pictorial blood assessment chart, PBAC), length of menstrual period, and a 6-item health-related quality of life (QOL) questionnaire were collected in the second week before, during each treatment cycle, follow-up cycle and were compared.

RESULTS: Both treatments led to significant decreases in mean PBAC scores, shorter durations of menstrual periods, and improved QOL rankings during both the first and second treatment cycles. The mean amplitude of PBAC decrement in the first cycle in the TA group was significantly greater than that in the NET group (29.31% vs. 10.64%, P=0.011); however, the difference did not continue into the second treatment cycle (39.87% vs. 28.9%, P=0.088). Improvement in four QOL items during the first cycle in the TA group was also significantly greater than that in the NET group (P=0.032−0.002). The incidence of adverse effects between the two groups was similar (12.72% vs. 15.78%, P=0.530), and was mostly comprised of gastrointestinal symptoms. The number of patients willing to continue the treatment in the second and follow-up cycles in the TA group, respectively, were significantly higher than those in the corresponding cycles of the NET group (96.30% vs. 76.6%, P=0.003; 77.36% vs. 55.56%, P=0.022, respectively).

CONCLUSIONS: TA 3g daily during cycle D1–5 is a more effective and tolerable treatment than NET on D19–26 in the first treatment cycle in patients with ovulatory menorrhagia.

Supported by: Daiichi Sankyo Co. Ltd.

MALE FACTOR: ART ABSTRACTS

Monday, October 15, 2007
3:00 pm

O-48

THE LEVEL OF SPERM VACUOLES IN THE FRESH POST-PROCESSED SPERM SAMPLE SIGNIFICANTLY AFFECTS IVF CYCLE OUTCOMES. N. Bar-Chama, J. Schiff, M. Luna, B. Dann, A. B. Copperman, J. Barritt. Reproductive Endocrinology and Infertility, Reproductive Medicine Associates of New York; Mount Sinai School of Medicine, New York, NY.

OBJECTIVE: Impaired sperm morphology has been shown to correlate with decreased fertilization in vitro. Numerous investigators have searched for other diagnostic markers and assays to assess sperm competency. Recently, sperm vacuoles have been suggested as a prognostic factor in IVF (1). We sought to determine whether the percentage of sperm with vacuoles in the fresh post-processed sample was associated with IVF clinical outcomes.

DESIGN: Retrospective data analysis of fresh sperm samples and subsequent IVF outcomes.

MATERIALS AND METHODS: A series of 1295 IVF cycles using fresh sperm samples from 11/2005- 03/2007 were included. Post-processed sperm samples were analyzed for sperm morphology (Kruger method) by a trained embryologist. The same samples were then used in the fresh IVF cycle. The percentage of sperm with vacuoles was recorded and classified as 0%, 1–4% and ≥5%. We investigated the correlation of vacuoles with fertilization, clinical pregnancy and loss rates. Chi-square and Student's t-tests were used where applicable.

RESULTS: Of the 1295 samples prepared for IVF, 1042 (80.5%) had no vacuoles, 202 (15.6%) had 1–4% vacuoles, and 51 (3.9%) had ≥5% vacuoles. Results are included in table 1. The mean oocyte age, fertilization and clinical pregnancy rates were not significantly different. Although the pregnancy rate declined as the percentage of vacuoles increased, a significant difference was not noted. A loss rate of 36.8% was found when the sperm sample contained ≥5% vacuoles which was significantly higher than those cases with <5% vacuoles (14.8%) (P<0.05).

TABLE 1. Results IVF Outcome

% Vacuoled Sperm	Oocyte Age	Fertilization Rate	Clinical Pregnancy Rate	Pregnancy Loss
0%	34.6 ± 5.4	57.1%	47.9% (499/1043)	14.8% (74/499)*
1–4%	34.5 ± 1.2	57.2%	43.6% (88/202)	14.8% (13/88)¶
≥5%	34.3 ± 1.2	55.8%	37.3% (19/51)	36.8% (7/19)*¶

*¶ P<0.05.

