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ASSESSMENT OF TOTAL ANTIOXIDANT CAPACITY, INTERFERON- GAMMA AND INTERLEUKIN-6 IN IDIOPATHIC INFERTILE MEN

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ABSTRACT

Infertility is an overall issue that portrayed by multifactorial etiology including physiological, hereditary, irresistible, ecological factors and in addition autoimmunity. The goal of this investigation is the connection between the aggregate cell reinforcement limit level with specific cytokines; interleukin-6 (IL-6) and interferon gamma (IFN-γ) levels in fundamental liquid and blood of idiopathic Infertility and ripe male relationship with original liquid level. Blood and fundamental liquids were gathered from 70 guys with idiopathic Infertility and 20 males with demonstrated Infertility and were broke down for the level of TAC, IL-6, IFN-γ and original liquid quality. Fundamental TAC levels were $p<0.05$ and lower in those with idiopathic Infertility. A negative relationship was seen between sperm quality parameters and TAC levels in idiopathic Infertility men. A fascinating noteworthy height in serum and semen IFN-γ level was recognized in idiopathic Infertility men. Furthermore, altogether lifted serum and semen IL-6 level was likewise seen in a gathering of idiopathic Infertility men. Inconclusion, TAC level has a critical negative impact on sperm quality and subsequently adds to male Infertility. Novel cytokine miniaturized scale designs for patients with various sorts of Infertility may be proposed and hence, cytokine level estimation of IL-6 and IFN-γ in fundamental plasma is recommended to be incorporated into assessment of particular kinds of male Infertility.

Keywords: Cytokines, Infertile, Total antioxidant capacity (TAC)

INTRODUCTION

The World Health Organization has characterized Infertility as the failure to imagine following one year of unprotected standard sex. Around 30% of cases include guys while around 35% include female factor and the rest of the speak to unexplained or idiopathic Infertility (WHO, 2010). The etiology of male Infertility is clearly a multifactorial one, could be physiological, neurotic, nourishing, inadvertent or even immunological variables (Khatoon et al., 2012). A high level of Infertility men neglects to impregnate their female partner as a result of oligozoospermia, asthenozoospermia tetratozoospermia or azoospermia (Feng et al., 2008). Mainly, the real wellsprings of cytokines in the human male conceptive tract are testicles and testicular macrophages, albeit a few cytokines, for example, IL-1 and IL-6 are additionally created by necessary testicular substantial cells, as Leydig and Sertoli cells (Fraczek et al., 2012). TNF-α, IFN-γ, IL-6, IL-8 notwithstanding a portion of their dissolvable receptors display in original liquid are likewise emitted by mesenchymal cells, safe cells, and spermatogonia (Qian et al., 2011). Aside from the part of cytokines in insusceptible adjustment, there is confirm that a portion of these polypeptides are specifically associated with the control of testicular capacity and furthermore might be strong modulators of steroid discharge from the testicles (Inhorn, 2003, Araoye, 2003). Interleukin-6 is a cytokine yield by both lymphoid tissue and in addition non-lymphoid cells (Hirano et al., 2011). The activity of IL-6 in testis is diminishes transferrin discharge in Sertoli cells and might represses spermatogenesis process and sperm motility. An expanded level of IL-6 in fundamental could be related in male Infertility. Interferon-γ is a professional fiery cytokine discharged by initiated T cells and additionally regular executioner cells, which can be raised in the fundamental liquid of patients with genitorurinary aggravation. IFN-γ is created by Sertoli, Leydig, and peritubular cells and has an administrative part in Leydig cell steroidogenesis process. What's more, IFN-γ is associated with ensuring the testis against viral diseases and plays an essential safe administrative part by empowering antigen-particular safe reactions (Carrasquel et al., 2003). Primarily, oxidative pressure may think about a fundamental danger of obscure men Infertility (Valko et al., 2007) which happen as a lopsidedness among the yield of responsive oxygen species (ROS) and their searching or balance by the cell reinforcement framework (Agarw et al., 2014,

