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Testicular morphometric studies in Nigerian males

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Summary

A total of 102 autopsy subjects found to have seemingly normal testicular picture were analysed to determine acceptable normal morphometric values. The mean tubular diameter in the last trimester of intrauterined life is 43.8μ in contrast to adult value of 173.0μ. The mean basement membrane thickness increases from 3.2μ in the last trimester to 6.9μ in the 10–14 years age range. The adult mean testicular score count is 19.3 in contrast to 5.3 observed in the last trimester. While Sertoli cells predominate in the last trimester of intrauterine life, many spermatozoa are seen during the period 15–19 years.

Résumé

Totalement 102 subjects, apres une examin post mortem qui ont eu des imâges tèsticulaires normales ètâit analysés. Le diametres tèsticulaire en evarage pendant le derniere trimestre de la vie intrauterine est 43.8μ contraire des numereux des grosses (173.0μ). Le forces des membranes basement en average est un increment de 3.2μ pendant le derniere trimestre à 6.9μ pendant le 10–14ans d'age. L'average bût compte des subjets groses est 19.3 contraire de 5.3 pendant le dernier trimestre. Les ceeles Sertoli sont plus pendant le derniere trimestre, mais c'est les spermatozoas qui sont plus pendant le 15–19 ams d'ages.

Introduction

Morphometric reference values for the testes amongst Nigerians are virtually unavailable. Surgical biopsy specimens of the testes are often read without reference to specific values but rather, a subjective assessment is made. The reports often depend on the individual pathologist's bias though he tries to highlight specific details. However, the provision of baseline data for supposedly normal Nigerian males of varying age groups will allow for comparative studies and also ensure a more objective assessment of testicular biopsies. Morphometric values when available are usually those of the caucasian population. Charny, Conston and Meranze documented morphometric values for the American population and also indicated age variations [1]. There are virtually no baseline data for our environment. Such values are likely to provide a scientific basis for assessment of the degree of variation from what might be considered normal. This study is aimed at determining supposedly normal values for Nigerian males, pattern of variation amongst the various age groups and compare results with reports from other parts of the world.

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Materials and methods

Materials were obtained from routine autopsies conducted on prematurely delivered or stillbirth male babies (28 weeks and above) and available male subjects seen at the Department of Morbid Anatomy, Lagos University Teaching Hospital. The mothers of the premature babies were of essentially good health status. During the period of study, 380 male subjects were examined out of which only 102 were analysed. The rest were discarded either due to organ autolysis or presence of testicular pathology. The period of study covered 3 years (January 1985 – December 1987). Volumetric studies were excluded because the emphasis is on the morphometric values for the seminiferous tubules. Sections of both left and right testes were taken and immediately fixed in Bouin's fluid to ensure better histological preservation [2,3] and processed by standard histological methods. The minimum size of specimen was 4.0 x 4.0 mm for good representation [3]. Four microns sections were later cut and stained with haematoxylin and eosin. For each age group, the following parameters were determined:

(a) Tubular diameter (μ)
(b) Basement membrane thickness (μ)
(c) Cell population
(d) Degree of spermatogenesis
(e) Mean testicular score count.

The above indices are based on the criteria used by Makler [4] which correlates well with that of Aafjes and Van der Vijver [5]. The latter is a modified form of the Johnson criteria [6]. All measurements were taken with a micrometer obtained from Graticles Ltd., Tombridge Kent, England. Only very round seminiferous tubules or those with very parallel walls when sectioned longitudinally were used for measurements [4]. The inner diameters of the tubules were measured as tubular diameter. One hundred tubules per section were scored. The mean score was determined by taking the average measurements from both testes to ensure adequate representation. The results were then subjected to statistical analysis to find the mean per age group and whether there were any significant statistical differences between the various groups.

Based on the Makler criteria [4], scoring for the various parameters are as follows:

(a) Scoring for inner diameter of the tubule

> 170μ	— 5
141–170μ	— 4
111–140μ	— 3
81–110μ	— 2
51 – 80μ	— 1
≤ 50μ	— 0

The largest or normal diameter is scored 5; completely obliterated tubules 0; any size between these poles — slightly reduced, moderately reduced, small and very small diameter are scored 1–4.

(b) Scoring for basement membrane thickness

≤ 8μ	— 5
9 – 11μ	— 4
12 – 14μ	— 3
15 – 17μ	— 2
18 – 20μ	— 1
> 20μ	— 0

Thin or normal thickness is scored 5; thickest or hyalinized membrane 0; thickness between these extremes — slight, moderate, severe and very severe are scored 1–4.

