MATERIALS AND METHODS: HuLM cells were cultured on cover slips and serum starved for overnight. Cells were treated with Vitamin D (1μ M and 5μ M) for 48 hours. Treated cells were fixed in 3.7% formaline, permeabilized in 0.1% Triton X-100 and stained with FITC-conjugated falloidin (2μ g/ml). Stained actin fibers were visualized using laser confocal microscopy.

RESULTS: We observed that at both concentrations of Vitamin D significantly decreased (1.6 and 3.5 folds) staining of actin fibers in HuLM cells when compared with untreated control cells (P < 0.05). Treatment of HuLM cells with Vitamin D showed shrinkage of cell size and disorganization of actin fibers as compared with untreated control cells. In addition, Vitamin D decreases fluorescent intensity of actin fibers in a concentration-dependent manner as quantified by image analyses. We are continuing to evaluate the effects of Vitamin D on other gene products that are involved in tumor fibrosis such as collagen type 1, TGF- β 1, PAI-1 and MMPs using immunofluorescence, western blot and RT-PCR analyses.

CONCLUSIONS: Our results showed that Vitamin D deregulates and disorganizes the structural actin fibers in HuLM cells suggesting a potential role in regulation of tissue fibrosis. Understanding the mechanisms by which Vitamin D regulates the fibrosis will help to determine the potential physiological contribution of Vitamin D deficiency in the increased risk of leiomyomas in African American women as well as provide novel therapeutic approaches.

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AN INJURY-INDUCED CELL MODEL OF EARLY UTERINE FIBROID DEVELOPMENT. L. Feng, F. L. Jayes, T. R. Greene, D. W. Schomberg, P. C. Leppert. OB-GYN, Duke University, Durham, NC.

OBJECTIVE: The etiology of uterine fibroids is unclear and there is no universally accepted model for this common condition often causing pain, bleeding and infertility. Recent studies indicate uterine fibroids are formed by accumulation of ECM rather than by cellular proliferation and that altered wound healing leading to fibrosis may be the underlying mechanism. Fibrogenesis is a complex process in which the transforming growth factor-beta (TGFb) pathway plays a major role. TGFb3 and an activator of TGFb, thrombospondin-1, were previously reported genes disregulated in uterine fibroids.

DESIGN: In an *in-vitro* cell culture model of myometrial cells we characterized fibrotic pathways activated due to injury.

MATERIALS AND METHODS: Smooth muscle cells from fresh myometrium obtained at hysterectomy from non-pregnant women were cultured in 10% serum until 70% confluent. Injury was simulated by 72 h serum-starvation (SS) followed by serum replacement (SB). Cells were evaluated for cell viability and examined by electron microscopy (EM) and confocal microscopy. We measured mRNA and/or protein expression of thrombospondin-1 after SB, and Smad3 phosphorylation after TGFb treatment.

RESULTS: Cell viability after 72 h SS did not differ from controls. Cells stained positive for human α -SMA. EM showed 60-80 % of SS cells had notched nuclei and a decreased cellular/nuclear ratio characteristic of myofibroblasts, cells active in wound repair and fibrosis. Thrombospondin-1 mRNA and protein expression were increased within 2 h after SB and remained elevated. TGFb treatment induced Smad3 phosphorylation.

CONCLUSIONS: We conclude that myometrial cells can be differentiated to myofibroblast-like cells and that wound healing and early fibrosis pathways with a potential role in early uterine fibroid development are inducible in this cell model. This model offers a promising approach for exploring the pathogenesis of uterine fibroids and for developing cell-based therapeutic strategies.

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DEVELOPMENT OF AN EVIDENCE-BASED CLASSIFICATION SYSTEM FOR UTERINE FIBROIDS. J. B. Davis, M. S. Broder, W. H. Catherino, R. W. Rebar, V. W. Pinn, J. H. Segars. Obstetrics and Gynecology, Akron General Medical Center, Akron, OH; Partnership for Health Analytic Research, LLC, Los Angeles, CA; Obstetrics and Gynecology, Uniformed Services University of the Health Sciences, Bethesda, MD; American Society for Reproductive Medicine, Birmingham, AL; OFC Research on Women's Health, National Institutes of Health, Bethesda, MD; The Eunice Kennedy Shriver National Institute of Child Health and Human Development, National Institutes of Health, Bethesda, MD.