Aitken et al., 2010). It is trusted that controlled levels (physiological levels) of these ROS are important for sperm typical physiology and preparation though, uncontrolled creation of ROS (neurotic levels) may take an interest in sperm dysfunctions (de Lamirande et al., 1997). In ripe male, add up to cancer prevention agent limit (TAC) and ROS creation stay in harmony or adjust (Saleh et al., 2002). Normally, cell reinforcements found in original liquid incorporate vitamins, glutathione, superoxide dismutase and additionally thioredoxin (Sanocka et al., 2004). A few examinations have alluded to the part of abnormal state fundamental ROS in male Infertility (Sharma et al., 1996 and Chen et al., 2012). Hence, the correct causes and pathway of the cytokines in the human men genital tract is still under examination. The relationship between original plasma TAC and cytokines, for example, IL-6 and IFN-γ and their relationship with sperm parameters are controversial, subsequently this investigation expected to investigate the TAC, IL-6 and IFN-γ levels in the fundamental liquid (and serum) in men with idiopathic Infertility and their relationship with fundamental liquid quality parameters.

MATERIALS AND METHODS

SUBJECTS: Nineteen guys were taken an interest in the examination; seventy members guys with age (18-50 years) were enrolled to the IVF and Infertility focus in Kamal Al-Samarae doctor's facility in Baghdad and Al-Sadder therapeutic city healing facility in Al-Najaf (Iraq) for the period from August 2016 to March 2017 griping of Infertility were incorporated into this investigation. These were separated into three gatherings; 25 oligozoospermia, 25 asthenozoospermia and 20 tetratozoospermia. Twenty guys of comparing ages to the patients with demonstrated richness were arbitrarily chosen as controls. All members were non-paid volunteers and a composed educated assent was acquired from each subject. Exclusion criteria were; particular genital diseases (irresistible or non-irresistible), un-slid testicles, foundational infection and testicular atrophy.

All original liquid examples were gathered by masturbation in sterile polypropylene holders following 3-5 long periods of restraint. Original liquid examples were condensed at 37°C for 30 minutes and routine semen examination was performed according to WHO rules (Khatoon et al., 2012). Fundamental plasma was gotten from full melted discharges and were centrifuged at 300 g for 20 minutes.

SPECIMEN COLLECTION: A standard semen liquid investigation was performed upon liquefaction as per WHO Manual (WHO, 2010) to quantify volume, appearance, pH, smoothness and essential sperm parameters. The routine original liquid investigation was done evaluating sperm thickness, motility, reasonability and in addition morphology. The rest of the semen test was centrifuged at 1000 ×g for 10 min; the original plasma was isolated for three equivalent amounts of and put away at - 70 °C until the point that further investigations.

Estimation of IL-6, IFN-γ and TCA in serum and fundamental plasma: Solid stage resistant examines were utilized for quantitative measure of IL-6, IFN-γ (sandwich ELISA units from Pepro-Tech, UK) and TAC (aggressive ELISA pack from My Bio-source, USA) in serum and original plasma.

Statistical Analysis: Statistical Package for Social Sciences (SPSS) variant 20 was utilized to gauge mean, standard deviation, standard Error, 95% certainty interim for mean qualities, and two outrageous qualities, least and most extreme. Chances proportion was connected for estimating quality of affiliation. We set P esteem ≤ 0.05 as noteworthy esteem.

RESULTS AND DISCUSSION:

The TAC level was resolved in original plasma though IL-6 and IFN-γ was resolved in serum and fundamental plasma from prolific and Infertility guys.

