Spontaneous Benign Prostatic Hyperplasia in the Beagle

AGE-ASSOCIATED CHANGES IN SERUM HORMONE LEVELS, AND THE MORPHOLOGY AND SECRETORY FUNCTION OF THE CANINE PROSTATE

C. B. BRENDLER, S. J. BERRY, L. L. EWING, A. R. MCCULLOUGH, R. C. COCHRAN, J. D. STRANDBERG, B. R. ZIRKIN, D. S. COFFEY, L. G. WHEATON, M. L. HILER, M. J. BORDY, G. D. NISWENDER, W. W. SCOTT, and P. C. WALSH, *The James Buchanan Brady Urological Institute, Department of Urology, Department of Population Dynamics, and Division of Comparative Medicine, Department of Pathology, The Johns Hopkins University, Baltimore, Maryland 21205*

ABSTRACT This paper is a cross-sectional study of spontaneous benign prostatic hyperplasia (BPH) in a single canine species. The effects of aging and hormonal changes on the growth, histology, and glandular secretory function of the canine prostate were studied in 42 male beagles ranging in age from 8 mo to 9 yr. The beagle prostate enlarges for at least 6 yr, whether normal or hyperplastic. In contrast, prostatic secretory function, determined by ejaculate volume and total ejaculate protein, declines markedly after 4 yr of age. These reciprocal growth and functional changes in the prostate are closely associated with a progressive increase in the incidence of BPH, which is already apparent in some dogs by age two. With age there is a modest decrease in serum androgen levels with no apparent change in serum 17β-estradiol levels. This suggests that the growth and functional changes that are associated with the development of BPH and are initiated very early in life reflect an altered sensitivity of the prostate to serum androgens or a response to the relative decrease in the serum androgen to estrogen ratio.

INTRODUCTION

Benign prostatic hyperplasia (BPH)[1] in the dog is at present the only familiar animal model for the human disease. Canine BPH is characterized by diffuse epithelial or glandular proliferation throughout the prostate (1) whereas human BPH is thought to arise specifically within the periurethral tissue and is characterized primarily by stromal hyperplasia (2, 3). Despite these differences, there are sufficient similarities between the two species to regard the canine condition as a useful model for comparison to the human disease (4–9).

Although the etiology and pathogenesis of the disease in both species is uncertain, it seems clear that hormonal factors are involved. In both man and dog, BPH is an age-related disease (10), and its development requires the presence of functioning testes (11). Walsh and Wilson (12) demonstrated that BPH could be induced in young castrated mongrels by the concomitant administration of androgens and estrogens, and this observation has been substantiated in subsequent studies (13, 14). To date, however, there have been no canine studies that have examined the relationships between age-associated endocrine changes and the spontaneous alterations in the morphology and secretory function of the prostate. In this study, untreated beagles of known age were used to perform a cross-sectional study investigating these interrelationships.

METHODS

Animals

42 healthy male beagles ranging in age from 8 mo to 9 yr were obtained from Laboratory Research Enterprises

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[1] *Abbreviations used in this paper:* BPH, benign prostatic hyperplasia; DHT, dihydrotestosterone.

Ltd., Kalamazoo, MI. The birth date and pedigree of each dog were known, and none of the dogs had been used previously for research purposes. The dogs were allowed 4 mo to adjust to housing conditions before starting the experiment.

Serum hormone determinations

Blood samples of 10 ml were collected weekly from the cephalic vein of each dog on three different occasions. All collections were made between 9:30 and 11:30 a.m. The three samples from each dog were centrifuged and the serum was pooled. Serum testosterone (17β-hydroxy-4-androstene-3-one), dihydrotestosterone (DHT, 17β-hydroxy-5α-androstane-3-one), 3α-androstanediol (5α-androstane-3α,17β-diol), and 17β-estradiol (1,3,5(10)-estratrien-3,17β-diol) levels were determined by the methods reported in detail by Cochran et al. (15).

Ejaculate collections

Since the dog possesses neither seminal vesicles nor bulbourethral glands, prostatic secretions account for >97% of the ejaculate volume (16). Using noninvasive techniques, we measured ejaculate volume and total protein content to assess prostatic secretory function.

Attempts were made to collect semen from all dogs on four different dates with 2–7-d intervals between collections. Semen was obtained by genital manipulation in the presence of a mongrel bitch in whom estrus had been induced with a subcutaneous silastic implant releasing 1 μg of 17β-estradiol/d. The ejaculates were collected through a funnel into a graduated cylinder containing the proteolytic inhibitors Trasylol (200 U/ml) and leupeptin (5 μg/ml). The ejaculate volumes were measured and recorded, and a 0.1-ml aliquot was removed for subsequent sperm count. The ejaculates were kept on ice and subsequently frozen in liquid nitrogen within 3 h after collection.

Protein determinations

The frozen ejaculates were thawed and a 0.2-ml aliquot was removed from each sample and centrifuged at 12,000 *g* for 5 min. The supernatant was withdrawn and total protein determined by the method of Lowry et al. (17).

Autopsy

All dogs were killed within 3 wk after the final ejaculate collection. The prostates and testes were removed rapidly and were measured and weighed.