(c) Scoring for cell population of the tubules

4 lines of cells	— 5
3 lines of cells	— 4
2 lines of cells	— 3
1 line of cells	— 2
Sertoli + Germinal cells	— 1
Sertoli cells only	— 0

(d) Scoring for degree of spermatogenesis

Spermatozoa observed — 5
Maturation to spermatids levels only — 4
Maturation to secondary spermatocytes level only — 3
Maturation to primary spermatocytes level only — 2
Only Sertoli cells and spermatogonia observed — 1
Sertoli cells only — 0

For each of the criteria indicated, the maximum score is 5. Therefore when all the criteria are considered together, the total maximum score obtainable would be 20. The mean testicular score for any subject will thus lie between 0 and 20. This score represents the fertility index for that individual.

Results

The results are as shown in Tables 1 and 2. During the last trimester of intrauterine life, the tubules are lined by primitive germ cells and Sertoli cells. The pubertal testes contain many secondary spermatocytes with patchy areas of Leydig cells. However, spermatozoa population become appreciable in the age range 15–19 years. The combined mean for subjects in the age range 15–65 years showed the tubular diameter to be 173.3μ (S.D. = 10.7); basement membrane thickness is 7.0μ (S.D. = 0.6) and mean score is 19.3 (S.D = 0.6).

The age range (15–65 years) represents the fertile age group comprising a total of 65 subjects from the entire population studied. In the last trimester of intrauterine life, the seminiferous tubules have a mean diameter of 43.8μ, the basement membrane is 3.2μ thick and the score representing fertility count is 5.3 (Fig. 1). The

predominant cell at this stage is the Sertoli cell. Within the first four years of life there is significant increase in the tubular diameter to 54.7μ while the score goes up to 7.7. Though there is an apparent increase in the basement membrane thickness, this is not statistically significant (Table 2). Between the ages of 5 and 9 years there is a significant rise in tubular diameter to 62.0μ and the basement membrane thickness to 5.2μ while the score is stagnant. A slightly sharp increase in tubular diameter (110.3μ), basement membrane thickness (6.9μ) and mean score (12.7) is observed in the 10–14 years age group. The basement membrane at this time approaches adult values and there is an accompanying increase in cell population.

During the period 15–19 years another significant increase in the tubular diameter from 110.0μ (seen in the 10–14 years age group) to 163.3μ occurs with the cell population in the 15–19 years group consisting of many spermatids and spermatozoa. The basement membrane increases to 7.3μ, though this is not statistically significant (Table 2). After 19 years, the tubular diameter increases to a mean of 176.3μ in the 20–24 years age group, and like the basement membrane thickness and score count it plateaus (Fig. 2). Table 2 shows that adulthood is fully achieved in the 20–24 years age range. During the last trimester of intrauterine life and infantile periods, mainly Sertoli cells are seen with few Leydig cells in the stroma. By 4 years, spermatogonia are seen and between the ages of 5 and 9 years, primary spermatocytes are observed. Suddenly between the ages 10–14 years secondary spermatocytes appear which show maturation to spermatids and spermatozoa and during the 15–19 years age range, spermatozoa population reaches adult values. The non availability of enough subjects in the age group over 65 years does not permit an assessment of what happens thereafter especially as it relates to tubular diameter (155.0μ) but this is not statistically significant.

Table 1: Age variation in relation to morphometric measurements

Age Group	No. of Subjects Studied	Tubular Diameter (μ)	Basement Membrane Thickness (μ)	Mean Score
Last trimester	8	43.8	3.2	5.3
0–4 years	13	54.7	3.6	7.7
5–9 "	9	62.0	5.2	7.8
10–14 "	7	110.0	6.9	12.7
15–19 "	6	163.3	7.3	18.7
20–24 "	8	176.3	7.1	19.1
25–29 "	6	172.5	6.8	19.3
30–34 "	7	169.7	7.2	19.4
35–39 "	9	177.8	7.1	19.3
40–44 "	6	171.0	6.8	19.5
45–49 "	5	177.0	6.5	19.4
50–54 "	5	172.0	7.0	19.4
55–59 "	6	174.0	7.0	19.5
60–64 "	5	177.0	7.3	19.0
+65–69 "	2	155.0	7.5	18.0

+ Results should be viewed with caution due to small sample size.