Table 1 speak to all-encompassing introduction of the investigation results in all examination gatherings (oligozoospermia, asthenozoospermia, teratozoospermia and in addition control) concerning semen quality parameters, TAC in original plasma, and IL-6, IFN-γ in serum and fundamental plasma. The mean qualities (U/ml) ± standard deviation (SD) of the TAC in these gatherings were 28.47 ±9.66, 23.46±6.62, 14.08±4.21 and 65.75±8.93 separately. Results demonstrates that the most elevated mean esteem was accounted in charge gathering, and the least mean esteem was in teratozoospermia gathering. A positive connection was seen between sperm quality parameters and TAC levels in all examination gatherings. For IL-6 results in serum, the most elevated mean focus (pg/ml) was seen in the asthenozoospermia subgroup of the idiopathic Infertility men 20.53±5.89, though the least fixation (pg/ml) was seen in the ripe men aggregate 9.04±1.10. Concerning the mean grouping of IL-6 in original plasma, the example resembled that of the IL-6 mean focus in serum. The example of the IFN-γ mean fixation (pg/ml) in both serum and fundamental plasma was comparative as the most noteworthy was seen in the oligozoospermia subgroup of the idiopathic Infertility men in

serum 20.71±7.60, and 13.36±3.46 in original plasma contrasted with the least focus which was seen in the ripe men gather 9.31±0.62 in serum and 7.56±0.85 in fundamental plasma.

Table 1: Semen parameters, TAC, IL-6 and INF- γ mean values in study groups.

Semen Parameters and study markers	Men with Idiopathic Infertility			Fertile Men (n=20) Mean±SD	*P-value*
	*Olig (n = 25) Mean± SD	**Asth(n = 25) Mean± SD	***Terato(n = 20) Mean± SD		
Volume (ml)	2.03±0.71	2.18±0.58	2.85±0.93	3.70±0.85	HS*
Sperm Count (×10^6)	5.32±2.08	47.16±15.97	41.55±12.28	48.60±12.95	HS*
Active linear progressive motility (%)	43.24±8.61	10.60±4.16	41.35±5.23	41.55±5.59	HS*
Sperm with normal morphology (%)	57.60±12.59	59.00±12.25	3.3±0.66	65.75±8.93	HS*
TAC in ****SP(U/ml)	28.47±9.66	23.46±6.62	14.08±4.21	60.36±10.73	HS*
IL-6 in serum (pg/ml)	15.11±6.77	20.53±5.89	14.63±8.01	9.04±1.10	
IL-6 in SP (pg/ml)	25.56±7.89	28.00±6.63	22.93±7.94	22.00±45.4	S**
INF-γ in serum (pg/ml)	20.71±7.60	16.60±6.89	17.92±7.02	9.31±0.62	HS*
INF-γ in SP (pg/ml)	13.36±3.46	13.28±3.07	12.76±4.51	7.56±0.85	HS

*Olig= Oligozoospermia, **Asth=Asthenozoospermia, ***Terato=Teratozoospermia, ****SP= Seminal plasma

Figures1, 2, 3, 4 and 5 represents graphically and stem-leaf plot the mean values for studied readings of TAC (U/ml) in seminal plasma, IL-6 (pg/ml) in serum, IL-6 (pg/ml) in seminal plasma, IFN-γ in serum and IFN-γ in seminal plasma respectively in idiopathic infertile subgroups and control.

