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Synopsis: XVIIth Testis Workshop, "Functional Genomics of Male Reproduction"

The XVIIth North American Testis Workshop held March 26–29, 2003, in Phoenix, AZ, was sponsored by the American Society of Andrology. The underlying theme of this successful conference was ''Functional Genomics of Male Reproduction.'' This program was developed to highlight the value of strategies that discover gene function in relevant developmental and physiological processes, all in the context of the testis, the germ cell, and male reproduction in general. Selection of the theme underscored the enthusiasm of the organizing committee for the innovative approaches of functional genomics that are bringing new understanding of the mechanisms underlying testis development and function. The program successfully showed how such knowledge may ultimately translate into clinical diagnostics and new treatment modalities.

From the outset, all participants were challenged by the innovative strategies discussed to stretch and think in new directions. In his keynote address, **Michael Griswold** (Washington State University) showed that the unbiased and global gene discovery facilitated by microarray analysis could provide new and valuable insight into the ''testicular transcriptome.'' The power of surveying changes in some 15 000 genes expressed in the testis was dramatically illustrated. Cultured rat Sertoli cells as well as mouse genetic models were used to detect hormonally responsive genes. As anticipated, FSH induced increase in transcription of a large number of specific genes, whereas, surprisingly, testosterone resulted in down-regulation of far more genes than were up-regulated. Developmental profiles are also taking shape, both whole testis and germ cell specific, and cluster analysis of the patterns of gene expression can ultimately elucidate pathways and regulatory networks controlling testis differentiation. Fortunately for all those who need time to think about the bewildering array of expression patterns, the information will soon be available at a Web site. This theme of gene discovery was continued in the first of 2 benchmark lectures, which was delivered by **Martin Matzuk** (Baylor College of Medicine). Gene discovery was not unbiased in these studies, but came from strategies (such as degenerate PCR, in silico subtractive hybridization, and genome database mining) designed to detect genes specific to the gonads. This tact has reaped several novel genes of importance for gametogenesis, including *Oosp1, Nobox, Obox, Tex4,* and *Zar1*; taken together the functional analysis of these genes has provided new insights into spermatogenesis, oogenesis, and early development. The second benchmark lecture, from **Susan Strome** (Indiana University in Bloomington), highlighted the importance of proteins controlling chromatin conformation and transcription. The MES proteins of the nematode *C elegans* act in a functional complex to control transcription from the X chromosome in the germline, where, for reasons that are not yet clear, the X chromosomes are globally silenced. MES-4 is a particularly interesting protein; it does not associate with the other known MES proteins and is, in fact, excluded from the X chromosome by the cooperative action of MES-2/MES-3/MES-6. Thus MES-4 is a mark of active chromatin. This fascinating talk set the concept of chromatin modification as a key element in the control of the program of gametogenic gene expression. These themes of novel gene identification, functional analyses, and the importance of chromatin unfolded in detail in the invited talks.

Gene function analysis has been particularly rewarding with respect to germ-line specification, development, and differentiation. **Haifan Lin** (Duke University) described genes known from mutational analysis to be required for germline stem cell division and differentiation of the gametogenic line. Interestingly, a hallmark of the *Drosophila* germline mitotic proliferation is asymmetric divisions involving a novel membrane skeletal organelle, the spectrosome that segregates the stem cell from the germ cell destined to differentiate. Although the precise spectrosomal structure may not be conserved, it is likely that its function is. Stem cell specification genes identified in *Drosophila* are conserved in mammals, and their function is being investigated by analysis of expression and a mutational (knockout) approach. **Mary Bedell** (University of Georgia) illustrated the value of an allelic series of mutations within a single gene by her analysis of the mouse Kit ligand gene, *Kitl* (also known as *Steel*), that encodes both membrane-bound and soluble forms of a growth factor. Mutations in this developmentally important gene can affect hematopoietic differentiation as well as spermatogenesis (and other processes). Unpredictably, mutations at different sites in the protein can affect the 2 processes with differing severity, providing fascinating insight into the complexity of protein structure-function relationships. **Guang-Quan Zhao** (University of Texas Southwestern Medical Center) presented a summary of the functional differences of different classes of BMPs (bone morphogenetic proteins) in germ-cell development and male re-

production. Mutations of the genes encoding these proteins can affect gonad and primordial germ cell formation spermatogenesis and epididymal function, suggesting their importance in several pivotal regulatory pathways in male reproduction. The analysis of the knockout of the aromatase gene (ArKO) by **Margaret Jones** (Prince Henry's Institute of Medical Research) served to illustrate how much we are learning about novel gene function in hormonal control of spermatogenesis. Ablation of gene function in the male causes age-related infertility and gametogenic arrest as well as dramatic effects on sexual behavior. Analysis of the gonadal phenotypes revealed effects on apoptosis of steroidogenic cells as well as possible sex reversal of ovarian follicles. **Gail Prins** (University of Illinois at Chicago) continued the theme of hormonal control of gene expression with an analysis of the effects of neonatal exposure to estrogens on differentiation of the prostate gland. This valuable model system has revealed hormonal effects on prostate morphogenesis through control of expression of both homeobox genes and genes encoding signaling factors that regulate epithelial-mesenchymal crosstalk.

