Dr. Robert Viger is the recipient of the 2005 SSR New Investigator Award (funded by the Virendra B. Mahesh New Investigator Endowment), which recognizes a member of SSR for outstanding research completed and published within 10 years after receiving the Ph.D. or other equivalent professional degree. Dr. Viger completed his Ph.D. in 1995 under the direction of Dr. Bernard Robaire at McGill University where he studied the regulation of 5α-reductase and received a number of awards based on these studies. Dr. Viger carried out postdoctoral studies in molecular biology with Dr. Mona Nemer at the Clinical Research Institute of Montreal. During his postdoctoral research, he determined that the transcription factor, GATA-4, was expressed in a sexually dimorphic fashion during gonadal differentiation in the mouse. Shortly thereafter, Dr. Viger obtained his current position in the Department of Obstetrics and Gynecology at Laval University where he continues to explore the importance of GATA-4. Below is a description of his research as provided by Dr. Viger.

**GATA Factors and Reproductive Function: More than Just a Question of Sex**

Like all organ systems, the proper development and functioning of the reproductive system requires that specific sets of genes be expressed at the right place and at the right time. There are many classes of genes have been shown to regulate these processes. These include signaling molecules, growth factors, hormones, and transcription factors. Transcription factors are nuclear proteins that modulate gene expression by binding to specific DNA sequences found in the 5' regulatory or promoter region of target genes. For the past several years, my laboratory has been studying the role played the GATA family of transcription factors in gonadal development and function. There are six vertebrate GATA factors (GATA1 to GATA6) that have been named based on consensus nucleotide sequence (A/T) GATA (A/G) to which they bind. These factors were originally identified as crucial regulators of cardiac development and hematopoietic cell differentiation. GATA expression and function, however, are not limited to these two systems. In 1998, we were the first to demonstrate that one member of the GATA factor family, namely GATA4, was prominently expressed in the gonads starting early in development. Therefore, our hypothesis at that time was that GATA4 might be a key regulator of mammalian sex determination and differentiation.

In mammals, the testis and ovary develop from a common gonadal primordium. Sex determination (gonadal differentiation) is triggered by expression of the Y chromosome-located testis determining gene, called SRY. Although SRY was discovered more than fifteen years ago, we still only know of a handful of genes that lie upstream or downstream of this factor. The fact that GATA4 gene is abundantly expressed in the somatic cell population of the genital ridge prior to and during the time of SRY expression provided strong circumstantial evidence that this factor was indeed implicated in the early steps of gonadal development and perhaps sex determination. This hypothesis has now been confirmed in the mouse, where in vivo disruption of GATA4 function via a mutation of the Gata4 gene has been shown to lead to a block in testis development and a marked down-regulation of Sry expression. Thus, GATA4 appears to function as a direct upstream regulator of SRY expression in the developing testis. Although the latter has yet to be conclusively demonstrated, we have obtained recent in vitro evidence that this is likely the case.

Following sex determination, the action of hormones produced by the newly formed testis is responsible for conveying the male sex differentiation signal to the rest of the developing embryo. These include testosterone, insulin-like 3 (INSL3) and Müllerian inhibiting substance...
(MIS or AMH). In humans, the MIS gene is tightly regulated during gonadal development; lack of expression causes persistent Müllerian duct syndrome, a form of male pseudohermaphroditism. Consistent with a role in male sex differentiation, we and others have shown GATA4 to be an important activator of both the mouse and human MIS promoters via a direct transcriptional cooperation with the nuclear receptor, steroidogenic factor-1 (SF-1). Although no human GATA4 gene mutations have yet been linked to gonadal defects, some of our recent work has suggested that disruption of this GATA4/SF1 transcriptional cooperation on the MIS promoter might account for some cases of human male to female sex reversal involving insufficient MIS expression.

Although it is becoming more and more undeniable that you "gotta have" GATA for sex, the story certainly does not end there. GATA factors are in fact expressed in multiple cell types of both the testis and ovary well beyond the sex determining period. This, combined with the fact that GATA factors can be found in the gonads of diverse species ranging from lower invertebrates to humans, prompted us to speculate that GATA factors may in fact be master regulators of gonadal gene expression and function. Indeed, we and others have broadened the scope of GATA action to potentially include the regulation of steroidogenesis and the physiopathology of some human endocrine disorders such as endometriosis, breast cancer, and polycystic ovarian syndrome (PCOS). To date, our insights into the roles played by GATA factors in the gonads have come from the identification of novel GATA-dependent genes. This growing list includes genes that code for hormones and/or their receptors (MIS, inhibin and FSH receptor), enzymes and proteins involved in steroidogenesis (StAR, 3-HSD, P450 aromatase), and transcription factors (SRY, DMRT1). The requirement of a single factor in the control of multiple target genes appears to be an important regulatory mechanism in many tissues, including the endocrine organs. Since GATA4 is expressed in multiple gonadal cell types, and can activate transcription of many target genes on its own and in association with other cell-restricted transcription factors, we like to view GATA4 as a "pan-gonadal" regulator of gonadal transcription. We have gone on to identify GATA4 as a novel target of hormone-induced cAMP/PKA signaling in gonadal cells where phosphorylation of GATA4 and recruitment of cofactors such as CBP is a key mechanism for conveying the cAMP responsiveness of certain genes. Thus, we believe that GATA factors (and in particular GATA4) represent the cornerstone of a large transcriptional complex involved in the activation of different sets of genes in response to hormonal and stress signaling in the gonads and other endocrine tissues.

Elucidation of the in vivo role played by GATA factors in the reproductive tract has been hampered by the embryonic-lethal phenotype of GATA knockout mice. To overcome this problem, we developed several years ago a truncated GATA4 protein, that when expressed in cells, could function as a GATA dominant negative (DN) competitor and thereby block endogenous GATA activity, at least in vitro. To knockdown GATA activity in vivo, we have since developed a transgenic mouse line that can conditionally express this GATA DN competitor in a tissue-specific manner. This technology is currently being applied to understand GATA function in the testis but hopefully will soon also be used to pin down the role of these factors in the ovary. Surprisingly, despite the importance of GATA factors in multiple developmental processes, we still know very little about how the different GATA genes are themselves regulated. To begin to map out the in vivo regulation of the GATA4 gene, we have also recently generated several transgenic mouse lines driving expression of different fluorescent markers (green, yellow, and red fluorescent proteins) under the control of the GATA4 gene promoter. Taken together, these in vivo models should allow us to make significant headway into our understanding of the role and regulation of these factors in the gonads in both health and disease, an important topic of which we have only begun to scratch the surface.