

Key Lectures *(Alphabetic order)*

K-1

The application of stem cell biotechnology based therapeutic in Asherman Syndrome

Akyash F¹, Aflatoonian R², Farashahi-Yazd E¹, Javidpou M¹, Golzadeh J¹, Aflatoonian A³, Eftekhari M³, Aflatoonian B^{1,4,5,*}.

1. Stem Cell Biology Research Center, Yazd Reproductive Sciences Institute, Shahid Sadoughi University of Medical Sciences, Yazd, Iran.
2. Department of Endocrinology and Female Infertility, Reproductive Biomedicine Research Center, Royan Institute for Reproductive Biomedicine, ACECR, Tehran, Iran.
3. Research and Clinical Center for Infertility, Yazd Reproductive Sciences Institute, Shahid Sadoughi University of Medical Sciences, Yazd, Iran.
4. Department of Reproductive Biology, School of Medicine, Shahid Sadoughi University of Medical Sciences, Yazd, Iran.
5. Department of Advanced Medical Sciences and Technologies, School of Paramedicine, Shahid Sadoughi University of Medical Sciences, Yazd, Iran.

Email: b.aflatoonian@ssu.ac.ir

Asherman syndrome (AS) is characterized by intrauterine adhesions (IUA) which associated with pregnancy complications and subsequent infertility disorder. Hormonal treatment and surgical operation used as traditional AS therapy. During surgery, deep damage of the basalis endometrium causes loss of stem/progenitor cells population which leading to amenorrhea, hypomenorrhea, dysmenorrhea and recurrent pregnancy loss (RPL). Nowadays, transplantation of mesenchymal stem/stromal cells (MSCs) due to their high proliferative activity, differentiation ability to various mesodermal cell lineages, immunomodulatory and secretion of the cytokines, chemokines and extracellular matrix (ECM) proteins to repair damaged tissues offer as suitable candidate to cure these patients.

Human MSCs can be derived from various sources for endometrial regeneration including, bone marrow, adipose tissue, umbilical cord, menstrual blood, amniotic membrane, human embryonic stem cells (hESCs) and endometrium.

Prianishnikov introduced endometrial stem cells (EnSCs) for the first time in 1978. Chan et al. isolated human EnSCs from endometrium in 2004. Later, in 2009 Gargett et al. extensively used the EnSCs in therapeutic applications. In our work, human endometrial tissue obtained following hysterectomy operation from a PCO woman after fully informed patient consent. Human EnMSCs were isolated, cultured and characterized, by immunofluorescence (IF) and flow cytometry techniques with CD105, CD90, CD73, CD44 and VIMENTIN as specific mesenchymal markers. Moreover, expression of CD166, CD10, CD105, VMENTIN, FIBRONECTIN, MHCI, CD14, MHCII genes was evaluated using RT-PCR. Our report confirms results of other studies regarding isolation, culture and characterization of human EnMSCs. In conclusion, endometrial tissue is a part of uterus with available source of MSCs with self-renewal and differentiation capacity that undergoes a cyclical

regeneration every month in normal women's life span. Therefore, human EnMSCs could be used for future novel therapeutic methods in regenerative medicine for treatment of uterine-factor infertile patients which can leads to RPL and finally resolve of surrogacy problems.

K-2

The use of advanced genetic technologies in the diagnosis of reproductive disorders

Al-Amri AH.

National Genetics Center, Royal Hospital, Muscat, Sultanate of Oman.

Email: ahmedamri@hotmail.com

Reproductive disorders and infertility are major health issues seen in about 10-15% of the couples. Such disorders are well-known for their impact on the social, medical, and economic burden of the individuals and the society as well. There is no doubt that, with the advances in technologies and the better understanding of the factors involved in developing these conditions, it has become possible to determine a great proportion of infertility causes in both men and women. However, still many couples receive the diagnosis of "idiopathic infertility", leaving them with uncertainty and unknown prognosis which results in performing unnecessary expensive testing and care. Moreover, the failure to precisely identify the causes has a huge impact on the families, causing a subsequent emotional instability. In fact, majority of the idiopathic infertility is due to some sort of genetic mutations, and revealing them would require special testings. Whole exome sequencing (WES) is one of the powerful techniques that allow the identification of the underlying mutations. This test is complex but it generally involves four main steps which are library preparation, sequencing, bioinformatics and Interpretation. Understanding the concept of these steps and knowing the applied strategies are important to be able to pick up the underlying mutations. Furthermore, the available up-to-date techniques, like whole genome sequencing (WGS), is something that also worth mentioning.

K-3

Platelet-rich plasma in poor responder management

Aflatoonian A.

Research and Clinical Center for Infertility, Yazd Reproductive Sciences Institute, Shahid Sadoughi University of Medical Sciences, Yazd, Iran.

Email: Abbas-aflatoonian@yahoo.com

Poor responders are defined as 9-24% of women undergoing in vitro fertilization (IVF) who do not respond well to controlled ovarian stimulation protocols. The approach and treatment of these patients is still a controversial subject in IVF. Different ovarian stimulation protocols have been assessed in terms of increase the number of oocytes retrieved and the clinical outcome. Similarly, alternative therapeutic managements such as growth hormone, testosterone, ASA and so forth

have been evaluated to improve the pregnancy outcome. Recently, Autologous (PRP) has been widely applied for numerous medical conditions without main complications. PRP is a high-concentrated of autologous human platelets comprising different kinds of adhesion molecules, cytokines, chemokines, coagulation factors, integrins, and growth factors. These elements induce cell proliferation, differentiation, and migration, angiogenesis, as well as extracellular matrix remodeling and tissue regeneration.

In the cases of ovarian failure in animals preliminary results of using Autologous platelet rich plasma (PRP) are encouraging. There are reports of successful pregnancies in cases of recurrent implantation failure and premature menopause after PRP treatment.

For the first time in human, three poor responder women with the history of IVF failure, poor oocyte and embryo quality were treated using multifocal intramedullary plus subcortical layers diffusion of PRP. The treatment resulted in two 17 and 24-week pregnant women and a successful live-birth (Sfakianoudis et al, 2019). In the same way, Farimani et al performed PRP injection into each ovary of 19 women underwent ovarian stimulation. The mean oocyte numbers was improved after PRP injection. Two women conceived spontaneous pregnancy and one case achieved clinical pregnancy and delivered a healthy baby girl (Farimani et al, 2019). A research project has been started in Yazd Reproductive Sciences Institute on 24 poor responder women and patients with premature ovarian failure (POF). The preliminary results showed Anti-Mullerian hormone elevation in two poor responder women and two POF patients after up to two PRP injections. Furthermore, one poor responder women achieved chemical pregnancy subsequent one administration of PRP. In addition one woman with ovarian failure experienced menstruation after second PRP injection. The final results may be published in future. It seems that PRP treatment could be effectively administered as an alternative successful and safe approach in the cases of poor responders due to improve ovarian infusion and functionality.

K-4

ART for infertile patient with endometriosis: A case review and a debate on optimal management

Akbari Sene A.

Shahid Akbar-abadi Hospital IVF Center, Iran University of Medical Sciences, Tehran, Iran.

Email: doctor_asturias@yahoo.com

During a case presentation session, we will have an interactive communication about the most important challenges for infertile patients with endometriosis who are candidates for ART cycles in order to reach consensus on the optimal management. Here are some practical and crucial suggestions:

- Detailed imaging evaluation replace invasive laparoscopic procedure for women with endometriosis.
- Avoiding unnecessary ovarian surgeries preserve a good ovarian reserve for ART cycles in women with endometriosis.

- Pre-treatment with GnRH-agonist before embryo transfer, improve the ART outcome for patients with endometriosis.

K-5

ART outcomes in endometrioma patients undergoing sclerotherapy versus laparoscopic cystectomy: A clinical trial

Alborzi S, Moradi Alamdarloo Sh, Poordast T, Keramati P, Shomali Z, Namavar Jahromi B, Zahiri Z, Ashraf MA.

Division of Infertility and Gynecologic Endoscopy, Department of Obstetrics and Gynecology, School of Medicine, Shiraz University of Medical Sciences, Shiraz, Iran.

Email: alborzisa@gmail.com

Background: Endometriosis is a common, chronic gynecologic condition associated with pelvic pain and infertility. Moderate to severe disease including endometrioma appears to negatively impact ART outcomes. Laparoscopic ovarian cystectomy is the treatment of choice, but it would result in reduction of ovarian reserve. Thus a less invasive procedure like ethanol sclerotherapy has been proposed before ART, but this technique may be associated with higher recurrence rate.

