



Impact of Reactive Oxygen Species Level in Serum and Culture Media on Pregnancy Rate after Day 3 and Day 5 Embryo Transfer in Iraqi Infertile Females Undergoing ICSI

Hanan Abdulrazzaq Abdulazeer¹, Muayad Sraibet Abbod¹, Mufeda Ali Jwad¹ 

¹High Institute of Infertility Diagnosis and Assisted Reproductive Technologies, Al Nahrain University, Baghdad, Iraq.
dr.hanan72@gmail.com

ABSTRACT

Reactive oxygen species (ROS) are active chemical compounds created by the body's metabolic process and are crucial in human reproduction. Excessive ROS can contribute to oxidative stress, which has been linked to poor fertility. Embryo culture medium plays an essential role in IVF/ICSI cycles. Eighty participants underwent ovarian stimulation and ICSI cycles; ROS levels were measured in blood samples on the ovum pickup (OPU) day and in each embryo culture dish on ET day using enzyme-linked immunosorbent assay (ELISA). These values were associated with the quality of generated embryos. Women were divided into two groups; the first had day three embryo transfers, whereas the second received blastocyst transfers on day 5. Grades I embryo count was shown to be inversely related to the serum and culture medium ROS levels. Pregnant women had significantly greater levels of GI embryos ($p=0.011$) and lower levels of GIII embryos ($p=0.024$). Pregnancy rates for the third- and fifth days of the embryo transfers were 24% and 50%, respectively, with ($p=0.045$). Pregnancy success rates were increased after ET day five and were strongly influenced by transfer day. Blastocyst-stage embryos with high serum ROS levels were found to be underdeveloped. Culture medium ROS levels can be used as an adjuvant criterion for embryo selection and metabolic marker for developing embryos.

Received:
28-Sep-2021
Accepted:
16-Nov-2021
Published:
17-Aug-2022

How to cite:

Abdulazeer HA; Abbod MS; Jwad MA; Impact of Reactive Oxygen Species Level in Serum and Culture Media on Pregnancy Rate after Day 3 and Day 5 Embryo Transfer in Iraqi Infertile Females Undergoing ICSI; Iraqi Journal of Embryos and Infertility Researches (IJEIR), (2022); 12 (2): 12-29.
Doi: <http://doi.org/10.28969/IJEIR.v12.i2.r2.22>

KEYWORDS

Culture media, Reactive oxygen species, Embryo development, Embryo transfer, Pregnancy rate

1. Introduction

Fertility is the ability to conceive and bear children (**Barbieri, 2019 [1]**) or to create a clinical pregnancy (**Vander Borgh and Wyns, 2018 [2]**). Infertility or subfertility affects a substantial proportion of the population in one manner or other. The World Health Organization defines infertility as "a disorder of the reproductive system defined by the failure to produce a clinical pregnancy following 12 months or more of frequent unprotected sexual intercourse," which is the most often used definition (**Szamatowicz and Szamatowicz, 2020 [3]**). In vitro fertilization/ intracytoplasmic sperm injection (IVF/ICSI) was the most often used ART therapy (52.2 percent). Only 33,1 percent of women who underwent ART therapy became pregnant as a result of it (**Lucas et al., 2021 [4]**). The need to identify the most viable embryo following in vitro fertilization (IVF) is crucial to enhance successful pregnancy rates. (**Lan et al., 2019 [5]**). The generation of reactive oxygen species from either intrinsic or extrinsic sources throughout ART's different stages may grow to dangerously high levels, resulting in oxidative

stress that harms gametes and subsequent embryos (**Gupta et al., 2017 [6]**). For many years, the link between infertility and oxidative stress has been extensively researched, with many studies examining the clinical impact of oxidative stress on both natural fertility and assisted reproductive techniques (**Ribas-Maynou and Yeste, 2020 [7]**). Increased levels of ROS have been linked to embryo fragmentation or blastocyst development in the used culture medium. Although ROS appears to have a role in early embryonic development, its specific function is yet unknown. According to (**Lan et al., 2019 [5]**), the last phase of IVF-ICSI is the transfer of the embryos generated by sperm oocyte fertilization after the controlled ovarian stimulation into the terminal. Embryo transfer (ET) is a significant step in assisted reproductive technology, and while early embryo transmission on day two or day three is widely preferred clinically, ET blastocyst is becoming increasingly important as it improves the synchronization of the embryo to the endometrium and the presentation of quality embryos in the embryo (**İnal,**

2021[8]). Therefore, the aim of this study was to determine the relationship between levels of ROS in serum and culture media with the pregnancy rate of embryo transfer in cleavage transfer on day three and blastocyst transfer on day five at different grades of embryo quality.

