a = mean ± S.D; b = $p < 0.5$; n = 10; NA = Number of profiles per unit area; NV = Numerical density; LV = Length density; SV = Star volume.

Numerical density of the seminiferous tubules: There was an increase in the numerical density of the seminiferous tubules of the Sprague-Dawley rats. The 4 weeks group increased from 55.0 ± 9.38 v. 68.9 ± 10.4 v. 97.2 ± 7.57. While the 8 weeks group increased from 70.9 ± 8.53 v. 89.3 ± 8.24 v. 130.7 ± 16.43. The increase was found to be dose and duration dependent. In the reversal group, there was increase from 18.5 ± 2.05 to 18.6 ± 1.30 to 19.9 ± 1.2 compared with the control (Table 2).

Length density of the seminiferous tubules: There was an increase in the length density of the seminiferous tubules of the Sprague Dawley rats for the experimental groups. For the 4 weeks group the increase was from 24.2 ± 2.03 v. 26.0 ± 2.63 v. 26.8 ± 3.61 and for the 8 weeks group was from 28.8 ± 3.39 v. 31.8 ± 4.38 v. 39.8 ± 6.23. This was found to be dose

and duration dependent. The reversal group also showed an increase in the length density from 30.4 ± 27.0 v. 49.8 ± 8.86 v. 60.2 ± 23.2 (Table 2).

Volume density of seminiferous tubules: There was an increase in the volume density of the seminiferous tubules of the experimental rats from 72.6 ± 2.1% v. 76.8 ± 2.5% v. 87.6 ± 1.6% for the 4 weeks group, from 71.4 ± 0.6 v.78.6 ± 3.4 v. 81.2 ± 0.7 for the 8 weeks group and a decrease observed for the reversal group from 68.1 ± 1.7 v. 64.3 ± 1.5 v. 62.5 ± 2.3 (Table 3).

Table 3: Effect of the extract of *Carica papaya* bark on the volume density of seminiferous tubules and interstitium of male Sprague-Dawley rats for 4wks, 8 wks and reversal groups

PERCENTAGE (%) VOLUME DENSITY		
	Dose (4 Weeks)	
DOSES	***Seminiferous tubule***	***Tubular interstitium***
Control	72.6 ± 2.1[a]	27.4 ± 1.2
Low dose	76.8 ± 2.5	23.6 ± 1.5
High dose	87.6 ± 1.6	12.4 ± 1.6
	Dose (8 Weeks)	
Control	71.4 ± 0.6[b]	28.6 ± 2.8
Low dose	78.6 ± 3.2	21.4 ± 3.2
High dose	81.2 ± 0.7	18.8 ± 1.5
	16 Weeks reversal	
Control	68.1 ± 1.7	31.9 ± 1.8
Low dose	64.0± 1.5	35.7 ± 2.5
High dose	62.5 ± 2.3	37.5 ± 3.6

Control: 5.0 ml of distilled water; Low dose: 50 mg/ml/kg/day of *Carica papaya*; High dose: 100 mg/ml/kg/day of *Carica papaya*; *a* = mean ± S.D; b = $p < 0.5$; n = 10.

Volume density of the testicular interstitium: There was a decrease in the volume density of the interstitium from 27.4 ± 1.2 v. 23.2 ± 1.5 v. 12.4 ± 1.6 for the 4 weeks group and from 28.6 ± 2.8 v. 21.4 ± 3.2 v. 18.8 ± 1.5 for the 8 weeks group. For the reversal group, there was an increase from 31.9 ± 1.8 of control to 35.7 ± 2.5 v. 37.5 ± 3.6 for the low and high dose treatment (Table 3).

Star volume of the seminiferous tubules: There was a reduction from 10.50 ± 0.22 v. 6.48 ± 0.19 v. 2.46 ± 0.17 in the star volume of the experimental rats for 4 weeks and from 8.13 ± 0.21 v. 5.44 ± 0.15 v. 3.35 ± 0.12 for 8 weeks group compared with the control. The reversal group showed an increase from 12.76 ± 4.3 v. 23.48 ± 4.8 v. 32.06 ± 3.5 compared with the control (Table 2).

DISCUSSION:

The morphometric analysis of the effect of CP bark extract on the seminiferous tubules is in concert with the histological result observed on the testis. The study demonstrated a dose and duration dependent decrease in the mean testicular volume, tubular diameter, cross sectional area and star volume of tubules; but an increase in the length density, numerical density, number of profiles per unit area and volume density of tubules.