Objective: In this study ART outcomes and recurrence rate after these two procedures in endometrioma patients are compared.

Materials and Methods: One hundred and sixteen infertile patients with endometrioma referring to two private and university hospitals affiliated to Shiraz University of Medical Sciences, Shiraz, Iran participated in this study. The patients were divided by simple random allocation in two groups. The first group underwent ART after one year of unsuccessful spontaneous pregnancy following laparoscopic ovarian cystectomy, and the second group had ethanol sclerotherapy following oocyte retrieval. Number of oocytes, cumulative pregnancy rate, and recurrence rate of endometrioma were evaluated.

Results: Thirty patients were analyzed in the laparoscopic cystectomy group and 34 in sclerotherapy group. Fourteen patients had been excluded (not meeting inclusion criteria; n=8, no oocytes; n=6), 9 had no embryos, and 18 lost to follow-up, 11 did not return for embryo transfer in the study period. Cumulative pregnancy rate was higher in the first (33.3%) than the second group (29.4%), but it was not statistically significant (p=0.95). The rate of recurrence was 10.7% in the first group, and 16.7% in the second group, which was not statistically significant (p=0.51).

Conclusion: No significant differences were detected in ART outcomes and recurrence rate comparing laparoscopic cystectomy and sclerotherapy of endometriomas.

K-6

Effect of progesterone level in ART

Aleysin A.

Department of Infertility, Tehran University of Medical Sciences, Tehran, Iran.

Email: ivfshariat69@gmail.com

Progesterone level in late follicular phase: Studies show that the actual stimulation protocol in IVF cycle induces supra physiological estradiol during follicular phase. Despite the administration of analogues GnRH many researchers have described premature luteinization about 5-30% premature luteinization is defined as raise in serum progesterone toward the end of the follicular phase. No clear association between progesterone elevation with fertilization rate and oocyte quality has been observed. However, progesterone elevation has negative impact on endometrial receptivity and embryo implantation.

Progesterone in luteal phase: Pregnancy in FET cycle under hormonal control (HRT) is highly dependent upon serum progesterone mid luteal (Optimal progesterone of 70-99 ng/ml). Other studies show that progesterone level in day of FET does not influence on implantation. Luteal phase serum progesterone level in fresh embryo transfer is a major prerequisite for successful implantation. progesterone level is a promising diagnostic and prognostic parameter for prediction of pregnancy in ICSI-ET cycles.

K-7

Laparoscopic myomectomy, Result of baseball suture

Asefjah H.

Parsian Hospital, Tehran, Iran.

Email: hossein.asefjah@gmail.com

Uterine fibroids, myomas, or leiomyomata are smooth muscle cell tumours and are the most common benign gynaecologic tumour in women of reproductive age. They are often found as part of the investigation of an infertile couple. They are rarely found before menarche and usually regress after menopause. They are hormonally responsive, and estrogens appear to promote their growth. Local estrogen concentrations have been shown to be higher in myomas than in the surrounding myometrium, possibly because of a higher concentration of aromatase. Hormonal responsiveness appears to be greater in submucosal than subserosal myomas.

The prevalence of fibroids in the infertile population of women is controversial. According to Donnez, approximately 5-10% of women presenting with infertility are found to have one or multiple fibroids. However, when all other causes of infertility are excluded, fibroids are found in only 1-2% of the remaining women. There have been no appropriately designed studies to demonstrate a direct causal relationship between the presence of fibroids and infertility. Six systematic reviews or meta-analyses published between 2001 and 2010 assessed whether fibroids have an impact on fertility. On the whole, it appears that women with fibroids have decreased fertility. The impact of fibroid number and size on fertility has not been clearly elucidated. Reproductive success does seem to be related to fibroid location.

Subserosal fibroids do not appear to have an impact on fertility; all systematic reviews and meta-analyses agreed on this point. Submucosal fibroids have been shown uniformly to have a negative impact on rates of

implantation, clinical pregnancy, miscarriage, and live birth/ ongoing pregnancy, The greatest debate remains on the impact and treatment of intramural fibroids. Ultrasound has been shown to be an adequate, rapid, safe, and cost-effective means of evaluating the size, number, and location of fibroids. Ultrasound may, however, be suboptimal for multiple fibroids, because of acoustic shadowing, and for the proper evaluation of endometrial impingement. Interobserver variation has also been found to be greater with this technique than with MRI MRI has been well studied in the evaluation of fibroid uteruses, especially for fibroid mapping and submucosal penetration. It was shown to be the most reliable method of evaluation when compared with vaginal ultrasound, hysterosonography, and hysteroscopy, with 100% sensitivity and 91% specificity (gold standard was pathological examination). The main drawbacks of MRI evaluation are lack of accessibility and high cost.

Submucosal fibroids are managed hysteroscopically. The fibroid size should be <5 cm, although larger fibroids have been managed hysteroscopically, but repeat procedures are often necessary (III-B). The first lesson physicians must learn is that if the patient is asymptomatic, no treatment is necessary. The presence of an abdominal mass is not an indication for hysterectomy or myomectomy unless it is of significant concern to the patient. There is no universally accepted criteria regarding number and size of myoma to be removed laparoscopically but as our techniques, especially suturing techniques and instruments for laparoscopy advance, our ability to do more complicated cases of laparoscopic myomectomy increase as well, Before laparoscopic myomectomy uterine mapping is mandatory, because the surgeon does not have sense of palpation during procedure, in order to have successful laparoscopic myomectomy the surgeon should answer the following questions before surgery,

- How many myomas are there?
- Where are the exact location of myomas?
- How is the distance of myoma from cavity?
- Is uterine cavity distorted?
- Are we able to perform operation?

Laparoscopic myomectomy is a challenging procedure and the most challenging part of this procedure is suturing. The goal of suturing is to restore myometrial integrity, prevent hematoma formation, prevention of defect & dehescence in myometrium and adhesion prevention . If any one of these goals are not met during procedure the future pregnancy would be in danger. Skill of surgeon is the most important factor for successful operation. In video clip base ball myometrial closure will be demonstrated, followed by cesarian section.

K-8

Diagnosis and treatment of uterine Isthmocele

Asgari Z.

Arash Womens Hospital, Tehran University of Medical Sciences, Tehran, Iran.

Email: dr.zahra.asgari@gmail.com

Isthmocele or cesarean scar defect is born due to the overwhelmingly increasing cesarean section (CS) rates all

over the world. It was an unknown entity in the last century. Cesarean sections are and can be responsible for short- and long-term maternal and fetal morbidity, mortality, and financial issues, directly and indirectly associated to the former. Out of the many problems that are caused by CS, isthmocele is a growing surgical concern that needs attention in identifying, diagnosing, managing, and treating this problem. Currently, treatments include medical and surgical approaches. Hysteroscopy as well as laparoscopy are used in the treatment. At this article we presented its symptoms, risk factors, effects on future fertility complications, diagnostic modalities, and choice of medical or surgical treatments,

K-9

Oocyte like cells derived from embryonic stem cells (New insight in reproduction and regenerative medicine)

Bahmanpour S.

Department of Reproductive Biology and Anatomical Sciences, Shiraz University of Medical Sciences, Shiraz, Iran.

Email: bahmans@sums.ac.ir

Although the field of stem cell biology has grown rapidly, there exists considerable confusion and disagreement as to the nature of stem cells. As the definitions can be restrictive, they are useful when they provide a basis for understanding and experimental standardization. A multipotent stem cell can generate multiple types of differentiated cells, the latter being cells with distinct morphologies and gene expression patterns. Due to their unlimited source and high differentiation potential, stem cells are considered as potentially new therapeutic agents for the treatment of infertility. Stem cells could be stimulated *in vitro* to develop various numbers of specialized cells including male and female gametes suggesting their potential use in reproductive medicine. Continuation of mammalian species requires the formation and development of the sexually dimorphic germ cells. Totipotent embryonic stem (ES) cells can readily be recovered from the inner cell mass or the germinal ridge of embryos. ES cells may be ideal for creating new organs through new protocols that will circumvent the current ethical and technical minefields. Some studies show that mouse embryonic stem cells in culture can develop into oogonia that enter meiosis, recruit adjacent cells to form follicle-like structures, and later develop into blastocysts. At the egg cylinder stage of embryonic development (embryonic day (E) 6.5 in mice), a population near the epiblast can be identified as primordial germ cells (PGCs). PGCs migrate to and colonize the genital ridges, where they produce mature germ cells and generate functional adult gametes. PGCs can be isolated either prior or subsequent to their arrival in the genital ridges and, when cultured with appropriate factors *in vitro*, can generate embryonic germ (EG) cells. EG cells have many of the characteristics of ES cells with respect to their differentiation potential and their contribution to the germ line of chimeric mice. The most notable difference between ES and EG cells is that the latter may display considerable imprinting of specific

genes. In addition, stem cell-based strategies for ovarian regeneration and oocyte production have been proposed as future clinical therapies for treating infertility in women. In this case, the summarized of current knowledge and future perspectives and challenges regarding the use of stem cells in reproductive medicine is needed to be discussed.

