

1-1-2008

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ÖZKAN ELMAZ

SERDAL DİKMEN

ÜMİT CİRİT

HIDİR DEMİR

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ELMAZ, ÖZKAN; DİKMEN, SERDAL; CİRİT, ÜMİT; and DEMİR, HIDİR (2008) "Prediction of Postpubertal Reproductive Potential According to Prepubertal Body Weight, Testicular Size, and Testosterone Concentration Using Multiple Regression Analysis in Kıvrıkcık Ram Lambs," *Turkish Journal of Veterinary & Animal Sciences*: Vol. 32: No. 5, Article 3. Available at: <https://journals.tubitak.gov.tr/veterinary/vol32/iss5/3>

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Prediction of Postpubertal Reproductive Potential According to Prepubertal Body Weight, Testicular Size, and Testosterone Concentration Using Multiple Regression Analysis in Kıvrıkcık Ram Lambs

Özkan ELMAZ^{1,*}, Serdal DİKMEN², Ümit CİRİT³, Hıdır DEMİR⁴

¹Department of Animal Husbandry, Faculty of Veterinary Medicine, Mehmet Akif Ersoy University, 15100 Burdur - TURKEY

²Department of Animal Husbandry, Faculty of Veterinary Medicine, Uludağ University, Bursa - TURKEY

³Department of Reproduction and Artificial Insemination, Faculty of Veterinary Medicine, İstanbul University, 34320, Avclar, İstanbul - TURKEY

⁴Department of Animal Husbandry, Faculty of Veterinary Medicine, İstanbul University, 34320 Avclar, İstanbul - TURKEY

Received: 31.07.2007

Abstract: The relationship between the prepubertal body weight, testicular size, testosterone concentration, and postpubertal reproductive function was investigated in Kıvrıkcık ram lambs. The body weight, testicular size, and testosterone concentration were measured every 20 days between 60 and 420 days of age. Semen was collected from the ram lambs at 7, 8, 9, 10, 11, 12, 13 and 14 months of age. Data obtained were analyzed by best subsets regression model. We determined that body weight, scrotal circumference, testicular length, and testosterone levels in the early prepubertal period (days 80, 100, and 120), when combined with each other, allow the determination of the postpubertal (days 220, 240, 260 and 280) testicular diameter.

Body weight and testosterone at prepubertal ages, either alone or in combination with other variables, were related to semen volume at postpubertal ages (at 7, 8, 9, 11, and 14 months of age).

It is concluded that there is a significant relation between postpubertal reproduction characteristics and the selection of Kıvrıkcık ram lambs during the prepubertal period (especially between days 80 and 120) according to body weight, testicular characteristics, and testosterone hormone concentrations. According to the relationship among these characteristics, selection of ram lambs at juvenile ages will improve the effectiveness of the selection and then the economic gain of sheep-breeding operations.

Key Words: Kıvrıkcık ram lambs, testicular size, semen, testosterone, prepubertal, postpubertal prediction

Kıvrıkcık Koç Kuzularda Çoklu Regresyon Analizi Kullanarak Puberta Öncesi Canlı Ağırlık, Testis Büyüklüğü, ve Testesteron Konsantrasyonuna Göre Puberta Sonrası Üreme Performansının Tahmin Edilmesi

Özet: Kıvrıkcık koç kuzularda puberta öncesi vücut ağırlığı, testis ölçümleri, testesteron hormon konsantrasyonu ile puberta sonrası üreme fonksyonları arasındaki ilişki incelendi. Vücut ağırlığı, testis ölçümleri ve testesteron konsantrasyonu 60. günden 420. güne kadar 20 günde bir alınmıştır. Sperma 7, 8, 9, 10, 11, 12, 13 ve 14. aylarda koç kuzulardan toplanmıştır. Elde edilen veriler best subsets regresyon modeline göre analiz edildi. Vücut ağırlığı çoğunlukla sperma motilitesi ve skrotum çevresi ile ilişkili, testesteron ile birlikte kombine edildiğinde puberta sonrasında testis çapı, sperma hacmi ve sperma konsantrasyonu üzerine çok sıkı bir ilişki belirlenmiştir. Bu araştırmada genel olarak Kıvrıkcık koçlarda puberta öncesi erken dönemde (80, 100 ve 120. günler) canlı ağırlık, skrotum çevresi, testis uzunluğu ve testesteron özellikleri birbirleriyle kombine edildiğinde puberta sonrası dönemde (220, 240, 260 ve 280. günler) testis çapı özelliğini önemli düzeyde belirlediği tespit edilmiştir.

