

**EFFECT OF TEX101 PROTEIN, VITAMIN D AND ZINC ON MEN FERTILITY**Hasan A. Farman<sup>1</sup>, Mohammed Abdulrazzaq Assi<sup>2\*</sup> and Mohammed Baqer M. Salih<sup>3</sup><sup>1</sup>Department of Anesthesia and Intensive Care, College of Health & Medical Techniques, Al-Furat Al-Awsat Technical University, Iraq.<sup>2</sup>Department of Community Health, College of Health & Medical Techniques, Al-Furat Al-Awsat Technical University, Iraq.<sup>3</sup>Department of Medical Lab Techniques, College of Health & Medical Techniques, Al-Furat Al-Awsat Technical University, Iraq.

\*e-mail : razaq\_assi@yahoo.com

(Received 23 February 2019, Revised 30 May 2019, Accepted 12 June 2019)

**ABSTRACT :** Approximately 15% of couples fail to have a child birth despite of regular intercourse for more than 12 month duration without the use of contraceptives. So that we evaluated some parameters that may affect the fertilization. To find the relation of TEX101, vitamin D and Zinc with fertility condition of men and to evaluate how their change affect the men fertility. In case-control study the 90 serum and seminal plasma sample which collected from fertile (n = 45) and infertile (n = 45) men were estimated for TEX101 and vitamin D by the use of ELISA technique but the element zinc was estimated by colorimetric method. Whole blood was collected via vein puncture. We were collect 90 sample of seminal plasma and serum from the fertility center which divided into two groups 45 fertile and 45 infertile men. In present study showed significantly (P<0.001) decrease in infertile group compare to fertile group in serum Vitamin D and seminal plasma TEX101protein and Zinc. This study was showed a significant (P <0.001) positive correlation between TEX101 protein, vitamin D and zinc. From this study can be concluded that zinc element and vitamin D changes affects the semen quality and as a result decrease fertilization rate in couples with male factor infertility. And can be concluded that TEX101testicular protein can be regarded as a good marker of male fertility.

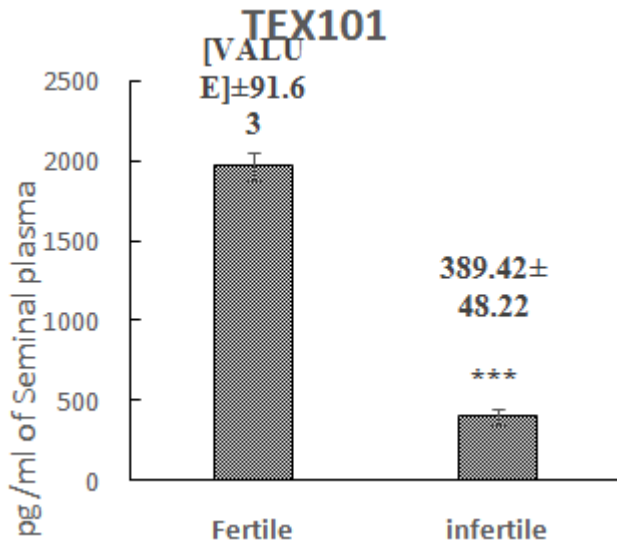
**Key words :** TEX101, vitamin D, zinc, infertile men.

**INTRODUCTION**

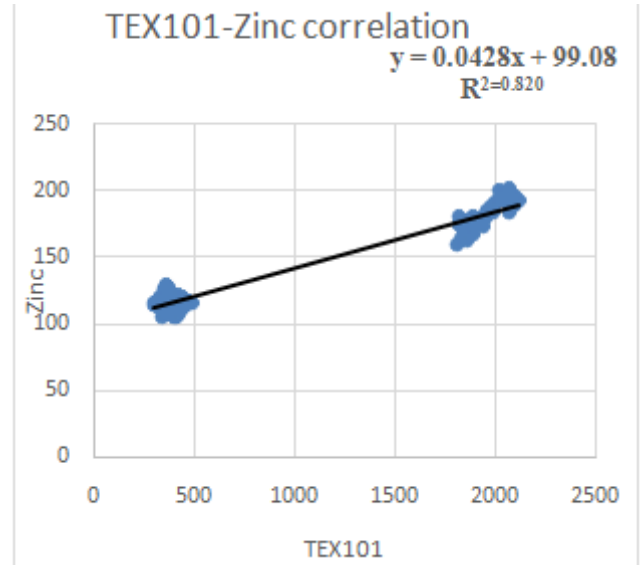
Approximately 15% of couples fail to have a child birth despite of regular intercourse for more than 12 month duration without the use of contraceptives. In fifty percent of couples without occurrence of pregnancy, is duo to abnormal semen quality so male cause infertility (Jungwirth *et al*, 2012). Male infertility may be duo to hypogonadism, varicocele, gonadotoxin expo-sure, testicular atrophy by mumps, obstruction of the genital tract or hormonal disturbance (McLachlan *et al*, 2007; Ghuman *et al*, 2018). Sometimes, it is impossible to treat men with azoospermia or oligospermia, this conditions are referred for IVF (*in vitro* fertilization) intrauterine insemination, duo to impaired sperm motility or quality (Wong *et al*, 2000; Rowe *et al*, 2000). However, infertility has been suggested to be at the upward push global and use of assisted reproductive strategies (ART) is monetary burden (Pedro *et al*, 2019).