CONCLUSIONS: Morphological analysis of the post-processed fresh sperm used for IVF cycles may allow for a correlation to outcome. We have described that the presence of vacuoles in ≥5% of the sperm used in IVF cycles impaired later embryonic development and significantly increased pregnancy loss rate. The relationship between abnormal sperm parameters and IVF outcomes must continue to be refined, but our findings demonstrate that a clinical pregnancy should not be the final outcome measure. Further investigation of the significance of sperm vacuoles is needed to confirm this predictive parameter and elucidate the pathophysiology of this observation. 1) Berkovitz, et al. Hum Reprod 2006.

Supported by: None.

Monday, October 15, 2007
3:15 pm

O-49

EVALUATION OF SPERM CHROMATIN INTEGRITY AND SEMINAL QUALITY IN HIV POSITIVE PATIENTS. F. Ayala A, A. Morales, M. Merino, E. Gonzalez, L. Sordia, V. Oscar. University Center of Reproductive Medicine, Hospital Universitario, UANL, Monterrey, NL, Mexico.

OBJECTIVE: To identify the effect of HIV infection on DNA maturation process and integrity of sperm cells, as well as the degree of alteration in seminal parameters using aniline blue and acridine orange tests.

DESIGN: Prospective, descriptive, comparative study.

MATERIALS AND METHODS: Twenty HIV positive male patients were included in the case group. Sperm chromatin was evaluated with acridine orange and aniline blue tests. Spermogram, hepatitis B, hepatitis C, Chlamydia and syphilis detection test were performed in every patient. Results were compared with an equal size group of healthy controls. Statistical evaluation was done using t test, simple correlation and logistic regression.

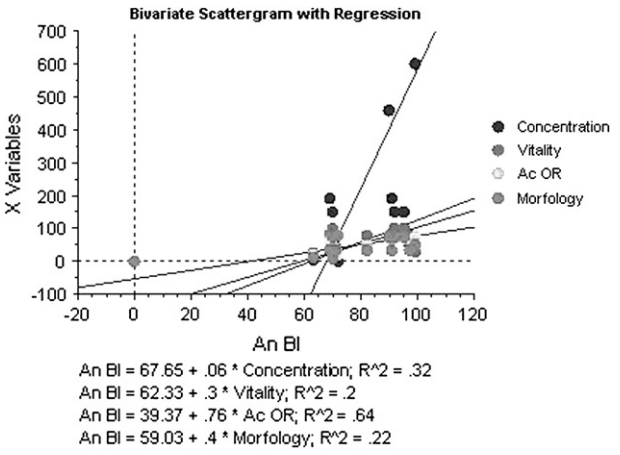

Figure 1.

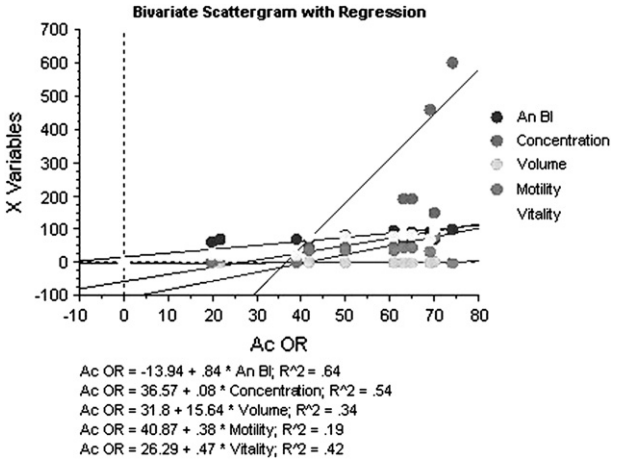

Figure 2.

RESULTS: There was no significant difference in aniline blue and acridine orange tests results in cases group compared with controls ($P>0.05$). Mean test results in HIV positive patients for aniline blue was 79.1% (±22.2), for acridine orange was 52.2% (±23.3). A statistically significant direct correlation was observed between aniline blue test result and acrinidine orange test result, sperm concentration, vitality and morphology in HIV patients (Figure 1). A direct correlation was observed between acridine orange test result and sperm concentration, semen volume, vitality and motility in the same group (Figure 2). There was no correlation between HIV viral load or CD4 count with chromatin integrity test results or spermogram parameters alterations.

