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Testicular Composition, Number of A Spermatogonia, Germ Cell Ratios, and Number of Spermatids in Three Different Breeds of Boars

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ABSTRACT: Seminal quality, including the number of spermatozoa in the ejaculate, varies among breeds of boars. Variation in seminal quality may be explained by breed differences in testicular size and composition. The objective of this study was to characterize testicular composition and population sizes of germ cells and somatic cells in three different breeds of boars. Testes from mature and normal Meishan (M, $n = 5$; age 225 days), Whitecross (WC, $n = 5$; age 346 days), and West African (WA, $n = 5$; age 322 days) boars were either fixed in Zenker-formol or perfused with glutaraldehyde, embedded in Epon, sectioned at 0.5 μm or 20 μm and evaluated stereologically for germ cells, Leydig cells, Sertoli cells, and other testicular structures. The paired parenchymal weight was higher ($P < 0.05$) in the WC (498 ± 35 g) than in the M (247 ± 17 g) or WA (133 ± 10 g). The nuclear volumes per boar of Sertoli cells, type A spermatogonia, leptotene, zygotene, pachytene, secondary spermatocytes, spermatids with spherical nuclei, and spermatids with elongated nuclei were higher ($P < 0.05$) in the WC than in the M or WA. The nuclear volumes per boar of blood vessels and Leydig cells were exceptionally higher ($P < 0.05$) in the M than in the WC or WA. The average germ cell nuclear diameters were similar ($P > 0.05$) for the M and WC but least ($P < 0.05$) in the WA breed. Also, the nuclear diameters of Leydig cells and Sertoli cells were higher ($P < 0.05$) in the M than in the WC or WA. The population sizes of all the intratubular components were positively correlated ($P < 0.05$) with testicular size (weight). Volume density of seminiferous tubules was similar in the WA and WC, but was lowest in the M, who had the highest density of Leydig cells. However, due to the larger testes, all germ cell types and Sertoli cells were higher in the WC than in the WA or M. The M and WA had similar low numbers of type A spermatogonia, but there was less germ cell degeneration in M than in WA, such that M spermatids were intermediate in number between WA and WC.

Key words: Breed, boar, testis composition.
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Morphometric analysis on the testis of any species or breed is necessary in assessing and estimating quantitative changes in testicular components and spermatogenic function arising from factors such as age (Johnson and Neaves, 1981; Jones and Berndtson, 1986), season (Hochereau-de Reviers and Lincoln, 1978), and nutrition (Johnson et al, 1992) as well as the effects of various treatments (Cosentino et al, 1986), toxins (Sinha Hikim and Hoffer, 1987), hormones (Berndtson and Igboeli, 1988), and drugs (Desouky et al, 1991). In the past, classical quantitative methods have been employed (Roosen-Runge and Giesel, 1950; Clermont and Morgentaler, 1955; Clermont and Perey, 1957) in assessing testicular structure and function under various physiological and pathological conditions. Recent studies on the structure of the testis have applied stereologic techniques, and results from these morphometric studies have been positively correlated with function of the testis (Johnson and Thompson, 1983; Mendis-Handagama et al, 1988; Hikim et al, 1989a,b). Thus, in the boar, Leydig cell volume has been shown to be strongly related to efficiency of steroid production (Lunstra et al, 1986).

In a recent study, baseline data on testicular composition in 12 species were reported (Russell et al, 1990). Similar data on the boar are either not comprehensive (Kennelly and Foote, 1964) or were not reported (Swierstra et al, 1974). Further, research on testicular composition of different breeds of boars has been limited. Differences in seminal quality of Meishan (M), Whitecross (WC), and West African (WA) breeds based on various seminal characteristics (Swierstra, 1973; Akinkpelumi, Personal communication; Borg et al, 1993) indicate that the testicular composition and ratios of germ cells may be different among breeds. Thus, investigation and comparison of testicular composition and ratios of germ cells

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in boars from these swine breeds of markedly divergent origins may provide new models and insights for studying mechanisms controlling boar testicular function. The objective of the present study was to characterize testicular composition and population sizes of germ cells and somatic cells in three different breeds of boars. Wide breed-related variations in the numbers of germ cells and Sertoli cells within the seminiferous tubules and interstitial components were found to be significant.