K-10

The effect of endometrial injury on pregnancy rate in frozen-thawed embryo transfer: A randomized control trial

Baradaran Bagheri R¹, Aflatoonian A¹, Hosseiniadat R².

1. Research and Clinical Center for Infertility, Yazd Reproductive Sciences Institute, Shahid Sadoughi University of Medical Sciences, Yazd, Iran.

2. Department of Obstetrics and Gynecology, School of Medicine, Kerman University of Medical Sciences, Kerman, Iran.

Email: rameshbaradaran1350@gmail.com

Background: Implantation failure is one of the most important factors limiting success in IVF treatment. The majority of trials have demonstrated favorable effect of endometrial injury on implantation success rate especially in women with recurrent implantation failure, while some studies failed to detect any benefit.

Objective: The purpose of our trial was to explore whether endometrial injury in luteal phase prior to frozen-thawed embryo transfer cycles would improve pregnancy outcomes?

Materials and Methods: We conducted a prospective controlled trial of 93 consecutive subjects at a research and clinical center for infertility. All women were undergone frozen-thawed embryo transfer (FTE) cycles. Women in the experimental group underwent endometrial biopsy with a Pipelle catheter in luteal phase preceding FET cycle. Primary outcomes were implantation and clinical pregnancy rates and secondary outcomes were chemical, ongoing and multiple pregnancy and miscarriage rates.

Results: 45 subjects who underwent endometrial injury (EI) were compared with 48 control group which did not include any uterine manipulation. There were no significant differences in baseline and cycle characteristics between two groups. The difference in implantation rate was trend to statistically significance, 11.8% in EI group vs. 20.5% in control group ($p=0.091$). The chemical, clinical and ongoing pregnancy rates were lower in EI group compared with control group but not statistically significant. The multiple pregnancy rate and miscarriage rate also were lower in EI group compared with control group.

Conclusion: Based on results of this study, local injury to endometrium in luteal phase prior to FET cycle had a negative impact on implantation and clinical pregnancy rates.

K-11

Management of endometriosis; past, present, future

Chaichian Sh^{1,2}.

1. *Minimally Invasive Techniques Research Center in Women, Tehran Medical Sciences Branch, Islamic Azad University, Tehran, Iran.*

2. *Pars Advanced and Minimally Invasive Medical Manners Research Center, Pars Hospital, Iran University of Medical Sciences, Tehran, Iran.*

Email: shchaichian@gmail.com

According to the literature, endometriosis was first explained in Egyptian scrolls in the sixteenth century BC. But the disease remained unknown in many human societies and patients underwent inappropriate treatment from incomplete drowning to even burning at the shadow of this belief that Satan has conquered their soles. Although Carl Freiherr von Rokitansky described the scientific and histological aspects of endometriosis in 1860, the disease was considered as an ambiguous disorder along with unclear pathology till about 1.5 century later. Despite this astonishing description, mismanagement of the endometriosis, continued to the extent that even Freud misdiagnosed the individuals with the disorder as hysteric patients and tried to treat them by hypnosis in the last decades of 19th and the first decades of the 20th century.

For the first time, the term "endometriosis" was issued by Sampson in 1925. With the continuation of his studies, his brilliant paper based on retrograde menstruation theory as a cause of peritoneal endometriosis was published in 1927. He also noticed the higher incidence of endometrioid and clear cell carcinoma in the endometrioma. It seems that the number of individuals with the risk of ovarian cancer approaches to 7 times greater than the normal population for these isolated types. So far according to Nezhat, the disease can be considered as a kind of screening tool for ovarian cancer. Georg Kelling carried out the first laparoscopic surgery on dogs in 1902. In addition, Hans Christian Jacobaeus applied the approach to operate on a human being in 1910. However, the procedure was modified and popularized by some pioneer people during the next couple of decades. At that time, surgeons were performing many kinds of surgeries on these women especially oophorectomy in the hope of overcoming symptoms, and reducing the risk of infertility and ovarian cancer. Over time and in the shadow of technical and instrumental progress in laparoscopy, this technique turned to the gold standard manner for the treatment of endometriosis.

As the time passed, it was cleared that, laparoscopic surgery is not the final solution for all endometriosis patients, and even worse, in some cases induces premature ovarian insufficiency and/or poor response to ART techniques. This is far beyond internal complications and hazards every surgery carries. Simultaneously, our insight into the pathophysiology of endometriosis were deepened through large number of studies. Based on the current evidence, various etiopathological factors such as genetic, epigenetic, environmental, immunological, stem cells and/or endocrine processes are involved. So far, no specific

susceptibility genes have been identified. Endometriosis is indeed a benign disorder. The hormonal aspect of endometriosis for many years has helped physicians to treat endometriosis. However, these numerous products have their own complications and restrictions, especially at the time of pregnancy desire and in menopausal patients.

In recent years, researchers have founded more sophisticated etiopathogeneses for the disease. According to a recent study, extrauterine stem cells originating from bone marrow are able to differentiate into endometriotic tissues. Despite confirmation of the role of molecular medicine as a new method for clarifying the pathogenesis of endometriosis, Sampson's implantation theory has never been discarded. Jones in 1998 indicated that the women with endometriosis reveal upregulation of the antiapoptotic gene BCL-2 in eutopic and ectopic endometrium. Moreover, he postulated that genetic alterations of endometrial cells which impact their tendency to implant may be hereditary. Linkage analysis done by Treloar in 2005 has revealed candidate genes with biological plausibility, as well.

The hypothesis of endometriosis as a pelvic inflammatory condition is accepted. In these patients, the peritoneal fluid and the number of activated macrophages are increased significantly. There is also a great variation in the cytokine/chemokine profile. Macrophages can produce and secrete various biologically active elements (i.e. cytokines, plasma proteins, coagulation and fibrinolytic agents, enzymes components of complement, and lipids). A unique protein similar to haptoglobin has been discovered by proteomics method in the peritoneal fluid of endometriosis patients. This protein decreases macrophage phagocytic activities after bounding process and enhances production of IL-6. Macrophage migration inhibitory factor, TNF- α , IL-1 β , IL6, and IL-8, and monocytes are in the peritoneal fluid of endometriosis patients. Integrin and E-cadherin and MMPs namely ICAM (Intracellular adhesion molecules) have been discovered in cells of menstrual effluent, endometrium, peritoneal fluid, peritoneum, and endometriotic cells. Langendonck in 2002 showed a significant role of hemoglobin in the pathogenesis of peritoneal endometriosis. This hypothesis confirms that, after RBC lysis, the resultant released hemoglobin in the peritoneal cavity induces activation of cell adhesion molecules, cytokine production, cell proliferation, and neovascularization. Hemoglobin molecules degradation to its product, heme, Iron, biliverdin and bilirubin may lead to oxidativ stress.

Despite these and other progresses in understanding the pathophysiology of this enigmatic disease and many compounds that their effectiveness has been postulated in several clinical trials, some, as gynecologic laparoscopists are continuing to transfer their habitual concepts that laparoscopy is the best treatment for endometriosis. With regard to ACOG guideline and much more scientific evidence, now it is the time to treat this malefic disease based on these causative etiologies and reserve surgery for the most important occasion during patient's lifetime.

K-12

Effects of Chlamydia trachomatis infection on sperm chromatin condensation and DNA integrity

Dehghan Marvast L^{1, 2}, Talebi AR², Ghasemzadeh J², Hosseini A², Pacey AA¹.

1. Department of Human Metabolism, Academic Unit of Reproductive and Developmental Medicine, The University of Sheffield, Sheffield, UK.

2. Research and Clinical Centre for Infertility, Yazd Reproductive Sciences Institute, Shahid Sadoughi University of Medical Sciences, Yazd, Iran.