2. Materials and Methods

2.1. Study participants and controlled ovarian stimulation

The current prospective study was conducted on 80 infertile women who were undergoing intracytoplasmic sperm injection (ICSI) at the High Institute for Diagnosis of Infertility and Assisted Reproductive Technology, Al Nahrain University, Iraq, Baghdad, from November 2020 to April 2021. The study was authorized by the Local Committee for Medical Ethics. The patient's fundamental demographic data, treatment details, and results are documented. The variables collected for this investigation included age, BMI, infertility type (primary infertility and secondary infertility), infertility cause (female factor, male factor, combined factor and

unexplained infertility), seminal fluid analysis for male partners in accordance with WHO 2010, hormonal analysis (FSH, LH, E2, Testosterone, Prolactin, TSH) for female partners on day 2 of the menstrual cycle, basal antral follicle count (BAFC), embryo quality (good or poor quality) and embryo development stage (at day 3 or 5). All participants in the current study were enrolled in flexible antagonist protocol, which started at day 2 of the menstrual cycle, controlled ovarian stimulation with Gonadotropin (rFSH) (Gonal-F; Serono Laboratories, Saint Cloud, France) followed by the administration of a GnRH antagonist (Cetrorelix acetate for injection 0.25 mg: Cetrotide®, Merk, Switzerland), according to the size of the largest follicles when they reach 13- 14 mm. Transvaginal ultrasound was done on the 5th day of stimulation, and subsequent scan was done every 2-3 days as necessary (Hu et al., 2016 [9]). Triggering of ovulation was induced when two to three leading follicles were observed by ultrasound at an average diameter of 17-18 mm by the administration of recombinant hCG (rhCG 6500 IU, Ovitrelle®;

Merck Serono) subcutaneously ([Majumdar. et al., 2019 \[10\]](#)) or r-hCG 250 µg and Decapeptyl® 0.2 mg (dual triggering) in a case with a high risk of OHSS ([Fabris et al., 2017 \[11\]](#)). Transvaginal ultrasound-guided Oocyte retrieval is done after triggering of ovulation with hCG about 35-36 hrs.

2.2. ICSI procedure

Immediately prior to ICSI, cumulus cells were removed. The denuded oocytes that demonstrated the first polar bodies were selected for ICSI, which was performed 0–3 hours after oocyte denuding. Motile husband's sperms were selected for intracytoplasmic sperm injection of mature (MII) oocytes.

2.3. Quality assessment of the in vitro development of embryos

After ICSI, embryo development was monitored and graded with grade I, grade II, and grade III. Four-cell phases (45–46 hours), eight-cell phases (69–70 hours), and the blastocyst phase (118–120 hours) was observed ([Shih et al., 2014 \[12\]](#)), quality of embryo was evaluated according to

microscopically morphological parameters. Three to five days later, embryo selection and embryo transfer were performed. Luteal support was started, and beta hCG determination was made (to document biochemical pregnancy). The ROS serum level on the day of ova pickup and the detection of ROS in every embryonic culture dish on the day of embryo transfer were done using an enzyme-linked immunosorbent assay (ELISA) kit at a private laboratory.

3. Statistical analysis

The data were analyzed using the Statistical Package for Social Sciences (SPSS) version.

22.0 and Microsoft Office 2007. The descriptive statistics, including frequency, range, mean, and standard deviation, were measured to describe the data. The groups were compared by applying an independent sample t-test (unpaired t-test between two groups), analysis of variance (ANOVA between three groups), and chi-square (for non-continuous data or percentage). The degree of association between continuous variables was calculated by Pearson's

Correlation coefficient (r) and the results were considered statistically significant when the p-value was less than 0.05.

4. Results

4.1. Correlations between serum and culture media ROS levels and embryo grading

The correlation between serum and culture media ROS and embryo grading was shown in **Table 1**. According to the results, there were a significant and inverse correlation between serum and culture media ROS in terms of the number of grades I embryos ($r = -0.230$, $p = 0.041$) ($r = -0.297$, $p = 0.048$), respectively. However, there was no significant correlation was established between serum and culture media ROS for Grade II and grade III ($r = 0.098$, $p = 0.406$), ($r = 0.067$, $p = 0.626$), ($r = 0.148$, $p = 0.208$) and ($r = 0.039$, $p = 0.778$) respectively.