Materials and Methods

Specimen and Histologic Preparations

Pairs of testes were obtained from five mature (332 days) indigenous WA boars from research farms in Nigeria. Within 20 minutes of slaughter, testes under ice were transported to the laboratory where they were weighed after the fascia and epididymis had been removed. Parenchymal weight was obtained by subtracting the weight of tunica albuginea from total testicular weight. Parenchymal volume was determined by dividing parenchymal weight by 1.05 g/ml. Slices of tissue (1–1.5 cm^3) from the equatorial region, and the dorsal and ventral poles were placed in freshly prepared Zenker-formol fixative for 24 hours, washed in running water for 24 hours, and stored in 70% alcohol until ready for processing (Humason, 1967). For comparative purposes, testes from five mature (225 days) M boars and five mature (346 days) WC boars (raised in Nebraska) were weighed and perfused with glutaraldehyde in cacodylate buffer and freed from fascia, and the parenchymal weight and volume were obtained as described above for the WA. Slices from different regions of the testes were fixed in 2% glutaraldehyde in 100 mM cacodylate buffer and subsequently stored at 5°C. Smaller slices (5 × 10 × 10 mm) of the glutaraldehyde-fixed parenchyma or Zenker-formol-fixed tissue were further fixed in 1% osmium in cacodylate buffer, dehydrated in alcohol, embedded in Epon 812, and sectioned at 0.5 μm or 20 μm (Johnson, 1985). All evaluations were carried out in one location (Texas) and by one technician.