Email: laleh.dm236@gmail.com

The present study was performed to investigate the relation of Chlamydia trachomatis infection to sperm chromatin/DNA integrity in a population of infertile men (male partner of infertile couples) from Iran. Blood, semen and first-void urine samples were obtained from 250 infertile men. Data were analysed with regard to the results of (i) serological analysis for specific antibodies to C. trachomatis in serum; (ii) the presence of C. trachomatis and DNA in first-void urine; and (iii) in the semen sample of the male partner, in addition to sperm analysis, four different tests (aniline blue, chromomycin A3, acridine orange and TUNEL) were used to detect sperm chromatin and DNA abnormalities. The main conclusions of the results were: (i) no evidence of C. trachomatis infection in semen samples was found; (ii) sperm DNA fragmentation and chromatin studies were not correlated with C. trachomatis diagnosis; (iii) the percentage of DNA fragmentation is positively correlated with the percentage of immotile sperm but negatively with semen volume, normal morphology; and (iv) in sperm chromatin evaluations, only the percentage of chromatin protamination was related to male age.

K-13

Does Time lapse monitoring (TLM) improve assisted reproductive outcomes?

Faramarzi A^{1,2}, Khalili MA².

1. Fertility and Infertility Research Center, Kermanshah University of Medical Sciences, Kermanshah, Iran.

2. Research and Clinical Center for Infertility, Yazd Reproductive Sciences Institute, Shahid Sadoughi University of Medical Sciences, Yazd, Iran.

Email: a.faramarzi90@gmail.com

High implantation rates following ART cycles are achieved via the transfer of embryos with the highest developmental competence. Multiple pregnancies as a result of the transfer of several embryos per cycle accompany with various complication. Therefore, single-embryo transfer (SET) is the preferred practice in ART program to avoid various complications of multiple pregnancies. For improving the pregnancy rates for SET, the embryologists need reliable biomarkers to aid their selection of embryos with the highest developmental potential. Embryo conventional morphological assessment limited to discrete snapshots once a day. Also, it exposes the embryos to suboptimal conditions regarding to pH, light and temperature outside the

incubators. Continuous observation of embryos via time-lapse monitoring (TLM) provides detailed information on embryo morphokinetics with stable culture condition. Moreover, TLM may introduce new dynamic parameters of embryo developmental potential as well as a versatile embryo assessment and provides new information regarding human embryo competence. Recently, several algorithms have been developed associated with the morphokinetics of early embryo development to formation of blastocyst, implantation potential, chromosomal content, pregnancy and live birth rates. We found that oocyte quality such as morphology, morphometry, presence of the meiotic spindle (MS), zona pellucida (ZP) birefringence and maturity might affect on the generated embryo morphokinetics. But, pronuclear pattern of fertilized oocytes was not related to embryo morphokinetics. However, there is not yet a universally consensus about them and there are significant knowledge gaps that can give opportunities for further studies in this field.

K-14

Fertility preservation in patients with cancer

Ghaffari F.

Department of Endocrinology and Female Infertility, Reproductive Biomedicine Research Center, Royan Institute for Reproductive Biomedicine, ACECR, Tehran, Iran.

Email: ghafaryf@yahoo.com

Approximately 10% of female cancer cases occur under the age of 45 years. Improvements cancer treatments, cancer patients living longer. The treatment for most of the cancer types in reproductive-age women involves removal of the reproductive organs or chemotherapy and/or radiotherapy that affect reproductive function. Advances in reproductive medicine now allow patients diagnosed with cancer during their reproductive years to undergo various fertility preservations treatments. Women of reproductive age who are scheduled to undergo cancer treatment that could lead to premature decline of ovarian function should be counseled regarding the possibility of oocyte or embryo cryopreservation.

These include: embryo cryopreservation, oocyte cryopreservation, cortical and whole ovary cryopreservation, ovarian transplantation, ovarian transposition, GnRH agonist protection. Patients be counseled all Fertility Preservation methods applicable to their specific circumstance. Ideally, this counseling should be performed by a physician specializing in reproductive endocrinology and infertility who has experience working with cancer patients. The most preferred methods for fertility preservation in cancer patients is Embryo cryopreservation and Mature oocyte cryopreservation, owing to its higher success rates compared with other, more experimental technologies. Therefore, it should be recommended as long as the patient's medical condition does not preclude safely performing COS or oocyte retrieval and the patient has adequate time to undergo COS and oocyte retrieval. Ovarian transposition prior to radiation and treatment with GnRHα coincident with gonadotoxic chemotherapy

are relatively simple, inexpensive, and readily available. This methods are experimental. Ovarian tissue cryopreservation is an option for patients who require immediate gonadotrophic treatment of malignancy. This procedure can be used when there is insufficient time to conduct ovulation induction, in patients with genetic mutations with a high risk for premature ovarian failure, in prepubertal girls or in women who have hormone-sensitive malignancies. prophylactically cryopreserve ovarian tissue can be used for hematopoietic stem cell transplantation for the treatment of benign hematologic diseases or autoimmune diseases that haven't respond to immunosuppressive treatment. blood-borne cancers have the highest of risk for malignancy following transplantation of ovarian tissue. whereas metastatic disease was less common in most other cancers. ovarian tissue transplantation is not recommended for patients with blood-borne malignancies or malignancies that metastasize to the ovary.

K-15

Fifteen years experiences of genetic evaluation of recurrent pregnancy loss and infertility

Ghasemi N, Kalantar SM, Dehghani MR, Sheikhha MH.

Abortion Research Center, Yazd Reproductive Sciences Institute, Shahid Sadoughi University of Medical Sciences, Yazd, Iran.

Email: n479g@yahoo.co.uk

Recurrent pregnancy loss (RPL) or recurrent IVF failure (RIF) and infertility are the main reasons for having no child. Recurrent pregnancy loss is an important reproductive health issue, affecting 2-5% of couples. The prevalence of infertility is between 7-15%. Among all causes of RPL and infertility, there is evidence to suggest contribution of genetic factors. The genetic causes of infertility are varied and include chromosomal abnormalities, single gene disorders and problems come with multifactorial inheritance. Some genetic factors influence males or females specifically, and others bring about both males and females affect fetuses. Several studies have been done in the field of reproductive genetic specifically in RPL and infertility considering both prevention and treatment in Yazd Abortion Research Center of Reproductive Sciences Institute. Evaluation of chromosomal abnormalities was performed in this center since 2001, perinatal diagnosis (PND) since 2013 and preimplantation genetic diagnosis (PGD) since 2015.

Single genes mutation or polymorphisms were evaluated in some studies according to previous evidence of genetic problems in RPL or RIF. Fifteen years' experience in this center were indicated that the frequency of the structural and numerical abnormalities in patients referring to this center are similar the other countries. Monogenic problem, in female with thrombophilia and male with AZF microdeletions indicated that they are the most important known causes. Fortunately, treatment with aspirin or aspirin plus heparin and hormonal therapy with PND or PGS were successfully raised normal child birth to 80% of

RPL cases. Frequency of the chromosomal abnormalities and the single-gene disorders in RPL and infertility or RIF, strongly suggested genetic counseling for success treatment.

K-16

Impact of intra-cytoplasmic morphologically selected sperm injection on ART outcome

Ghazali Sh¹, Khalili MA², Talebi AR².

1. Islamic Azad University, Sanandaj Branch, Sanandaj, Iran.

2. Research and Clinical Center for Infertility, Yazd Reproductive Sciences Institute, Shahid Sadoughi University of Medical Sciences, Yazd, Iran.

Email: sh_ghazali@yahoo.com

Selecting best spermatozoa with good morphology during intra-cytoplasmic sperm injection (ICSI) is an important issue. In recent seventeen years improving ICSI using high magnification (x6000) results in establishing intra-cytoplasmic morphologically selected sperm injection (IMSI) method utilizing Nomarsky optic system for real time selecting free vacuole spermatozoa. Therefore, new criterion, motile sperm organelle morphology sperm examination (MSOME), had been developed to identify high quality spermatozoa based on cephalic vacuole and its number and size, head shape and size, acrosome integrity and irregularity of mid-piece under high magnification prior to oocyte injection. Some investigations claim that IMSI can improve embryo quality, implantation rate and pregnancy rate, and decrease miscarriage rate especially in patients with ART failure. Different types of opposition opinion have been emerged about the clinically advantages of IMSI, but almost believe that severe male factor and repeated implantation failure (RIF) patients could benefit from this method. Time consuming, high budget necessity and high skill for performing IMSI are the challenging items should be mentioned.

K-17

Cleavage-stage embryo micromanipulation in the ART program

Halvaei I.

Department of Anatomical Sciences, Faculty of Medical Sciences, Tarbiat Modares University, Tehran, Iran.

