A CRISPR VEZT Knock-in mouse model of endometriosis increases lesion and adhesion formation reflective of Stage IV disease

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Introduction/Background
Endometriosis is an estrogen dependent gynaecological disease where endometrial-like tissue implants in locations ectopic to the uterus. *VEZT*, an adherens junction protein coding gene, is an endometrial risk gene of unknown function. In this study we characterised peritoneal lesion and adhesion formation and impacts on fertility in a CRISPR *VEZT* knock-in mouse model of endometriosis.

Materials and Methods
VEZT-Cre mice induced with tamoxifen daily (5x75mg/kg). Reproductive and liver tissue was collected at 4 and 12 weeks to verify global expression (N=6). Endometriosis was established with the intraperitoneal injection of minced donor uterine tissue (40 mg) into *VEZT*-Cre and C57/BL6 wildtype (WT) recipient mice (N=6 each). Adhesions were established through the injection of 17β-estradiol valerate weekly (4x10 ug/ml). Fertility was observed with crossing induced male and female *VEZT*-Cre mice (N=7).

Results
Tamoxifen induced *VEZT*-Cre mice overexpressed *VEZT* in liver, uterus and testis by 12 weeks. Large cystic, highly proliferative, inflamed and vascularised lesions were identified in all grafted *VEZT*-Cre mice from 4-8 weeks in liver, pancreas, ovary, bowel, peritoneum and inguinal fat pads surrounded by fibrosis and adhesions. WT mice failed to produce lesions beyond 4 weeks. *VEZT*-Cre uteri significantly upregulated TGFβ, TNFα (inflammation) and CTGF (fibrosis) mRNA expression in eutopic endometrium compared to WT mice. When *VEZT*-Cre mice (but not WT mice) were treated with 17β-estradiol-valerate to mimic the endometriotic environment, adhesions formed throughout the peritoneal cavity binding organs along with inguinal fat pad adipogenesis and cystic ovaries. When *VEZT*-Cre mice were mated, they were successfully impregnated with live birth rates and maternal morbidity pending.

Conclusion
This is the first endometriosis mouse model to demonstrate a significant role for *VEZT* in the development, adhesiveness and invasiveness of endometriotic lesions and in the presence of estradiol, the development of significant peritoneal adhesions. These findings identify *VEZT* is a potential therapeutic target or biomarker in the future.

Key words: Endometriosis, Mouse, Model