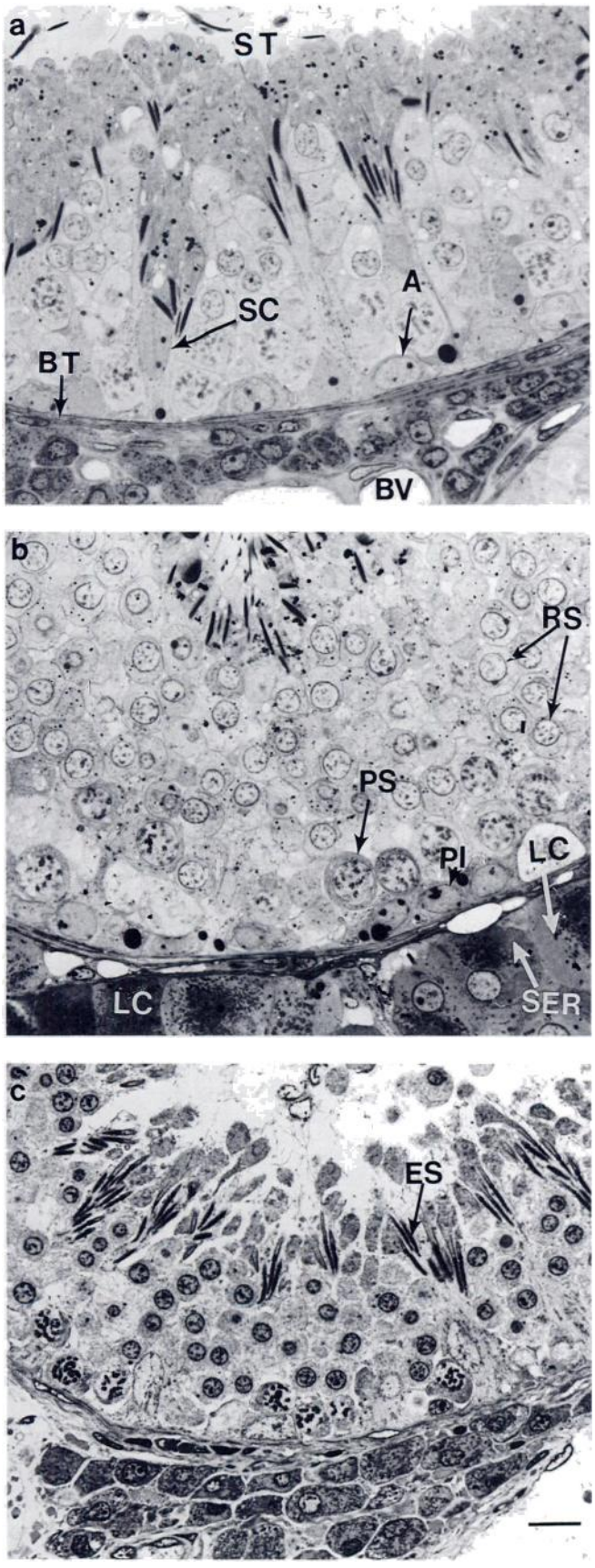
Stereologic Evaluation of Tissues

The 0.5-μm Epon sections stained with toluidine blue were examined under brightfield microscopy at ×1,000 magnification. The volume densities or percentages of parenchyma occupied by different structures including seminiferous tubules, Sertoli cells, Leydig cell nuclei, and other interstitium components such as the lymphatics and blood vessels were determined by means of a 50-point ocular grid employing Chalkley's (1943) point-counting method. Briefly, cell types/structures of interest as well as all other testicular structures have an equal chance of being scored by the 50-point grid under view. Thus, the total number of times the points land on the structure of interest divided by the total possible number of "hits" gives the volume density of the structure in the testis. A total of 10,000 hits were scored for each boar, which was sufficient to achieve a high precision, with average coefficients of variation of 8, 11, 24, 16, and 15, respectively, for pachytene spermatocytes, spermatids with spherical nuclei, Leydig cell nuclei, Leydig cell cytoplasm, and Sertoli cell nuclei. Randomness and sufficient sampling were achieved by the use of three or four sections on a slide and by random movements across each section in both axes during microscopic examination. The tubular staging system of Swierstra (1968) was employed to identify cell types within given stages. Germ cells counted included type A spermatogonia (Stages I–VIII), type B spermatogonia (Stages VI–VIII), leptotene primary spermatocytes (Stages I and II), zygotene primary spermatocytes (Stages II–IV), pachytene primary spermatocytes (Stages I–III and V–VIII), spermatids with spherical nuclei (Stages I and V–VIII), and spermatids with elongated nuclei (Stages II–VIII). The volume density of each component measured above multiplied by total parenchymal volume (parenchymal weight/1.05 g/ml) yielded the total volume of that component per boar or testis. The average specific gravity of 1.05 g/ml was obtained by dividing the weight of a piece of tissue when immersed and allowed to drop to the bottom of water by the weight of the same piece when suspended in water. The values for different breeds were WC: 1.04 ± 0.01 g/ml; M: 1.07 ± 0.01 g/ml; and WA: 1.05 ± 0.01 g/ml.

The unstained 20-μm Epon sections were examined under Normaski optics to determine the maximum diameter of all round or oval nuclei of cells mentioned above (Johnson et al, 1980; Johnson and Neaves, 1981). By focusing up and down on the specimen, the maximum diameters of nuclei embedded in the thick Epon were located and measured. For the Sertoli cells and type A spermatogonia, the nuclei diameters were estimated as the average of the height and width measurements due to their irregular and oval shapes respectively (Johnson, 1985). At least 50 measurements per nucleus per boar were measured out using a computerized digitizing unit directly attached to the microscope with Normaski optics. Measurements were randomized among the stages where each germ cell appeared. The number of cells of a given cell type per testis was calculated as the product of percentage nuclei, parenchymal volume, and histologic correction factor for section thickness and nuclei diameter (Weibel and Paumgartner, 1978) divided by the average volume of a single nucleus of that cell type (Kennelly and Foote, 1964; Amann, 1970; Johnson et al, 1984). The average volume of a single nucleus was calculated by the formula for a sphere (Johnson, 1985). No correction factor for shrinkage was applied because glutaraldehyde-fixed and osmicated Epon sections had previously been reported to undergo negligible shrinkage during histological processing (Johnson and Neaves, 1981; Johnson et al, 1983), and no evidence of shrinkage (e.g., no obvious tubules separated from the interstitium, Fig. 1) was noted in these specimens regardless of initial fixative used. The ratio of the number of different germ cell types to that of preceding cell type was calculated to estimate indices of degeneration during the spermatogenic cycle. Given the seminiferous tubular diameter and tubular volume, the total length of seminiferous tubules per boar was calculated using the formula for the volume of a cylinder (Johnson and Neaves, 1981).

Statistical Analyses

Differences between breeds in all the components estimated were tested with one-way analysis of variance and the Student-



Newman-Keuls test (Sokal and Rohlf, 1969; SAS Institute, 1985). All values derived for volume densities were subjected to arcsine transformation prior to analyses. Correlation coefficients between parameters of spermatogenesis were determined for each breed. The coefficients for most parameters were similar between breeds and data were pooled across breeds (Sokal and Rohlf, 1969).

Results

Breed Variation in Body and Testicular Weights

Physical examination of all the animals involved in this study showed no gross pathology of the testis. All were mature boars that had previously been used as either control animals in semen studies or for breeding purposes. As a result, their spermatogenic capabilities were confirmed prior to the time of slaughter.

Data presented in Table 1 on the main parameters reveal an interesting trend. Though their ages did not differ significantly ($P > 0.05$), values for body, paired testicular, paired parenchymal, and tunic weights recorded for the WC were almost three and two times higher ($P < 0.05$) than those for the WA and M, respectively. The testes of the WC had heavier tunica albugineae ($P < 0.05$) than those of the M or WA. However, there were no significant differences ($P > 0.05$) in the testis/body weight ratio and tunic/testicular weight ratio in the three breeds. Quantitatively, the tunica albuginea accounted for 8.0%, 8.3%, and 6.8% of the paired testes weight in the WC, M, and WA, respectively. Across breeds, body, testicular, and parenchymal weights were correlated positively ($P < 0.05$) with the volume densities and total volume per boar of various germ cell nuclei as well as Sertoli and Leydig cell nuclei.