Email: ihalvaei@modares.ac.ir

Embryo micromanipulation is one of the routine tasks which performs at any ART clinic worldwide. Embryo micromanipulation was developed specially after introduction of intracytoplasmic sperm injection. Maybe more micromanipulations are performed at cleavage-stage embryos. A wide range of techniques belongs to embryo micromanipulation including assisted hatching, removing detrimental components from healthy embryos, blastomere biopsy, and embryo splitting. The main question may raise is when these techniques should be used and which side effects may they have. Besides, the safety of micromanipulation techniques should be taken into account.

K-18

Potential of human twin embryos generated by embryo splitting in assisted reproduction and research

Ilic D.

King's College London, Faculty of Life Sciences and Medicine, London, UK.

Email: duscko.ilic@kcl.ac.uk

Embryo splitting has been introduced successfully to the veterinary medicine several decades ago and today is a part of standard practice. A very limited number of studies have been conducted in humans and non-human primates. The published material, especially studies with human embryos, is controversial. Some reports suggest that twinning technology will find clinical use in reproductive medicine in the future, whereas others conclude the opposite, that human twin embryos created in vitro are unsuitable not only for clinical but also for research purposes. In this talk, I will present an overview of embryo splitting experiments in humans and non-human primates and discuss the potential of this technology in assisted reproduction and research.

K-19

Obstetrics and perinatal complications in Frozen Embryo transfer Versus Fresh transfer

Karimzadeh MA.

Madar Hospital, Yazd, Iran.

Email: makarimzadeh@yahoo.com

The aim of this presentation is to compare the pregnancy and neonate outcome after fresh and frozen-thawed embryo transfer in the assisted reproductive technology (ART). The study focused on clinical and laboratory factors affecting the abnormal placentation, especially placenta Previa, in patients conceiving in the IVF program. The available evidences confirm that neonates born after frozen thawed embryo transfer had significantly higher mean birth weight than after fresh embryo transfer (ET). Moreover, the birth weight distribution in singletons was found to shift towards "large for gestation" (LGA) after frozen-thawed ET. On the other hand, the pregnancies after fresh ET were characterized by a higher incidence of placenta praevia and 3rd trimester bleeding. Placenta praevia was more common in IVF patients with fresh ET in a stimulated cycle than in patients with ET in a natural cycle. An important issue arose of how the ovarian hormonal stimulation relates to abnormal placentation and if the serum hormone levels interfere with in the IVF treatment results. There is higher risk of placenta praevia in patients with endometriosis and in those that transfer the embryo in prepared endometrium for FET or Fresh compared with the natural cycles, though what causes this risk remains largely unknown. The animal studies showed that impaired vasculogenesis early in gestation may lead to underdevelopment of placental vessels and cardiovascular development and developmental programming problems, such as intrauterine growth

restriction of the fetus, resulting in long-term health consequences for the offspring, such as cardiovascular diseases that is higher in ART cycles.

K-20

Adenomyosis: a neglected disease

Mehdizadeh A.

Iran University of Medical Sciences, Tehran, Iran.

Email: amehdizadahkashi@yahoo.com

Adenomyosis is a heterogenous gynecologic condition. Patients with adenomyosis can have a range of clinical presentations. The most common presentation of adenomyosis is heavy menstrual bleeding and dysmenorrhea; however, patients can also be asymptomatic. Currently, there are no standard diagnostic imaging criteria, and choosing the optimal treatment for patients is challenging. Treatment including Medical and Surgical. The medications most commonly used to treat bleeding and pain in adenomyosis are hormonal treatments that induce an endometrial atrophy either by a local action (levonorgestrel intrauterine system) or by a systemic action both on the endometrium and on the hypothalamic-pituitary-ovarian axis (progestogens, danazol, GnRH agonists). Adenomyosis is characterized by a decreased expression of progesterone receptors A and B in ectopic endometrial lesions, possibly related to epigenetic changes. This progesterone resistance in adenomyosis could potentially lead to an abnormal expression of progesterone receptor-related genes, to a reduced expression of implantation-related genes, and to a resistance to progestogens treatment.

In symptomatic young women desiring to conceive, the concept of conservative, uterine-sparing surgery for adenomyosis is acquiring more and more consensus; nevertheless, conservative surgery has not become the standard treatment for adenomyosis yet. This is mainly because adenomyotic tissue invades the uterine muscle layer with unclear borders, determining the absence of a surgical cleavage plane, so complete excision of the affected area remains inaccurate and often causes heavy blood loss. Moreover, the excision of adenomyotic tissue is always accompanied by excision of myometrium, so it is partly destructive for the uterine wall. Therefore, the advantages of removing an affected area must be balanced against the disadvantages of leaving a possibly defective uterine wall.

In 1952 the term "hysteroplasty" was used to describe uterine-sparing surgery in young women with extensive adenomyosis. The currently available uterine-preserving surgical options for adenomyosis could be classified as complete excision of adenomyosis (preferably used in case of localized adenomyosis), debulking surgery/partial adenomyomectomy (preferably used in case of diffuse adenomyosis), and nonexcisional techniques, used when removal of adenomyotic tissue is not included (i.e., uterine artery ligation, electrocoagulation of myometrium, hysteroscopic and nonhysteroscopic endometrial resection/ablation). A recent review concluded that uterine-sparing surgery for adenomyosis appears to be feasible and satisfactory: After complete

excision, the dysmenorrhea reduction, menorrhagia control, and pregnancy rates were found to be 82.0%, 68.8%, and 60.5%, respectively. After partial excision, the dysmenorrhea reduction, menorrhagia control, and pregnancy rates were 81.8%, 50.0%, and 46.9%, respectively.

K-21

The webs we don't know we weave": Sex therapy protocol in infertility settings

Merghati Khoei ES.

Tehran University of Medical Sciences, Tehran, Iran.

Email: effat_mer@tums.ac.ir, effat_mer@yahoo.com

Sexual problems in a couple with infertility are a complex area of specialty. Reciprocal association between sexual problems (SP) and infertility has argued worldwide. Infertility has a negative impact on the sexual relationship of infertile couples. The adverse impact is essentially associated with the sexual self-concept of infertile individuals and their partners. Sexual self-concept is manipulated through the treatments an infertile couple receives. Evaluation process and treatments are offered in infertility clinics influences the ways couples organize their intimacy, particularly their sexual life. Communication between partners is altered and affectionate exchanges lose their spontaneity. Consequently, sexual function is affected by infertility due to limited or avoided sexual relationships. Risk of sexual dysfunctions, primary vs secondary to infertility is high (OR 9.53; 2.27-40.01). In infertile men, low sexual desire and lack of sexual satisfaction are the most prevalent types of sexual dysfunction, ranging from 8.9-68.7%. 1/6 and 1/10 infertile men experience erectile dysfunction and/or premature ejaculation, and orgasmic problem respectively. Women with infertility have a high rate of sexual dysfunction compared with men; 65% with primary and 77% with secondary infertility. Genitopelvic pain/penetration disorder (GPPPD) and low sexual desire (32%) are commonly diagnosed in women with infertility issues.

In sum, sexual difficulties often remain or get worse after treatment ends successfully or unsuccessfully. Often, health providers and clinicians ignore or restrain the SPs of infertile couples. Lack of consultative support from specialized sex therapist alters the quality of care in infertility settings. One of the main reasons for this gap can be the question that "how should we take care of these couples through their sexual lives?" Adoption of a patient-centered framework for evaluation and treatment is acknowledged as the essential concepts underlying the management of sexual problems in infertility clinics. Consideration of treatment principles is the second seminal concept. After complete assessment, the first step in the management is case formulation. Understanding the contribution of each partner to the problem is vital second step, a sex therapist must take to achieve the goal of balancing partners. Determination of treatment options is the third step. Treatments for SP can be broadly classified into general and specific approaches.

Sex education and relaxation exercises are utilized as the general approach. The specific approach can be either pharmacological or non-pharmacological or a combination of both. The final step is the selection of treatment which is accorded to patient/couple's choice. Follow-up assignments enable the therapist to evaluate the treatment effectiveness and its impact on infertility treatment plan.

K-22

Epigenomic study of early mammalian development

Na J.

School of Medicine, Tsinghua University, Beijing, China

Email: jie.na@tsinghua.edu.cn

Histone modifications are fundamental epigenetic regulators that dictate stem cell potency. We surveyed the landscape of H3K4me3, a histone hallmark for transcription initiation, from developing gametes to post-implantation embryos using a highly sensitive approach, STAR ChIP-seq. Upon fertilization, extensive reprogramming occurs on the paternal genome, as H3K4me3 peaks are depleted in zygotes but are readily observed after major zygotic genome activation at the late two-cell stage. On the maternal genome, we found a non-canonical form of H3K4me3 (ncH3K4me3) in full-grown and mature oocytes, which exists as broad peaks at promoters and a large number of distal loci. Such broad H3K4me3 peaks are in contrast to the typical sharp H3K4me3 peaks of promoters. The ncH3K4me3 in oocytes then is inherited in zygotes and early 2-cell embryos, before being erased in the late 2-cell embryos, when canonical H3K4me3 starts to be established. Interestingly, downregulation of H3K4me3 in full-grown oocytes by overexpression of the H3K4me3 demethylase KDM5b lead to reactivation of transcription in GV oocytes.

