

## Functional Anatomy of the Testicular Vascular Pedicle in the Rhesus Monkey: Evidence for a Local Testosterone Concentrating Mechanism<sup>1</sup> (38513)

D. J. DIERSCHKE, S. W. WALSH, R. J. MAPLETOFT, J. A. ROBINSON,  
AND O. J. GINTHER

*Wisconsin Regional Primate Research Center, Department of Veterinary Science and Endocrinology-Reproductive Physiology Training Program, University of Wisconsin, Madison, Wisconsin 53706*

The intimate anatomical relationship which exists between veins and arteries within the testicular vascular pedicle has been described for many species but has received only limited attention in the primate (1). It has been generally assumed that the regulation of testicular temperature through a countercurrent heat exchange system is the major functional role of this vascular arrangement (2). Recent reports in the rat (3-5) and ram (6), however, have suggested another role, i.e. to maintain a high concentration of testosterone in the testis and epididymis through a local transfer of hormone from veins to arteries. The present study was conducted to describe the anatomical association between these vessels in the testicular vascular pedicle of the rhesus monkey (*Macaca mulatta*) and to evaluate the possibility of a vascular transfer mechanism in this species.

**Materials and Methods.** The vascular anatomy was studied in six adult (5.8-14.4 yr, 6.5 to 9.5 kg) and two immature (3.3 and 4.0 yr, 4.1 and 5.4 kg) male rhesus monkeys. The methods for preparing and photographing the specimens are described in detail elsewhere (7, 8). In brief, the animals were killed by exsanguination and the appropriate major arteries or veins were cannulated, flushed with saline and injected with colored latex. The reproductive tracts were subsequently dissected from the carcass, fixed, dehydrated and cleared in a step-wise manner. The arterial system alone, venous system alone and

the combined arterial and venous systems were studied in three, one and four monkeys respectively. Photography was performed utilizing various combinations of transillumination and surface lighting. All pictures presented herein are unretouched photographs of actual specimens.

Hormonal transfer in the testicular pedicle was evaluated in 11 adult (8.3-14.5 yr, 6.6-9.7 kg) rhesus monkeys. Anesthesia was induced with sodium thiamylal (Surital, Parke Davis & Co.) and maintained with halothane (Fluothane, Ayerst Laboratories, Inc.). An incision was made through the scrotum, tunica vaginalis and tunica albuginea at the distal pole of the testis (right testis in six animals and left testis in five) and approximately one cm of the testicular artery on the lateral surface was dissected free (Fig. 1). Heparin (1000 units) was administered iv and a polyethylene cannula (i.d. 0.034 in., o.d. 0.050 in.) was inserted retrograde to the direction of flow and tied in place. The contralateral femoral artery had been previously dissected at a level approximately midway between the pelvis and knee and cannulated as above. Testicular and femoral arterial blood was collected simultaneously into heparinized tubes which were immediately placed in an ice bath. The plasma was separated within 1 hr of collection and stored at -20°.

Plasma concentrations of testosterone were quantitated in 50 or 100 µl aliquots utilizing a radioimmunoassay which was described previously (9) but independently validated in our laboratory. Sephadex LH-20 columns (Pharmacia Fine Chemicals) were substituted for thin-layer chromatography to separate testosterone from dihydrotestosterone and other cross-reacting compounds. For this purpose, 1.0 cm (diam) columns were packed with 1.0 g of Sephadex in benzene:

<sup>1</sup>Supported by Grant No. RR-00167 from the National Institutes of Health to the Wisconsin Regional Primate Research Center, Grant No. MH-21312 from the National Institute of Mental Health, Public Health Service Training Grant No. 5-T01-HD-00104-07, and Grant No. 630-0505A from the Ford Foundation. Publication No. 14-007 of the Wisconsin Regional Primate Research Center.