Therefore, the goals of pathophysiology precise therapy of male infertility are to acquire spontaneous

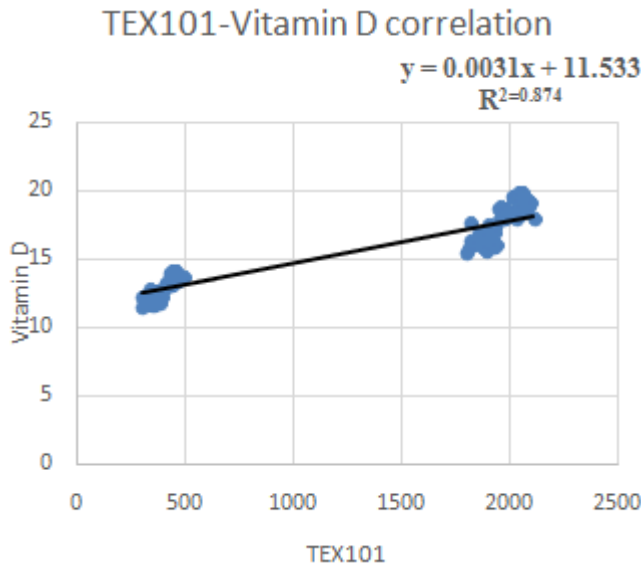
being pregnant and child bearing, to remove the need of ART, to downstage the extent of ART had to pass male factor infertility and additionally to raise the level of pregnancy with ART in the ones who've carried out unassisted replication (Efesoy *et al*, 2009). In some cases that there is biochemical disorder the sperm cannot fertilize the ova despite of having typical shape and normal count. This means seminal plasma with typical properties don't assured the fertilization. Also defects in semen quality can identify the causes of infertility to avoid the difficulty of treatment (Iyad *et al*, 2009). On the opposite hand, majority of researchers' referred to importance of hormonal evaluation, apart from screening of semen, the position features of hormones with the aid of complicated functions that is essential for spermatogenesis (Ali *et al*, 2010). Numerous factors affecting male infertility, for instance, varicocele that found in approximately 2-22% of men with infertility is as a result of decreased testosterone level in serum (Hala *et al*, 2010). Other than hormonal defects the genetic factors, environmental



**Fig. 1 :** Seminal plasma concentration of TEX101 in fertile and infertile men. Data are the mean ± SD (n = 45 in each group). \*\*\* indicates significance (p<0.001) compare to fertile group (independent T-test).



**Fig. 3 :** Correlation between TEX101 and zinc. n = 90, each group has 45 participate.



**Fig. 2 :** Correlation between TEX101 and Vitamin D. n = 90, each group has 45 participate.

factors, coital factors and idiopathic factors accounts for approximately 25% of infertility in men (Iqbal *et al*, 2009). Human TEX101 is a membrane GPI-anchored protein encoded by the *TEX101* gene, located in the 19q13.31 region of chromosome 19. According to the Human Protein Atlas, TEX101 expression is restricted to testicular tissue and male germ cells, with no evidence of expression in any other human tissue or cell type (Djureinovic *et al*, 2014). TEX101 is testis specific protein that exclusively expressed in germ cell of men so that it is an important biomarker of men infertility (Schultz *et al*, 2003; Drabovich *et al*, 2018).