Sperm development, more than almost any other biological process, involves protein variants, and function of these variants is being revealed by genomic and structural approaches. **Deborah O'Brien** (University of North Carolina) presented an elegant and comprehensive mutational, structural, and cellular analysis of the function of the spermatid-specific glycolytic enzyme glyceraldehyde phosphate dehydrogenase (GAPDS). This work has fueled an emerging ''map'' of structural localization of sperm tail glycolytic enzymes to the scaffold of the outer dense fibers (ODF)—a truly novel concept that illuminates the function of the heretofore enigmatic ODF. **Gail Cornwall** (Texas Tech Health Sciences Center) discussed a large gene family, the cystatin-related proteins, specifically those of the epididymal spermatogenic (CRES) subgroup. The role of these unique proteins is not yet fully revealed, but they are cell-specific and highly regulated, suggesting novel roles for these protease inhibitors.

During the course of the conference, it became abundantly clear that DNA-binding and chromatin-modifying proteins play important roles in spermatogenesis and the development of male fertility. **Paula Cohen** (Albert Einstein College of Medicine) used her elegant mutational and developmental protein localization studies to illustrate novel roles for proteins in the DNA mismatch repair pathway in the processes of meiotic recombination and spermatogenesis. These studies are challenging current paradigms about protein interactions and function and will thus go a long way toward unraveling pathways of recombination. **Rolf Jessberger** (Mt Sinai School of Medicine) directly addressed roles of ''structural maintenance of chromosomes'' (SMC) proteins in spermatogenesis, highlighting the importance of chromosome cohesion to both meiotic recombination and chromosome segregation. Continuing on the theme of chromatin modification, **Willy Baarends** (Erasmus University) presented a functional analysis designed to determine the role of protein ubiquitination in spermatogenesis and particularly in setting the ''histone code'' modifications that are essential in regulating gene expression and chromatin conformation. Knockouts of genes in the ubiquitination pathway have revealed their importance to spermatogenic success and their roles in meiotic chromosome dynamics. Proteins that control chromatin conformation and completion of the meiotic divisions are under tight regulation, and **Joel Richter** (University of Massachusetts Medical School) described universal pathways of translational-level regulation that have been revealed by his studies of proteins regulating meiotic maturation of frog oocytes. As an example of perhaps unanticipated conservation of protein function, he has shown that a protein (CPEB) regulating cytoplasmic polyadenylation in oocytes also functions in regulation of translation of synaptonemal complex proteins during meiotic prophase in mouse spermatocytes. Surprisingly, nutritional studies illustrate the importance of chromatin and DNA integrity at the other end of the spermatogenic pathway. **Lynn Wallock** (Children's Hospital of Oakland Research Institute) gave tantalizing glimpses into effects of nutrition and micronutrients (vitamin C and folate) as well as behavior (smoking) on spermatogenic output, which may reflect DNA integrity and overall sperm quality.

All of these studies show the importance of both genomic and phenotype analysis in clarifying mechanisms of development of male reproductive function. **John Schimenti** (The Jackson Laboratory) showed the value of phenotype-driven mutagenesis to identify novel genes involved in processes of spermatogenesis. The studies he presented have provided insights into molecular mechanisms of meiotic recombination as well as having revealed complexity of genetic control of spermatogenesis in general. **Jeremy Wang** (University of Pennsylvania) presented results of imaginative genome-wide experimental and in silico analyses to detect genes that function in spermatogenesis. Coupling of gene discovery in mouse models with polymorphism association studies in humans translates basic biology into human diagnostic clinical applications. On the same theme, **Paul Turek** (University of California at San Francisco) showed the importance of thorough and informed phenotype analysis of human male infertility as a prerequisite for a functional genomic understanding. These studies captured the theme of functional genomics in revealing the power of cross-species genetic analyses, revealing function of a human gene

BOULE in rescuing *Drosophila* infertility due to mutation in the fly orthologous gene.

Participants at the Testis Workshop were further enriched by the presentation of nearly 100 posters exploring various facets of male reproduction and the role of functional genomic analyses. Taken together, the oral and poster presentations expanded the horizons of the conference attendees, prompting considerable curiosity as to how the new approaches and strategies might play out at the XVIIIth North American Testis Workshop in 2 years!

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Mary Ann Handel
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Matthew P. Hardy
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