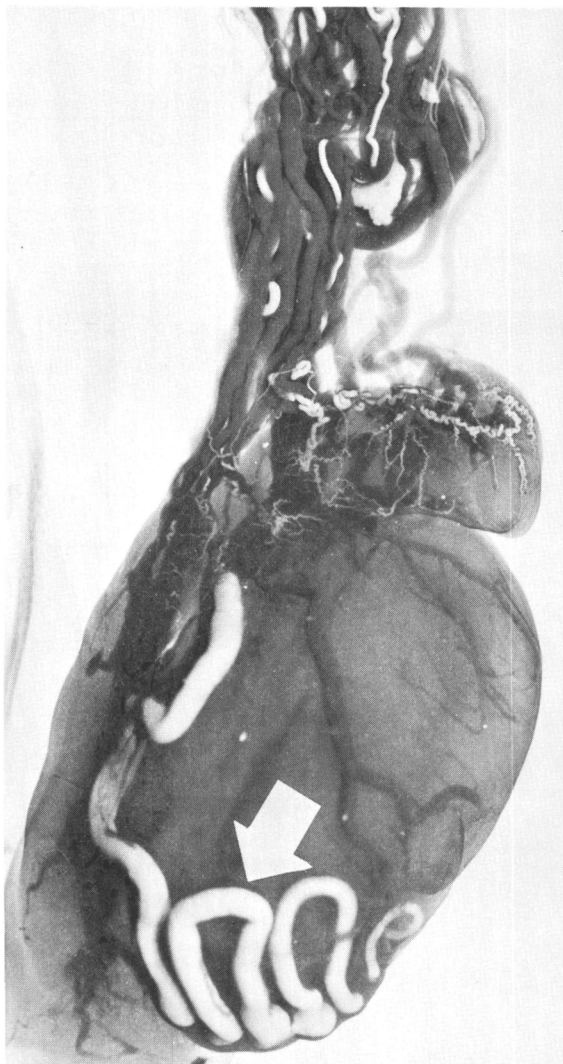


FIG. 1. Arteries (light) and veins (dark) on posterior side of right testis from an adult animal. The major structures from top to bottom are: pampiniform plexus, head of the epididymis, testis. The arrow indicates the approximate site for the collection of testicular arterial blood.

methanol (17:3) and rinsed with the elution solvent, trimethyl pentane:benzene:methanol (18:1:1). Testosterone, containing less than 5% dihydrotestosterone, was eluted in the 15.5–22.5 ml fraction. The average internal recovery of testosterone, estimated by the addition of 1000 cpm of  $^3\text{H}$ -testosterone (New England Nuclear, sp act 100 Ci/mole), was  $89\% \pm 4$  (SD). The range of the standard curve was 10–500 pg and blank values of  $2.4 \text{ pg} \pm 2.6$  (SD) were attained. The sensitivity of the assay was determined to be 7.5 pg of testosterone (10). Duplicate

determinations of 37 individual samples containing 30–325 pg of testosterone resulted in a within-assay variation of  $\pm 8.3$  pg (SD) and the between-assay variability, determined by assaying aliquots from a pool of male rhesus serum in each assay, was calculated to be  $\pm 13.8$  pg (SD) (11).

Total plasma protein concentrations were determined by a modified biuret method (12).

*Results. Vascular anatomy.* The internal spermatic arteries originated from the abdominal aorta between the origins of the



FIG. 2. Anterior view of the right testicular vascular pedicle from an adult male. Arteries (light) and veins (dark) were injected with colored latex. Note the close relationship between the internal spermatic artery and spermatic vein throughout the length of the spermatic cord and the increased complexity of this relationship near the testis (bottom of Figure).

renal and inferior mesenteric arteries. The abdominal portion of this artery was located between channels of the spermatic vein and was relatively straight while that portion located between the inguinal canal and the testis was highly convoluted (Figs. 2 and 3). The testicular extension of the spermatic artery passed across the posterior medial surface of the testis, alongside the body of the epididymis (Figs. 1 and 3). It then followed a tortuous course forming a series of S-shaped curves as it passed around the distal pole to the lateral ventral surface of the testis where it branched and passed into the deeper portions of the testis. Arteries to the epididymis originated from the convoluted portion of the spermatic artery (Figs. 1 and 3).

The testicular veins rose to the surface of the testis where they entered the pampiniform plexus (Figs. 1, 2, and 4). Veins from the epididymis also drained into the pampiniform plexus (Fig. 1). The pampiniform plexus consisted of numerous veins which were intertwined with the convolutions of the spermatic artery (Fig. 5). Many venous anastomoses covered the surface of the spermatic artery resulting in a large area of surface contact between veins and arteries. The number of veins of the plexus gradually decreased to two or three channels which passed through the inguinal canal. The channels were interconnected by many anastomoses (Fig. 2) and merged to form one vessel just prior to entering the vena cava on the right side or the renal vein on the left side.

*Plasma determinations.* The average concentration of testosterone was significantly greater in samples obtained from the testicular artery than in those from the femoral artery (Table I). In only one animal (No. 1153) was the concentration gradient clearly in favor of the femoral arterial sample. Total plasma protein concentrations, on the other hand, were not statistically different in the same paired samples. The mean total protein in all samples was  $5.56 \text{ g/100ml} \pm 0.08$  (SEM) which is similar to that previously reported for the rhesus monkey (13).

