

Blepharophimosis, Ptosis, and Epicanthus Inversus

FOXL2 Abnormal Gene Product

Polled intersex syndrome (PIS) in goat. In mammals, the Y-located *SRY* gene is known to induce testis formation from the indifferent gonad. A related gene, *SOX9*, also plays a critical role in testis differentiation in mammals, birds, and reptiles. It is now assumed that *SRY* acts upstream of *SOX9* in the sex determination cascade, but the regulatory link which should exist between these two genes remains unknown. Studies on XX sex reversal in polled intersex syndrome (PIS) goats have led to the discovery of a female-specific locus crucial for ovarian differentiation. In the PIS goats, dominant polledness is associated with recessive XX intersexuality. The PIS locus was shown to be located at goat chromosome 1q43, a region syntenic to human chromosome band 3q23 [Vaiman et al 1999]. Despite important phenotypic differences, the PIS goat has been considered to be an animal model for BPES. Pailhoux et al [2001] have shown that PIS is caused by the deletion of a critical 11.7-kb DNA element devoid of coding sequences and containing several repetitive elements. However, it was demonstrated that the deletion affects the transcription of at least two genes: the non-coding *PISRT1* gene (PIS-regulated transcript number 1), and *FOXL2*, located at 20 kb and 200 kb with respect to the deletion, respectively. This suggested a common long-range transcriptional regulation of *PISRT1* and *FOXL2* expression in the goat by the PIS locus and is suggestive of a putative disease-causing mechanism for BPES in humans.

Mouse models. A recent study presenting homozygous *Foxl2lacZ* mutant mice has shed light on the function of *FOXL2* during folliculogenesis in vivo. In *Foxl2lacZ*, granulosa cells from homozygous mutant ovaries do not complete the squamous-to-cuboidal transition, leading to the absence of secondary follicles and oocyte atresia. Activin-bA and anti-Müllerian inhibiting hormone expression is absent or strongly diminished in *Foxl2lacZ* homozygous mutant ovaries. Two weeks after birth, most, if not all, oocytes expressed Gdf9 in *Foxl2lacZ* homozygous mutant ovaries, indicating that nearly all primordial follicles have already initiated folliculogenesis at this stage. This activation, in the absence of functional granulosa cells, leads to oocyte atresia and progressive follicular depletion. In addition to providing a molecular mechanism for premature ovarian failure in BPES, this study showed that granulosa cell function is not only crucial for oocyte growth but also for ovary maintenance in vivo [Schmidt et al 2004]. A second report confirmed that mice lacking *Foxl2* recapitulate features of human BPES and that granulosa cell development fails in *Foxl2*-null animals from the time of primordial follicle formation [Uda et al 2004].

Structure, evolution, and expression. Crisponi et al [2004] reported that three translocation breakpoints, located at more than 171 kb 5' of the transcription start

of *FOXL2* causing BPES, fall within intron 6 of *MRPS22*, a large gene consisting of 20 exons. Sequence comparisons revealed conserved segments in introns 6, 11, and 12 of human and mouse. The intron 11 sequence is also deleted in the PIS goat. The authors stated that the conserved sequences are candidates to be distant enhancers or otherwise to affect higher-order chromatin structure to impose long-range *cis*-regulation of *FOXL2* expression.

A comparative human/mouse analysis of the orthologous region around the PIS locus by Nikic & Vaiman [2004] permitted the targeting of genes in the 1-mb environment, and identification of previously unknown mouse orthologues for *Pisrt1*, *Bpesc1*, and *Chr3syt*, and a human orthologue for *PISRT1*. Tissue-specific gene expression in mice and goats was assessed for ten and eight genes, respectively, located in a 1-mb DNA region surrounding the PIS locus. It was shown that gene expression is essentially regulated in a similar manner in goat and mouse tissues in the PIS vicinity.

Beysen et al [2005] identified novel microdeletions outside *FOXL2* in simplex and familial BPES cases. Specifically, four rearrangements with an overlap of 126 kb are located 230 kb upstream of *FOXL2*, telomeric to the reported translocation breakpoints. Interestingly, the human orthologous region of a 12-kb sequence deleted in the polled intersex goat is contained in this SRO, providing evidence of human-goat conservation of *FOXL2* expression and of the mutational mechanism.

Literature Cited

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