**MATERIALS AND METHODS**

This study was conducted in the fertility Center’s of AL-Sader Medical city, Najaf Province, Iraq during the period from 1 Nov. 2018 to 31 Mar. 2019. Semen and blood specimens were collected from infertile patients in addition to control group (Normozoospermia) that attended to Al-Sader fertility center. 90 sample was collected from that 45 was infertile and 45 normal fertile men. Average age of patients is (34±9.5) years.

**Semen collection**

Normally human semen can be separated into seminal plasma and spermatozoa. For obtain seminal plasma depleted from sperm the semen was centrifuged which collected from healthy person with proved fertility and from men with infertility. By using ELISA kit to estimate the seminal plasma TEX101 while using colorimetric method of zinc estimation.

**Blood collection**

Blood samples were collected by vein puncture technique obtain 5 ml of blood from both infertile patient and fertile men as control. After that hold the blood in room temperature so as to clot than centrifuge it in 5000 round for 5 minutes to obtain serum which by using ELISA technique will estimate the concentration of vitamin D.

**RESULTS AND DISCUSSION**

The results in Table 1 were showed significant decrease in infertile group compare to fertile group in vitamin D, TEX101 and zinc.

**Biochemical and hormonal markers in infertile men  
Seminal plasma Testis expressed 101 in infertile men**

This study was showed significantly decrease

**Table 1** : Hormonal and Biochemical markers in fertile and infertile men.

Group	Fertile n = 45 Mean ± SD	Infertile n=45 Mean ± SD	P-value
Vitamin D (ng/ml)	17.58±1.35	12.84±0.78***	< 0.001
TEX101 (pg/ml)	1961.6±91.63	398.42±48.22***	<0.001
Zinc (µmol/L)	32.88±5.89	15.13±4.27***	< 0.001

(\*\*\*) indicates significance ( $p < 0.001$ ) compare to fertile group (independent T-test).

( $P < 0.001$ ) in seminal plasma concentration of TEX101 in infertile group ( $398.42 \pm 48.22$  pg/ml) compare to fertile group ( $1961.6 \pm 91.63$ ) as shown in Figure 1.

In a study indicated that TEX101 present in seminal plasma in free soluble. In healthy subjects TEX101 concentration was 5436 pg/ml, but in azoospermia was 500 pg/ml, oligospermia 450 pg/ml, fertile post vasectomy men, patient with sertoli- cell only syndrome and obstructive azoospermia had undetectable levels of TEX101. The study was demonstrated TEX101 as a reliable test for evaluation vasectomy success and azoospermia. Our current study can confirm that TEX101 decreases significantly in infertile men compare to fertile men (Korbakis *et al*, 2017).

In another study was demonstrated that TEX101 and epididymis-expressed ECM1 can be used as a differential marker for OA (obstructive azzospermia) and NOA (non-obstructive azoospermia). They proposed, this study can be used as replace must of testicular biopsies (Drabovich *et al*, 2013).

Considerable evidence suggest that vitamin D deficiency has adverse effect on fertility. The study done by Rehman *et al* (2018) stated that sufficient vitamin D has positive impact on spermatogenesis, total sperm count, motility and male reproductive function. And patients with infertility has lower serum concentration of vitamin D. In the regression model, one unit motility is related with 0.15 unit impact on vitamin D (Rehman *et al*, 2018). In the present study demonstrated that patients with infertility has a lower level of vitamin D concentration. Then our finding close to this study.

There was a study suggested that seminal plasma zinc level measurement is one of important marker for evaluation of infertility in men, and demonstrate that seminal plasma zinc level had significant positive relation with spermogram parameters such as sperm motility, sperm morphology and sperm concentration. low seminal plasma zinc level may be the cause of impaired sperm

function and as a result cause male infertility. This study was in accordance by Abed *et al* (2013), Kothari *et al* (2016).

### Correlation between TEX101 and other variables

#### Correlation between TEX101 and vitamin D

Fig. 2 showed significant ( $P < 0.001$ ) positive Pearson correlation ( $r = 0.935$ ) between TEX101 and Vitamin D.

#### Correlation between TEX101 and zinc

Fig. 3 showed significant ( $P < 0.001$ ) positive Pearson correlation ( $r = 0.906$ ) between TEX101 and zinc.