*Discussion.* The results confirm that a close

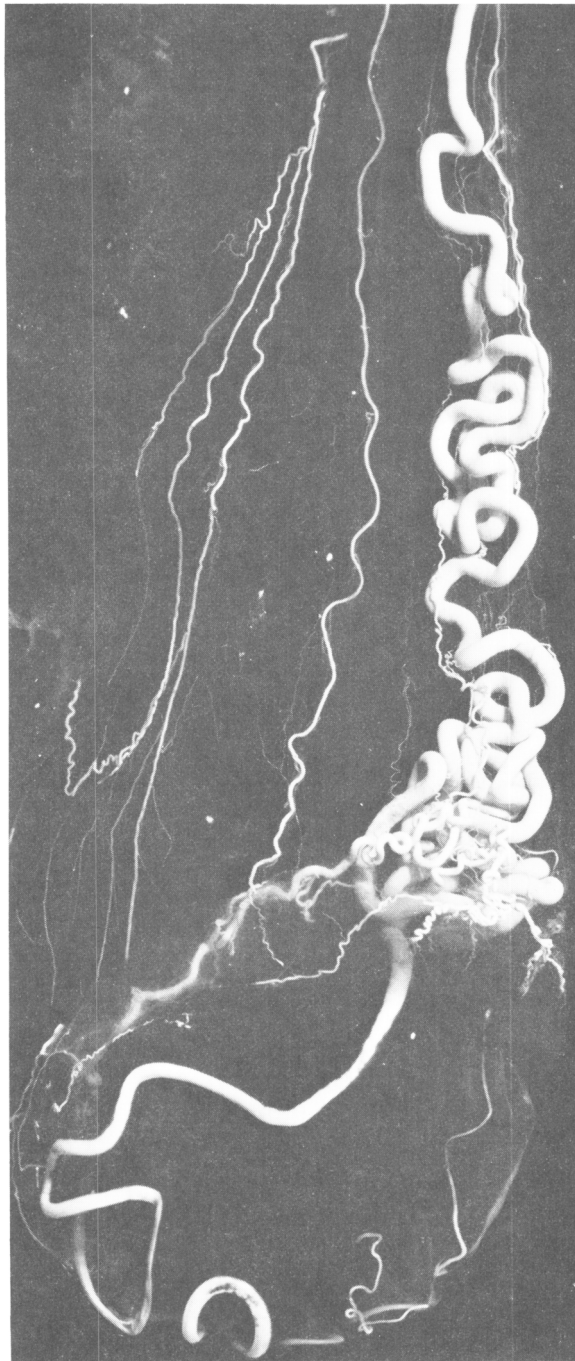


FIG. 3. Arteries on posterior side of right testis from an adult male. The internal spermatic artery is shown above and its extension (testicular artery) over the surface of the testis below. The outline of the epididymis is seen along the upper portion of the testis.



FIG. 4. Veins on posterior side of right testis from an adult male. Note the anastomoses between venous channels in the pampiniform plexus and the origins of these vessels on the testis (bottom of Figure).

anatomical relationship exists between the internal spermatic artery and the pampiniform plexus in the rhesus monkey, thus providing the morphological basis for the hypothesis that hormones may pass between the vessels. The preliminary functional evidence reported herein that arterial blood which has passed through the area of the pampiniform plexus contains a higher concentration of testosterone than arterial blood from a peripheral site supports this hypothesis and corroborates similar findings in rats (3-5) and rams (6). That this observation in

the monkey likely results from a selective transfer mechanism, rather than from local hemoconcentration or other general hemodynamic phenomena is suggested by the additional finding of no difference in total plasma protein concentration between samples from the testicular artery and the femoral artery.

While it is probable that a hormone concentrating mechanism of this type could play a physiologically important role in testicular and/or epididymal function, whether or not the relatively small additional quantities of



FIG. 5. Close-up of the pampiniform plexus (dark) and the internal spermatic artery (light) of the specimen shown in Fig. 1. Note the large area of surface contact between veins and arteries in this region of the testicular vascular pedicle.

testosterone demonstrated in this study are critical to this function is unknown. Nevertheless, these observations in the male monkey provide the basis for similar studies in the female monkey. In females of many non-primate species, considerable evidence has been accrued which supports the concept that a substance produced within the uterus is transferred to the ovary through local venoarterial pathways (14), and similar vascular arrangements involving extensive areas of surface contact between veins carry-

ing uterine and ovarian blood and vessels carrying ovarian arterial blood have recently been described for the rhesus monkey (7).