We also investigated chromatin landscape during human embryo preimplantation development, using an improved assay for transposase-accessible chromatin with high-throughput sequencing (ATAC-seq). We find widespread accessible chromatin regions in early human embryos that overlap extensively with putative cis-regulatory sequences and transposable elements. There were both conservation and difference in developmentally significant transcription factor expression in human and mouse preimplantation embryos. In addition, we find widespread open chromatin regions before zygotic genome activation (ZGA), many reside in distal regions that are enriched for transcription factor-binding sites. A large portion of these regions then become inaccessible after ZGA in a transcription-dependent manner. Such extensive chromatin reorganization during ZGA is conserved in mice and correlates with the reprogramming of ncH3K4me3. Taken together, these data not only reveal a conserved principle that underlies the chromatin transition during mammalian ZGA, but also help to advance our understanding of epigenetic reprogramming during human early development and in vitro fertilization.

K-23

Assisted reproduction and sperm DNA integrity, how can we obtain sperm with better DNA quality?

Narimani N.

Research and Clinical Center for Infertility, Yazd Reproductive Sciences Institute, Shahid Sadoughi University of Medical Sciences, Yazd, Iran.

Email: Nima_dr2001@yahoo.com

Male fertility potential is assessed traditionally with conventional semen analysis, but recently, with growing popularity of assisted reproduction techniques (ART) sperm DNA integrity gained increasing attention, since it may better predict the chance for spontaneous pregnancy and ART outcome. A proportion of apparently normospermic but infertile men, show to have abnormal levels of sperm DNA fragmentation. Studies found that an increased sperm DNA fragmentation index (DFI) leads to less spontaneous pregnancies and lower ART results. DFI-measurement has been recommended in unexplained infertility, recurrent abortion and in ART failure. In this mini review we briefly describe sperm chromatin structure, mechanisms of sperm DNA injury and various type of sperm DFI assays. In addition, we have categorized our strategies for obtaining sperm with better quality into three groups based on the moment of sampling.

K-24

LED irradiation and male gametogenesis

Nematollahi-mahani SN.

Department of Anatomy, Afzalipour School of Medicine, Kerman University of Medical Sciences, Kerman, Iran.

Email: nnematollahi@kmu.ac.ir

Physical factors including light of different wavelengths could contribute in various molecular phenomenon in the body. Among different types of light, light emitting diodes (LED) have received special attention, having in mind the simple apparatus which is needed to produce LED irradiation and their narrow wavelength. Studies on different tissues including bone, cartilage, hematopoietic cells and nervous cells have shown the proliferative and differentiation capacity of LED on these type of stem cells. However, the intensity of the LED irradiation and the wavelength have been resulted in different and sometimes paradoxical results. The effects of light on gametogenic differentiation of stem cells from various sources have to be cleared through future researches. In the current presentation I will discuss this issue and present the recent findings on the impact of LED on male gametogenesis.

K-25

Reproductive medicine in female reproductive system

Nikukar H^{1,2}.

1. Medical Nanotechnology and Tissue Engineering Research Center, Yazd Reproductive Sciences Institute, Shahid Sadoughi University of Medical Sciences, Yazd, Iran.

2. Department of Advanced Medical Sciences and Technologies, School of Paramedicine, Shahid Sadoughi University of Medical Sciences, Yazd, Iran.

Email: habibnik@ssu.ac.ir

Repair, regeneration and replacement of organs and tissues in an augmented mode are the main goal of regenerative medicine. In reproductive system, regeneration of female and male genital tissues is a hope for fateful treatment of infertility in special cases. Main female genital organs are programmed to save, carry, fertilize, implant and grow germinal cells and fertilized egg/eggs toward a mature healthy neonate. Any interruption in this complex pathway could introduce infertility. Tissue engineering is trying to treat special situations of infertility with abnormal or absent the tissue or related organs by a wonderwork between cells, scaffolds and growth factors. This is a collaborative art between cell biologists, medical doctors and engineers by application of advanced recent technology.

K-26

Surgical or non-surgical treatment of uterine fibroids? That's the question

Rahimi MA.

Chatswood Medical Center, Chatswood, Sydney, Australia.

Email: drmahmoud.rahimi@gmail.com

Women with uterine fibroids (Myomas) have a stronger desire for non-surgical treatment of their fibroids to preserve their uterus and potential fertility. What are the pros and cons of such non-surgical treatments and what effects do these treatments have on the patient's long-term prognosis? How do we, as gynaecologists, treat patients with such requests? What about the lack of histology and risk of fibrosarcoma? What are the questions, answers, and controversies to non-surgical treatments of uterine fibroids?

The most common benign tumour of the uterus are Fibroids. Studies have shown that uterine fibroids are prevalent in 50% of women at the reproductive age and the cumulative incidence of fibroids by the age of 50 are 70% in white women and >80% for African Americans. Usually the symptomatic patients seek treatment. The current treatment options are; medical and minimally invasive surgical (Laparoscopy myomectomy), and non-surgical procedures.

The non-surgical procedures consist of:

➤MRgFUS (Magnetic resonance guided focused ultrasound): using ultrasound energy to heat and destroy the fibroids.

➤UAE (Uterine artery embolization): where an interventional radiologist uses a catheter to deliver small particles that block the blood supply to the uterine body.

These treatment options, and their complications and controversies, will be discussed in full at the congress with attendees.

K-27

Imaging findings in Adenomyosis (MRI and Ultrasound)

Rasekhi AR.

Department of Radiology, Shiraz University of Medical Sciences, Shiraz, Iran.

Email: dr.alirezarasekhi@gmail.com

Uterine adenomyosis is a common gynecologic condition that is characterized by the presence of heterotopic endometrial glands and stroma in the myometrium with adjacent smooth muscle hyperplasia. The histopathologic features of adenomyosis are varied and contribute to its imaging appearance. The accompanying smooth muscle hyperplasia produces the typical gross appearance of adenomyosis and corresponds to areas of decreased echogenicity at endovaginal ultrasonography (US) and areas of decreased signal intensity at magnetic resonance (MR) imaging. Endovaginal US also shows heterogeneity of the myometrial echotexture, which corresponds to small echogenic islands of heterotopic endometrial tissue surrounded by the hypoechoic smooth muscle.

On T2-weighted MR images, bright foci are seen in areas of abnormal low signal intensity within the myometrium in approximately 50% of patients. These foci correspond to islands of heterotopic endometrial tissue, cystic dilatation of heterotopic glands, or hemorrhagic foci. With the advent of high-resolution imaging techniques, signs associated with the presence of heterotopic endometrial tissue are being detected with increasing frequency. These signs include myometrial cysts, myometrial nodules, linear striations, pseudowidening of the endometrium, and poor definition of the endomyometrial junction. Pitfalls in diagnosis of uterine adenomyosis include leiomyoma, endometrial carcinoma, myometrial contractions, and muscular hypertrophy.

K-28

Usage of human endometrial mesenchymal stem cells in in vitro implantation model

Salehnia M, Rahimipour M.

Department of Anatomy, Faculty of Medical Sciences, Tarbiat Modares University, Tehran, Iran.

Email: mogdeh@dr.com; Salehnia@modares.ac.ir

To establish an in vitro implantation model using endometrial W5C5+ mesenchymal stem cells and mouse blastocysts as a surrogate embryo this study was designed. The W5C5+ mesenchymal stem cells were isolated from human endometrial cell suspension (ECS) at 4th passage by magnetic activated cell sorting method. ECS and W5C5+ cells were separately co-cultured with human myometrial muscle cells for five days. After collection of mouse blastocysts, the embryos were transferred on the top of co-cultured cells for 48 hrs. The interaction of embryo and cultured cells was assessed by morphological and ultrastructural evaluations and analysis of the expression of genes related to implantation (α V and β 3 integrins,

interleukin-1 receptor; IL-1R, leukemia inhibitory factor; LIF and leukemia inhibitory factor receptor; LIFR). The micrographs showed trophoblastic cells were outgrowth around the embryonic cells and they attached to the ECS and W5C5+ cells. Ultrastructural observation revealed the appearance of pinopode and microvilli-like structures on the surfaces of both ECS and W5C5+ endometrial cells.