4.2. Comparison of ROS levels in serum and culture media between pregnant and non-pregnant women at day 3 and day 5 of embryo transfer

Serum and culture medium ROS levels were compared between pregnant and non-pregnant females at days 3 and 5 of the embryo transfer, as illustrated in Table 2 and Figure 1. The results showed higher serum ROS levels in non-pregnant women at day 5

of ET. However, there were no significant differences in ROS levels in serum and culture media between pregnant and non-pregnant women at day three and day 5 of embryo transfer ($p > 0.05$), meaning that the day of embryo transfer had no effect on serum and culture media ROS level.

4.3. Comparison of embryo characteristics between pregnant and non-pregnant women

The comparison of embryo characteristics between pregnant and non-pregnant women were demonstrated in **Table 3** and **Figure 2**. According to the results, there were significant differences between pregnant and non-pregnant patients in the number of GI embryos ($p = 0.011$) with higher levels in pregnant women and a significant difference in the number of GIII embryos ($p = 0.024$) with lower levels in pregnant women. However, there were no.

Table (1): Correlations between serum and culture media ROS levels and embryo grading.

Serum ROS levels	GI embryos	GII embryos	GIII embryos
r	- 0.230	0.098	0.067
p	0.041*	0.406	0.626
Culture media ROS levels	GI embryos	GII embryos	GIII embryos
r	- 0.297	0.148	0.039
p	0.048*	0.208	0.778

Table (2): Comparison of ROS levels in serum and culture media between pregnant and non-pregnant women at day 3 and day 5 of embryo transfer

Parameter (Mean $\pm$ SD)	Pregnant women	Nonpregnant women	p-value
Serum ROS (U/l) at day 3 of ET	630 $\pm$ 193	619 $\pm$ 243	0.877
Serum ROS (U/l) at day 5 of ET	740 $\pm$ 352	1231 $\pm$ 1265	0.279
Culture media ROS (U/l) at day 3 of ET	299 $\pm$ 89	282 $\pm$ 55	0.343
Culture media ROS (U/l) at day 5 of ET	269 $\pm$ 47	281 $\pm$ 48	0.601

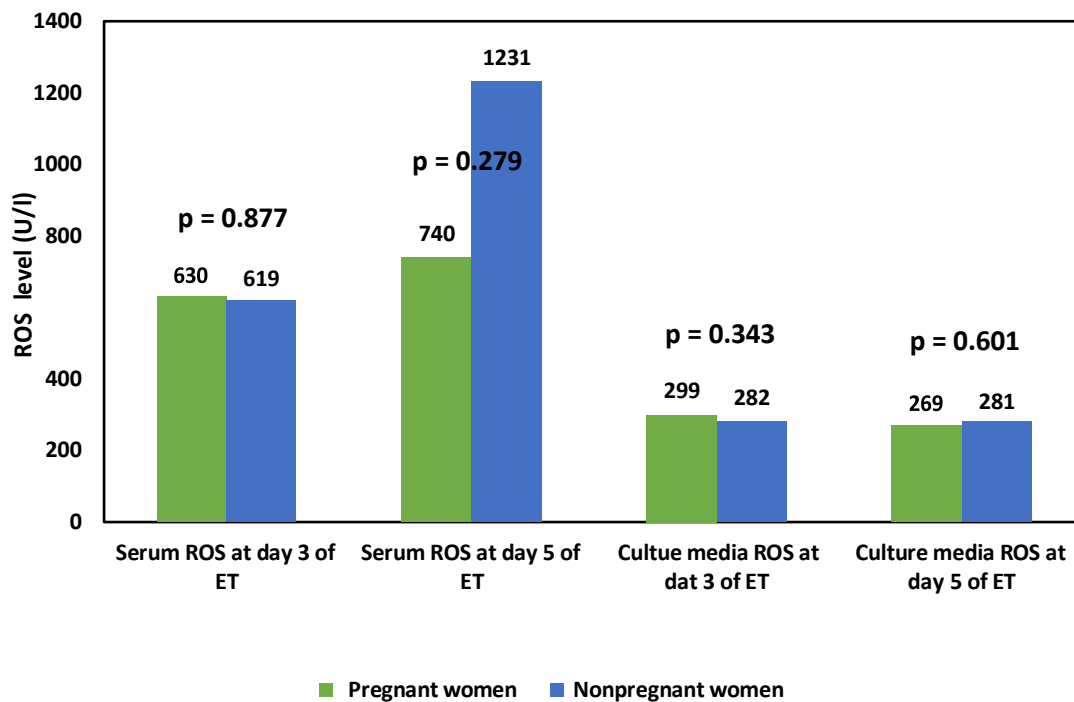


Figure (1): Comparison of ROS levels in serum and culture media between pregnant and non-pregnant women at day 3 and day 5 of embryo transfer

