Crystals of Reinke in the Nuclei of Human Testicular Interstitial Cells

Crystals of Reinke represent a characteristic feature of the cytoplasm of the Leydig cells in the human testis. Although the lattice structure of these crystals has been described in considerable detail in recent ultrastructural studies[1-3], the manner in which they are formed is still in doubt. In an earlier study[3], it was suggested that the crystals were formed by aggregation and condensation of small tubular inclusions found in the cytoplasm of the interstitial cells. More recent investigations[4,5] revealed that nuclei of some interstitial cells also contained tubular inclusions which are identical to those found in the cytoplasm (Figure 1). This observation suggested that if the crystals were indeed derived from the tubular inclusions they might well be found in the nuclei of human interstitial cells.

During further studies, structures morphologically identical to the cytoplasmic crystals of Reinke were seen in the nuclei of interstitial cells in human testicular material fixed in osmium tetroxide (Figure 2). The nuclear crystals, similar to their cytoplasmic counterpart were not membrane-bounded and were clearly separated from the nuclear membrane by chromatin material (Figure 3). The nuclear crystals appeared singly and were present in only a small number of interstitial cells.

The finding of nuclear crystals of Reinke further supports the hypothesis that the small tubular inclusions are precursors of the crystals both in the nucleus and cytoplasm. The functional significance of the crystals remain obscure. Their appearance at puberty suggests a link with the onset of androgen biosynthesis in the interstitial cells but their absence in the syndrome of testicular femization[6] where androgen biosynthesis is normal[7], is against this view.

Testicular interstitial cell nuclei containing tubular inclusions have been shown to exhibit decreased ATPase activity[4], an observation which suggests that the formation of inclusions may be related to metabolic alterations in the nuclei of these cells. These conclusions were derived from a study of material from 60- to 70-year-old men in which it was postulated that both the alteration in enzymatic activity and tubular inclusion formation were age-dependent phenomena. However, other studies[5] show that the inclusions are also present in the nuclei of testicular interstitial cells in material from men aged between 25–45 years. Nevertheless, the possibility of enzymatic metabolic alterations as a cause of crystal

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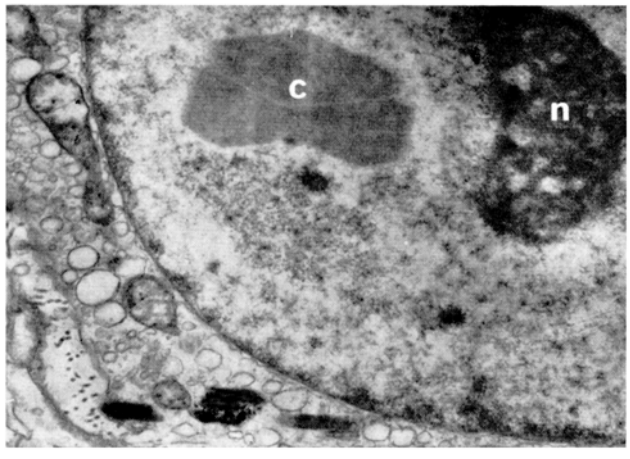

Fig. 2. Crystal of Reinke (c) in the nucleus of an interstitial cell. A nucleolus (n) is indicated. × 7000.

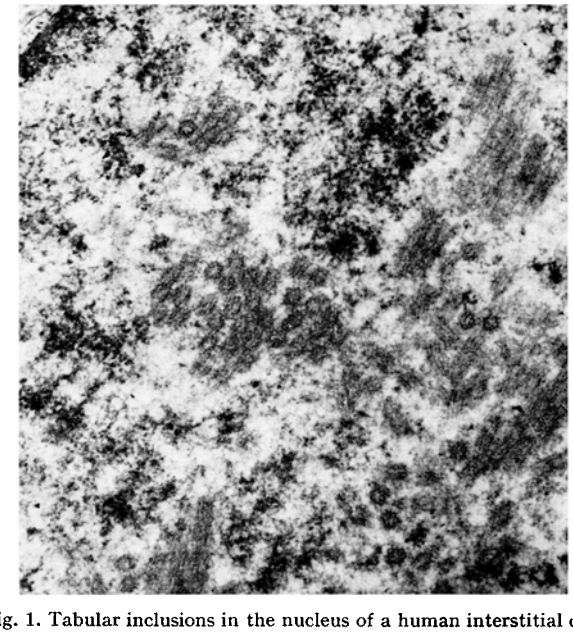

Fig. 1. Tabular inclusions in the nucleus of a human interstitial cell. × 14,000.