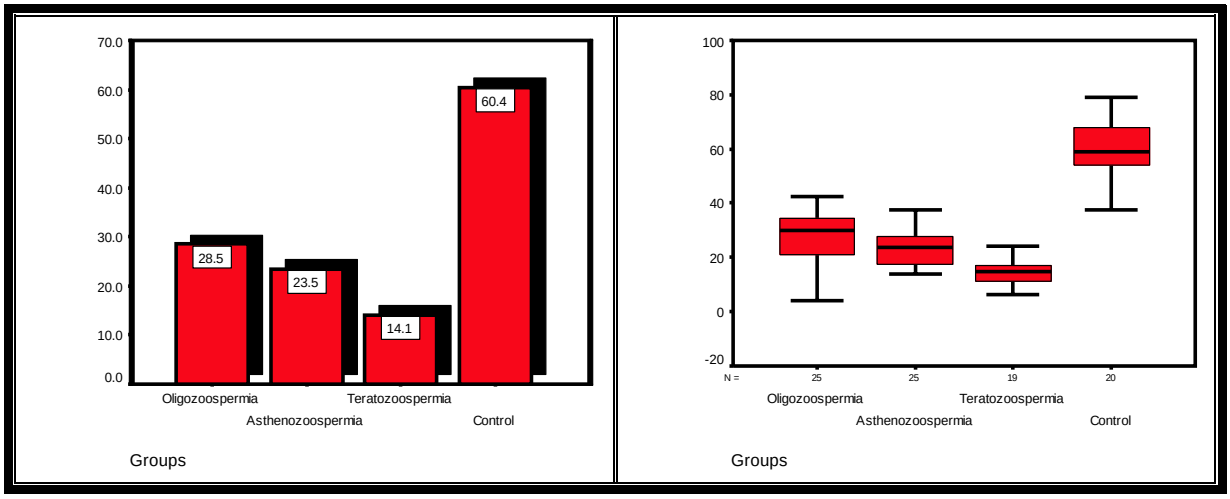

Figure 1: Mean values and Stem – Leaf plot of TAC (U/ml) in seminal plasma of different groups

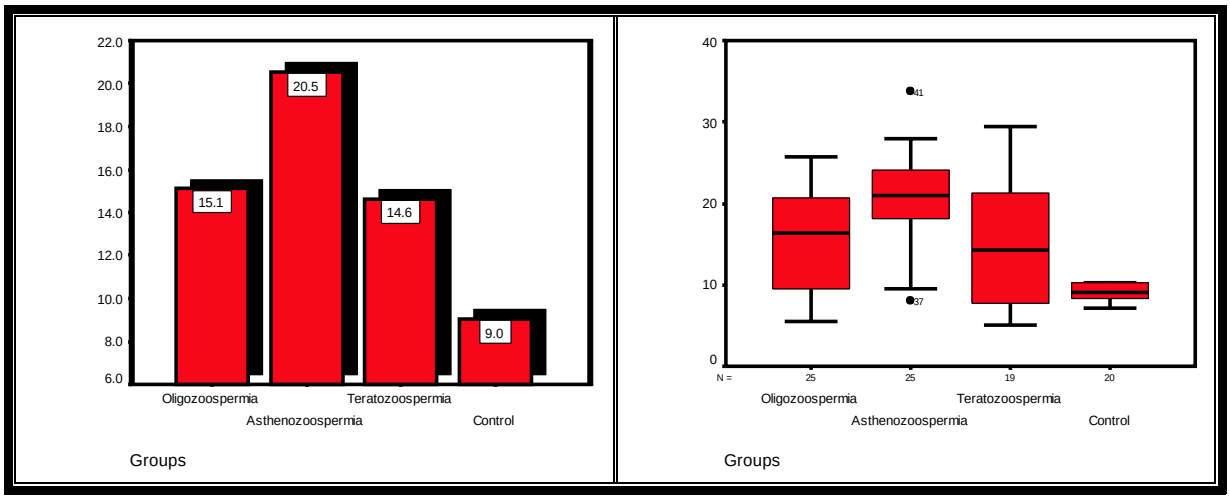

Figure 2: Mean values and Stem – Leaf plot of IL-6 (pg/ml) in serum in different groups