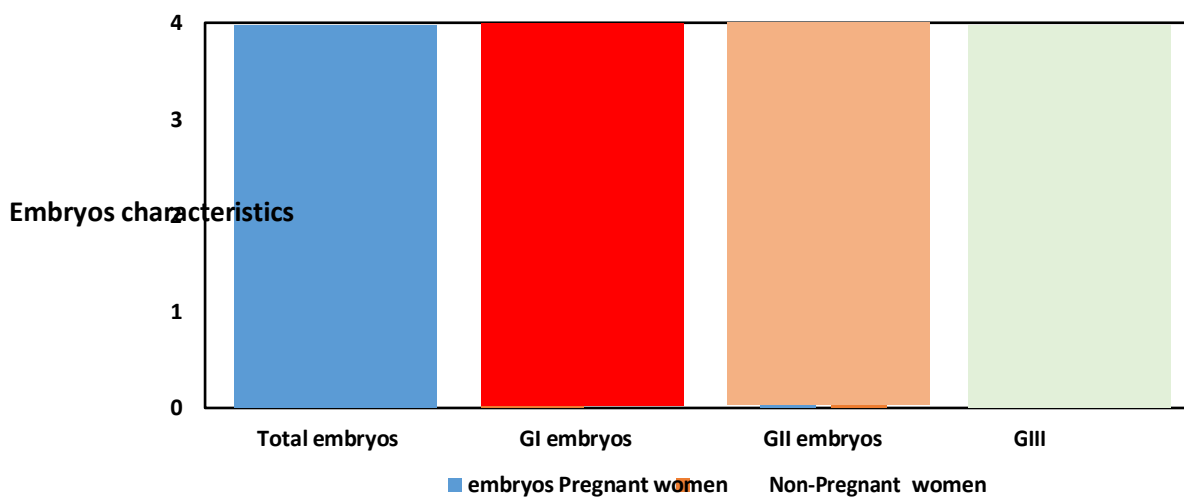


Figure (2): Comparison of embryo characteristics between pregnant and non-pregnant women

Table (3): Comparison of embryos characteristics between pregnant and non-pregnant women

<i>Parameter</i>	<i>Pregnant women</i>	<i>Non-pregnant women</i>	<i>p-value</i>
Total embryo count (Mean $\pm$ SD)	3.08 $\pm$ 0.97	2.86 $\pm$ 1.03	0.365
Grade I embryo (Mean $\pm$ SD)	2.67 $\pm$ 1.09	1.88 $\pm$ 1.29	0.011*
Grade II embryo (Mean $\pm$ SD)	0.42 $\pm$ 0.88	0.68 $\pm$ 1.04	0.288
Grade III embryo	0	0.68 $\pm$ 1.42	0.024*

*: p value < 0.05 (significant)

Table (4): Comparison of the day of embryo transfer between pregnant and non-pregnant women

Day of embryo transfer (ET) n. (%)	Day 3	Day 5	p-value
Pregnant	15/62 (24%)	9/18 (50%)	p = 0.045*
Non-pregnant	47/62 (76%)	9/18 (50%)	P = 0.045*