Puberta öncesi yaşta canlı ağırlık ve testesteron ya tek başlarına yada diğer değişkenlerle kombine edildiğinde puberta sonrasında (7, 8, 9, 11 ve 14. aylar) sperma hacmi ile ilişkilidir.

Kıvrıkcık koç kuzularda puberta öncesi dönemde (özellikle 80-120 günler arası) canlı ağırlık, testis özellikleri ve testesteron hormon özelliklerine göre yapılacak seleksiyon ile puberta sonrası üreme özellikleri arasında önemli bir ilişkinin olduğu sonucuna varılmıştır. Bu özellikler arası ilişkiye bağlı olarak koç kuzularda genç yaşta yapılacak seleksiyon ile koyunculuk işletmelerinin hem ekonomik karlılığı hem de seleksiyonda isabet derecesi artırılmış olacaktır.

Anahtar Sözcükler: Kıvrıkcık koç kuzular, testis ölçümleri, sperma, testesteron, puberta öncesi, puberta sonrası tahmin

* E-mail: elmaz@mehmetakif.edu.tr

Introduction

Most rams reach puberty between 5 and 9 months of age depending on body weight, nutrition, breed, and various environmental factors (1). Improvement of sheep productivity requires attention on various components, one of which is the early selection of the rams. The statistical analysis of testicular size is of importance, since it is significantly correlated with reproductive activity (2). In Kivircik rams, it was determined that the body weight at the first semen collection is 20.6 kg, the first semen production age is 144 days, and the fertile semen production age is 195.36 days (3).

Generally in rams, testicular measurements are easily measured characteristics at juvenile ages. The most important ones of these features are testicular diameter, testicular length, testicular volume, and scrotal circumference. Testicular diameter has a direct relationship with sperm production. The rams with bigger testes produce more sperm than the rams with smaller testes (4). Prepubertal testicular growing rates have been used to predict adult testicular size and spermatogenic function in bulls (5). Regression equations have revealed that testicular size was positively related to body weight and age in Awassi ram lambs (6). Positive correlations between prepubertal ram lamb hormone levels and subsequent testes size, sperm output and mating frequency have also been reported in Suffolk rams (7).

Considering the morphological and physiological breed characteristics in sheep breeding operations, determination of breeding value of candidate breeder rams at juvenile age is very important. Selection of ram lambs at a juvenile age is an advantage for improvement of both economic gain of operations and effectiveness of selection. This study was carried out in order to demonstrate the relationship between some reproductive characteristics (body weight, testicular size, and testosterone concentration) at juvenile age and postpubertal reproductive features in Kivircik rams.

Materials and Methods

The materials of the study were 15 ram lambs born from 4-5-year-old Kivircik sheep. The study was carried out in clinical boxes of the Faculty of Veterinary Medicine at Istanbul University. The ram lambs were weaned at a mean of 100 days of age. During the suckling period, lambs were supplemented with lucerne and concentrate feed.

Data collection commenced at 60 days of age. Afterwards, measurements of body weight and testicular size, and collection of blood samples for testosterone hormone level determination were performed once every 20 days until the end of the trial (420 days). Semen was collected from the ram lambs at 7, 8, 9, 10, 11, 12, 13, and 14 months of age using an electro-ejaculator. Body weight was recorded in the morning, before feeding. Testicular diameter was recorded with a caliper on the left and right testicles as the widest anteroposterior diameter. Testicular length was also measured with a caliper on both left and right testicles as the distance between the top of the tail and the head of the epididymis. Scrotal circumference was measured with a flexible tape at the point of maximum circumference of paired testes. Paired testes volume were calculated as $0.0396 \text{ (average testis length)} \times \text{(scrotal circumference)}^2$ (8).