CONCLUSIONS: There is no difference in sperm chromatin alterations was observed in HIV positive patients compared with healthy controls. There is no correlation of the degree of sperm chromatin or semen parameters alterations with HIV viral load or CD4 count in HIV positive patients. Analine blue and orange acridine tests results are correlated with most semen parameters results in HIV patients.

Supported by: None.

Monday, October 15, 2007
3:30 pm

O-50

CHARACTERIZATION OF MALE GERM-LINE STEM CELLS OBTAINED FROM HUMAN ADULT TESTICULAR TISSUES. D. R. Lee, J. J. Lim, S. Y. Kim, S. H. Song, T. K. Yoon, K.-S. Kim. Fertility Center of CHA Hospital, CHA Research Institute, Pochon CHA University, Seoul, Korea; Department of Anatomy and Cell Biology, College of Medicine, Hanyang University, Seoul, Korea.

OBJECTIVE: Male germ-line stem cells (GSCs, spermatogonial stem cells) self-renew and produce large numbers of differentiating germ cells that become spermatozoa throughout the adult life. GSCs have unipotency and become only germ cells (Buehr et al., 1997). However, Guan et al. (2006) recently reported that mouse adult GSCs can be transformed in culture to embryonic stem cell (ESC)-like cell lines displaying pluripotency both in vitro and significantly in vivo.

DESIGN: This study was to find and characterize pluripotent GSCs from human testicular cells of non-obstructive azoosperimia (NOA).

MATERIALS AND METHODS: Testicular tissues were obtained from 35 NOA patients (maturation arrest and Sertoli cell-only syndrome) under informed consent were dissociated and plated into gelatin-coated dishes. After passaging, dissociated cells were analyzed by RT-PCR, immunocytochemistry, flow cytometry using markers of ESC and SSC. Also, telomerase activity was analyzed.

RESULTS: In the presence of glial cell line-derived neurotrophic factor (GDNF), epidermal growth factor (EGF), basic fibroblast growth factor (bFGF) and leukemia inhibitory factor (LIF), multi-cellular colonies were formed from testicular cells of 10 patients after 2–4 weeks of culture. Colonies were dissociated and passaged every 2 weeks. At 2nd passages, markers of ESC (Oct-4, Rex-1, SSEA-4, TRA 1–60, TRA 1–80) was highly expressed and then revealed decreased expression after 4 passages. Markers of GSC (integrin $\alpha 6$, and $\beta 1$) were highly expressed until 5 passages. c-Kit receptor, a marker of post-meiotic germ cell was more highly expressed at 5th passages. Finally, telomerase activity (DNA laddering) were peaked in 2–3 passages and then decreased. Around 6 passages, proliferating activity was quickly decreased, and most cells were attached onto the culture dish and differentiated into fibroblast-like cells.

CONCLUSIONS: This result revealed partial presence of pluripotent SSCs in the cultured human adult testicular tissues. Purification and long term-proliferation of pluripotent SSC from human testicular tissue may allow individual cell-based therapy without the ethical and immunological problems.

Supported by: This work was supported by a grant from the National R&D Program (2006–04127) of the Ministry of Science & Technology, Republic of Korea and the Korea Science and Engineering Foundation.

Monday, October 15, 2007
3:45 pm

O-51

EVALUATION OF MALE TRANSLOCATION PATIENTS UNDERGOING ART. A. L. Davis, M. A. Witt, C. W. Elsner, D. Mitchell-Leef, Z. P. Nagy, T. C. Callaghan. Reproductive Biology Associates, Atlanta, GA.

OBJECTIVE: To review the clinical outcomes for male translocation carriers undergoing ART with variable sperm parameters. To determine if there was any association with clinical outcome to the type of translocation, the degree of disruption to spermatogenesis and the level of abnormal sperm morphology.

DESIGN: Five male patients with a robertsonian translocation and 3 male patients with reciprocal translocations were reviewed for semen parameters, embryo quality and PGD results.