In addition the embryos were developed to egg-cylinder stage in two groups. The analysis of genes expression showed no significant differences in genes expression in both groups in the presence of embryo but an increased expression of α V was detected in W5C5+ cells in comparison with ECS cells in the absence of embryo. Thus, this study showed W5C5+ cells during co-culture with smooth muscle cells could be differentiated to epithelial-like cells and interacted with mouse embryo and this co-cultured cells has a potential as an implantation model.

K-29

Using human induced pluripotent stem cells (hiPSCs) as a new therapeutic agent for the treatment of infertility: Current progress, future perspectives and challenges

Shahsavarani H^{1,2}.

1.Lab of Regenerative Medicine and Biomedical Innovations, Pasteur Institute of Iran, Tehran, Iran.

2.Department of Cellular and Molecular Sciences, Shahid Beheshti University, Tehran, Iran.

Email: hosein.shahsavarani@gmail.com

Infertility is a global public health concern with a high prevalence in around 15% of couples at reproductive age. The successful generation of induced pluripotent stem (iPS) cells from somatic cells brought about new hope for regenerative therapies including fertility and genetic disorders. Due to their few ethical issues, abundance of the sources and high differentiation potential, induced pluripotent stem cells are recently has attracted high attentions as potentially new therapeutic agents for the treatment of infertility. These type of stem cells could be stimulated in vitro to differentiate into almost all kinds of cells including male and female gametes suggesting their potential use in reproductive medicine. Moreover, it just surprisingly report of making sperm from female cells or eggs from male cells in mammals using CRISPR technology have revolutionized this field of research. Here, we summarize recent developments, challenges and future prospective towards treating infertility and genetic disease with eggs and sperm using induced pluripotent stem cells. Additionally, the barrier to the development of aforementioned techniques for making healthy viable and fertile mature individuals without any chromosome disorders will be discussed.

K-30

Knowledge of fertility decline in women via natural pregnancy and Assisted Reproductive Technologies

Lucas L, Roseario R, Shelling A.

Department of Obstetrics and Gynaecology, Faculty of Medical and Health Sciences, University of Auckland, Auckland, New Zealand.

Email: a.shelling@auckland.ac.nz

Female fertility declines with age. University students are the group of people most likely to postpone parenthood, yet several international studies have shown that they overestimate their fertility. We designed a questionnaire based on a previous study where University students were asked to answer questions related to their awareness of fertility decline in spontaneous and IVF pregnancies, and methods they considered would prolong their reproductive lifespan. Our study has shown that University students overestimated the rates of pregnancy for both spontaneous natural and IVF pregnancies. Students are mainly aware of the availability of Assisted Reproductive Technologies, however overestimate their effectiveness. Few students mentioned non-medical or well-being initiatives as measures to prolong parenthood. It is important University students are aware of the rate of fertility decline in women, as although Assisted Reproductive Technologies can be effective at times, they are not a guaranteed solution to an aging woman's fertility. University students, like other cohorts, demonstrated an overestimation regarding the chances of a female's pregnancy and predicted the fertility decline to occur much later than it does in reality.

K-31

Thin endometrium and treatment modalities

Taheripناه R.

Infertility and Reproductive Health Research Center, Shahid Beheshti University of Medical Sciences, Tehran, Iran.

Email: taheripناهf@gmail.com

Endometrium is one of the most important factors in assisted reproductive technology cycles. Thin endometrium with frequency of 2.4% in assisted reproductive technology cycles is associated with lower implantation and pregnancy rate. The appropriate endometrium is 7-10 mm for embryo transfer. Inflammatory, iatrogenic and idiopathic causes lead to thin endometrium. Thin endometrium is characterized by poor growth of glandular epithelium, high uterine blood impedance and low VEGF that causes of poor vascular development and further decrease of blood flow. Chronic endometritis with destroy of the estrogen receptors has a crucial role in refractory endometrium to hormonal preparing. Different modalities were explained to improve the endometrial growth and higher thickness such as intrauterine injection of G-CSF, extended higher dose of estragon support, adding the low dose hCG during the hormonal preparing for frozen embryo transfer. Other modalities such as intrauterine autologous platelet rich plasma infusion, luteal phase support with GnRH agonist was reported but none of them have been substantiated. Recently, intrauterine administration of bone marrow stem cell,

progenitor cells was reported from the IVI center but their efficacy is unknown. Also, Sildenafil, tocopherol, aspirin, and Larginine, pentoxifylline have been used to improve endometrial vascularity in unresponsive endometrium. In addition to lower implantation, the process of invasion may be hindered due to the lack of an adequate endometrial bed and increases the placenta accrete rate. Bone marrow mesenchymal stem cells (BMSCs) improved endometrium thickness, probably via their migration and immunomodulatory. The best protocol for embryo transfer in thin endometrium cases is the FET due to better endometrial thickness without fair of the embryo destroys.

K-32

Marine stem cells: A new approach for human reproductive tissue regeneration

Tamadon A.

Persian Gulf Marine Biotechnology Research Center, The Persian Gulf Biomedical Sciences Research Institute, Bushehr University of Medical Sciences, Bushehr, Iran.

Email: amintamadon@yahoo.com

Seventy-five percentage of earth's surface is covered by water and is habitat of countless and diverse resource of marine invertebrates. Such as animals living on land, marine invertebrates' reproduction, development and regeneration are dependent on their stem cells. Marine invertebrates constitute the largest biodiversity from morphologically simple organisms (e.g., cnidarians, sponges), to the more complex echinoderms, mollusks, protochordates, and crustaceans. They all have marine invertebrate stem cells (MISC) produce a large number of novel bioactive-molecules, many of which are of significant potential interest for human reproductive regeneration. In the current decade, the positive effect of extracts of some marine invertebrates (e.g., sea cucumber) have been studied in mammalian reproduction. On the other hand, marine drugs extracted from marine invertebrates (e.g., sponges, mollusks, soft corals), acting on microfilaments and microtubules, have a wide range of impacts on reproductive events (e.g., sperm maturation and motility, oocyte maturation, fertilization, and early embryo development). MISCs have been detected or cultured in some marine invertebrates (e.g., cnidarians, crustaceans). Regenerative or proliferative effects of activated MISCs have been also shown on the other tissues (e.g., bone, skin). Merging the available evidences can speculatively suggest the MISCs of marine invertebrates can be applied for reproductive organ (e.g., ovary, uterine, cervix, and testis) engineering or tissue (e.g., endometrium, seminiferous tubules) regeneration. By the way limitations such as different factors in MISC culture in comparison with mammalian cell culture and low number of available antibodies for MISC characterization may affect the progress of this approach for future research. In conclusion, large biodiversity of marine invertebrates living in Persian Gulf, Oman Sea, and Caspian Sea open a new window for future studies can be focused on the application of MISCs for reproductive tissue engineering.

K-33

Effects of the hydroalcoholic extract of the *Psidium guajava* fruit on osteoporosis prevention in ovariectomized rats

Diyanat S¹, Salehi M¹, Koohi-Hosseinabadi O², Tanideh N^{3,4}, Dehghani F⁵, Koohpeyma F⁶, Daneshi S³.

1. School of Nutrition and Food Science, Shiraz University of Medical Sciences, Shiraz, Iran.
2. Center of Comparative and Experimental Medicine, Shiraz University of Medical Sciences, Shiraz, Iran.
3. Stem Cells Technology Research Center, Shiraz University of Medical Sciences, Shiraz, Iran.
4. Department of Pharmacology, School of Medicine, Shiraz University of Medical Sciences, Shiraz, Iran.
5. Histomorphometry and Stereology Research Center, Shiraz University of Medical Sciences, Shiraz, Iran.
6. Department of Endocrinology, Endocrinology and Metabolism Research Center, Shiraz University of Medical Sciences, Shiraz, Iran.

Email: tanidehn@sums.ac.ir

Background: Several plants have been shown to possess antioxidant and estrogenic properties that can be useful in postmenopausal bone-loss prevention.

Objective: The present study aimed to investigate the anti-osteoporotic effects of the hydroalcoholic extract of the *Psidium guajava* (PG) fruit in ovariectomized (OVX) rats.