Breed Variation in Testicular Composition

The volume density of seminiferous tubules in the testicular parenchyma was less ($P < 0.05$) in the M (48.6%) than in the WC (68.7%) or WA (60.4%) but the M had higher ($P < 0.05$) interstitial components (Table 2). Also,

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FIG. 1. Portions of cross sections of seminiferous tubules in different breeds of boars. Tissues were perfused in glutaraldehyde, osmicated, embedded in Epon, sectioned at 0.5 μm and stained with toluidine blue. Cellular and nuclear morphology of germ and somatic cells were similar among breeds. (a) Whitecross, (b) Meishan, (c) West African. Testicular components enumerated and shown include seminiferous tubules (ST), A spermatogonia (A), preleptotene spermatocyte (PI), pachytene spermatocyte (PS), spermatids with round nuclei (RS), spermatids with elongated nuclei (ES), Leydig cells (LC), blood vessels (BV), Sertoli cell nuclei (SC), and tubular boundary tissue (BT). The Leydig cells are largest in the Meishan (b). Even under the light microscope, the smooth endoplasmic reticulum (SER) appears as clear cytoplasm or swirls of cytoplasm and mitochondria as dark dots in the clear cytoplasm of abundant SER. Bar length = 20 μm.

Table 1. *Body weights and testicular weights (mean ± SE) in adult boars of three breeds*

Parameters	Whitecross	Meishan	West African
n	5	5	5
Age (days)	346.4 ± 1.89*	225 ± 2.88	332.1 ± 2.75
Body weight (kg)	133.45 ± 5.46[a]	77.76 ± 2.02[b]	34.01 ± 0.96[c]
Paired testes weight (g)	541.10 ± 35.23[a]	269.48 ± 18.93[b]	148.44 ± 6.98[c]
Paired parenchymal weight (g)	497.78 ± 34.74[a]	247.24 ± 17.17[b]	133.27 ± 10.05[c]
Tunica albuginea weight (g)	43.32 ± 1.57[a]	22.24 ± 1.84[b]	10.13 ± 0.72[c]
Relative testicular weight† (g/kg)	4.1 ± 0.3	3.4 ± 0.2	4.2 ± 0.2
Relative tunic weight‡ (%)	8.12 ± 0.15	8.23 ± 0.24	7.09 ± 0.43

Means for a given parameter with different superscripts (a, b, c) are significantly different ($P < 0.05$).
* Mean ± SEM.
† Relative testicular weight = paired testis weight (g)/body weight (kg).
‡ Relative tunic weight = (paired tunic weight/paired testis weight)100.

the M has the shortest ($P < 0.05$) seminiferous tubule length. While there were no differences in the luminal diameter, seminiferous tubular diameter, and seminiferous epithelial height between the WC and M, these values were particularly low ($P < 0.05$) in the WA (Table 3). Boundary tissue thickness of seminiferous tubules was similar ($P > 0.05$) for the three breeds.

The percentages of the testis occupied by various germ cells, Sertoli cells, and several interstitial components are shown in Table 2. The volume density of type A spermatogonia was significantly ($P < 0.05$) higher in the WC than in the M or WA. With the exception of type B spermatogonia, leptotene and zygotene primary spermatocytes, secondary spermatocytes, and spermatids with elongated nuclei, the volume densities of nuclei of other germ cells differed significantly ($P < 0.05$) among breeds. For these germ cells, that is, pachytene spermatocytes including spermatids with spherical nuclei, volume densities did not differ between the M and WC, and the values obtained for the W were always lower. For Sertoli cells, the M has the lowest volume density ($P > 0.05$) while values for the WC and WA were similar. The portion of the interstitium occupied by blood vessels and Leydig cell cytoplasm was highest ($P < 0.05$) in the M, but the volume densities of the lymphatics and Leydig cell nuclei did not differ ($P > 0.05$) among breeds. Nuclear diameters and the volume of a single nucleus of various germ cell types were similar for the M and WC (Table 3) and values for WA were lower ($P < 0.05$). However, values for the Leydig cell and Sertoli cell were higher ($P < 0.05$) in the M than in the WC or WA.

The total volume per boar of germ cell nuclei and Sertoli cell nuclei or interstitial structures (Fig. 1) varied significantly (Table 2) among breeds. For the number of type A spermatogonia, leptotene and zygotene primary spermatocytes, secondary spermatocytes, and Sertoli cells, the WA did not differ from the M, but these values were higher in the WC. However, variations among breeds in the total volume per boar of pachytene spermatocyte nuclei and spermatids with spherical and elongated nuclei were very large (Table 2). Values for the WC were always high while those for the M were intermediate. For the interstitial components, the total volume of blood vessels and Leydig cell nuclei per boar was higher ($P < 0.05$) in the WC than in the M or WA while values for blood vessels were lowest in the WA. No significant breed differences ($P > 0.05$) existed in total volume of lymphatics, but the M was exceedingly higher ($P < 0.05$) in Leydig cell cytoplasm total volume than the WC or WA.

