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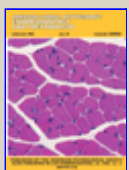
JOURNAL ARTICLE

In vitro differentiation of rat seminiferous tubular segments from defined stages of the epithelial cycle morphologic and immunolocalization analysis

J. Toppari and M. Parvinen

Rat seminiferous tubule segments have been cultured in chemically defined medium (F12/DMEM 1:1) without added hormones or growth factors. The segments (1-2 mm) were isolated from defined stages of the cycle of the seminiferous epithelium (VIII and XII) by transillumination-assisted microdissection. The precise stages were examined by phase contrast microscopy of live cells squashed carefully out from the adjacent segments between glass slides. The squash technique was also used for a primary screening of the cultured tubules. Pachytene primary spermatocytes from stages VIII to XII of the cycle were able to complete meiotic divisions in vitro. From stage XII, they differentiated up to step 5 spermatids, expressed their specific antigens, and developed characteristic movement patterns of the flagellum and of the chromatoid body. Preleptotene and zygotene spermatocytes from the same cell association differentiated synchronously, as judged by chromosome morphology, characteristic chromosome rotation in zygotene and early pachytene, and by development of specific antigen expression. The elongation phase of spermiogenesis did not proceed normally in vitro. The rate of differentiation was the same as observed earlier in vivo. Earlier studies with [3H]thymidine labeling and autoradiography only permitted follow-up of the development of preleptotene spermatocytes. With the present method, all stages of spermatogenesis can be traced in culture with great accuracy in experiments relating to local regulation of spermatogenesis.

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Endocrinology, December 1, 2005; 146(12): 5164 - 5175.

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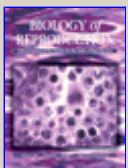
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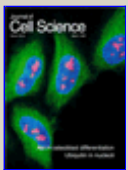
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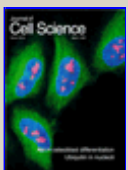
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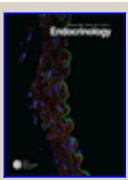
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