### CONCLUSION

This study showed that zinc element and vitamin D decrease affects the semen quality and as a result decrease fertilization rate in couples with male factor infertility. However, the TEX101 testicular protein can be regarded as a good marker of male fertility. Therefore, this study recommended on importance of performing TEX101 test for men who suffering from male cause infertility along with vitamin D and the element zinc as biochemical markers for men fertility condition.

### REFERENCES

- Abed A (2013) Essence of some trace elements in seminal fluid and their role in infertility. *Essence of some trace elements in seminal fluid and their role in infertility* 2(6).
- Ali F (2010) Prevalence and pattern of endocrinological abnormalities in oligospermic and azoospermic patients. *Fac Med Baghdad* 52, 4.
- Djureinovic D, Fagerberg L, Hallstrom B, Danielsson A, Lindskog C, Uhlen M and Ponten F (2014) The human testis-specific proteome defined by transcriptomics and antibody-based profiling. *Mol. Hum. Reprod.* 20, 476-488.
- Drabovich A P (2013) Differential diagnosis of azoospermia with proteomic biomarkers ECM1 and TEX101 quantified in seminal plasma. *Science Translational Medicine* 5(212), 212ra160-212ra160.
- Drabovich A P, Zhang J, Kanoatov M, Moskovtsev S and Librach C L (2018) Identification of testis- and germ cell-specific proteins as biomarkers of spermatogenesis and targets for sperm selection. *Fertility and Sterility* 110(4), e306-e307.
- Efesoy O, Çayan S and Akbay E (2009) The efficacy of recombinant human follicle stimulating hormone in the treatment of various types of male factor infertility at a single university hospital. *Journal of Andrology* 30(6), 679-684.
- Ghuman N and Ramalingam M (2018) Male infertility. *Obstetrics, Gynaecology & Reproductive Medicine* 28(1), 7-14.
- Hargreave T B and Mahmoud A M (2000) *WHO manual for the standardized investigation and diagnosis of the infertile male*. Cambridge University Press.
- Hala I, Abdul-Wahab R H, Muna A, Mohammad A and Mohammad A (2010) Evaluation of Serum Testosterone, Progesterone, Seminal Antisperm Antibody, and Fructose Levels among Jordanian Males with a History of Infertility. Article p: 8 pages.

- Iqbal A J (2009) Study of the hormonal change and the physiological criteria for patients with infertility. University of Kerbala. Pp: 70-88.
- Iyad F D (2009) Study effect of some chemo-biological changes in seminal plasma in patients with oligospermia. University of Inbar ISSN, 1; 3, 64- 67.
- Jungwirth A, Giwercman A, Tournaye H, Diemer T, Kopa Z, Dohle G and EAU Working Group on Male Infertility (2012) European Association of Urology guidelines on Male Infertility: the 2012 update. *European Urology* **62**(2), 324-332.
- Korbakis D (2017) Preclinical evaluation of a TEX101 protein ELISA test for the differential diagnosis of male infertility. *BMC Medicine* **15**(1), 60.
- Kothari R P and Chaudhari A R (2016) Zinc levels in seminal fluid in infertile males and its relation with serum free testosterone. *JCDR* **10**(5), CC05.
- McLachlan R I, Rajpert-De Meyts E, Hoesli-Hansen C E, de Kretser D M and Skakkebaek N E (2007) Histological evaluation of the human testis—approaches to optimizing the clinical value of the assessment: mini review. *Hum Reprod.* **1**, 2–16.
- Pedro J, Frederiksen Y, Schmidt L, Ingerslev H J, Zachariae R and Martins M V (2019) Comparison of three infertility-specific measures in men and women going through assisted reproductive technology treatment. *Journal of Health Psychology* **24**(6), 738-749.
- Rehman R, Lalani S, Baig M, Nizami I, Rana Z and Gazzaz Z J (2018) Association Between Vitamin D, Reproductive Hormones and Sperm Parameters in Infertile Male Subjects. *Frontiers in Endocrinology* **9**, 607.
- Schultz N, Hamra F K and Garbers D L (2003) A multitude of genes expressed solely in meiotic or postmeiotic spermatogenic cells offers a myriad of contraceptive targets. *Proceedings of the National Academy of Sciences* **100**(21), 12201-12206.
- Wong W Y, Thomas C M, Merkus J M, Zielhuis G A and Steegers-Theunissen R P (2000) Male factor subfertility: possible causes and the impact of nutritional factors. *Fertility and Sterility* **73**(3), 435-442.