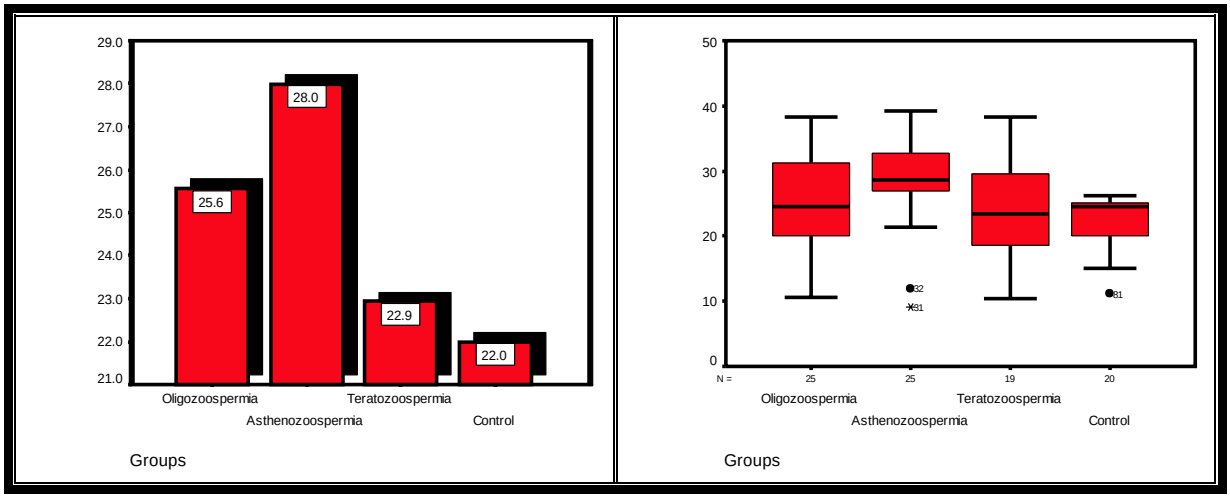

Figure 3: Mean values and Stem - Leaf plot of IL-6 (pg/ml) in seminal plasma in different groups

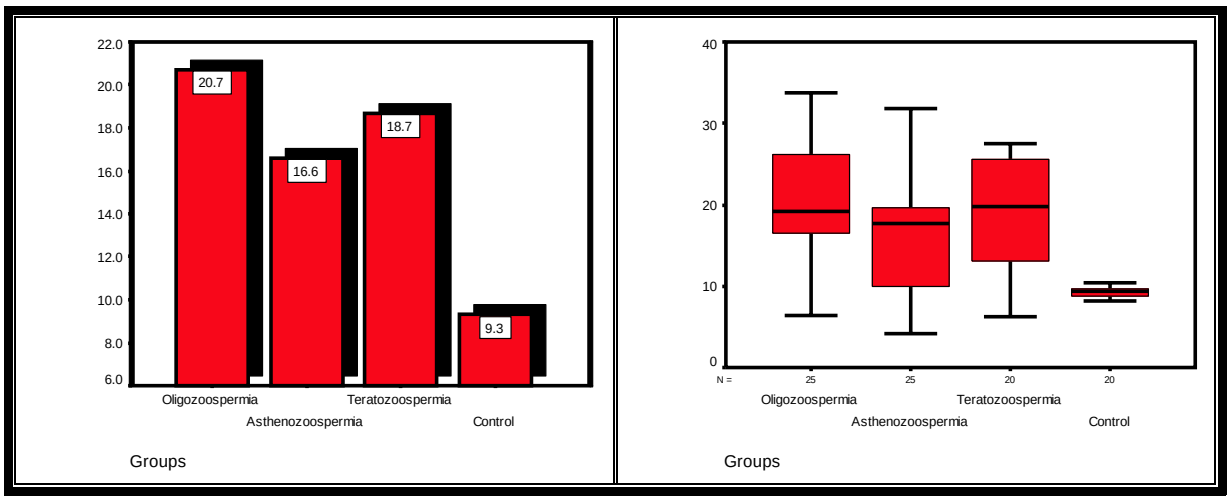

Figure 4: Mean values and Stem - Leaf plot of IFN-γ (pg/ml) in serum in different groups

