## Origin of the rete testis in bovine embryos

By Wangari M. Mathai

Department of Veterinary Anatomy and Histology University of Nairobi

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## Introduction

Several different opinions are expressed in the literature with respect to the origin of the rete testis: 1. According to WALDEYER (1908), BRAUN (1877), SEMON (1877) and PETER (1904) (all quoted after WILSON 1926), the rete testis (and also the sex cords) are derived from evaginations from the Malpighian corpuscles, the rete testis being the proximal portions of such evaginations. 2. ALLEN (1904) is of the opinion that the rete testis arises from invaginations of the peritoneal epithelium in a pregonadal region at the anterior end of the gonad. Eventually these invaginations grow into the gonad region and establish a network of tubules whose end branches (the tubuli recti) communicate with the sex cords. NELSEN (1944) derives the rete testis from coelomic epithelium through cellular invaginations and inward proliferation. He also notes that the region where the rete develops is possibly an aberrant region which becomes secondarily incorporated into the hilar region of the developing gonad to provide ducts for the male genital system. 3. In their reports, GRUENWALD (1942) and GILLMAN (1948) indicated that the rete testis developes from the free ends of the more cranial sex cords with which the rete tubules maintain direct continuity. 4. FELIX (1912) and WILSON (1926) are of the opinion that the rete testis differentiates in situ from mesenchymal cells which rapidly develop into cord-like structures. The same view is shared by VAN VLOTEN (1932) and KREHBILE (1963).

## Materials and methods

Twenty-five bovine embryos of 24, 25, 26, 27, 32, 34, 35, 37, 38, 40, 45, 46, 48, 49, 51, 52, 53, 55, 56, 60, 61, 61, 64, 65 and 66 mm crown-rump length (CRL) were used in this study. Gonads and mesonephros (still attached) were removed and fixed in Helly's solution or in  $10 \, \frac{9}{0}$  formalin. Paraffin sections about 8 to 10 microns thick were cut and stained with haematoxylin and eosin (or phloxin) or with the PAS reaction for a general survey. Microphotographs were taken using a Leitz Ortholux microscope with the orthomat automatic camera attached to it.

## Observations

The gonad anlage develops along the ventro-medial aspect of the mesonephros to which it is directly attached. At the cranial and caudal ends the anlage is attached to the body wall through a cranial and a caudal ligament, respectively. The development of the gonad *per se* will not be described here but it should be noted that the germinal epithelium becomes highly proliferated and cellular ingrowths (not invaginations) into the underlying mesenchyme are observed in embryos of 8 to 15 mm CRL. These epithelial cells make the bulk of the gonad stroma from which the different gonadal tissues are differentiated.

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Fig. 1. A cross-section through the urogenital region of a bovine embryo of 37 mm showing the darkly stained cells of the rete blastema (H. and  $E. \times 43$ )

In embryos of about 25 mm CRL a cylindrical cord of darkly-staining cells is noted extending from the gonad onto the Malpighian corpuscle (Fig. 1). The cord is composed of small epithelioid cells whose nuclei stain strongly with haematoxylin. The cells are tightly packed and irregularly arranged. They are all of about the same size and shape and their small, rod-like nuclei are oriented parallel to the long axis of the rete blastema. The cytoplasm of these cell is also more acidophilic than the cytoplasm of the surrounding tissue and it stains lightly with the PAS reaction and toluidine blue (Fig. 2). This serves to demarcate the area of the rete blastema. The long axis of both the intra- and extragonadal components of the rete testis forms an acute angle with the cranio-caudal axis of the gonad.

The cells of the rete blastema are rapidly developed into a network of simple

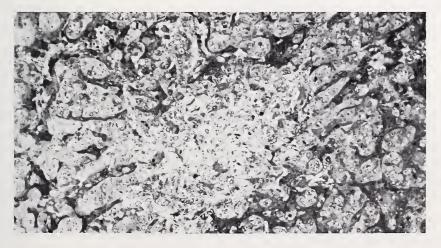


Fig. 2. Part of a cross-section through the rete blastema from a bovine embryo of 34 mm CRL (Epon-araldite, Toluidine blue,  $\times$  264)

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cords of cells. These cords have already developed lumina in embryos of 55 mm CRL. The rate of development in the rete testis is the same both in the intra- and in the extra-gonadal components.

