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CHARACTERISTICS OF THE NICHE REQUIRED FOR HUMAN SPERMATOGONIAL STEM CELL EXPANSION. P. L. Yango,[a] J. F. Smith,[b] E. Altman,[a] A. Poelzl,[a] P. V. Lishko,[c] N. D. Tran.[a] [a]Department of Obstetrics, Gynecology and Reproductive Sciences, University of California San Francisco, San Francisco, CA; [b]Department of Urology, University of California San Francisco, San Francisco, CA; [c]Department of Molecular & Cell Biology, University of California Berkeley, Berkeley, CA.

OBJECTIVE: Spermatogenesis is maintained by an appropriate interaction between spermatogonial stem cells (SSCs) and the various types of somatic cells within the testicular niche. However, little is known about this dynamic relationship. The aim of this study is to investigate the unique cellular niche required for SSC expansion.

DESIGN: An in vitro study with human testicular SSC culture.

MATERIALS AND METHODS: Normal adult human testicular tissues were collected and a portion was sectioned for analysis by immunofluorescence (IF). The remaining tissue was digested into single cell suspensions. SSCs and somatic cell subpopulations were isolated by fluorescence activated cell sorting (FACS), analyzed, co-cultured, and evaluated for their role in supporting SSC growth.

RESULTS: SSCs, spermatogonia, spermatocytes, Sertoli cells and stromal cells were individually isolated, using a distinct set of extracellular markers. When purified SSCs were cultured in the presence of different populations of testicular supporting cells or mouse embryonic fibroblasts, testicular stromal cells were found to be absolutely essential for appropriate SSC binding and growth. Without the niche provided by stromal cells, SSCs failed to bind and growth was inhibited. Specifically, these stromal clones were shown to originate from a population of testicular mesenchymal stem cells (MSCs). Interestingly, Sertoli cells were not an essential component of the niche required for SSC growth.

CONCLUSION: SSC growth and maturation requires physical interaction with testicular stromal cells. Testicular MSCs are essential by providing and maintaining the stromal cells needed in this niche. These data serve as a foundation necessary for future work aimed at in vitro expansion and maturation of SSCs.

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ASSISTED REPRODUCTIVE TECHNOLOGIES - CLINICAL I

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PATIENTS QUALIFYING FOR SINGLE EMBRYO TRANSFER (SET) HAVE EXCELLENT CRYO-AUGMENTED CUMULATIVE ONGOING PREGNANCY RATES AND LOW MULTIPLE BIRTH RATES. J. Kresowik, A. Sparks, G. Ryan, E. Duran, B. Van Voorhis. Obstetrics & Gynecology, REI Division, University of Iowa, Iowa City, IA.

OBJECTIVE: We sought to evaluate fresh and frozen cycle outcomes of all patients undergoing policy SET. At our institution SET is mandatory for all women < 38 years of age with at least 7 zygotes, no history of a failed fresh cycle at our center, and at least one good quality blastocyst available for transfer.

DESIGN: Retrospective cohort study.

MATERIALS AND METHODS: Data from our program's clinical database were reviewed. We included all women undergoing policy SET in a fresh cycle and subclassified them as donor oocytes, age < 35, and age 35-37. If patients did not achieve live birth, we analyzed subsequent frozen cycle ongoing pregnancy/live birth rate (OP/LBR). During frozen embryo transfer (FET) cycles, patients were eligible for double blastocyst transfer. We calculated overall cryo-augmented OP/LBR and multiple gestations.

RESULTS: Analysis included 712 fresh transfer cycles. Nearly 92% of these patients had frozen embryos. The LBR of first fresh cycle was 63.1%. Of those not conceiving, 83% returned for FET. Patients underwent one, two, or three FETs to attempt pregnancy and achieved an OP/LBR of 64.8%, 57.6%, and 33.3% respectively in these cycles. The average number embryos transferred in FET cycles were 1.8, 2.3, and 2 respectively. The multiple gestation rate from fresh and subsequent FET cycles was 9%.