The numbers of each germ cell type, Sertoli cells, and Leydig cells per testis were significantly ($P < 0.05$) higher in the WC than in the M or WA (Fig. 2). There was an abundant population of type A spermatogonia and remarkably high numbers of spermatocytes and spermatids with spherical nuclei in the WC. Despite the larger testicular size of the M, type A spermatogonial population did not differ significantly ($P > 0.05$) between the M and WA. However, for other advanced germ cell types, the M had intermediate numerical values between the WC and WA. Using the number of germ cells at different steps in spermatogenesis per A spermatogonium, there was a trend for spermatids with spherical nuclei per A spermatogonium to be higher for the M (31 ± 5) than for the WC (18 ± 3) or WA (21 ± 3) (Table 5).

Discussion

Puberty, defined as the time of first appearance of spermatozoa in the seminiferous tubules, occurs in the European breeds of boars at an average age of 147 days (Phillips and Andrews, 1936) and in the West African boars at about 120 days (Egbunike, 1980). Though the European and West African breeds could be used for breeding at 6–7 months of age, maturity may not be attained until over 8–10 months of age (Egbunike and Steinbach, 1972). On the contrary, Meishan boars reach puberty as early as 75 days of age though the testis does not acquire the ability to produce morphologically normal spermatozoa until about 120 days (Harayama et al, 1991).

Table 2. *Breed variation in volume density and total volume of nuclei of germ and Sertoli cells including the interstitial structures in testicular parenchyma of adult boars*

Seminiferous tubule compartment	Percentage nuclei/structures in parenchyma			Total volume of nuclei/structures (ml) per testis		
	Whitecross	Meishan	West African	Whitecross	Meishan	West African
Cell types						
Type A spermatogonia	0.58 ± 0.09[a*]	0.32 ± 0.03[b]	0.32 ± 0.03[b]	2.68 ± 0.38[a*]	0.75 ± 0.11[b]	0.41 ± 0.07[b]
Type B spermatogonia	0.39 ± 0.05	0.36 ± 0.07	0.22 ± 0.03	1.79 ± 0.18[a]	0.88 ± 0.22[b]	0.28 ± 0.06[c]
Leptotene spermatocytes	1.03 ± 0.23	0.73 ± 0.25	0.34 ± 0.13	4.99 ± 1.17[a]	1.70 ± 0.55[b]	0.42 ± 0.16[b]
Zygotene spermatocytes	1.08 ± 0.22	0.78 ± 0.16	1.14 ± 0.32	5.34 ± 1.30[a]	1.80 ± 0.35[b]	1.40 ± 0.35[b]
Pachytene spermatocytes	4.93 ± 0.21[a]	5.31 ± 0.11[a]	3.80 ± 0.16[b]	23.56 ± 2.44[a]	12.48 ± 0.89[b]	4.88 ± 0.57[c]
Secondary spermatocytes	0.25 ± 0.05	0.23 ± 0.60	0.22 ± 0.08	1.21 ± 0.29[a]	0.54 ± 0.15[b]	0.27 ± 0.11[b]
Spermatids with spherical nuclei	4.46 ± 0.23[a]	4.10 ± 0.18[a]	2.73 ± 0.25[b]	21.03 ± 1.63[a]	9.63 ± 1.07[b]	3.49 ± 0.44[c]
Spermatids with elongated nuclei	1.42 ± 0.13	1.59 ± 0.06	1.37 ± 0.12	6.83 ± 1.01[a]	3.72 ± 0.25[b]	1.72 ± 0.18[c]
Sertoli cells	1.80 ± 0.10[a]	1.27 ± 0.10[b]	2.18 ± 0.17[a]	8.48 ± 0.81[a]	2.97 ± 0.28[b]	2.71 ± 0.15[b]
Interstitial structures						
Leydig cell nuclei	1.20 ± 0.16	1.36 ± 0.22	0.93 ± 0.08	5.55 ± 0.62[a]	3.15 ± 0.42[b]	1.17 ± 0.12[c]
Leydig cell cytoplasm	5.92 ± 0.20[c]	17.11 ± 0.58[a]	8.87 ± 1.07[b]	27.81 ± 1.39[b]	40.44 ± 3.72[a]	11.20 ± 1.64[c]
Blood vessels	4.65 ± 0.37[b]	7.08 ± 0.97[a]	2.53 ± 0.32[c]	21.85 ± 1.66[a]	16.73 ± 2.59[a]	3.11 ± 0.31[b]
Lymph vessels	1.84 ± 0.89	2.94 ± 0.28	1.16 ± 0.22	9.39 ± 5.16	6.87 ± 0.71	1.42 ± 0.24
Seminiferous tubules	68.66 ± 1.65[a]	48.55 ± 1.71[c]	60.37 ± 1.31[b]	324.13 ± 20.98[a]	114.38 ± 9.30[b]	76.34 ± 5.52[b]

Means for a given parameter within rows with different superscripts (a, b, c) are significantly different ($P < 0.05$).
* Mean ± SEM.