Spermatological characteristics were examined at Istanbul University, Faculty of Veterinary Medicine, Department of Artificial Insemination and Reproduction Laboratory. Semen was collected from rams using a manually controlled electro-ejaculator (P-T Electronics, Model 304, USA) with a rectal probe with 3 electrodes. The rectal probe was lubricated and gently inserted into the rectum, and the electrodes were oriented ventrally. Electric current was applied starting from 1 V for 2 sec with 2-sec rest intervals between stimuli, increasing the voltage stimuli by 1 V at a time. The penis was prolapsed beyond the prepuce, and semen was collected into a graduated collection vial attached to an artificial vagina at room temperature. Collected semen was immediately transported to the laboratory and immersed in a water bath at 30 °C. Volume of the ejaculates was read directly from the graduated collection tube (0.1 ml accuracy). The sperm concentration was determined by optical density with a spectrophotometer (Photometer SDM4, Minitüb, Germany) calibrated for ram species (1:1000 dilution rate). A small subsample of semen was diluted with physiological saline on a slide, covered with a coverslip and placed on a microscope stage at 37 °C. The percentage of progressively motile sperm was estimated qualitatively by examining approximately 8 fields at a magnification of 400× (9). To avoid variance, all semen samples were analyzed by a single researcher in this study.

All ram lambs were blood-sampled by jugular venipuncture. Collection of blood samples was completed in 1 h in the morning. After collection, blood samples were

centrifuged at 1200× g for 15 min, and serum samples were stored at -20 °C until assayed. Serum testosterone concentrations were measured in a double antibody radioimmunoassay (RIA) using a testosterone RIA Kit (DSL-4000 ACTIVE® Testosterone Coated-Tube Radioimmunoassay Kit, Diagnostic Systems Laboratories, Inc., USA).

Statistical Analysis: Multiple regression model was used to determine the relationship between the traits. Best subsets regression analysis generated many significant equations at particular juvenile ages perhaps indicating that at these ages reproductive function of lambs may relate strongly to their postpubertal reproductive function. Significant prediction equations involving specific combination of juvenile variables were always applied to only 1 postpubertal age, although some variables were present in a number of different models that were applied collectively to all postpubertal ages.

Prediction equations between combinations of juvenile traits and individual postpubertal reproductive features were generated by the best subset regression (10). Measurements of body weight (BW), testis diameter (TD), testis length (TL), scrotum circumference (SC), testis volume (TV) and testosterone (Tst) at various juvenile ages (between 60 and 120 days) were set as independent variables. Measurements of testicular characteristics (testis diameter and scrotum circumference from 220 to 420 days) and semen characteristics (semen volume, spermatozoa concentration, and motile spermatozoa, from 7 to 14 months of age) at postpubertal ages were set as dependent variables. In the best subset regression method, VIF values were taken into consideration as described (Personal communication).

Significant 1, 2, 3, and 4 variable models were determined for each lamb age, and the models producing largest R^2 , smallest S (standard deviation) and with a CP value less than $p + 1$ were chosen. The maximum number of independent variables (p) included in an equation was 4. The estimate of Mallows' Cp (11) was also considered in the evaluation of the equations since Mallows' Cp is a more reliable statistic than R^2 when the number of observations (n) is small. An equation was considered to be reliable when the R^2 was high and Cp was low, and when Cp is $\leq p+1$ (Personal communication).

1. CP - We want the value of CP to be less than or equal to $p + 1$. (Note: CP is a measure of the fitness of the model.)

2. S - We want our estimate for the standard deviation to be as small as possible. Note: $S = \sqrt{MSE}$, so when S is large, the denominator of our F-ratio, $F = MSR/MSE$, is also large, which makes the F-ratio smaller and possibly statistically insignificant.

3. Radj - We want the value of R_{adj} to be as close to 1 (100%) as possible without creating other problems such as multicollinearity.

4. Parsimony Principle - We balance the above 3 statistics with the parsimony principle stating that a model with fewer variables is preferred to one with many variables. In essence, a slightly lower value of S or a slightly higher value of R^2_{adj} does not justify a model with additional variables. Once we selected our model, we needed to check the departure model assumptions. Interpretability is another criterion in choosing a model. If transformations are used, the model may be hard to interpret.