Histology

Two cross-sections were taken from each prostate for histological examination; one section from the left midportion and a second from the right midportion of the gland. All sections included the urethral mucosa and the outer margin of the prostate. The tissues were fixed in 10% neutral buffered formalin, embedded in paraffin, mounted, and stained with hematoxylin and eosin. A study of histological sections by light microscopy was made by a veterinary pathologist (J.D.S.) without prior knowledge of the age of the dog or the weight of the gland from which each specimen was obtained.

Four histologic patterns were identified: immature, normal, benign glandular hyperplasia, and benign complex hyperplasia. Complex hyperplasia was formerly termed cystic hyperplasia (14). We feel that complex hyperplasia is a more accurate term, since it represents a histologic pattern in which cystic elements predominate but areas of glandular hyperplasia and foci of atrophy can be observed as well. Detailed descriptions of the four histologic types follow and representative photomicrographs are shown in Fig. 1.

Immature pattern. Glands from sexually immature animals are characterized by a branching ductular system. In contrast to the normal adult gland, the alveolar portions of the gland are not developed, and there is no evidence of secretory activity by the ductal cells. The epithelial cells are small and cuboidal with a relatively high nuclear to cytoplasmic ratio. The cytoplasm is pale and amphiphilic. There is no evidence of eosinophilic secretion within these cells. There is relatively more connective tissue stroma in the immature gland than in the fully developed gland. This stroma is composed principally of delicate fibrous connective tissue, but smooth muscle is found in the outer capsule and in major trabeculae radiating into the gland. There is considerable variation in the tubular development in the glands of the immature dogs. Some have only very rudimentary branchings of the tubular system, while others show greater development and differentiation. In some animals, there is also evidence of early secretory activity in a few of the epithelial cells.

Normal pattern. Normal prostates are characterized by compound tubular alveolar glands that radiate from their urethral duct openings. The cells lining all portions of the duct are columnar to cuboidal with basally located nuclei that are round to oval in shape. The cytoplasm of the cells is intensely eosinophilic; this is especially marked in the apical portions of the cells. The alveolar portions of the gland contain primary and secondary infoldings of secretory epithelium that project into the alveolar lumen. The lumens of the glands are usually not dilated and they do not contain obvious secretory material. The alveoli are separated by a delicate fibrous connective tissue stroma. This stroma also has larger elements composed of smooth muscle that divide the gland into irregular lobules. These smooth muscle septa are contiguous with the capsular stroma which surrounds the gland and are also connected with the smooth muscle and fibrous tissue located in the periurethral area. Occasionally mononuclear inflammatory cells (lymphocytes and plasma cells) are seen in the stroma, most commonly in the periurethral area.

Benign glandular hyperplasia. In benign glandular hyperplasia there is an obvious increase in the amount of secretory epithelium. Each of the lobules is larger and has more elaborate branching than in the normal gland. The alveoli are larger and contain more cells. As a result, the papillary projections of secretory epithelium into the alveoli are more elaborate. In addition, the size of the secretory epithelial cells is increased due principally to an increase in the amount of cytoplasm. In many of these glands, the amount of stroma is relatively less than in the normal gland, and the septa appear somewhat attenuated. In this form of BPH, the glandular proliferation occurs in all portions of the gland but often assumes a more nodular pattern in the periurethral areas.

Benign complex hyperplasia. In the complex form of BPH in the dog, there are a variety of changes occurring within the affected glands. Areas of glandular hyperplasia are intermingled with foci in which the secretory epithelium is atrophic and attenuated. In these atrophic areas, there is a relative increase in the stroma, which is composed of both collagen and smooth muscle. Some of the alveoli are dilated

