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Breakthroughs in Andrology

Expression of Testicular Germ Cell Genes Identified by Differential Display Analysis

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Using differential display reverse transcriptase–polymerase chain reaction (DDRT-PCR) we identified transcripts encoding for the RNA helicase mDEAH9, Ran binding protein 5 (RanBP5), and 3 novel complementary DNAs designated GC3, GC12, and GC14 in developing testicular germ cells. Sources of RNA for the initial DDRT-PCR screen were purified mouse type A spermatogonia, adult mouse wild-type testis, and W/W mutant mouse testis. We identified cDNA fragments for mDEAH9, RanBP5, GC3, GC12, and GC14 in testis and type A spermatogonia samples from wild-type mice, but not in samples from the W/W mouse testis. These same transcripts were absent in Northern blots of testis RNA from mice treated with busulfan 30 days prior, but were present in testis RNA from wildtype mice at 5, 15, 25, and 40 days of age. The mDEAH9 gene was expressed in many tissues, whereas RanBP5 and GC12 genes were expressed predominantly in the testis with much lower expression in other tissues. The expression of GC3 and GC14 were limited to the testis as evidenced by Northern blot and RT-PCR analyses. The mDEAH9 transcript was not detected in cultured interstitial cells but was found at low levels in cultured immature Sertoli cells, whereas the RanBP5, GC3, GC12, and GC14 transcripts were not detected in either cultured testicular interstitial cells or cultured Sertoli cells. RT-PCR analyses of isolated spermatogonia, pachytene spermatocytes, and round spermatids revealed that mDEAH9, RanBP5, GC3, GC12, and GC14 genes were expressed in all 3 cellular populations. In situ hybridization analyses of testis samples from 40-day-old mice localized expression of mDEAH9, RanBP5, GC3, GC12, and GC14 to the seminiferous tubules. RanBP5 expression appeared to be regulated during the cycle of the seminiferous epithelium, with the highest expression in stages III through VII. Expression of GC14 was greatest in the meiotic germ cell populations.

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