MATERIALS AND METHODS: Semen parameters were assessed according to World Health Organization, 1999, criteria using Kruger strict morphology. Oligozoospermia was classified as extreme if spermatozoa were less than 1 million/ml, severe 1–5 million/ml and moderate 5–20 million/ml. Embryos were assessed at 68 to 72 hours after insemination. Several parameters including cell number, extent of fragmentation and cell size were taken into account and assigned to four grades; A to D (corresponding to excellent; good; medium and poor quality) Preimplantation Genetic Diagnosis was carried out on day 3 cleavage stage embryo biopsy, using fluorescence in situ hybridization (FISH), on single interphase nuclei with locus specific probes that allowed the discrimination between the normal and balanced form of the translocation. Where possible aneuploidy screening of chromosomes unrelated to the translocation was performed.

RESULTS: As follows;

TABLE 1.

PATIENT #	KARYOTYPE	SPERM CLASSIFICATION	KRUGER STRICT MORPHOLOGY	# EMBRYOS TESTED	NORMAL OR BALANCED FOR TRANSLOCATION	NORMAL FOR OTHER CHROMOSOMES	# CHAOTIC PER ABNORMAL EMBRYOS	# OF EMBRYOS TRANSFERRED	EMBRYO GRADE TRANSFERRED	AGE OF FEMALE	HCG	# OF FCA
1	45,XY,der(13;14)(q10;q10)	Extreme	NR	8	0	0	3/8	0		32		
		TESA										
2	45,XY,der(13;14)(q10;q10)	Extreme	2	11	0	0	11/11	0		30		
3	45,XY,der(13;14)(q10;q10)	Moderate	0	15	3	3	3/12	2	AA	29	+	0
4	45,XY,der(13;14)(q10;q10)	Normal	5	11	7	7	2/4	3	AAA	32	+	3
5 cycle1	45,XY,der(13;15)(q10;q10)	Severe	1	3	2	0	1/3	0		37		
5 cycle2	45,XY,der(13;15)(q10;q10)	Severe	1	5	5	3	0/2	3	AAB	37	+	3
6	45,XY,t(2;8)(q37.3;q23.1)	Extreme	0	11	0	NR	NR	0		28		
7	45,XY,t(11;21)(p15.1;q22.1)	Normal	8	4	0	NR	NR	0		31		
8	46,XY,t(7;12)(q34;q24.1)	Normal	NR	15	1	NR	NR	1		35	-	0

CONCLUSIONS: The number of abnormal embryos was shown to be directly associated with the degree of disruption of spermatogenesis and an increase in abnormal morphology. It was seen in those embryos that were tested for further chromosomal aneuploidy, of chromosomes unrelated to the translocation, there was a higher degree of aneuploidy in patients with suboptimal sperm parameters, in particular an increase in chaotic embryos. Where translocation couples need assisted reproductive technology for sub-fertility, PGD is a valuable screen for imbalance even for those couples where the risk of viable chromosome abnormality is assumed to be low. Furthermore, for those patients with poor sperm parameters there is a need to consider aneuploidy screening for chromosomes unrelated to the translocation carried.

Supported by: None.

Monday, October 15, 2007
4:00 pm

O-52

FREQUENT EJACULATION. A PILOT STUDY OF CHANGES IN SPERM DNA DAMAGE AND SEMEN PARAMETERS USING DAILY EJACULATION. D. J. Greening. Medicine, Sydney IVF, Sydney, NSW, Australia.

OBJECTIVE: To assess the effect of frequent ejaculation on sperm DNA damage and on semen parameters.

DESIGN: Prospective study.

MATERIALS AND METHODS: 42 men with a raised SCSA level >15% were enrolled in the pilot study from a cohort of 138 couples investigated for either recurrent miscarriage of repeated IVF failure. Initial SCSA levels and semen parameters were assessed on all men after 3 days abstinence (D−3). <15% SCSA was considered normal, 15–27% low abnormal SCSA and >27% as high abnormal. Previous SCSA studies suggested a robustness of SCSA results on repeat testing and thus only one initial SCSA >15%