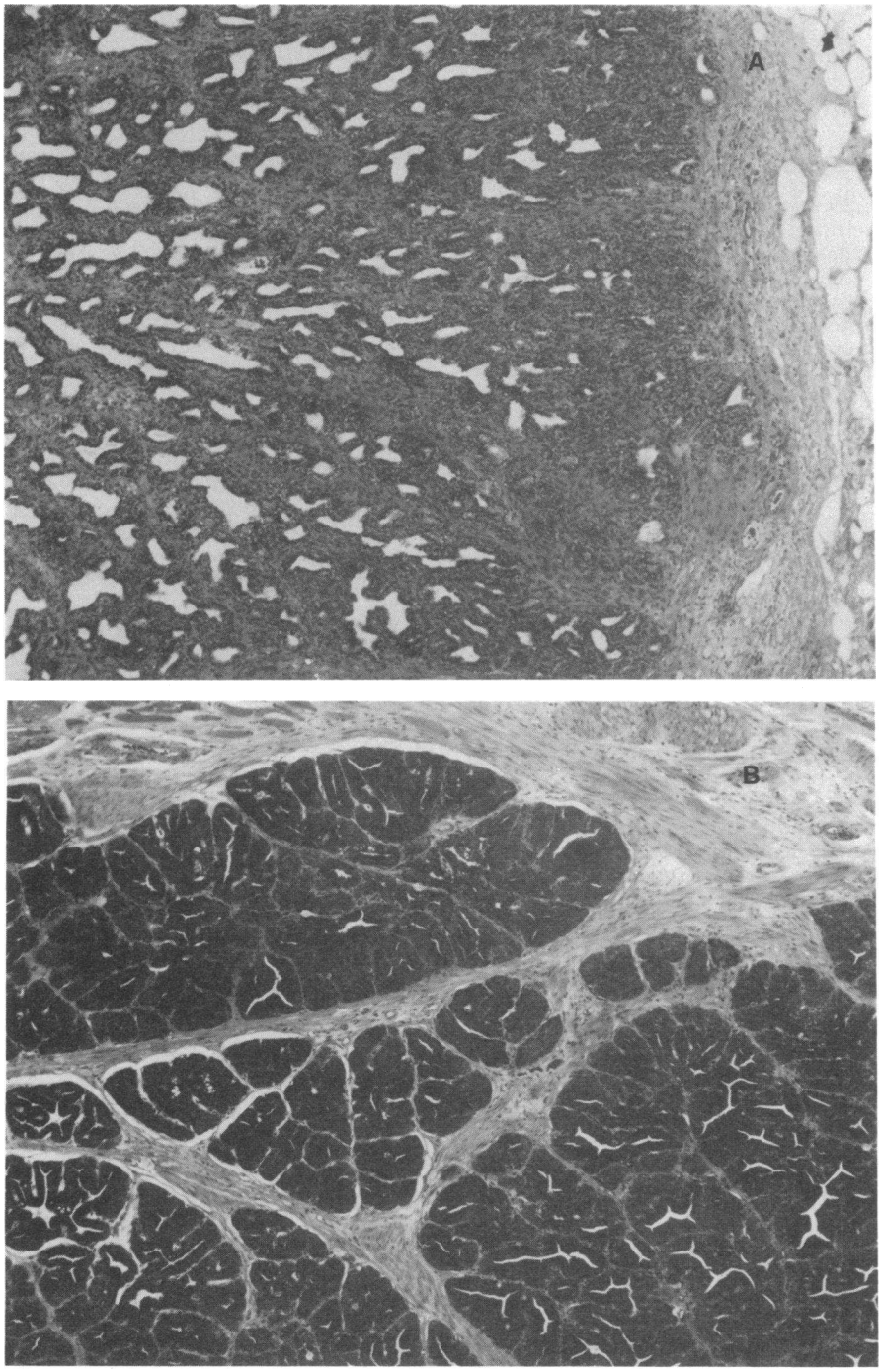

FIGURE 1 (A) Immature prostate gland with poorly developed alveoli showing no evidence of secretory activity by the ductal cells. ×70. (B) Normal prostate gland with well-developed secretory epithelium. Alveoli are separated by delicate bands of connective tissue stroma. ×70. (C) Benign glandular hyperplasia. The alveoli are larger and the papillary projections of secretory epithelium are more elaborate than in the normal prostate. ×70. (D) Benign complex hyperplasia. Areas of glandular hyperplasia are intermingled with foci of atrophy. Many alveoli are cystically dilated with flat cuboidal epithelium showing no evidence of secretory activity. Amount of stroma is increased. ×70.