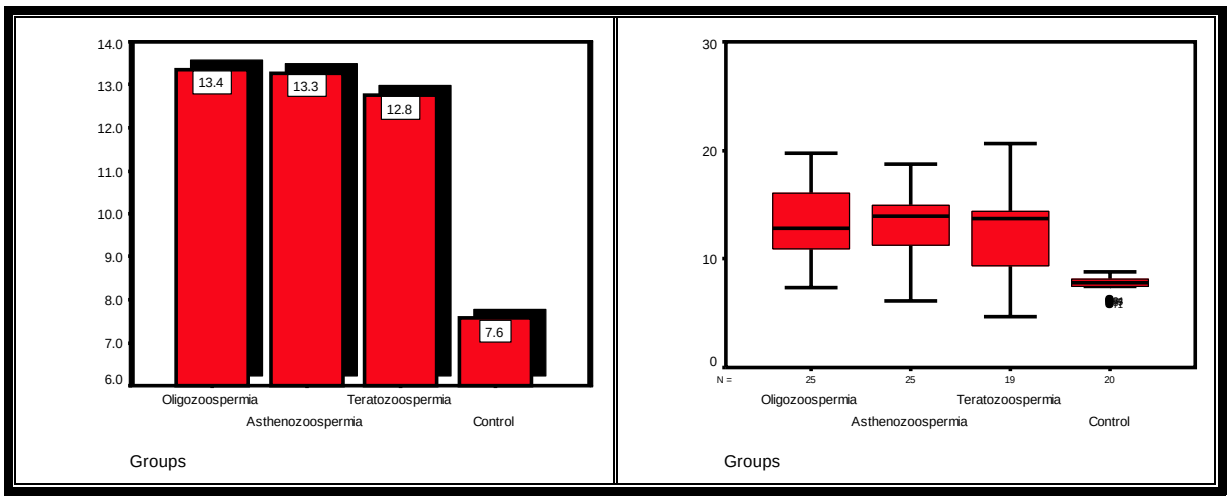

Figure 5: Mean values and Stem – Leaf plot of IFN-γ (pg/ml) in seminal plasma in different groups

In the present data, bring down levels of TAC were seen in men with idiopathic infertility than in fruitful men. A positive connection was appeared between ordinary sperm, motility, morphology, and TAC levels in all investigation gatherings. This outcome would affirm numerous other prior investigations which likewise finished up the positive relationship between the TAC and the semen quality parameters (Hosseinzadeh et al., 2013, Mahfouz et al., 2009, Pahune et al., 2013, Koca et al., 2003, Appasamy et al., 2007). Such consistency in this issue let us to report it as a reality and to consider it as the main factor in the inclination towards men idiopathic Infertility. As

needs be, TAC might be utilized as one of a kind biomarker for examining the oxidative worry in sperms.