	Fresh LBR	Frozen embryos	1 FET OP/LBR	2 FET OP/LBR	3 FET OP/LBR	Cryo-augmented OP/LBR	% multiple rate of OP/LB
Donor n = 78	55%	100%	16/28	3/5	0/1	79%	21%
<35 n = 504	66%	92%	81/117	12/22	2/7	85%	8%
35-37 n = 130	58%	85%	19/34	2/3	1/1	75%	7%
ALL n = 712	63.1%	91.7%	64.8%	56.7%	33.3%	82.2%	9%

CONCLUSION: In a favorable prognosis patient group, cumulative cryo-augmented OP/LBRs are very high, despite mandatory SET in fresh cycle. Although double embryo transfer is done in subsequent FET cycles, the overall multiple gestation rate is relatively low. This information is useful in counseling patients on the significant advantages of SET when compared to the risk of not achieving a pregnancy from fresh cycle embryo cohort.

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THE NUMBER OF BABIES BORN GLOBALLY AFTER TREATMENT WITH THE ASSISTED REPRODUCTIVE TECHNOLOGIES (ART). G. D. Adamson,[a] M. Tabangin,[b] M. Macaluso,[b] J. de Mouzon.[c] [a]Fertility Physicians of Northern California, Palo Alto, CA; [b]Division of Biostatistics and Epidemiology, Cincinnati Children's Hospital Medical Center, Cincinnati, OH; [c]INSERM, Le Kremlin Bicetre, France.

OBJECTIVE: To estimate the global number of live-born infants delivered after ART treatment since Louise Brown in 1978.

DESIGN: Retrospective analysis of data from international reports.

MATERIALS AND METHODS: We reviewed 10 world reports published by the International Working Group for Registers on Assisted Reproduction (IWGROAR, 1989-2000) and the International Committee for Monitoring Assisted Reproductive Technology (ICMART, 2003-2007). Within reporting years, we used a hierarchical imputation method to address incompleteness arising from pregnancies with unknown outcomes and from non-reporting IVF programs: we used country-specific, region-wide or global data to estimate missing numbers. For many countries and reporting periods, we computed a lower and an upper boundary to allow for sensitivity analyses. We also interpolated the numbers of infants born in each country between reporting years.

RESULTS: The estimates of the number of infants born in the reporting countries increased from 11,323 in 1989 to 210,408 in 2007. Among the 74 countries that ever reported data, the cumulative number of babies born after IVF increased to 89,000-95,000 in 1990, to 887,000-999,000 in 2000, and 2.2-2.4 million (M) in 2007. An additional 1.2-1.7M are likely to have been born through 2012, bringing the total to 3.3-4.1M. This does not include at least 3 dozen non-reporting countries. Among these, China alone has been estimated from Internet and other sources to have added over 0.9 M babies.

CONCLUSION: Approximately 3.3-4.1M babies have been born after ART in the countries reporting data to the international registry. These numbers do not include countries that never reported results. The upper boundary of these estimates is compatible with the statement released by ICMART in 2012 that, accounting for missing information, 5 million babies have been born from ART. ICMART will continue to update this estimate as annual global ART data are collected and analyzed.

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AURICULAR ACUPRESSURE REDUCES ANXIETY LEVELS AND IMPROVES OUTCOMES OF IN VITRO FERTILIZATION: A PROSPECTIVE, RANDOMIZED AND CONTROLLED STUDY. F. Qu, L.-F. Xing, H. Huang. Women's Hospital, School of Medicine, Zhejiang University, Hangzhou, China.

OBJECTIVE: To explore whether auricular acupressure (AA) can relieve anxiety during the period from trans-vaginal oocyte retrieval (TVOR) to the embryo transfer (ET) in in vitro fertilization (IVF) treatment and whether AA can improve clinical pregnancy rate (CPR), implantation rate (IR), and live birth rate (LBR) of IVF.

DESIGN: A prospective, randomized and controlled study was conducted in the Reproductive Medicine Center of Zhejiang Province, China.

MATERIALS AND METHODS: 305 infertile patients with tubal blockage who were referred to our center for IVF were included. The women were randomized into a control group with 102 cases, a Sham-AA group with 102 cases and an AA group with 101 cases using a randomization chart constructed by randomizing numbers with Microsoft Excel. The anxiety levels were rated with Spielberger's State Trait Anxiety Inventory (STAI) and the Amsterdam Preoperative Anxiety and Information Scale (APAIS). Data of CPR, IR and LBR for all the women were obtained. The levels of neuropeptide Y (NPY) and transforming growth factor alpha (TGF-alpha) in the follicular fluids were detected with ELISA. The trial was registered at Australian New Zealand Clinical Trials Registry (ANZCTR) with the ACTRN 12611000899943 (http://www.ANZCTR.org.au/ACTRN12611000899943.aspx).