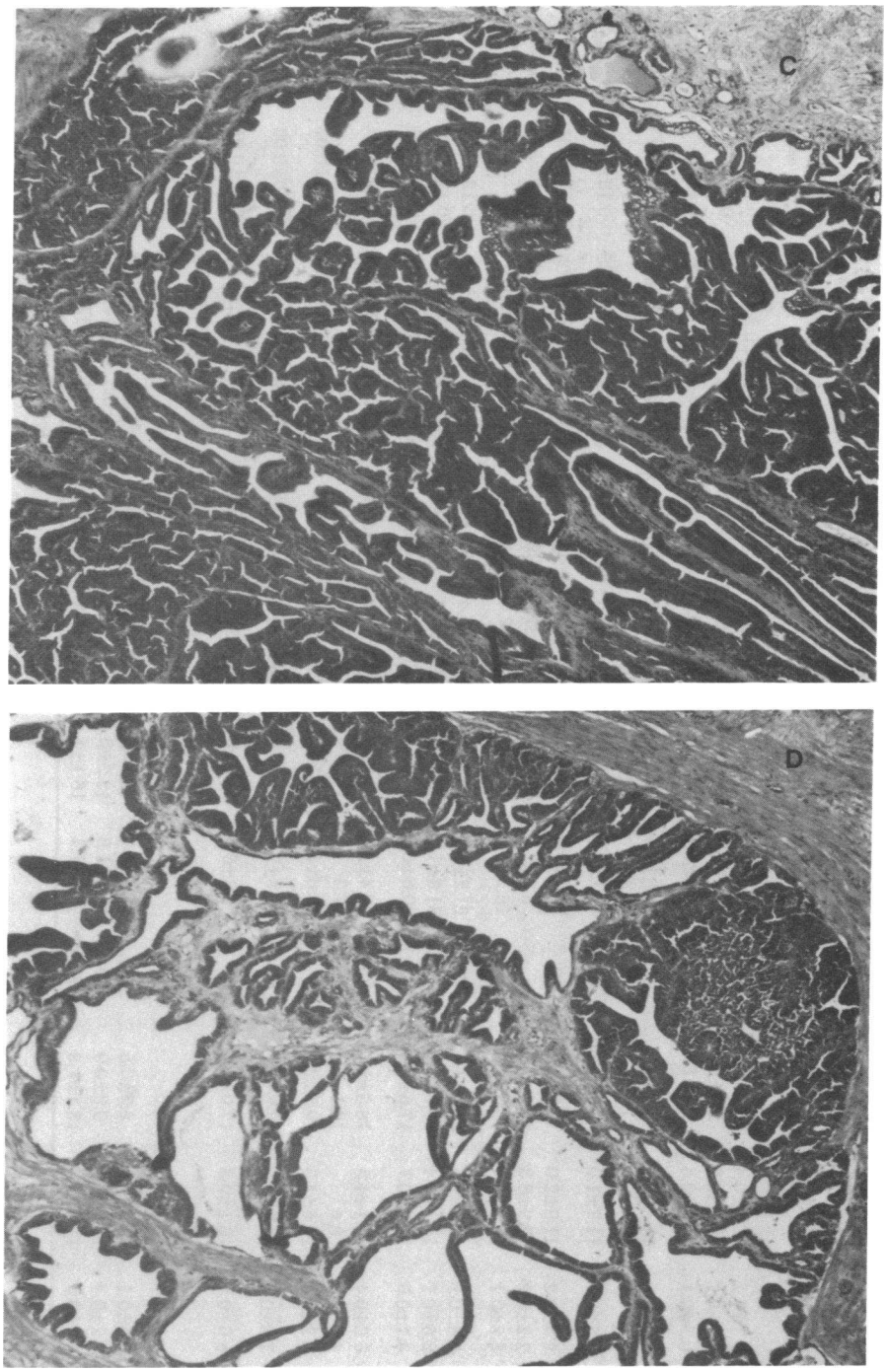

FIGURE 1 (*Continued*)

and cystic and filled with eosinophilic material; it is these cysts that characterize this condition. The nature of the epithelium lining the cysts varies from plump columnar cells with eosinophilic cytoplasm to thin cuboidal cells without obvious morphologic evidence of secretory activity. Cysts can be present anywhere in the gland, although they are most often found in the periurethral area. Chronic inflammation is common and usually located in the stromal bands. Occasionally in areas of marked inflammation there is squamous metaplasia of the secretory epithelium.

TABLE I

Mean Body Weights, Prostatic Weights, Ejaculate Volumes, Total Ejaculate Proteins, Sperm Counts, and Serum Hormone Concentrations for All Age Groups of Dogs

Group	No. of beagles	Mean age	Body wt	Prostate		Ejaculate*				Serum hormones§			
							Protein		Sperm/ ejaculate‡				
				Weight	Histology (% BPH)	Vol	mg/ml	Total	($\times 10^{-6}$)	Testo‖	DHT	3a Diol¶	Estradiol
		yr	*kg*	*g*		*ml*				*ng/ml*	*ng/ml*	*ng/ml*	*pg/ml*
I. All	7	0.7±0.1	9.1±0.4	3.6±1.0	0	4.1±2.1	9.5±0.9	42±23.2	99±41	2.2±0.7	0.25±0.04	0.18±0.03	8.6±1.3
Immature	5	0.7±0.1	8.7±0.2	2.1±0.3		1.3±0.2	9.5±1.4	12±3.9	59±13	1.2±0.2	0.21±0.03	0.17±0.04	9.8±1.0
Normal	2	0.7±0.1	10.1±0.8	7.3±1.4		8.4±3.4	9.5±1.6	85±46.2	136±70	4.5±0.3	0.37±0.01	0.19±0.03	5.5±3.5
II. All	7	1.3±0.1	11.6±0.8	7.3±1.1	0	10.8±2.4	14.1±1.1	146±29.7	372±29	3.3±0.6	0.49±0.05	0.12±0.03	7.8±0.9
Immature	1	1.4	10.4	4.3		9.3	17.8	166	419	2.4	0.33	0.09	6.0