Men Infertility thought about a genuine medical issue and, in spite of significant advances in its conclusion and treatment, its specific etiology stays obscure. Be that as it may, it is trusted that original oxidative pressure could be one of the primary factors in the pathogenesis of idiopathic Infertility (komran et al., 2013, Yousif et al., 2011). Primarily, high levels of fundamental ROS may prompt sperm DNA harm and lipid peroxidation, and in the end loss of sperm motility (Pasqualotto et al., 2008, Khosrowbeygi et al., 2004). For this situation high measures of polyunsaturated unsaturated fats are gathered in the spermatozoa plasma films, which would predispose to oxidative damage. Not with standing that, ROS could likewise prompt base modifications, DNA cross-joins, DNA strand breaks and in addition chromosomal adjustments (Zini et al., 2009, Iwasaki et al., 1992). Thus, a few investigations have endeavored to think about the impacts of cancer prevention agent supplements on male Infertility (Kemal et al., 2000, Keskes-Ammar et al., 2003). In spite of the fact that the impacts of various dosages and sorts of oral cancer prevention agents in endeavors to enhance original liquid parameters have not been set up, it is trusted that cell reinforcement treatment has valuable effects on male Infertility (Dawson et al., 1992, Greco et al., 2005). Additionally, the consequences of cell reinforcement treatment alone or in blend are controversial. For instance, a few examinations have watched that vitamin C, vitamin E, folic corrosive, selenium, and zinc alone or in blend could enhance fundamental liquid parameters, especially sperm motility and sperm DNA uprightness (Wong et al., 2002, Havrylyuk et al., 2015). The outcomes alluded to general height in the mean focus worried of both IL-6 and IFN-γ in all idiopathic Infertility men bunches in correlation with the rich men gathering. This height was clearer in the serum for IL-6 chiefly in the asthenozoospermia gathering, while this rise was seen in both serum and original plasma mostly in the oligozoospermia gathering. Around comparable outcomes were accounted for by different investigations (Paradisi et al., 1996, Dousset et al., 1997). that affirm the nearness of these cytokines in fundamental plasma and alluded to a conceivable relationship between these professional incendiary cytokines and expanding levels of oxidative worry and also sperm quality parameters. The relationship amongst cytokines and disabled proliferation speaks to an intriguing issue in light of their application in different parts of fruitfulness control, including gonadal and sperm capacities. The human original plasma contains a collection of cytokines including epidermal development factor (EGF), TGF, TNF-α, different interleukins and huge numbers of their solvent receptors. The sufficiency of negative relationship between's original plasma cytokines and sperms quality parameters including lessen the ova-entering capacity of spermatozoa and ROS is as yet subject to discuss (Hedger et al., 2003, Yousif et al., 2013). Then again, a conceivable part of cytokines in nearby direction of sex hormones and paracrine control of proliferation forms, including spermatogenesis process is still under scrutiny. One illustration is the impact of IL-6 on spermatogonial multiplication; Sertoli cells stereidogenesis, germ cells separation (Hukkanen et al., 1995). Some in vitro ponders, confirmed that cytokines can prompt ROS generation as delivered of inducible NOS by mouse, rodent and human osteoblast-like cells with no obviously characterized instrument (Martínez et al., 2007, Slimani et al., 2014). Other examination (Naz et al., 2000), demonstrates that cytokines can influence straightforwardly on spermatozoa to animate their level of lipid peroxidation process, showing a direct receptive oxygen animal varieties creation by the spermatozoa. The intriguing finding is that two receptors for cytokines were situated on human spermatozoa, IFN-γ receptors (Laflamme et al., 2005), and additionally IL-6 receptors, both particular ligands for the authoritative of the gp80 subunit (IL6-Ra) (Yoshida et al., 2004), and for the flag transducing protein gp-130 (IL-6Rb) (Havrylyuk et al., 2015). A few creators expressed that the impact of cytokines on ROS level and in the long run sperm quality parameters is a measurements subordinate system (Tsuchimoto et al., 2015).

To distinguish the real effect of cytokines in male idiopathic Infertility, facilitate pre-clinical and creature display contemplates are required inside the field of immunology and sub-atomic science to research the impact of cytokine-hindering on enhancing the sperms quality and execution in such patients. A few creators recommending the expansion of these aftereffects of immunological information tending to the discovering whether these elements may profile male Infertility with particular cytokine micropattern for every Infertility issue enabling them to be better separated and all the more unequivocally analyzed and after that treated utilizing more refresh way (Camejo et al., 2001). Appraisal of original plasma cytokine level is as yet not a normal practice (Arunkumar et al., 2018).

All in all, a positive connection was uncovered between sperm check, sperm motility, and ordinary sperm morphology, with TAC levels in both idiopathic Infertility men and prolific men. In this way,

TAC could be utilized as oxidative pressure markers, and their tests could be valuable in the assessment of idiopathic infertility. What's more, these markers may manage us in antioxidant treatment. What's more, this information may endorse a novel cytokine miniaturized scale designs for male diverse reasons for Infertility. Original plasma cytokines; IL-6 and IFN-γ ought to be incorporated into assessment of particular kinds of male Infertility. By and large, this investigation has watched noteworthy control in cytokines small scale design concerning diverse companions of Infertility patients, with idiopathic Infertility.

CONCLUSION

In light of the outcomes acquired, the information proposes broadening infertility research by assessing IL-6 and IFN-γ in blood serum and in fundamental plasma that clear up whether these elements may have novel key to the clinical work-up of male infertility administration.

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COMPETING INTEREST: Authors declare that they have no competing interest.

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