Table 2: Determination of *P*-values to test for significant statistical differences between adjacent age groups

Age Groups	Tubular Diameter	Basement Membrane Thickness	Mean Score
0–4 yrs/foetal	$P < 0.01$	$P > 0.1$	$P < 0.01$
5–9/0–4	$P < 0.01$	$P < 0.01$	$P > 0.5$
10–14/15–19	$P < 0.02$	$P < 0.01$	$P < 0.01$
15–19/10–14	$P < 0.1$	$P > 0.2$	$P < 0.01$
20–24/15–19	$P < 0.1$	$P > 0.5$	$P > 0.2$
25–29/20–24	$P > 0.2$	$P > 0.2$	$P > 0.5$
30–34/25–29	$P > 0.5$	$P > 0.1$	$P > 0.5$
35–39/30–34	$P > 0.1$	$P > 0.5$	$P > 0.5$
40–44/35–39	$P > 0.2$	$P > 0.1$	$P > 0.5$
45–49/40–44	$P > 0.2$	$P > 0.2$	$P > 0.5$
50–54/45–49	$P > 0.1$	$P > 0.1$	$P > 0.5$
55–59/50–54	$P > 0.2$	$P > 0.5$	$P > 0.5$
60–65/55–59	$P > 0.5$	$P > 0.2$	$P > 0.2$

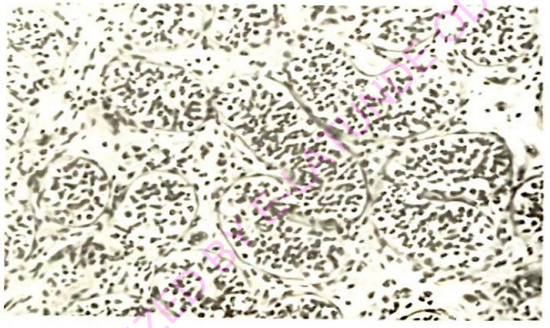

Fig. 1: The testis in the last trimester of pregnancy. Note the basement membrane thickness, tubular diameter, primitive germ cells and presence of some foetal Leydig cells (H & E x 300).

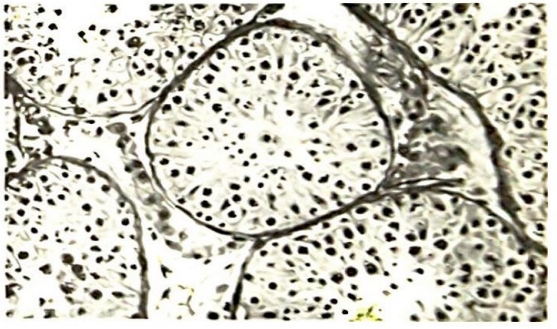

Fig. 2: Adult testis. Spermiogenesis is evident. Note the basement membrane thickness, tubular diameter and presence of mature Leydig cells (H & E x 300).

Discussion

The results show the testicular morphometric values with age variation, and what might be considered acceptable as normal morphometric values for reproductive Nigerian males. A striking observation however is the sharp and sudden increase during puberty (10–14 yrs), in the values obtained for the parameters studied. The tubular diameter increases from 62.0μ in the 5–9 years age group to 110.0μ in the 10–14 years group. Similarly, the basement membrane thickness increases from 5.2μ to 6.9μ and the mean score from 7.8 to 12.7. Immediately after, that is, in the 15–19 years age group these parameters have values of 163.3μ, and 18.7 respectively which approximates the adult values. This observation almost coincides well with that of Charny, Conston and Meranze who reported in the Caucasian population, a sudden increase in tubular diameter from 60.0μ at puberty to 150–180μ in adults [1]. We can thus safely conclude that there is no racial variation as regards this sudden maturation. It is also pertinent to note that the maximum fertility index once established during the 20–24 years age range is maintained up to at least the seventh decade provided all other factors are normal. Probably if the sample size had included more subjects aged 70 years and above further conclusions would have been drawn. The sudden rise in morphometric values during puberty is due to increase in plasma levels of luteinizing hormone (LH) and follicle stimulating hormone (FSH) and this hormonal increase in fact precedes phenotypic alterations by some months [7]. Macini and associates [8] showed that increased FSH levels cause Sertoli cells maturation whilst LH causes Leydig cell maturation. Germ cell maintenance and initiation of spermatogenesis are the responsibilities of FSH in addition to increasing testicular size. Follicle stimulating hormone however does not affect spermiogenesis, that is maturation of the secondary spermatocytes to spermatozoa [9]. This function is the preserve of LH which stimulates the Leydig cells to secrete testosterone. This hormone apart from exerting peripheral effects of secondary sexual characteristics acts locally on the seminiferous tubules to ensure spermiogenesis. Apart from the age variations evident in the result, an important finding in this study is the establishment of what might be considered as acceptable normal morphometric values for the reproductive Nigerian males. The tubular diameter is 175.0μ ± 10.7, basement membrane is 7.0μ ± 0.6, and the mean testicular score count is 19.3 ± 0.6. It is hoped that this study will provide a good basis for future and more meaningful studies of the testes amongst Nigerians.

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