A reduction in seminiferous tubule star volume can be interpreted in several ways. For instance, it might signify that certain seminiferous tubules are smaller (thinner or shorter) or that all tubules are smaller (i.e. each tubule is simply scaled down in size) or that the coiled pattern is different. There is morphometric evidence that the total length of tubule is reduced by administration of CP bark aqueous extracts and this seems to affect many tubules. This impoverished linear growth could produce the drop in star volume seen in the present study. However, the possibility cannot be excluded that all tubules are reduced in size. In assessing the scale of tubular reduction, it would be imprudent to draw firm conclusion by regarding star volume as summative units of space because they are, in reality locally defined, point-sampled regions of arbitrary space. Therefore, dividing total volumes by star volumes merely indicates the theoretical numbers of star volume which can be contained in total volume. However, such numbers do give an idea of the impact of tubular shrinkage on tubular number and size. The number rose between 4 and 8 weeks. Tubular star volume could be imagined as the volume through which spermatogenic cells travel to reach the *rete* testis, if its progress in all possible direction was rectilinear. This volume would depends not only on the total volume of tubular space which itself varies due to episodic flow but also the number of Sertoli cells which project into it. The star volume of tubules is therefore determined by length, diameter, curvature and incidence of Sertoli-

Sertoli cell bridges. Star volume is a volume-weighted mean average and therefore highly sensitive to seminiferous tubular size distribution [20]. However gross volume is a result of parameters which are independent of tubular size distribution. Thus the data may suggest that only the gross volume summarizes the data on total testicular volume while star volume indicates that by the 4th week following administration of CP bark there are many significant small seminiferous tubules. Thus the size decrease and tubular cellular changes suggest that the small tubules represent tubular reduction in size.

The use of star volume to determine the size of seminiferous tubules has benefits and disadvantages. Although providing a useful way of realizing the concept of inter-Sertoli-Sertoli cell spaces and the adluminal compartment, star volume is a statistically noisy variable. The Noisiness of these estimates stem from the fact that the adluminal compartment is a point-sampled local region of arbitrary space; therefore the star volume depends on a variety of factors including the number, size, and arrangement of Sertoli cells in the seminiferous tubules. The rational for measuring star volume is that this variable is influenced more directly by differences in total tubular area. By focusing on the relationship between tubular surface area and volume the relevance to issues associated with transport in the tubules is more obvious. Unfortunately like volume-weighted mean volume, the star volume is a noisy variable. Present results suggest that it may be even nosier and so its application in future studies will need to balance this precision against the potential benefits of being able to characterize tubular size more comprehensively. Its usefulness lies in providing a direct and unbiased estimate of volume which has a strict mathematical definition. For the tubules the star volume will be less than the total volume. The number of seminiferous tubules might also alter. A 25% decrease in tubular volume would correspond to a roughly 10% reduction in linear dimensions if all overall size altered isomorphically.

Results obtained in the present study showed that tubular length actually decrease between 40 to 80 % suggesting that there was an overall decrease in tubular length. This might also reflect the stunted growth of tubules. This suggestion might represent a general reduction in the proportion of parenchymal tissues which is supported by volumetric analyses.

In terms of tubular function, volume and surface area are influential in determining production and transport of spermatogenic cells whilst the total volume influences supply. However other factors are clearly important. These include number of spermatogenic cells in the basal compartment, the Sertoli-Sertoli cell barrier which determines the number of cells in the adluminal compartment. It is not

possible to predict the likely consequences of these changed geometric relationships on tubular sperm production or flow because the present morphometric data form only part of the complete picture. The increase in the volume density of the seminiferous tubules despite a reduction in the diameter of the tubules only suggest that there was a significant reduction in the interstitium due to the destruction of the Leydig cells. These cells are responsible for the production of testosterone. The size of the testicular interstitium normally correlates positively with the number of Leydig cells, which in turn correlates positively with the germ and testicular level of testosterone [21, 22].

For the reversal group, the parameters remained reversed for the 4 weeks group and remained the same for the 8 weeks group showing an evidence of substantial recovery using a lower dose of the aqueous extract of the bark.

CONCLUSION:

The administration of the CP bark aqueous extract reduces the total length of the tubules thereby producing a drop in the star volume. The effect is more pronounced for the high dose than in the low dose. This suggests that the extract could impair spermatogenesis thereby acting as a potential contraceptive.

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