*Summary.* The detailed anatomy of arteries and veins in the testicular pedicle of the rhesus monkey, with special emphasis on an area of extensive surface contact between these vessels in the spermatic cord, is described. The mean plasma testosterone concentration in blood from the testicular artery was significantly greater than the mean for samples collected simultaneously from the

TABLE I. COMPARISON OF PLASMA CONCENTRATIONS OF TESTOSTERONE IN TESTICULAR AND PERIPHERAL ARTERIES

| Animal number     | Testosterone (ng/ml) |                 |
|-------------------|----------------------|-----------------|
|                   | Testicular artery    | Femoral artery  |
| A79               | 2.57                 | 2.18            |
| B66               | 3.15                 | 2.51            |
| F46               | 5.36                 | 4.49            |
| 530               | 8.22                 | 7.70            |
| 1012              | 12.01                | 10.66           |
| 1016              | 1.40                 | 1.36            |
| 1152              | 11.16                | 9.38            |
| 1153              | 2.77                 | 3.08            |
| 1154              | 5.14                 | 4.73            |
| 1156              | 0.67                 | 0.72            |
| 1158              | 3.68                 | 3.49            |
| Mean ( $\pm$ SEM) | 5.10 $\pm$ 1.15*     | 4.57 $\pm$ 1.00 |

\*  $P < 0.02$  (paired  $t$  test).

contralateral femoral artery in 11 monkeys. These observations are interpreted as supportive of the hypothesis that a hormone concentrating mechanism involving the local transfer of testosterone between the vessels of the pampiniform plexus and the spermatic artery exists in the male rhesus monkey.

The authors gratefully acknowledge the contributions of Dr. P. T. K. Toivola in developing surgical procedures and his suggestions in preparing the manuscript. Photographs were taken by Mr. W. P. Steffen-

hagen. Expert technical assistance was provided by R. Abrams, G. Scheffler, J. Scheffler, and A. Mitchell.

1. Setchell, B. B., in "The Testis" (A. D. Johnson, W. R. Gomes, and N. L. Vandemark, eds.), Vol. 1, p. 101. Academic Press, New York (1970).
2. Waites, G. M. H., in "The Testis" (A. D. Johnson, W. R. Gomes, and N. L. Vandemark, eds.), Vol. 1, p. 241. Academic Press, New York (1970).
3. Free, M. J., Jaffe, R. A., Jain, S. K., and Gomes, W. R., *Nature New Biol.* **244**, 24 (1973).
4. Einer-Jensen, N., *J. Reprod. Fert.* **37**, 145 (1974).
5. Free, M. J., and Jaffe, R. A., *Proceedings of the Annual Meeting of the Society for the Study of Reproduction, Ottawa, Canada*, pp. 167, (Abstract No. 172) (1974).
6. Ginther, O. J., Mapletoft, R. J., Zimmerman, N., Meckley, P. E., and Nuti, L., *J. Animal Sci.* **38**, 835 (1974).
7. Ginther, O. J., Dierschke, D. J., Walsh, S. W., and Del Campo, C. H., *Biol. Reprod.* **11**, 205 (1974).
8. Del Campo, C. H., Steffenhagen, W. P., and Ginther, O. J., *Amer. J. Vet. Res.* **35**, 303 (1974).
9. Resko, J. A., Malley, A., Begley, D., and Hess, D. L., *Endocrinology* **93**, 156 (1973).
10. Ekins, R. and Newman, B., in "Steroid Assay by Protein Binding" (E. Diczfalusy and A. Diczfalusy, eds.) p. 11. Second Karolinska Symposium on Research Methods in Reproductive Endocrinology, Bogtrykkeriet Forum, Copenhagen (1970).
11. van der Molen, H. J. in "The Androgens of the Testis" (K. B. Eik-Nes, ed.) p. 195. Marcel Dekker, Inc., New York (1970).
12. Werchselbaum, T. E., *Amer. J. Clin. Pathol.* **16**, 40 (1946).
13. Petery, J. J., *Lab. Animal Care* **17**, 342 (1967).
14. Ginther, O. J., *J. Animal Sci.* **39**, 550 (1974).

Received September 16, 1974. P.S.E.B.M. 1975, Vol. 148.