The acquisition of lumina is a gradual process in which the cells rearrange themselves and the cords become lined by a simple low columnar or cuboidal epithelium which rests on a basal lamina. The terminal differentiation of the rete tubules involves their union with the ends of the sex cords (the tubuli recti) on the gonad and the mesonephric tubules at the Malpighian corpuscle.

## Discussion

Local differentiation of the rete testis is suported by FELIX (1912), WILSON (1926), VAN VLOTEN (1932) and KREHBILE (1963), who however, did not study the development of this tissue in detail. In this study it has been confirmed that the rete tissue differentiates in close proximity with the sex cords and some of the cranial Malpighian corpuscles. Eventually a direct contact is established between the sex cords

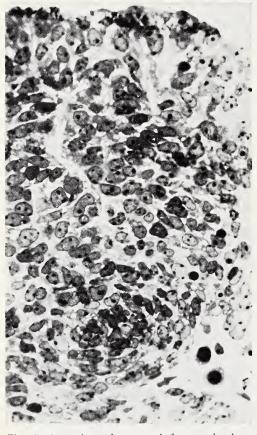


Fig. 3. A section of a gonad from a bovine embryo of 13 mm CRL showing thick radiating cords of epithelioid cells which grow into the underlying mesenchyme (Epon-araldite, Methylene blue,  $\times$  1,300)

and the rete testis at one end and the rete testis and the neighbouring mesonephric tubules at the other end. If the development of this tissue is not followed closely, it is quite possible to assume that the rete tubules are the end portions of the sex cords (AL-LEN 1904); that the capsule of the Malpighian corpuscles has given rise to these neighbouring rete tubules through a simple process of evagination (Waldeyer, Braun, Semon, Pe-TER after WILSON 1926) or that the rete testis represents a sort of vestigial tissue at the cranial end of the gonad which, failing to form sex cords, is secondarily utilized for the formation of duct systems for the male genital ract (Nelsen 1944).

The opinion of COERT (after AL-LEN 1904) and ALLEN (1904) that evaginations from the capsule of the Malpighian corpuscles unite with the rete testis is shared neither by DE BURTET and DE RINTER (1920–21), VAN BEEK (1924), VAN VLOTEN (1932) nor by the present author. The results of this investigation agree with the latter two investigators that the Malpighian corpuscles have nothing to do with the origin of the rete testis.

In bovine embryos of 8 to 15 mm CRL cellular ingrowths are observed originating from a proliferated and stratified germinal epithelium. These

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ingrowths are arranged in columns which are perpendicular to the surface of the developing gonad (Fig. 3). Such ingrowths may have been mistaken for actual invaginations of the thickened germinal epithelium and may have drawn some investigators to the conclusion that both the sex cords and the rete testis originate from these investigations. Similarly, since the rete tubules develop in close proximity with some of the cranial Malpighian corpuscles and because eventually the lumina of both tubular systems communicate, it is possible to assume that the capsule of the Malpighian bodies gives rise to the rete tubules through simple evaginations.

It is important that the early embryonic development of organs should be studied using very close series of specimens. Lack of such a close series result in incomplete information which may be responsible for incorrect conclusions and assumptions. By using a close series of bovine embryos, it has been possible in this study to clearly confirm that the rete testis does not only differentiate in situ but that it has also a dual origin.

#### Summary

The early development of the rete testis was studied in thirty five bovine embryos of between 24 and 66 mm crown-rump length. The rete testis differentiates *in situ* and has a dual origin. One component originates from the gonad blastema and communicates with the tubuli recti. The other component differentiates from local mesenchyme of the mesonephrogenic tissue and communicates with some of the mesonephric tubules (the future Ductuli efferentes).

#### Zusammenfassung

#### Die Entstehung des Rete testis bei Rinderembryonen

Die Frühentwicklung des Rete testis wurde an 35 Rinder-Embryonen von 24 mm bis 66 mm Scheitel-Steiß-Länge untersucht.

Das Rete testis differenziert sich in situ und hat doppelten Ursprung. Eine Komponente entsteht aus dem Gonaden-Blastem und verbindet sich mit den Tubuli recti. Die andere Komponente entsteht aus dem Mesenchym des mesonephrogenen Gewebes und verbindet sich mit einigen der Tubuli des Mesonephros, den zukünftigen Ductuli efferentes.

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- Author's address: Dr. WANGARI M. MATHAI, Department of Veterinary Anatomy and Histology, University of Nairobi, P. O. Box 30197, Nairobi, Kenya

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