Normal	6	1.3±0.1	11.7±1.0	7.8±1.1		11.1±2.8	13.4±1.1	143±34.8	370±56	3.5±0.7	0.52±0.05	0.13±0.03	8.2±1.0
III. All	5	2.4±0.2	11.9±2.0	13.1±1.6	40	11.4±2.3	15.4±1.1	181±46.2	226±139	2.9±0.6	0.41±0.05	0.11±0.01	8.4±0.9
Normal	3	2.2±0.2	12.9±3.3	11.2±1.9		8.2±2.1	14.6±1.1	118±31.5	221±133	3.6±0.7	0.44±0.05	0.11±0.01	9.0±0.6
BPH	2	2.6±0.1	10.3±1.0	15.8±1.7		16.2±1.5	16.7±2.3	274±62.3	314±178	1.9±0.9	0.36±0.08	0.11±0.01	7.5±2.5
IV. All	5	3.8±0.2	11.9±0.6	15.5±1.4	40	21.2±1.1	15.7±1.3	333±29.4	204±20	2.9±0.4	0.41±0.05	0.12±0.03	8.4±0.5
Normal	3	3.6±0.1	11.1±0.4	14.0±1.0		21.6±1.9	14.1±0.6	304±24.2	191±17	2.8±0.4	0.38±0.06	0.11±0.02	8.6±0.7
BPH	2	4.1±0.6	12.9±0.7	17.6±2.7		20.6±0.4	18.2±2.4	376±61.2	223±9	3.1±1.0	0.44±0.09	0.12±0.04	8.0±1.0
V. All	8	6.2±0.1	12.1±1.3	18.3±2.4	88	8.4±1.4	15.2±1.7	133±26.4	174±84	2.4±0.5	0.36±0.07	0.09±0.01	6.8±0.5
Normal	1	6.0	8.6	12.1		7.3	11.8	86	7	3.0	0.31	0.10	6.0
BPH	7	6.2±0.1	12.6±1.3	19.2±2.5		8.6±1.6	13.5±2.8	121±32.2	276±82	2.3±0.6	0.36±0.07	0.09±0.01	7.0±0.5
VI. All BPH	6	7.2±0.1	11.4±0.5	18.5±2.7	100	6.8±1.5	18.5±3.0	137±40.0	96±25	2.1±0.4	0.39±0.03	0.10±0.02	9.0±0.5
VII. All BPH	4	8.6±0.3	10.1±0.3	17.0±2.5	100	2.3±0.3	10.0±0.4	22±2.3	177±55	2.1±0.5	0.32±0.07	0.12±0.01	8.5±1.0
Group totals**													
All immature	6	0.8±0.1	9.0±0.3	2.5±0.5	0	3.3±2.0	11.6±2.3	51±38.4	166±66	1.4±0.3	0.23±0.03	0.16±0.04	9.2±1.0
All normal	15	2.2±0.4	11.4±0.7	9.9±0.9	0	12.0±1.8	13.2±0.6	159±25.4	253±45	3.5±0.3	0.44±0.03	0.13±0.01	8.0±0.6
All BPH	21	6.4±0.4	11.6±0.5	18.1±1.2	100	9.1±1.4	16.1±1.3	160±28.2	199±25	2.2±0.3	0.37±0.03	0.11±0.01	8.0±0.4