Table 3. *Average germ cell, Leydig cell, and Sertoli cell sizes in adult boars of three breeds*

Cell type	Nuclear diameter (μm)			Nuclear volume (fl)		
	Whitecross	Meishan	West African	Whitecross	Meishan	West African
Type A spermatogonia	9.51 ± 0.18[a*]	9.58 ± 0.16[a]	8.39 ± 0.27[b]	451.72 ± 25.35[a*]	462.46 ± 22.78[a]	312.96 ± 32.29[b]
Type B spermatogonia	9.70 ± 0.18[a]	10.14 ± 0.21[a]	8.69 ± 0.19[b]	480.22 ± 27.42[a]	549.43 ± 35.93[a]	345.70 ± 21.29[b]
Leptotene spermatocytes	9.25 ± 0.12[a]	9.05 ± 0.07[a]	6.91 ± 0.42[b]	414.76 ± 16.96[a]	388.86 ± 8.56[a]	179.95 ± 29.55[b]
Zygotene spermatocytes	9.87 ± 0.16[a]	9.32 ± 0.15[b]	7.69 ± 0.21[c]	507.43 ± 29.08[a]	425.56 ± 21.41[b]	240.29 ± 20.14[c]
Pachytene spermatocytes	10.70 ± 0.04[a]	10.73 ± 0.13[a]	9.28 ± 0.06[b]	642.99 ± 7.77[a]	648.31 ± 23.18[a]	418.93 ± 8.06[b]
Secondary spermatocytes	7.98 ± 0.21[a]	8.43 ± 0.10[a]	7.32 ± 0.15[b]	267.79 ± 19.51	304.47 ± 10.80	205.88 ± 12.42
Spermatids with spherical nuclei	7.15 ± 0.03[a]	7.04 ± 0.08[a]	6.07 ± 0.05[a]	191.28 ± 2.72[a]	183.13 ± 6.30[a]	117.31 ± 2.87[b]
Leydig cell nuclei	7.01 ± 0.07[b]	7.60 ± 0.17[a]	6.98 ± 0.07[b]	180.30 ± 5.78[b]	231.19 ± 15.45[a]	177.99 ± 5.74[b]
Sertoli cell nuclei	11.32 ± 0.31[b]	12.50 ± 0.48[a]	11.09 ± 0.20[b]	766.89 ± 61.78[b]	1,039.69 ± 119.95[a]	718.54 ± 39.28[b]

Means within rows with different superscripts (a, b, c) are significantly different ($P < 0.05$).
* Mean ± SEM.

Table 4. *Breed variation in basal membrane thickness, luminal diameter, seminiferous tubular diameter, length, and epithelial height in adult boars of three breeds*

Parameter	Whitecross	Meishan	West African
Basal membrane thickness (μm)	4.62 ± 0.23*	4.89 ± 0.20	5.04 ± 0.20
Luminal diameter (μm)	86.69 ± 10.59[a]	93.77 ± 8.44[a]	59.90 ± 2.07[b]
Seminiferous tubular diameter (μm)	257.44 ± 7.61[a]	287.32 ± 14.89[a]	132.96 ± 2.47[b]
Seminiferous tubular length (km)	6.26 ± 0.39[a]	1.78 ± 0.12[b]	6.02 ± 0.69[a]
Seminiferous epithelial height (μm)	85.37 ± 4.92[a]	96.78 ± 8.07[a]	36.53 ± 0.78[b]

Means for a given parameter within rows with different superscripts (a, b, c) are significantly different ($P < 0.05$).
* Mean ± SEM.

Given that the three breeds evaluated in this study belonged to the same age group (as no significant difference was detected in their age ranges), the variation in their testicular parenchymal weights cannot be explained by the different ages at which they attained puberty alone. Rather, as has been suggested (Green and Winters, 1945; VanDemark, 1956), testicular growth is considered to be highly associated with body weight. The highly significant correlation ($P < 0.01$) between testis weight and body size obtained in this study ($r = 0.94$) is similar to the value of $r = 0.90$ reported for bulls by VanDemark (1956) but much higher than $r = 0.42$ presented by Almquist and Amann (1961) for dairy bulls.

