mild male factor, minimal to mild endometriosis and unexplained infertility problems. Intrauterine insemination (IUI) and donor insemination (DI) gained popularity because of simplicity, non-invasiveness and cost-effectiveness. There is however no consensus of opinion regarding at what stage couples should be offered *in vitro* fertilization (IVF). The aim of this study is to determine the optimum number of SIUI or DI cycles that can be offered to a selected population of subfertile couples.

Materials and methods: The electronic database (ACUSys) of the IVF Unit at St Mary's Hospital was searched for all S-IUI or DI cycles carried out between January 2001 and December 2005. During this period, the couples with unexplained infertility, mild male factor (defined as sperm count of 10–19 million/ml and >20% progressive motility) or mild endometriosis were offered up to 7 cycles of S-IUI. All women were between 22–38 years old, with less than 4 years history of subfertility and body mass index (BMI) between 19–30 kg/m^2. They all had normal baseline hormone profile and bilateral tubal patency on hysterosalpingography or laparoscopy and dye test. Controlled ovarian stimulation was achieved by daily subcutaneous injections of human menopausal gonadotrophin (hMG) in mean doses of 75 IU, ranging from 37.5 IU to 150 IU, until transvaginal sonography showed one follicle greater than 16.5 mm in diameter. Ovulation was then induced with 5000 IU of human chorionic gonadotrophin (hCG) and insemination carried out 36 hours later. The cycle was cancelled if there were more than three follicles ≥14 mm diameter. The DI programs were performed after thawing frozen donor sperm that was then used in un-stimulated cycles. Serum LH levels were measured daily starting from day 3 of a spontaneous menstrual cycle and plotted on a chart. Insemination was performed 24 hours after the LH surge (>25 IU/ml). The study received approval from the Local Research Ethics Committee.

Results: One hundred sixty-two couples underwent a total of 410 cycles of S-IUI with an average of 2.5 cycles per couple. The cumulative live birth rate of SIUI showed a progressive increase from 8.0% after the first cycle to 26.1% after cycle 4. No pregnancies occurred in couples who had more than 4 cycles of S-IUI. There were no significant differences with respect to woman's age, BMI, duration, type and cause of subfertility between patients who had 1 to 4 cycles and those who had >4 cycles. Two hundred thirty-five couples underwent a total of 716 DI cycles with an average of 3.0 cycles per couple. Contrary to S-IUI, the cumulative LB rate of DI increased from 44.8% after 4 cycles to 79.2% in cycle 9. A beta-binomial selection model showed no significant heterogeneity between patients for both S-IUI and DI, and the data was consistent with the cycles being independent.

Conclusions: The probability of live birth after the fourth cycle of SIUI is very small. Whilst it is beneficial and probably cost effective to offer couples with male factor infertility more than 6 cycles of DI, this may not be the case for S-IUI. Noteworthy, the apparent decline in LB rate for S-IUI over time could simply be an artifact of the very small number of couples getting many treatment cycles.

SELECTED ORAL COMMUNICATION SESSION

Session 43: Endometriosis: basic research

08 July 2008 **15:15–16:30**

O-159 Oral Combating endometriosis by blocking proteasome and nuclear factor-kappab pathways

S. Hascalik[1], O. Celik[1], K. Elter[2], M.E. Tagluk[3], B. Gurates[4], N.E. Aydin[5]
[1]*Inonu University Medical Faculty, Obstetrics and Gynecology, Malatya, Turkey,* [2]*EuroFertil, Center for Human Reproduction, Istanbul, Turkey,* [3]*Inonu University, Electric and Electronic Engineering, Malatya, Turkey,* [4]*Firat University Medical Faculty, Obstetrics and Gynecology, Elazig, Turkey,* [5]*Inonu University Medical Faculty, Pathology, Malatya, Turkey*

Introduction: Endometriosis is associated with systemic subclinical inflammation. Nuclear Factor-kappaB (NF-κB) is a ubiquitously expressed transcription factor that causes inflammation in endometriotic cells. The inhibition of NF-κB within the endometrial implant may be achieved either by NF-κB inhibitors or by prevention of releasing of NF-κB precursors using proteasome inhibitors. The objective of this study was to investigate the effect of pyrrolidine dithiocarbamate (PDTC; an NF-κB inhibitor) and bortezomib (Velcade; a proteasome inhibitor) on the development of experimental endometriotic implants in rats.