* Average ejaculate values of four collections.

‡ Average sperm count of four collections.

§ Average of three collections.

‖ Testosterone.

¶ 5α-Androstane-3α,17β-diol.

** Values from groups I through VII separated by histologic diagnosis.

TABLE II

Histologic Patterns for All Age Groups of Dogs

Group no.	Average age	Percentage of dogs with prostate pathology			
		Immature	Normal	Glandular BPH	Complex BPH
	yr				
I.	0.7	72	28	—	—
II.	1.3	14	86	—	—
III.	2.4	—	60	40	—
IV.	3.8	—	60	40	—
V.	6.2	—	12	38	50
VI.	7.2	—	—	50	50
VII.	8.6	—	—	25	75

RESULTS

Dogs of approximately the same age were grouped together to form seven groups (I–VII) covering the age range from 0.7 to 8.6 yr. Table I details the number of dogs in each group and their mean ages and body weights. Data relating to prostatic weight, histology, secretory function, sperm counts, and serum hormone levels are presented. Mean values±SE are given for all dogs in each group and then subdivided into histologic categories within each group.

Prostatic weight. The beagle prostate appeared to mature by 1.5 yr of age, and this event was associated with a threefold increase in prostatic weight. Thereafter, prostatic weight continued to increase until age

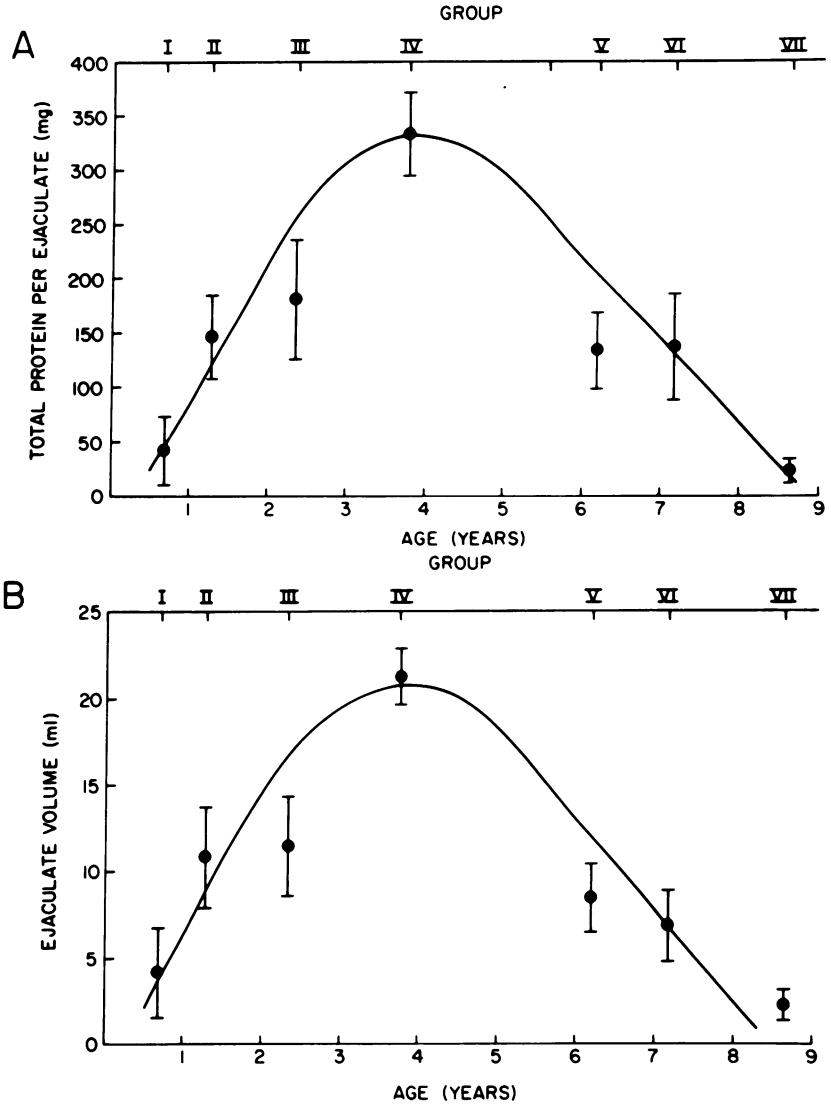

FIGURE 2 Mean (±SE) total ejaculate protein (A) and mean ejaculate volume (B) are plotted for each age group of dogs.

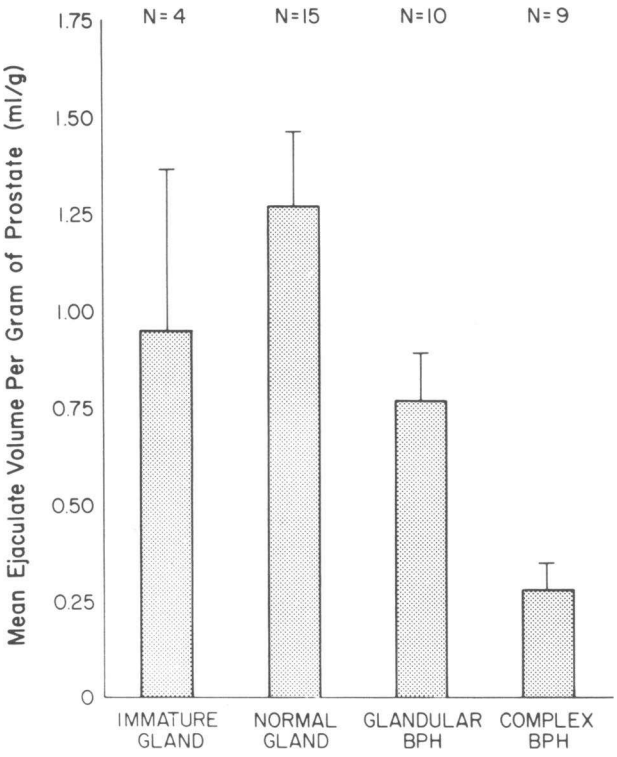

FIGURE 3 Mean (±SE) ejaculate volume per gram of prostate is plotted for each histologic group of dogs regardless of age.

six. This growth was observed in both normal and hyperplastic glands, although the hyperplastic glands in age groups III, IV, and V averaged slightly more than the normal glands. No further increase in mean prostatic weight was noted after age six.

Prostatic histology. Table II shows the histologic classification within each age group. Over three-fourths of the prostates had matured by age 1.3 yr. Thereafter, beginning with group III (mean age 2.4 yr) there was a progressive increase in the incidence of BPH. By age 7 yr (group VI), all dogs showed histologic evidence of the disease. Glandular hyperplasia was the predominant pattern observed in dogs <5 yr of age, and preceded the changes of complex hyperplasia seen in older dogs.

Prostatic secretory function. Fig. 2 shows the relationship between prostatic secretory function and age. As the prostate matured, there was a threefold increase in secretory function determined by both total ejaculate protein and ejaculate volume (group I vs. group II). Secretory function continued to increase until age four (group IV), after which there was a marked decline with further aging.

The decline in secretory function was associated with the development of BPH. In Fig. 3 the secretory efficiency of the prostate, expressed as ejaculate volume per gram of prostate, is shown for each histologic group regardless of age. Secretory efficiency decreased 80% comparing complex BPH to normal prostates.

Sperm counts. Sperm counts were maximal in group II (mean age 1.3 yr) and increased threefold over group I (mean age 0.7 yr), undoubtedly reflecting testicular maturation. Thereafter, although there was considerable variation among dogs in each age group, sperm counts tended to decrease with age (Table I).

Serum hormone levels. Serum testosterone was highest in group II (mean age 1.3 yr) and was increased threefold over the mean level of testosterone in immature dogs in group I (mean age 0.7 yr). Thereafter, there was a gradual 35% decline in serum testosterone with further aging. Serum levels of DHT followed a similar pattern with aging. 3α-Androstanediol levels decreased by one-third as the dogs matured and subsequently remained constant with aging. No significant changes in 17β-estradiol levels were observed with aging. Thus, the serum testosterone/17β-estradiol ratio gradually declined with aging reflecting the decrease in serum testosterone.