The breed-related variation in parenchymal weights was also reflected in their testicular composition. Thus, the volume density of seminiferous tubules was highest in the WC but lowest in the M, the latter also having the least seminiferous tubular length but higher tubular diameter (Table 4). This result is consistent with the observations of Bascom and Osterud (1925) that the volume

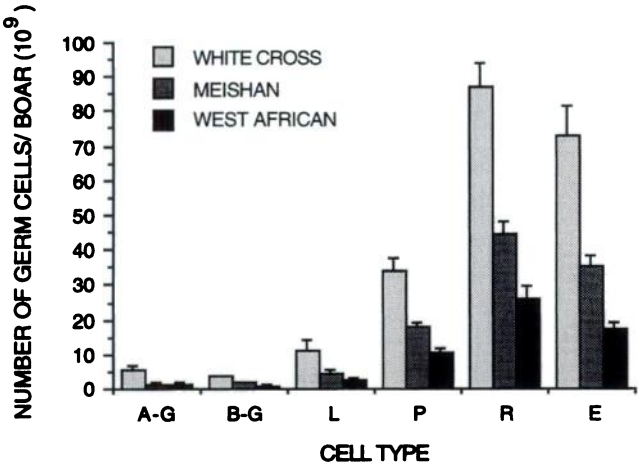

FIG. 2. The numbers of germ cell types per boar ($\times 10^9$) in 15 adult boars. All respective germ cell numbers were higher ($P < 0.05$) in the Whitecross (WC) than in the Meishan (M) or West African (WA) due to its larger parenchymal size. Number of A spermatogonia was similar between M and WA despite differences in testis size. Other advanced germ cell populations were intermediate in the M. A-G: type A spermatogonia; B-G: type B spermatogonia; L: leptotene spermatocytes; P: pachytene spermatocytes; R: round spermatids; E: elongated spermatids.

of the seminiferous epithelium is determined largely by the length of seminiferous tubules and not the diameter. However, considering that other extratubular components contribute to the absolute weight of the testis, the M, having the highest volume densities of interstitial components (Fig. 1) coupled with its enhanced seminiferous tubular diameter, compensated for this and was intermediate in testis size between the WC and WA (Table 1). Such compensatory interactions have also been observed in age-related studies in boars (Dierichs et al, 1973; van Straaten and Wensing, 1977, 1978; Allrich et al, 1983) where an inverse relationship between the seminiferous tubules and Leydig cell volume densities was reported. However, despite this variation, the volume density of seminiferous tubules recorded for our boars are within the range of values (62–73%) reported by Bascom and Osterud (1925) and by Kenelly and Foote (1964) (70%) for 2-year-old Yorkshire boars. Though somewhat similar to 61.3% reported for the stallion (Swierstra et al, 1974), they are clearly smaller than values for most ruminant testes such as 76% for the ram (Hochereau-de Reviers et al, 1993), 82% for the water buffalo (Pawar and Wrobel, 1991), and 76% for the shorthorn bull (Swierstra, 1966).

The length of the seminiferous tubules in the WC (6.3 ± 0.4 km) and WA (6.0 ± 0.7 km) are comparable to values (6.3 km) reported by Bascom and Osterud (1925) for boars. However, the low correlations (0.28 and 0.11; $P > 0.05$) between the seminiferous tubular length with parenchymal and body weights, respectively, show no direct relationship between these parameters. Thus, factors other than body size and parenchymal weight (such as tubular diameter and volume density) must be responsible for differences in the tubule lengths observed in these breeds. Basal membrane thickness was similar for the three breeds, indicating that under normal physiological conditions, breed has no effect on this seminiferous tubule component. Our range of values for tubular diameter is comparable to 223–225 μm, reported by Bascom and Osterud (1925), but the low correlation coefficient ($r = 0.37$; $P > 0.05$) indicates no direct relationship between tubular diameter and body or testis size.