Results

In this study, the determinative degree of reproduction characteristics of Kıvrıkcık ram lambs at juvenile age on the testicular diameter at various postpubertal periods is given in Table 1. Strong and consistent indicators of testicular diameter (TD) were body weight (BW) and testosterone (Tst). BW and Tst were also components of 3 multi-variable models, with the model at day 100 (BW, scrotal circumference (SC), and Tst) having the strongest relationship ($R^2 = 0.85$, $P < 0.001$) with TD at 220 days of age. Body weight alone provided medium degree indication of TD postpubertal, but when combined with SC and Tst (100 days), it provided an excellent indication ($R^2 = 0.63 - 0.62$ respectively, $P < 0.01$) of TD at 260 and 280 days. Body weight together with testosterone levels on day 80, in prepuberty, allowed a significant determination degree on testicular diameter on days 220, 260, 280 and 420 in postpubertal rams ($R^2 = 0.72, 0.58, 0.63, \text{ and } 0.42$; $P < 0.001, P < 0.01, P < 0.01, \text{ and } P < 0.05$, respectively). It has been determined that body weight, scrotal circumference, and testicular length, when each of the 3 combined or when the 3 were combined with testosterone in a 4 variable system, significantly determined the testicular diameter of rams at 220, 240, 260, 380, and 420 days of age.

The relationship between the important reproduction characteristics at juvenile age and the postpubertal scrotal circumference in Kıvrıkcık rams is shown in the Table 2.

Table 1. Significant prediction equations between postpubescent testicular diameter (dependent variable) and juvenile reproductive traits (independent variables).^{a,b}

| Juvenile Age (days) | Juvenile Trait Regression Coefficients | | | | | | R ² | Cp | p |
|---------------------------------------|----------------------------------------|-------|--------|--------|--------|--------|----------------|-----|---|
| | BW | TD | SC | TV | TL | Tst | | | |
| Testicular diameter at Day 220 | | | | | | | | | |
| 60 | | 0.826 | | | 0.841 | -0.187 | 0.71** | 4.0 | 3 |
| 80 | 0.142 | | | | | 0.339 | 0.72*** | 2.5 | 2 |
| 100 | 0.102 | | 0.214 | | | -0.343 | 0.85*** | 4.0 | 3 |
| 120 | 0.0749 | | 0.253 | | -0.279 | | 0.79*** | 3.1 | 3 |
| 140 | 0.0985 | | | | | 0.255 | 0.60** | 2.2 | 2 |
| Testicular diameter at Day 240 | | | | | | | | | |
| 60 | | 1.47 | | | | -0.246 | 0.57** | 2.1 | 2 |
| 80 | 0.11 | | | | | | 0.47** | 0.2 | 1 |
| 100 | 0.126 | | | | | | 0.48** | 2.0 | 1 |
| 120 | 0.0846 | | 0.232 | | -0.48 | -0.118 | 0.78*** | 5.0 | 4 |
| Testicular diameter at Day 260 | | | | | | | | | |
| 60 | | | | | 0.905 | | 0.41** | 1.6 | 1 |
| 80 | 0.0868 | | | | | 0.556 | 0.58** | 2.5 | 2 |
| 100 | 0.117 | | 0.113 | | | -0.359 | 0.63** | 4.0 | 3 |
| 120 | 0.118 | | 0.183 | | -0.58 | | 0.69** | 3.0 | 3 |
| Testicular diameter at Day 280 | | | | | | | | | |
| 60 | | | | | 0.717 | -0.196 | 0.38** | 2.5 | 2 |
| 80 | 0.0509 | | | | | 0.685 | 0.63** | 2.6 | 2 |
| 100 | 0.104 | | 0.0847 | | | -0.461 | 0.61** | 4.0 | 3 |
| 120 | 0.095 | | | | | | 0.39** | 0.9 | 1 |
| Testicular diameter at Day 300 | | | | | | | | | |
| 80 | | | | | | 0.809 | 0.69*** | 0.8 | 1 |
| 100 | 0.0993 | | | | | -0.354 | 0.38* | 2.0 | 2 |
| Testicular diameter at Day 340 | | | | | | | | | |
| 80 | | | | 0.0444 | | | 0.52** | 2.9 | 2 |
| 100 | | | 0.222 | | | | 0.38** | 0.0 | 1 |
| Testicular diameter at Day 360 | | | | | | | | | |
| 60 | | | | | 0.665 | | 0.30* | 0.8 | 1 |
| 80 | -0.085 | | | 0.0715 | | 0.813 | 0.77*** | 4.0 | 3 |
| 100 | | | 0.209 | | | -0.216 | 0.47** | 2.4 | 2 |
| 120 | | | 0.104 | | | 0.208 | 0.44* | 1.9 | 2 |
| 140 | -0.041 | | | 0.0116 | | 0.318 | 0.62** | 4.0 | 3 |
| Testicular diameter at Day 380 | | | | | | | | | |
| 60 | | | 0.229 | | | | 0.27* | 0.3 | 1 |
| 80 | | | | 0.0343 | | 0.738 | 0.52** | 2.2 | 2 |
| 100 | | | 0.205 | | | | 0.42** | 0.8 | 1 |
| 120 | 0.0913 | | 0.163 | | -0.661 | 0.167 | 0.42** | 5.0 | 4 |
| Testicular diameter at Day 420 | | | | | | | | | |
| 60 | | | 0.313 | | | | 0.37* | 0.1 | 1 |
| 80 | 0.11 | | | | | 0.424 | 0.42* | 2.0 | 2 |
| 100 | 0.151 | | | | | | 0.38** | 0.8 | 1 |
| 120 | 0.152 | | 0.158 | | -0.65 | | 0.53* | 3.1 | 3 |
| 140 | 0.0973 | | | | | | 0.25* | 1.1 | 1 |