Relationships between age, serum hormone levels, prostatic weight, secretory function, and the incidence of BPH. The relationships between age, serum hormone levels, prostatic weight, and the incidence of BPH are shown in Fig. 4. This shows that the increase in prostatic weight with aging was paralleled by a progressive increase in the incidence of BPH, and that this occurred while serum testosterone levels were declining. This suggests that with aging the prostate develops an increased sensitivity to serum testosterone or responds to a shift in the testosterone/17β-estradiol ratio in terms of growth, such that lower levels of serum testosterone maintain larger glands in the advanced age groups.

The reciprocal relationships of prostatic growth response and prostatic secretory response to serum testosterone with aging are shown in Fig. 5. This shows that prostatic growth becomes more sensitive to serum testosterone with aging, while in contrast, secretory response to serum testosterone declines markedly.

DISCUSSION

The growth pattern observed in the beagle prostate in this study suggests that the normal prostate continues to enlarge in adult life, and that hyperplasia appears before the gland has reached full development. O'Shea (18) noted a similar pattern in his study of several canine species, and suggested that the endocrine changes responsible for prostatic hyperplasia may be initiated early in adult canine life.

The present study also suggests that canine prostatic

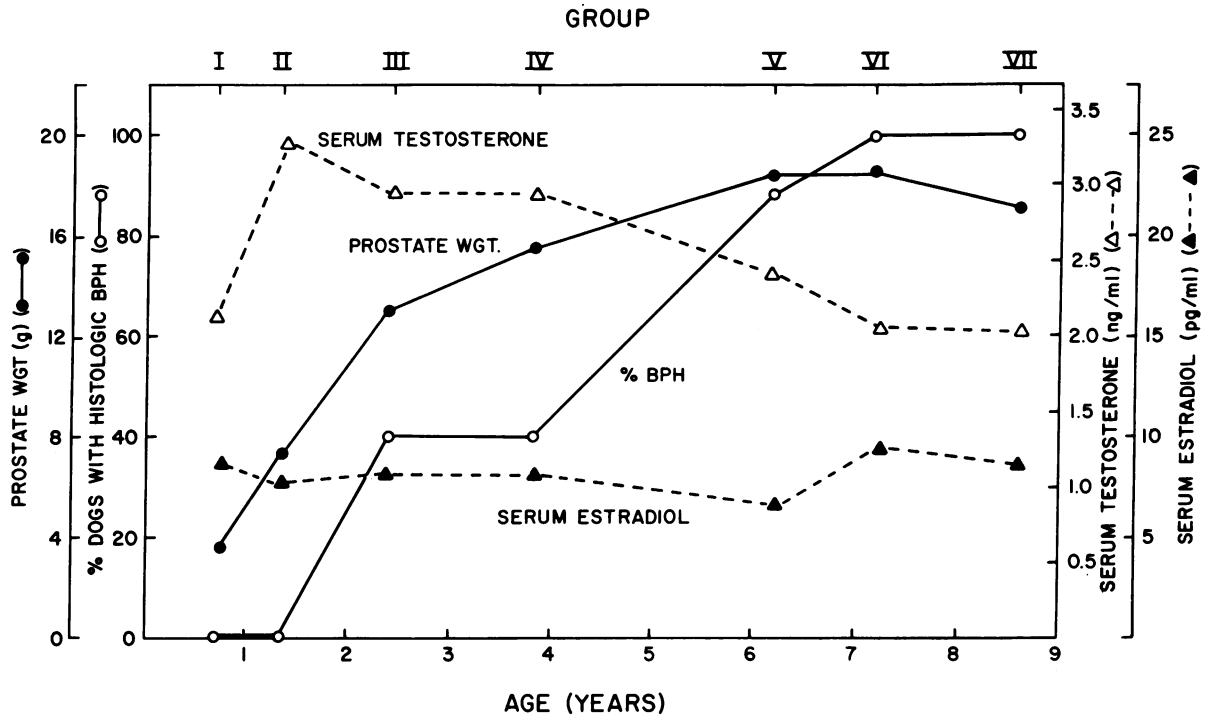

FIGURE 4 Mean serum testosterone, serum 17β-estradiol, prostatic weight, and the percentage of dogs with histologic BPH are plotted for each age group of dogs.