Materials and Methods: Sixty female Sprague-Dawley rats were randomly divided into 6 groups: a control positive group, a sham-operated group, an OVX group given normal saline (OVX-only group), and 3 treatment groups comprising 2 OVX groups treated orally with 500 and 1000 mg/kg/d of the hydroalcoholic extract of the PG fruit respectively and an OVX group treated with an injection of 0.15 mg/kg of estradiol. The study was conducted over a 12-week period. Samples from the animals' blood, femoral bones, and uteri were collected for stereological and biochemical analyses. The data were analyzed using SPSS, version 19. A P value equal to or less than 0.05 was considered statistically significant.

Results: The results revealed a significant decrease in the levels of calcium, total antioxidant capacity, and phosphorus as well as uterus weight, femoral ash density, femoral volume and weight, and numbers of osteocytes and osteoblasts. Moreover, there was an increase in the levels of alkaline phosphatase and urine deoxypyridinoline together with a rise in the number of osteoclasts in the OVX-only group compared to the control and treatment groups ($P \leq 0.05$). The hydroalcoholic extract of the PG fruit increased femoral weight and volume, femoral ash density, numbers of osteocytes and osteoblasts, and trabecular volume of the bones in comparison with the OVX-only group in a dose-dependent manner. No significant difference was observed between the groups in the levels of malondialdehyde and interleukin-6.

Conclusion: The hydroalcoholic extract of the PG fruit prevented OVX-induced bone loss in the rats, with no proliferative effect on atrophic uteri; it should, therefore, be considered for treatment purposes.

K-34

Successful laparoscopic treatment of Chylous Ascites after pelvic lymphadenectomy: A case report and peanut oil application

Shu zhong Y.

Department of Obstetrics and Gynecology, The First Affiliated Hospital of Sun Yat Sen University, Guangzhou, China.

Email: yszlfy@163.com

A 23-year-old female patient with refractory chylous ascites was successfully treated with laparoscopic ligation of the ruptured lymphatic vessel. The young patient developed abdominal distention after right-side pelvic lymph node dissection for dysgerminoma of the right ovary. Conservative managements failed to control the symptoms. Laparoscopic surgery was performed after oral administration of peanut oil, revealing the presence of a whitish fluid in the abdominal cavity. The responsible lesion of the chylous ascites was detected in the right obturator fossa and ligated with the HEM-O-LOK System (Kangji Medical Instrument Co., Ltd., Hangzhou City, Zhejiang Province, China). The patient experienced an uneventful recovery and has been completely free of symptoms for 1 year. Laparoscopic surgery should be considered as a treatment of choice for intractable chylous ascites, and peanut oil could be used before surgery as an effective way to facilitate detection of the leakage during surgery.

K-35

Individualization of controlled ovarian Stimulation

Vaiarelli A.

Infertilità di coppia, Tecniche di PMA, Italy.

Email: alberto.vaiarelli@gmail.com

Ovarian stimulation is defined as pharmacological treatment with the intention of inducing the development of ovarian follicles. The number of oocytes is a key factor to maximize CLBR in each group of patients. This is possible by fully exploiting the ovarian reserve. The evidence of multiple follicular waves during a single menstrual cycle in women opened important clinical implications for the treatment of poor prognosis patients (advance maternal age and poor ovarian reserve). In poor prognosis patients and in all patient with sub-optimal response undergoing *non-embryo transfer cycle*, double stimulation in one ovarian cycle might be considered an important tool for increasing the number of oocytes retrieved and embryos obtained in a short time frame. Moreover, collecting more oocytes increases the higher chance to complete an average-sized family. Exogenous gonadotropins did not significantly modify the likelihood of aneuploidy embryos in patients undergoing controlled ovarian stimulation. Indeed, the increased number of oocyte retrieved does not compromise oocyte competence. Finally, the phase of the ovarian cycle when Controlled Ovarian Stimulation is started does not affect the euploidy rate.

K-36

Optimized oocyte numbers

Vaiarelli A.

Infertilità di coppia, Tecniche di PMA, Italy.

Email: alberto.vaiarelli@gmail.com

The main goal of any assisted reproductive technique is having live birth of a healthy child. Today the measure of success in IVF must be the Cumulative Live Birth rate (CLB) per started cycle. The ability to predict the ovary response is the priority to obtain the right number of oocytes and to define the right individual treatment for the right patients. Although the number of the oocytes retrieved is poorly correlated to the quality and to the competence of the oocytes, but if we have more eggs (>15), we increase the chance to obtain a baby. Indeed, the woman age is the only factor affecting embryo quality. Many factors can be used as predictors of ovarian response: age, biochemical parameters (FSH and AMH), morphological parameters (antral follicular count) but also some clinical conditions like polycystic ovary syndrome (PCOS) and low BMI. AMH and AFC may be considered a very good predictor of the hyper or hypo response. In accordance with ovarian reserve we can individualize four main categories of patients: high responder, normal responder, moderate/low responders and poor responders. For each group we can have different treatment strategies and different objectives.

K-37

Spermatogenesis after allotransplanting of adipose tissue-derived and bone marrow-derived mesenchymal stem cells in seminiferous tubules of azoospermic mice

Zhanbyrbekuly U¹, Dorvash MR², Khoradmehr A³, Zare Sh², Rahmanifar F⁴, Dianatpour M^{2, 5}, Tanideh N^{2, 6}, Khodabandeh Z², Razeghian Jahromi I⁷, Koochi-Hoseinabadi O⁸, Aynaiev Y¹, Tamadon A⁹.

1. Department of Urology and Andrology, Medical University Astana, Astana, Kazakhstan.

2. Stem Cells Technology Research Center, Shiraz University of Medical Sciences, Shiraz, Iran.

3. Department of Basic Sciences, School of Veterinary Medicine, Shiraz University, Shiraz, Iran.

4. Research and Clinical Center for Infertility, Yazd Reproductive Sciences Institute, Shahid Sadoughi University of Medical Sciences, Yazd, Iran.

5. Department of Basic Sciences, School of Veterinary Medicine, Shiraz University, Shiraz, Iran.

6. Department of Medical Genetics, School of Medicine, Shiraz University of Medical Sciences, Shiraz, Iran.

7. Department of Pharmacology, School of Medicine, Shiraz University of Medical Sciences, Shiraz, Iran.

8. Cardiovascular Research Center, Shiraz University of Medical Sciences, Shiraz, Iran.

9. Central Lab, Shiraz University of Medical Sciences, Shiraz, Iran.

10. The Persian Gulf Marine Biotechnology Research Center, The Persian Gulf Biomedical Sciences Research Institute, Bushehr University of Medical Sciences, Bushehr, Iran.

Email: Ulanbek.amu@gmail.com

Background: Adipose tissue-derived mesenchymal stem cells (AT-MSCs) and bone marrow-derived mesenchymal stem cells (BM-MSCs) could be used for cell therapy of azoospermia due to their differentiation and therapeutic potential. AT-MSCs and BM-MSCs have proliferative induction potential and immunomodulatory effects, and can secrete growth factors and cytokines. In this experimental study the ability of allotransplantation of AT-MSCs and BM-MSCs in recovery of fertility in busulfan-induced azoospermic testes of mice were evaluated using histomorphometric indices and flow cytometry. The aims of the present study were also to evaluate the potential of differentiation of AT-MSCs and/or BM-MSCs into germinal layer cells of seminiferous tubules.

Materials and Methods: Adult male mice were randomly divided into four groups including the intact control, azoospermia-induced, AT-MSCs therapy, and BM-MSCs therapy groups. Except intact control, the other groups were intraperitoneally received two doses of 10 mg/kg of busulfan with 21 days' interval in order to induce azoospermia. AT-MSCs and BM-MSCs were isolated from subcutaneous fat and bone marrow tissues of two donor eGFP^{+/+} mice. The MSCs were injected into the efferent ducts of testes of cell therapy mice 35 days after the last busulfan injection. The cell therapy groups were sampled 60 days after cell therapy and the other groups were sampled in the same time. The histomorphometric indices of testes were evaluated. Flow cytometry was used to compare expression of eGFP in the produced sperm in epididymis of all groups.

Results: Histomorphometric evaluation of seminiferous tubules of cell therapy groups showed that most of the tubules had degrees of spermatogenesis. Histopathologic evaluation of testes showed that most of the seminiferous tubules of cell therapy group had normal cell morphology. Spermatogenesis was not seen in the azoospermia group. Production of some eGFP sperm showed evidence of probable differentiation of MSCs into germinal cell layers.

Conclusion: Allotransplanted AT-MSCs and BM-MSCs could successfully induce spermatogenesis in azoospermic seminiferous tubules of mice. AT-MSCs are easier and safer to obtain for stem cell therapy of azoospermia in comparison with BM-MSCs. Therefore, AT-MSCs and BM-MSCs can be injected in cell transplantation of azoospermia in human.