^a BW: body weight, TD: testicular diameter, SC: scrotum circumference, TV: testicular volume, TL: testicular length, Tst: testosterone, R²: coefficient of determination, Cp: Mallows' Cp, p: number of predictors in equation. ^b Regression coefficients with *: P < 0.05, **: P < 0.01, ***: P < 0.001.

Table 2. Significant prediction equations between postpubescent scrotum circumference (dependent variable) and juvenile reproductive traits (independent variables).^{a,b}

| Juvenile Age (days) | Juvenile Trait Regression Coefficients | | | | | | R ² | Cp | p |
|----------------------------------|----------------------------------------|----|-------|-------|--------|--------|----------------|-----|---|
| | BW | TD | SC | TV | TL | Tst | | | |
| Scrotum circumference at Day 220 | | | | | | | | | |
| 60 | | | 1.059 | | 2.728 | | 0.50** | 2.0 | 2 |
| 80 | 0.634 | | | | | 2.303 | 0.62** | 2.3 | 2 |
| 100 | 0.572 | | 0.873 | | | -1.565 | 0.68** | 4.0 | 3 |
| 120 | 0.532 | | 1.354 | | -3.049 | | 0.74*** | 3.2 | 3 |
| 140 | 0.467 | | | | | 1.356 | 0.52** | 2.0 | 2 |
| Scrotum circumference at Day 240 | | | | | | | | | |
| 60 | | | 1.659 | | | | 0.52** | 0.4 | 1 |
| 80 | 0.522 | | | | | 2.425 | 0.54** | 2.0 | 2 |
| 100 | 0.555 | | 0.757 | | | -1.973 | 0.62** | 4.0 | 3 |
| 120 | 0.532 | | 1.216 | | -3.198 | | 0.68** | 3.5 | 3 |
| Scrotum circumference at Day 260 | | | | | | | | | |
| 80 | | | | 0.243 | | 2.689 | 0.50** | 2.3 | 2 |
| 100 | 0.457 | | 0.732 | | | -1.424 | 0.57** | 4.0 | 3 |
| 120 | 0.516 | | 1.209 | | -3.393 | | 0.77*** | 3.0 | 3 |
| Scrotum circumference at Day 280 | | | | | | | | | |
| 80 | 0.376 | | | | | 4.907 | 0.78*** | 2.0 | 2 |
| 100 | 0.431 | | 0.936 | | | -2.063 | 0.53* | 4.0 | 3 |
| Scrotum circumference at Day 300 | | | | | | | | | |
| 80 | 0.238 | | | | | 4.628 | 0.72*** | 2.4 | 2 |
| 140 | | | 0.829 | | -2.713 | 2.814 | 0.59** | 4.0 | 3 |
| Scrotum circumference at Day 320 | | | | | | | | | |
| 80 | 0.334 | | | | | 3.238 | 0.64** | 2.0 | 2 |
| Scrotum circumference at Day 340 | | | | | | | | | |
| 80 | | | | 0.209 | | 3.28 | 0.81*** | 2.1 | 2 |
| 100 | 0.288 | | 0.631 | | | | 0.58** | 2.0 | 2 |
| 120 | 0.472 | | | | -1.12 | 1.541 | 0.67** | 3.1 | 3 |
| 140 | | | | 0.038 | | 1.366 | 0.57** | 2.1 | 2 |
| Scrotum circumference at Day 360 | | | | | | | | | |
| 80 | | | | 0.25 | | 2.189 | 0.60** | 2.4 | 2 |
| 100 | | | 1.157 | | | -0.429 | 0.60** | 2.1 | 2 |
| 120 | 0.353 | | 0.788 | | -2.421 | 1.096 | 0.64** | 5.0 | 4 |
| Scrotum circumference at Day 380 | | | | | | | | | |
| 80 | | | | 0.174 | | 3.245 | 0.59** | 2.2 | 2 |
| 100 | | | 1.125 | | | -0.905 | 0.52** | 2.0 | 2 |
| 120 | 0.339 | | 0.937 | | -3.158 | 0.835 | 0.56* | 5.0 | 4 |
| Scrotum circumference at Day 400 | | | | | | | | | |
| 120 | 0.532 | | 0.841 | | -2.92 | | 0.51* | 3.1 | 3 |
| Scrotum circumference at Day 420 | | | | | | | | | |
| 80 | 0.522 | | | | | 3.132 | 0.56** | 2.5 | 2 |
| 100 | 0.951 | | | | | -1.378 | 0.51** | 2.4 | 2 |
| 120 | 0.952 | | 0.645 | | -3.769 | | 0.72** | 3.0 | 3 |