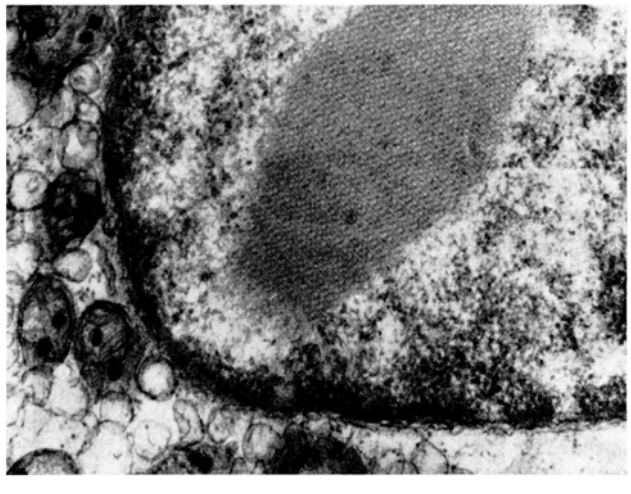

Fig. 3. Nucleus of interstitial cell containing a crystal of Reinke which shows features of its lattice structure. × 28,000.

formation needs further study together with an investigation of the etiological factors involved. It is known that crystal formation occurs as a feature of viral-induced damage in cells infected with Adenovirus type 5[8]. Although unlikely as an etiological factor because of the wide-spread occurrence of Reinke's crystals in human testicular interstitial cells, viruses remain to be excluded as possible agents[9].

Zusammenfassung. Es wird gezeigt, dass in den Leydig'schen Zwischenzellen die Reink'schen Kristalle auch im Zellkern vorkommen und darauf hingewiesen, dass eine Modifikation des Zellkernstoffwechsels zu diesen Bildungen führen könnte.

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Morphology of Muscle Spindles in the Rat

Some morphological features of muscle spindles in the rat have been described (BARKER and HUNT, BRIDGMAN, LANDON, MERILLEES, ZELENA and HNIK[1-5]) but no detailed description exists which is comparable with those available for the cat (BARKER, BOYD[6,7]), rabbit (BARKER[8]), and oppossum (JONES[9,10]). In particular, no information is available on the motor innervation of rat muscle spindles.

In the present investigation we studied the anatomy of muscle spindles in the plantar lumbrical muscles to the second, third, fourth and fifth digits. Rats of Wistar origin were used. Muscles from normal animals were treated as follows: one group was embedded in paraffin and spindles were reconstructed from serial transverse sections stained with hematoxylin and eosin; a second group was stained with gold chloride (COLE[11]), the teased specimens being used for spindle counts and for measuring total spindle length; a third group was stained by a silver technique (WINKELMANN and SCHMIT[12]) and the innervation of teased spindles was examined; a fourth group of muscles with attached nerves was stained with osmium tetroxide and used for the measurement of myelinated nerve fibre diameters in cross sections of the nerve trunk to each muscle. The latter 2 procedures (silver stains and OsO_4 stains) were also carried out on animals in which 1 hind limb had been either de-afferented or de-efferented by selective spinal root section for a period of 35 days.

Myelinated nerve fibre diameters to the lumbrical muscles ranged from 1–10 μ in normal and de-efferented nerves and from 1–7 μ in de-afferented nerves. The total number of motor nerve fibres to any muscle ranged from 5–9, and the sensory nerve fibres from 18–23. From 3–8 muscle spindles/muscle were found in muscles having a wet weight of 4–6.5 mg. No tandem muscle spindles were found.

Reconstructions of spindles from serial cross sections showed that both nuclear chain (NC) and nuclear bag (NB) muscle fibres were present. The mean number of intrafusal muscle fibres in 14 specimens was 3.5, most spindles containing 2 NC and 2 NB fibres. The mean diameter, measured at one pole of the capsule, of the NC fibres was 7.7 ± 1.0 μ ($n = 26$), and the NB fibres 10.0 ± 1.6 μ ($n = 24$), these measurements are uncorrected for shrinkage. The mean length of NB muscle fibres measured from the same paraffin-embedded material was 1.5 mm ($\pm$ 0.5) and that of the NC fibres was 1.2 mm ($\pm$ 0.6). Since the capsule length in all cases was less than 1.0 mm, and most of the muscle fibres were symmetrically placed with regard to the capsule, then the usual situation was that both NB and NC fibres extended beyond each pole of the capsule. The equatorial nucleated region of NB fibres had a mean length of 490 μ, or about $^1/_3$ the length of the NB fibre. This total nucleated length includes both the 'nuclear bag' proper, which contained 2 or 3 nuclei in any cross section, and its flanking myotube regions; the nuclear bag accounted for about half the total figure. The equatorial nucleated region in NC fibres had a mean length of 310 μ, or about $^1/_4$ the length of the NC fibre. The nuclei themselves differed in NB and NC fibres, those in the NC fibres generally being longer and narrower than in the NB fibres. At the equator of the fibres NC nuclei were about 3 μ wide and 8 μ long while the NB nuclei measured 4–6 μ in both the transverse and longitudinal directions. No intrafusal muscle fibre was seen to branch.

The sensory innervation of the spindles was investigated in silver-stained specimens of normal and de-efferented muscles. 50% of the spindles studied contained one primary ending alone. This occupied the central 130–240 μ of the intrafusal muscle bundle and usually took the form of tightly wound spirals on both NB and NC muscle fibres. The remaining 50% of the preparations contained 1 primary sensory ending and 1 or 2 secondary endings. The secondary endings were seen as fine sprays of nerve terminals mainly on the juxta-equatorial regions of NC fibres and occupied from 100–260 μ of the muscle fibre.

The motor innervation of the spindles was studied in normal and de-afferented preparations stained with silver. The most striking feature was that each spindle received

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