Materials and methods: Endometriosis was surgically induced in 30 rats using the method of Vernon and Wilson. Three weeks later the viability and volume of the implants were recorded and classified as described by Ingelmo. Six of 30 rats were not considered satisfactory because the implants of four were nonviable and of two were covered by omentum. Other remaining rats were put into three groups with equal numbers: the control (0.1 mL of isotonic saline/day per rat, i.p), the PDTC (100 mg/kg of body weight, i.p) and the bortezomib (0.2 mg/kg of body weight, i.p) groups. Seven days after treatment, a third laparotomy was done and the volume of implants was measured again using the ellipsoid formula. The animals were then sacrificed, and the implants were processed for microscopic examination. Five micrometer sections of paraffin blocks were stained with hematoxylin and eosin, Masson's trichrome, Ki67 which is expressed in the cells during M, G1, S and G2 phases of cell cycle, proliferating cell nuclear antigen (PCNA), a nuclear protein whose expression peaks during the S phase of the cell cycle and CD34 a stain used for endothelial cells. Histologic changes of the implants, vascularity, PCNA, Ki67 and CD34 immunoreactivity was assessed under light microscopy and scored using a semi-quantitative grading system.

Results: In 80.0% of implanted rats vesicle at the sutures region was observed; the vesicle diameters were <2 mm (grade 2) in 41.6%, between 2 mm and 4.5 mm (grade 3) in 33.3% and >4.5 mm (grade 4) in 25.0%. After treatment with PDTC or bortezomib these percentages were decreased in grade 3 and grade 4, and increased in grade 1 and 2. In the control group, on the contrary, these percentages increased in grade 4 and decreased in grade 2. The implant volumes were $72.5 \pm 28.7\ mm^3$, $64.6 \pm 24.7\ mm^3$ and $67.3 \pm 35.1\ mm^3$ before treatment, which did not show a statistically significant difference ($P > 0.05$), and were $80.4 \pm 30.1\ mm^3$, $31.9 \pm 20.0\ mm^3$ and $32.8 \pm 20.1\ mm^3$ after the treatment in the control, PDTC and bortezomib groups, respectively. After the treatment, while the volumes in the control group were higher then others ($P < 0.000$ and $P < 0.000$), the volumes in the PDTC and bortezomib groups remained comparable ($P = 0.798$). As the pre- and post-treatment volumes were compared in each group, the posttreatment volumes were decreased in the PDTC and bortezomib groups ($P < 0.002$ and $P < 0.001$), and slightly increased in the control group ($P = 0.279$). Immunohistochemical stainings for PCNA, Ki67 and CD34 has revealed an intense proliferation of stromal, glandular and endothelial cells in implants of control animals, but only a minor proliferation of this cells which were mainly localized within the endometrial stroma in implants of PDTC and bortezomib groups. In PDTC and bortezomib groups, CD34, PCNA and Ki67 expressions were similar but were significantly reduced as compared to control group ($P < 0.05$). Also the mean score of vascularity, stromal and glandular tissue in the PDTC and bortezomib groups were lower than in the control group ($P < 0.05$).

Conclusions: Selective NF-κB inhibition induces regression on endometriotic implants in rats. Accordingly, PDTC and bortezomib may represent a novel therapeutic strategy for treatment of endometriosis.

O-160 Oral Tunicamycin enhances tumor necrosis factor-related apoptosis-induced ligand-induced apoptosis in endometriotic stromal cells

A. Hasegawa[1], Y. Osuga[1], Y. Hirota[1], K. Hamasaki[1], A. Kodama[1], T. Yano[1], Y. Taketani[1]
[1]*University of Tokyo, Obstetrics and Gynecology, Tokyo, Japan*

Introduction: Failure of apoptosis of refluxed endometrial cells within the peritoneal cavity is a possible etiologic factor for development of endometriosis. It has been demonstrated that osteoprotegerin (OPG) concentration in the peritoneal fluid is increased in women with endometriosis. Because OPG has an antagonistic effect against tumor necrosis factor-related apoptosis-induced ligand (TRAIL), attenuation of TRAIL-inducing apoptosis in endometriotic cells has been suggested to subserve the development of endometriosis. Recently, enhancement of TRAIL-induced apoptosis by tunicamycin, via induction of endoplasmic reticulum (ER) stress, has