^a BW: body weight, TD: testicular diameter, SC: scrotum circumference, TV: testicular volume, TL: testicular length, Tst: testosterone, R²: coefficient of determination, Cp: Mallows' Cp, p: number of predictors in equation.

^b Regression coefficients with *: P < 0.05, **: P < 0.01, ***: P < 0.001.

Scrotum circumference measurement and in combination with other traits in prepuberty was prevalent in many of the equations providing strong indications of postpubertal scrotum circumference. Both body weight and serum testosterone concentrations on day 80 of prepuberty, were found to be of high importance in determining the scrotal circumference on days 220, 240, 280, 300, 320, and 420 ($R^2 = 0.62, 0.54, 0.78, 0.72, 0.64$ and 0.56 ; $P < 0.01, P < 0.01, P < 0.001, P < 0.001, P < 0.01,$ and $P < 0.01,$ respectively). In addition, when the testicular volume and testosterone level on day 80 of prepuberty were assessed together, the scrotal diameter determination degree at days 260, 340, 360 and 380 was slightly higher ($R^2 = 0.81 - 0.60$ range, $P < 0.001 - P < 0.01$). A significant relationship was found between body weight, scrotal circumference, and testosterone concentration on day 100, and the scrotal circumference on days 220, 240, 260, 280, 340 and 420 of postpubertal rams ($R^2 = 0.68, 0.62, 0.57, 0.53, 0.58$ and 0.51 ; $P < 0.01, P < 0.01, P < 0.01, P < 0.05, P < 0.01$ and $P < 0.01,$ respectively). Body weight, scrotal circumference and testicular length on day 120 (prepuberty), when combined, strongly indicate the scrotal diameter on day 220 and 260 (postpuberty) ($R^2 = 0.74$ and 0.77 ; $P < 0.001,$ respectively).

Determination degrees of body weight, testicular size, and testosterone levels on postpubertal semen characteristics are given in the Table 3. Body weight and testosterone at different juvenile ages, either alone or in combination with other variables, were related to semen volume at all 6 postpubertal ages. Scrotum circumference alone on day 60 was related ($R^2 = 0.59, P < 0.01$) to semen volume at 7 months of age. A significant correlation was found between the selection of ram lambs on day 100, according to body weight and testosterone concentration, and postpubertal semen volume at 7, 8, and 14 months of age ($R^2 = 0.49, 0.48$ and 0.51 ; $P < 0.01, P < 0.05$ and $P < 0.01,$ respectively). The combination of body weight and testicular length on 100 days of age provided a strong indication ($R^2 = 0.49$ and $0.51,$ respectively; $P < 0.01$) of the semen volume at 7 and 14 months of age. Body weight and testosterone, either alone or in combination with other variables, were related to sperm concentration at all 5 postpubertal ages (Table 3). Body weight together with testicular length and serum testosterone concentration (days 100 and 120), gave a reasonably good indication ($R^2 = 0.51, P < 0.05$ and $R^2 = 0.64, P < 0.01,$ respectively) of sperm concentration at 7 months of age. At 60 days of age, the scrotum circumference, in combination with