The boars used for this study were raised in different locations (WC and M, Nebraska; WA, Nigeria). As such

Table 5. *Yield of germ cells per type A spermatogonia at various points in spermatogenesis in adult boars of three breeds*

Parameter (ratio)	Whitecross	Meishan	West African
Type B spermatogonia/type A spermatogonium	0.67 ± 0.08*	1.04 ± 0.24	0.61 ± 0.08
Pachytene spermatocytes/type A spermatogonium	7.20 ± 2.63	12.64 ± 1.59	9.12 ± 1.44
Spermatids with spherical nuclei/type A spermatogonium	17.61 ± 3.91	31.69 ± 4.80	21.22 ± 3.05
Elongated spermatids/type A spermatogonium	14.59 ± 3.05	24.42 ± 2.60	14.84 ± 3.27

* Mean ± SEM.

the effect of environmental factors (temperature, humidity, etc.), management, nutrition, and disease load on testicular functions cannot be ignored. Furthermore, due to the difficulty encountered in procuring these samples, a different fixation method was used for the WA than for the WC and M. While these factors likely did not alter the results, the possibility that they may have had impact on some of the testicular parameters could not be eliminated totally. The abundance of type A spermatogonia (Fig. 2) in WC would appear to confer breed advantage on it especially in relation to initial steps of spermatogenesis. In horses the observed seasonal fluctuations in quantitative production of spermatozoa have been related to the size of type A spermatogonial population (Johnson, 1985), which is in turn dependent on the number of stem cells per testis, the scheme or pattern of stem cell renewal (Johnson, 1991), the number of cell divisions from stem cells to the primary spermatocytes (Hochereau-de Reviers, 1981), and the amount of degeneration of specific subtypes of spermatogonia (Huckins, 1978; Johnson, 1995). Thus, it is believed that the highest number of spermatids in the WC was established at the spermatogonial level by the largest population size of A spermatogonia. However, the ratio of the number of type B to type A spermatogonia was slightly higher in the M (1:1.04) than in the WC (1:0.67) or WA (1:0.61). Given that the scheme of stem cell renewal and spermatogonial divisions are thought to be similar for the three breeds, variation in spermatogonial production may be ascribed to a lower index of cellular degeneration affecting spermatogonial mitosis or differentiation in the M.

In spite of the heavier testes of the Meishan breed, number of type A spermatogonia was similar between the M and WA breeds. However, as spermatogenesis progressed, the M seemed to assume a number of germ cells (Fig. 2) intermediate between the WC and WA. This trend appears to have arisen from less germ cell degeneration in the M, thereby resulting in a higher cellular yield. Considering spermatogenesis from spermatocytogenesis to the end of the second meiotic division, the number of spermatids with spherical nuclei per type A spermatogonium ($\times 10^9$) was greater in the M (31.7 ± 4.8) than in the WC (17.6 ± 3.9) or WA (21.2 ± 3.1). This implies that the M has the least degeneration of germ cells during this period. The yield of spermatids per type A spermatogonium is one of the reliable measures of efficiency of spermatogenesis (Berndtson and Igboeli, 1989). This greater number of spermatids with spherical nuclei per type A spermatogonium in the M would suggest minimal germ cell degeneration occurring between A spermatogonia and spermatids with spherical nuclei and is indicative of a trend for a more efficient spermatogenic process in this breed. In conclusion, seminiferous tubules occupied the same volume density in the WA and WC, but were lowest in the M who had the highest density of Leydig cells. Due to larger testes, the numbers of all germ cell types and Sertoli cells were higher in the WC than in the WA or M who had similar numbers of A spermatogonia. Since the WA was less efficient in producing subsequent germ cell progeny than was the M, the M had a number of spermatids intermediate to those of the WA and WC.

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Announcement

Nominations Are Now Being Accepted for the 1997 Distinguished Andrologist, Distinguished Service, and Young Andrologist Awards

Members of the American Society of Andrology are invited to nominate individuals for our most prestigious awards.

The *Distinguished Andrologist Award* is presented to a senior investigator who has made outstanding contributions to the progress of Andrology; the recipient does not have to be a Member of the American Society of Andrology.

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1977	Roberto E. Mancini	1987	Emil Steinberger
1978	Robert J. Hotchkiss	1988	Yves W. Clermont
1979	Thaddeus Mann	1989	C. Alvin Paulsen
1980	John MacLeod	1990	Marie-Claire Orgebin-Crist
1981	Alexander Albert	1991	Philip Troen
1982	Eugenia Rosemberg	1992	C. Wayne Bardin
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The *ASA Distinguished Service Award* recognizes an individual who has provided distinguished service to the American Society of Andrology. The recipient must have been a member of the Society for at least ten years, must have no direct affiliation with the organizational sponsors of the award, and must have served the Society in at least three of the following categories: (1) Chair of a standing committee; (2) Member of the Executive Council; (3) Editor of the *Journal of Andrology;* (4) Chair of an Annual Meeting; (5) Officer of the Society. Sponsored by the Genetics and IVF Institute.

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Your letter of nomination should include the specific reason for nominating the individual for the selected award and the address and phone number of your nominee. It is *not* necessary to include a curriculum vita.

The Awards Committee of the American Society of Andrology thanks you for your assistance. If you have any questions, please contact Peter Schlegel at the above address, by telephone (212) 746-5491, or Email: pnschleg@mail.med.cornell.edu.

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