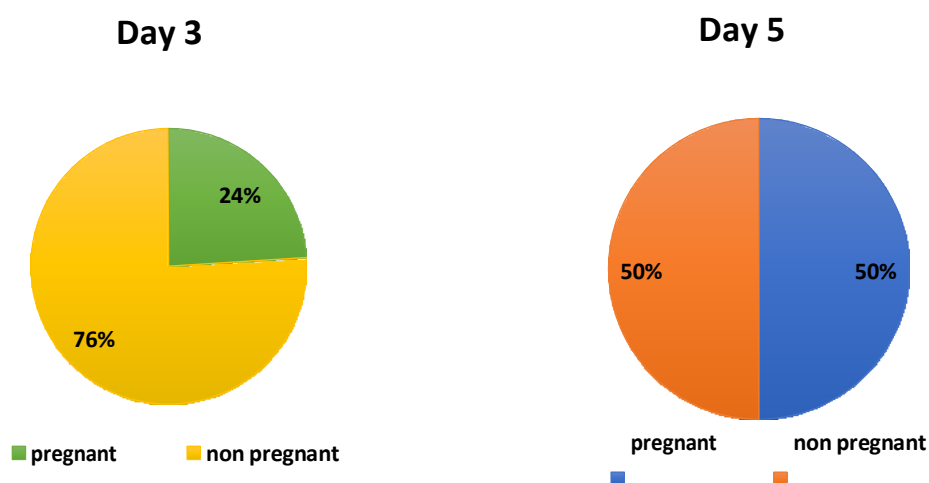


Figure (3): Comparison of the day of embryo transfer between pregnant and non-pregnant women

Significant differences in total embryo count and numbers of GII embryos with a p-value greater than 0.05. Embryo transfer was performed three days for 62 females (77,5%) and days for 18 females (22.5%) after oocyte retrieval. There was a significantly higher pregnancy rate on day 5 of embryo transfer (50%) compared to day 3 of embryo transfer (24%) with (p value=0.045%), as shown in **Table 4** and **Figure 3**.

5. Discussion

In this prospective cohort research, data from 80 patients were examined. Determination of ROS level and direct comparison with the quality grades of the embryos retrieved were made in the serum and cultural media of infertile women undergoing ICSI. Since its debut, significant improvements to its delivery have been developed to enhance pregnancy rates and to promote patient safety, the ART methods remain changed and updated (**Thoma et al., 2021 [13]**).

5.1. Correlations between serum and culture media ROS levels and

embryo grading

Although morphological scores are the best technique available for embryonic picking, measurement of oxidative stress indicators like ROS levels can be a non-invasive alternative in culture medium or can be coupled with traditional morphological scores to make embryo selection more accurate. ROS and antioxidants are stable under physiological circumstances. Excess oxidative stress might theoretically destroy the cellular environment, adversely influence fertilization, impede cellular embryo development, or cause apoptosis, leading to embryo fragmentation (**Lan et al., 2019 [5]**). The research in the current study has found that the ROS level measured on OPU day is significantly correlated with the number of Grade I embryos ($r=-0.230$, $p=0.041$). Also, substantial negatively related intercultural media (ROS) to embryos of grade 1 were observed ($r=0.297$, $p=0.048$). Many studies have shown that high ROS levels are detrimental to the embryo; others have shown that lower levels indicate lowered fertilization potential,

therefore reducing the oocyte capacity and likely.

Resulting in poor embryo quality in fertilization (Siristatidis et al., 2016 [14]).

5.2. Comparison of ROS levels in serum and culture media between day 3 transfer day 5 of embryo transfer in pregnant and non-pregnant women

Reactivity Oxygen Species have an impact on several physiological ovary activities, including ovarian steroidogenesis, oocyte maturation, ovulation, blastocyst development, implantation, luteolysis, and pregnancy luteal support (Lu et al., 2018 [15]). Excessive oxidative stress can have detrimental consequences on the cellular environment, adversely impact fertilization, impede cell development in the embryo, or lead to embryo fragmentation (Lan et al., 2019 [5]). The ROS serum and cultural media were compared between pregnant and non-pregnant women on day 3 and day 5 of embryo transfer. The results demonstrated that the day of the embryo was not affected by serum- and culture-related medium ROS,

nor did the date of the embryo transfer have any connection. In non-pregnant women, serum ROS levels were greater. However, another study showed that ROS levels in culturally-based media are not linked significantly to the score, embryonic quality, formation, arrest, and/or design opportunities, so reactive oxygen species levels in culture media could not distinguish between conception and non-conception cycles (Lan et al., 2019 [5]). Many variables influence ROS generation in the culture media, such as concentration of oxygen, light, treatment of oocytes, and general physicochemical characteristics that might alter the overall cell metabolism (Lin and Wang, 2021 [16]). These results were already reached via earlier trials showing fewer clinical PRs in both the traditional IVF and the ICSI cycles with greater ROS levels on day 3. (Bedaiwy et al., 2010 [17]).