testicular length and testosterone, was related ($R^2 = 0.47, P < 0.05$ and $R^2 = 0.50, P < 0.05,$ respectively) to sperm concentration at 7 to 9 months of age. In all periods of prepuberty, the combination of different characteristics allowed significant determination of motile spermatozoa rate at month 7 ($P < 0.01$) (Table 3). Body weight, together with testicular volume on days 80 and 140, was related to motile spermatozoa $R^2 = 0.50$ and 0.63 ($P < 0.01$ and $P < 0.01,$ respectively) at 7 months of age. Juvenile reproductive traits had no significant effect at 9, 10, and 14 months of postpubertal age.

Discussion

According to the data obtained in this study, the prepubertal (days 80, 100, and 120) body weight, scrotal circumference, testicular length, and testosterone levels allowed significant determination of testicular diameter at various postpubertal periods (days 220, 240, 260, and 280) in Kivircik rams when combined in 2, 3 or 4 variables. These results are in agreement with previous findings (12-15). This study suggests that body weight and testosterone may provide a good indication of postpubertal semen features. Some other combination of variables is related to specific functions more strongly at specific postpubertal ages. For instance, the relationship between body weight and testosterone at 80 days of age and at a postpubertal age of 280 and 300 days was also strong ($R^2 = 0.78$ and $0.72,$ respectively). Yarney and Sanford (7), working with Suffolk ram lambs raised in natural lighting, reported a positive correlation between testosterone level at 5 months with testes size of yearling rams. Phenotypic correlations between body weight and testicular traits were also found to be high in Turkgedi male lambs (16). Similarly, Salhab et al. (6) found that the correlation between scrotum circumference and body weight was 0.91 for Awassi ram lambs. Selection of Kivircik rams according to prepubertal body weight, scrotal circumference, and testosterone level and testes volume will improve the effectiveness of determination of postpubertal scrotal circumference.

It has been shown that testicular diameter and along with scrotal circumference are excellent indicators of spermatogenic function (17,18). Body weight and testosterone at different juvenile ages in our study, either alone or in combination with other variables, were related to semen volume at all 5 postpubescent ages (7, 8, 9, 11, and 14 months).

Table 3. Significant prediction equations between postpubescent semen characteristics (dependent variable) and juvenile reproductive traits (independent variables).^{a,b}