OBJECTIVE: Uterine fibroids are common benign tumors in women. To provide optimal care physicians and researchers need an evidence-based sys-

tematic approach to describe fibroids, but no universally accepted clinical scoring system currently exists. The objective of this effort was to develop an evidence-based fibroid classification system.

DESIGN: A modified Delphi process.

MATERIALS AND METHODS: A literature search was used to develop a theoretical framework that included goals, characteristics, and measurement domains that led to the development of a 203-item rating form. Ten expert panelists involved in the study and treatment of uterine fibroids including: an interventional radiologist, two general gynecologists, a basic researcher, an epidemiologist, an academician, and a reproductive endocrinologist met in Bethesda. Panelists completed the rating form, which used a Likert scale to determine the criteria deemed essential for a fibroid classification system. Individual responses were discussed and scored based on the level of agreement among all participants. A modified questionnaire was developed, completed and the responses used to frame a consensus classification system at a follow-up meeting.

RESULTS: The new 'Bethesda' classification system defines fibroids by location in comparison to the uterine cavity on a 0 to 6 point scale, with zero being completely in the cavity and six being pedunculated. The system further defines the location of fibroids as fundal (upper 2/3 of the uterus), isthmic (lower 1/3 of the uterus), or cervical (below the internal os). The fibroid size is determined by ultrasound clinically and/or by MRI for research.

CONCLUSIONS: The Bethesda scoring system for uterine fibroids provides a framework for both clinicians and investigators. The system incorporates evidence-based research and a feasibility test and validation study are planned.

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SEXUALITY

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HYPOACTIVE SEXUAL DESIRE DISORDER (HSDD) IN WOMEN WITH AND WITHOUT COMORBID CONDITIONS. D. Foley, B. Johnson, K. Foley. Health Economics & Outcomes Research, Boehrin ger-Ingelheim.com, Ridgefield, CT; Pharmaceutical Research Operations, Thomson-Reuters Healthcare, Newtown, PA; Department of Epidemiology, Thomas Jefferson University, Philadelphia, PA.

OBJECTIVE: To describe clinical characteristics & comorbid conditions of women with commercial insurance diagnosed with HSDD.

DESIGN: Retrospective U.S. claims database analysis.

MATERIALS AND METHODS: The Marketscan® Commercial Database of adjudicated insurance claims was used to identify women aged 18-64 diagnosed with HSDD (ICD-9 code: 302.71) from 1/98-9/06 (prior to most studies of HSDD). Clinical characteristics & comorbid conditions associated with sexual dysfunction were examined in the year prior to diagnosis.

RESULTS: 2,870 women were diagnosed with HSDD, 70% of whom were between the ages of 18 & 49 (younger women) while 30% were between the ages of 50 & 64 (older women). In the 12 months prior to diagnosis with HSDD, younger women were more likely than older women to have had gynecological surgery (3.6% vs. 1.7%, p=0.0064), or fill a prescription for birth control (25.4% vs. 4.1%, p<0.0001). Older women were more likely to have non-reproductive associated conditions that could affect sexual dysfunction including diabetes (5.0% vs. 1.9%, p<0.0001), thyroid abnormality (11.2% vs. 7.0%, p=0.0002), cancer or history of cancer (7.8% vs. 1.8%, p<0.0001). Approximately two-thirds of women in each age group had at least one condition possibly correlated with sexual dysfunction. Deyo Charlson Comorbitity scores indicated that women without correlates of sexual dysfunction were significantly healthier relative to those with at least one correlate (0.06 vs. 0.18, p<0.0001 in younger women & 0.07 vs. 0.41, p<0.0001 in older women).

CONCLUSIONS: Approximately one-third of women with HSDD have no diagnosed or treated medical conditions known to affect sexual functioning. For these women, HSDD may be their only medical diagnosis. Of the women with comorbid conditions, younger women were more likely to have conditions directly affecting their reproductive systems, while older women were more likely to have non-reproductive conditions.

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