5.3. Comparison of embryo characteristics between pregnant and non-pregnant women

To increase pregnancy success, we must select the more viable embryo following in vitro fertilization (IVF) (Lan et al., 2019 [5]). The main purpose of IVF is to choose the best. Embryos to transfer to produce a healthy single pregnancy (Li et al., 2020 [18]). Two key aspects impacting live birth results were embryonic culture and embryo quality (Wang et al., 2021 [19]). The findings from the current study indicated that the pregnancy rate for embryo transfers was substantially greater on Day 5 (50 percent) compared to Day 3 (24 percent) with ($p=0.045$). The researchers have ascribed superior outcomes to better embryo selection, greater synchronization, and lower cervical mucus with the transfer of blastocyst. The embryo that can grow into the blastocyst is, therefore, the embryo that is better implanted. (Bhandari, et al., 2017 [20]). The findings of this study corroborate those of a prior study, which found that the clinical pregnancy rate for blastocysts was greater than for cleavage-stage embryos (Kirillova et al., 2020 [21]). Coskun et al. and Levron et al., on the other hand, found no difference in implantation

and pregnancy rates between patients who received day three and day five embryo transfers. While Karaki et al. found that embryo culture to the blastocyst stage resulted in a better implantation rate than embryos transplanted on day 3 (Alfaraj et al., 2017 [22]). In vitro fertilization cycle results are heavily influenced by the quantity and quality of transferred embryos. Bad-quality embryo transfers have a possibility of clinical pregnancy and live birth rate, whereas good-quality embryo transfers have a higher chance of clinical pregnancy and live birth (Aldemir et al., 2020 [23]). There were no significant variations in the mean total transferred embryo count between pregnant and non-pregnant females in the current research. According to Pinborg et al., the obstetric result of a surviving singleton is negatively influenced when the number of fetal/embryos increases. They have also backed up the theory that early implantation site overcrowding causes first-trimester losses. According to a meta-analysis, the number of embryos transferred had no effect on the outcome of a singleton

pregnancy conceived as a consequence. This suggests that there may be variables intrinsic to the ART technology or the infertile patient herself that contribute to worse results (Bhandari et al., 2017 [20]). In the current study, there were highly significant Differences in mean grade I embryo number and mean grade III embryo number between the two groups, being higher levels of mean grade I embryo number in and lower levels in the number of GIII embryos in pregnant women. This may, in part, explain the significantly higher pregnancy rate in association with good-quality embryo transfers in the current study. These findings were consistent with previous research, which found that the rates of clinical pregnancy and live birth in the best-quality embryo group were considerably higher than those in the bad-quality embryo group (Sun et al., 2020 [24]).

5. Conclusions

Infertile individuals who had embryo transfer on the fifth day had a greater rate of pregnancy success. Increased levels of ROS production in

embryo culture medium may harm embryo growth metrics as well as clinical PRs since ROS levels in serum and prepared culture media are linked to embryo development outcomes. As a result, ROS level measurement might be utilized as an additional quality control technique in IVF labs.

Acknowledgment

We would like to acknowledge Al Nahrain University, Baghdad, Iraq.

Funding

This work received no funding.

Author Contribution

Khalaf, NM, performed the study, examined and reviewed results, and manuscript writing with the help and supervision of Mohammed, AA, and Rahim, AI.

Conflict of Interest

The authors declare no conflict of interest.

Ethical Clearance

The study was approved by the Ethical Approval Committee.

References

- [1] Barbieri RL. Female infertility. In Yen and Jaffe's Reproductive Endocrinology 2019 Jan 1 (pp. 556-581). Elsevier. [[Online article link](#)]
- [2] Vander Borcht M, Wyns C. Fertility and infertility: Definition and epidemiology. Clinical Biochemistry. Elsevier BV; 2018 Dec;62:2–10. [[Online article link](#)].
- [3] Szamatowicz M, Szamatowicz J. Proven and unproven methods for diagnosis and treatment of infertility. Advances in Medical Sciences. Elsevier BV; 2020 Mar;65 (1):93–6. [[Online article link](#)]
- [4] Lucas AF, Gemechu DB, Du Plessis SS, Aboua YG. Fertility and pregnancy outcome among women undergoing assisted reproductive technology treatment in Windhoek, Namibia. Journal of Assisted Reproduction and Genetics. Springer Science and Business Media LLC; 2021 Feb 8;38 (3):635–43. [[Online article link](#)]
- [5] Lan K-C, Lin Y-C, Chang Y-C, Lin H-J, Tsai Y-R, Kang H-Y. Limited relationships between reactive oxygen species levels in culture media and zygote and embryo development. Journal of Assisted Reproduction and Genetics Springer Science and Business Media LLC; 2018 Nov 10;36 (2):325–34. [[Online article link](#)]
- [6] Gupta S, du Plessis SS, AlQasem S, Nouh M, Cakar Z. Therapeutic Role of Antioxidants (AOX) in the Treatment of Infertility. In Oxidative Stress in Human Reproduction 2017 (pp. 129-150). Springer, Cham. [[Online article link](#)]
- [7] Ribas-Maynou J, Yeste M. Oxidative stress in male infertility: causes, effects in assisted reproductive techniques, and protective support of antioxidants. Biology. 2020 Apr;9 (4):77. [[Online article link](#)]
- [8] İnal HA, İnal, ZÖ. Early-cleavage versus blastocyst stage embryo transfer: a prospective comparative study. Journal of the Turkish German Gynecological Association. 2021 Dec