| Juvenile Age (days) | Juvenile Trait Regression Coefficients | | | | | | R ² | Cp | p |
|----------------------------------------|----------------------------------------|--------|--------|--------|--------|--------|----------------|------|---|
| | BW | TD | SC | TV | TL | Tst | | | |
| Semen volume at month 7 | | | | | | | | | |
| 60 | | | 0.186 | | | | 0.59** | 0.6 | 1 |
| 80 | 0.0706 | | | | | | 0.47** | 0.9 | 1 |
| 100 | 0.0464 | | 0.0671 | | | | 0.49** | 2.6 | 2 |
| 120 | 0.0398 | | 0.0578 | | | | 0.41* | 1.5 | 2 |
| Semen volume at month 8 | | | | | | | | | |
| 80 | | | | 0.023 | | | 0.47** | 2.0 | 2 |
| 100 | 0.04 | | 0.077 | | | -0.176 | 0.48* | 4.0 | 3 |
| 120 | | | 0.11 | | | | 0.52** | -0.6 | 1 |
| 140 | 0.034 | | | | | 0.144 | 0.48** | 2.3 | 2 |
| Semen volume at month 9 | | | | | | | | | |
| 60 | | -0.316 | | | 0.165 | -0.071 | 0.67** | 4.0 | 3 |
| Semen volume at month 11 | | | | | | | | | |
| 120 | 0.023 | | | | | -0.091 | 0.51** | 2.0 | 2 |
| Semen volume at month 14 | | | | | | | | | |
| 100 | 0.062 | | -0.092 | | | | 0.51** | 2.2 | 2 |
| 120 | 0.04 | | | | -0.149 | -0.59 | 0.51* | 3.6 | 3 |
| 140 | 0.043 | | | -0.005 | | | 0.42* | 2.6 | 2 |
| Sperm concentration at month 7 | | | | | | | | | |
| 60 | | | -0.362 | | 1.886 | 0.523 | 0.47* | 4.0 | 3 |
| 80 | -0.18 | | | 0.164 | | | 0.36* | 2.1 | 2 |
| 100 | -0.285 | | | | 1.765 | 0.864 | 0.51* | 3.2 | 3 |
| 120 | -0.098 | | | | 0.541 | 0.689 | 0.64** | 3.0 | 3 |
| Sperm concentration at month 8 | | | | | | | | | |
| 100 | -0.238 | | | | 1.559 | | 0.51** | 1.4 | 2 |
| Sperm concentration at month 9 | | | | | | | | | |
| 60 | | | -0.387 | | 0.83 | -0.377 | 0.50* | 4.0 | 3 |
| 80 | -0.089 | | | | | 0.587 | 0.31* | 2.3 | 2 |
| 100 | -0.105 | | 0.144 | | | -0.51 | 0.49* | 4.0 | 3 |
| 140 | -0.15 | | | | 0.288 | 0.259 | 0.49* | 4.0 | 3 |
| Sperm concentration at month 10 | | | | | | | | | |
| 80 | | | | -0.027 | | 0.559 | 0.35* | 2.4 | 2 |
| 140 | -0.066 | | | | | 0.409 | 0.44* | 2.0 | 2 |
| Sperm concentration at month 13 | | | | | | | | | |
| 60 | -0.248 | | | | 1.485 | -0.163 | 0.57** | 4.0 | 3 |
| 80 | -0.206 | | | | 1.164 | | 0.44* | 2.4 | 2 |
| 140 | -0.121 | | | | 0.377 | | 0.32* | 2.0 | 2 |
| Motile spermatozoa at month 7 | | | | | | | | | |
| 60 | | | | | 17.949 | 3.472 | 0.47** | 2.1 | 2 |
| 80 | 0.583 | | | 1.032 | | | 0.50** | 2.2 | 2 |
| 100 | 1.357 | | 2.416 | | | | 0.49** | 2.1 | 2 |
| 120 | 0.817 | | 1.475 | | 6.114 | | 0.55** | 3.6 | 3 |
| 140 | 0.888 | | | 0.197 | | | 0.63** | 2.2 | 2 |
| Motile spermatozoa at month 12 | | | | | | | | | |
| 60 | | -42.05 | 5.818 | | 22.202 | | 0.58** | 3.1 | 3 |
| Motile spermatozoa at month 13 | | | | | | | | | |
| 60 | | 3.641 | | | | -14.75 | 0.49** | 1.3 | 2 |

^a BW: body weight, TD: testicular diameter, SC: scrotum circumference, TV: testicular volume, TL: testicular length, Tst: testosterone, SV: semen volume, MS: motile spermatozoa, R²: coefficient of determination, Cp: Mallows' Cp, p: number of predictors in equation.

^b Regression coefficients with *: P < 0.05, **: P < 0.01, ***: P < 0.001.

Body weight in combination with other variables at prepubertal ages has a moderate effect on motile spermatozoa rate at onset of puberty (7 months). This is in agreement with the findings of Yarney and Sanford (12) and Langford et al. (19) who reported that the combination of body weight and testosterone at day 150 provided a strong indication ($R^2 = 0.79$) of the daily sperm output at 13 to 14 months of age. A significant relationship was found between testosterone levels, scrotal circumference, body weight, the prepubertal morphological and physiological characteristics, and the postpubertal reproductive performance. Fernandez-Abella et al. (20) reported that in autumn, the testosterone levels in Corriedale and Merino rams were positively correlated with sperm production ($r = 0.74$; $P = 0.01$), and that the best multiple correlation that allowed prediction of about 40% of the variation in sperm production in autumn was

obtained when scrotal circumference and testosterone levels in the late spring were taken into account ($r = 0.66$; $P < 0.01$). The results of this study led us to postulate that prepubertal body weight and testosterone concentration provide good indication for postpubertal reproductive function of Kivircik ram lambs. However, the combination with other traits in the best subsets regression analysis improved the prediction value at a given postpubertal age. These best subsets regression analyses can be used for ram evaluations to improve the economic gain at central ram test stations and effectiveness of selection.

Acknowledgements

The authors would like to thank Prof. John Climent for his great support to this study.

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