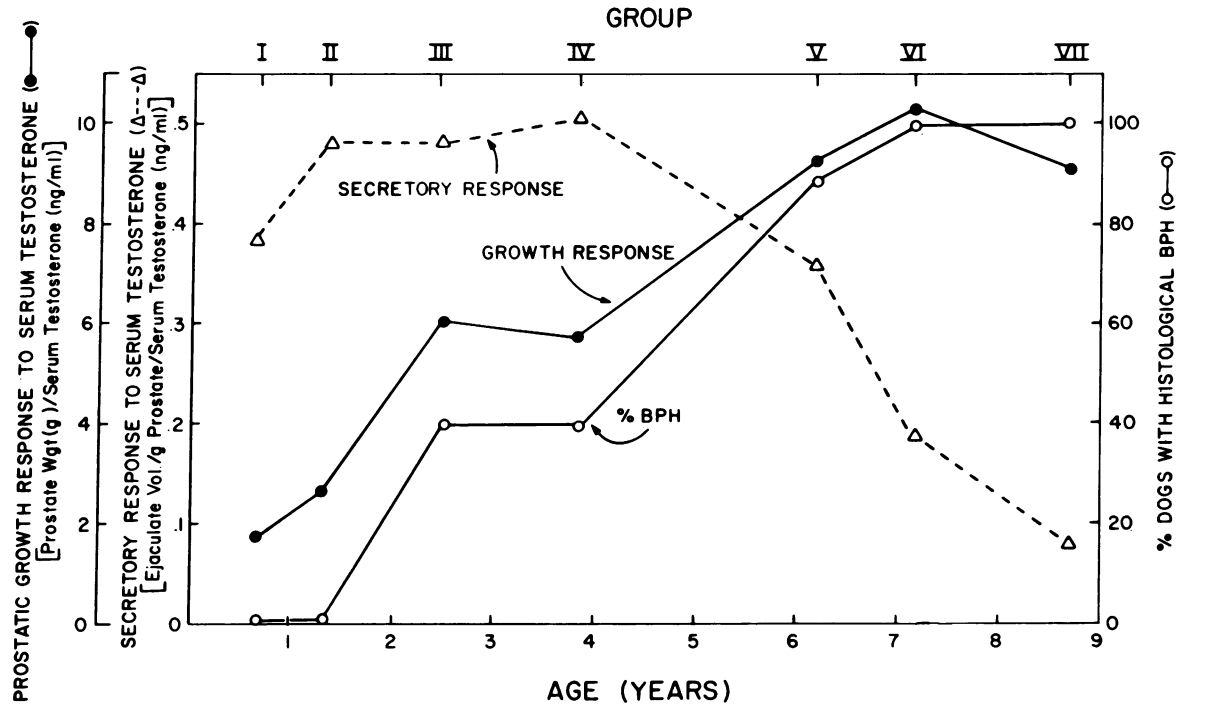

FIGURE 5 Mean prostatic growth and secretory response to serum testosterone and the percentage of dogs with histologic BPH are plotted for each age group of dogs.

growth may diminish in the later years of life. Zuckerman and McKeown (10) observed a similar phenomenon in their study of 243 dogs of many breeds. They concluded that after the 8th yr of life the canine prostate may either undergo involution or continued growth, and that the two possibilities occur with equal frequency.

In this study, early histologic changes of BPH were observed in 40% of dogs as early as 2–3 yr of age. O'Shea (18) observed in his canine study that between 3 and 6 yr of age the normal histologic pattern was frequently replaced by one of glandular hyperplasia. In their human study, Harbitz and Haugen (19) reported a 60% incidence of histologic BPH in men during the fifth decade of life. They did not report the incidence of histologic BPH in men younger than 40, which would have been of interest.

This study demonstrates that prostatic secretory function peaks in the beagle at ~4 yr of age and then declines dramatically. Thus, the secretory capacity of the prostate diminishes as the gland continues to grow and the histologic changes of BPH become more evident (Figs. 3 and 5). The lowest secretory function was observed in dogs with complex (cystic) BPH, which agrees with our earlier results reported by Wheaton et al. (20).

The decline in secretory function is not believed to be due to ductal obstruction (21, 22), and it has been suggested that estrogen can block prostatic fluid and electrolyte transport even in the presence of androgen stimulation (23). Isaacs et al. (23) reported that total ejaculate protein was maintained in dogs with BPH as total ejaculate volume declined; this would support the concept of a selective estrogen blockade of fluid and electrolyte transport. However, in the present series, we found that protein and volume varied similarly, and that as volume declined so did total protein. Thus, whether the decline in prostatic secretory function observed with age is secondary to a specific hormonal effect or whether it is related to prostatic enlargement remains to be determined.

In this study we observed a gradual decline throughout adult life in the mean serum testosterone and DHT concentrations. Mean serum testosterone values also decline in man, but this appears to occur late in life after the age of 60 yr (24, 25). Nawata et al. (26) reported a similar decline in DHT in aging men, but Pirke and Doerr (27) have reported that serum levels of DHT are unchanged with age.

In this study serum 17β-estradiol levels did not change with age, while in man there has been reported to be a slight increase both in serum 17β-estradiol (28) and estrone (29) with age. Nevertheless, we did observe a decline in the serum testosterone to 17β-estradiol ratio with age as observed in man due to the gradual decrease in serum testosterone levels of beagle dogs.

Altered sensitivity of the prostate to serum testosterone or to the testosterone to 17β-estradiol ratio may be a critical factor in the pathogenesis of BPH. The continued growth of the prostate despite decreasing serum testosterone concentrations may reflect a metabolic shift within the prostate favoring the production of the active androgen 5α-DHT. Such a metabolic alteration has been reported previously in the canine studies of Shain and Nitchuk (30) and of Isaacs and Coffey (31). Prostatic DHT levels have been reported to be severalfold greater in hyperplastic tissue compared with normal prostate in both man (32–35) and dog (36, 37), thus providing further evidence for such a metabolic shift.

In summary, this report relates alterations in the morphology and secretory function of the canine prostate to endocrine changes with age. The development of BPH in the dog with the associated changes in prostatic growth and function seem closely related to similar endocrinologic events that are thought to be associated with the pathogenesis of the human disease.

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