6;22 (4):279-285. [[Online article link](#)]

- [9] Hu X, Luo Y, Huang K, Li Y, Xu Y, Zhou C, et al. New Perspectives on Criteria for the Determination of HCG Trigger Timing in GnRH Antagonist Cycles. *Medicine*. Ovid Technologies (Wolters Kluwer Health); 2016 May;95 (20):e3691. [[Online article link](#)]
- [10] Mayekar R, Majumdar A, Sachan R, Nandanwar Y, Soni N, Banker M, et al. A multicenter, randomized, equivalence trial of a new recombinant human chorionic gonadotropin preparation versus ovitrelle® for ovulation in women undergoing intrauterine insemination following ovarian stimulation. *Journal of Human Reproductive Sciences*. Medknow; 2019;12 (1):53. [[Online article link](#)]
- [11] Fabris AM, Cruz M, Legidos V, Iglesias C, Muñoz M, García-Velasco JA. Dual Triggering With Gonadotropin-Releasing Hormone Agonist and Standard Dose Human Chorionic Gonadotropin in Patients With a High Immature Oocyte Rate. *Reprod Sci*.

2017 Aug;24 (8):1221- 1225. [[Online article link](#)]

- [12] Shih YF, Lee TH, Liu CH, Tsao HM, Huang CC, Lee MS. Effects of reactive oxygen species levels in prepared culture media on embryo development: a comparison of two media. *Taiwan J Obstet Gynecol*. 2014 Dec;53 (4):504-8. [[Online article link](#)]
- [13] Thoma M, Fledderjohann J, Cox C, Kantum Adageba R. Biological and Social Aspects of Human Infertility: A Global Perspective. *Oxford Research Encyclopedia of Global Public Health*. Oxford University Press; 2021 Mar 25. [[Online article link](#)]
- [14] Siristatidis C, Vogiatzi P, Varounis C, Askoxylaki M, Chrelias C, Papantoniou N. The Effect of Reactive Oxygen Species on Embryo Quality in IVF. *In Vivo*. 2016 Mar-Apr;30 (2):149-53. PMID: 26912827. [[Online article link](#)]
- [15] Lu J, Wang Z, Cao J, Chen Y, Dong Y. A novel and compact review on the role of oxidative stress in female reproduction. *Reproductive Biology and Endocrinology*.

Springer Science and Business Media LLC; 2018 Aug 20;16 (1):80. [[Online article link](#)]

[16] Lin J, Wang L. Oxidative Stress in Oocytes and Embryo Development: Implications for In Vitro Systems. Antioxidants & Redox Signaling. Mary Ann Liebert Inc; 2021 Jun 10;34 (17):1394–406. [[Online article link](#)]

[17] Bedaiwy MA, Mahfouz RZ, Goldberg JM, Sharma R, Falcone T, Abdel Hafez MF, Agarwal A. Relationship of reactive oxygen species levels in day three culture media to the outcome of in vitro fertilization/intracytoplasmic sperm injection cycles. Fertility and sterility. Elsevier BV; 2010 Nov;94 (6):2037-42. [[Online article link](#)]

[18] Li H, Xu X, Jing Y, Liu L, Wang Y. Associations between a new day four embryo grading system and implantation rates in frozen embryo transfer (FET) cycles. Medicine (Baltimore). 2020 Oct 16;99 (42):e22676. [[Online article link](#)]

[19] Wang N, Zhao X, Ma M, Zhu Q, Wang Y. Effect of Day 3 and Day 5/6 Embryo Quality on the Reproductive

Outcomes in the Single Vitrified Embryo Transfer Cycles. Front Endocrinol (Lausanne). 2021 Apr 23;12:641623. [[Online article link](#)]

[20] Bhandari S, Ganguly I, Agarwal P, Munaganuru N, Gupta N, Singh A. Relationship of Number of Embryos Transferred with Perinatal Outcome of Singleton Pregnancy. J Reprod Infertil. 2017 Jan-Mar;18 (1):179-184. [[Online article link](#)]

[21] Kirillova A, Lysenkov S, Farmakovskaya M, Kiseleva Y, Martazanova B, Mishieva N, Abubakirov A, Sukhikh G. Should we transfer poor quality embryos? Fertility research and practice. 2020 Dec;6 (1):1-7. [[Online article link](#)]

[22] Alfaraj S, Alzaher F, Alshwaiaer S, Ahmed A, et al. Pregnancy outcome of the day Three versus day five embryo transfer: A retrospective analysis. Asian Pacific Journal of Reproduction [Internet]. Asian Pacific Journal of Tropical Medicine Press; 2017 Mar 1;6 (2):89–

92. [[Online article link](#)]

[23] Aldemir O, Ozelci R, Baser E, Kaplanoglu I, Dilbaz S, Dilbaz B, Tekin OM. Impact of Transferring a Poor-Quality Embryo Along with a Good Quality Embryo on Pregnancy Outcomes in IVF/ICSI Cycles: a Retrospective Study. Geburtshilfe Frauenheilkd. 2020 Aug;80 (8):844-850. [[Online article link](#)]

[24] Sun Y, Li E, Feng G, Li M, Fu Y, You J, Liu X, Zhu Y. Influence of cleavage- stage embryo quality on the in-vitro fertilization outcome after single embryo transfer in fresh cycles. Taiwan J Obstet Gynecol. 2020 Nov;59 (6):872-876. [[Online article link](#)]

Peer Review Information

Double-blind peer Review in which both authors and reviewers does not know each other.

This work was reviewed by

[Asst. Prof. Dr. Khulood Majid Saeed Alsaraf](#)

[Lec. Dr. Ahmed Naser](#)

Editorial Policy

The editorial policy at IJEIR ensured that this article fit the standards of scientific publications.

This work was copyedited by

[Dr. Taif Alaws](#)

Authors at OrcID

Mufeda Ali Jwad



<https://orcid.org/0000-0002-7995-6755>

Authors Biographies

Hanan Abdulrazzaq Abdulazeez

She received the M.B.Ch.B from the College of Medicine, Al-Anbar University, in 1996. She worked as a rotator house officer in the Al-Ramady hospital from 1996-1998. She had a senior resident (permanency) in Gynecology and Obstetrics at Heet Hospital from 2007 – 2009. Then, she received DGO (College of Medicine, Baghdad University) in 2012/ 2013. Since 2019, she is attending the High Institute for Infertility Diagnosis and Assisted Reproductive Technologies, Al-Nahrain University, Baghdad, Iraq.

Dr. Muayad Sraibet Abbod

He received the M.B.CH.B. From Al-Kufa University, the M.Sc. and the Ph.D. in Pharmacology from Al Nahrain University in 1998, 2008, and 2014, respectively. He occupied several academic positions, including the dean assistant for scientific affairs at Al Nahrain University. He has more than ten published papers, both local and international.

Dr. Mufeda Ali Jwad

She received her MBChB. From the College of Medicine at the University of Baghdad in 1996. Her M.Sc. in Applied Embryology and her Ph.D. in Infertility and Clinical Reproduction were from the High Institute of Infertility Diagnosis and Assisted Reproductive Technologies, Al Nahrain University, in 2007 and 2018,

respectively. She worked as a rotator in the Baghdad health department from 1996-1999. She worked in Gyn. & Obs. in Alsamawa general hospital and Babylon hospital from 2000-2003. She worked at the Babylon University, College of Medicine, anatomy and embryology department from 2003-2004. She has been working as a specialist physician and a consultant clinic at the High Institute of Infertility Diagnosis and Assisted Reproductive Technologies, Al Nahrain University, from 2008-2015. Currently, she is an assistant professor and specialist in infertility and clinical reproduction. She is the head of the clinical reproductive physiology department from 2019 till now. She has more than 25 published articles in national and international journals.



© 2022 Author (s)

This article is licensed under a

Creative Commons Attribution 4.0 International License, which permits use, sharing, adaptation, distribution, and reproduction in any medium or format, as long as you give appropriate credit to the original author (s) and the source, provide a link to the Creative Commons license, and indicate if changes were made.

<http://creativecommons.org/licenses/by/4.0/>.