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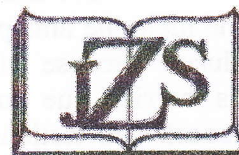
Randomized Blind Controlled Study of the Incidence of Seminal Antisperm Antibodies in Male with Primary Infertility in Kirkuk Governorate.



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Abstract

This is a randomized blind controlled prospective study included 123 males with primary infertility referred from private clinics from May 2004 to May 2005. ELISA was used to detect the antisperm antibodies in the seminal fluid of infertile males (60 patients with normal seminal fluid analysis and 63 patients with abnormal seminal fluid analysis). It was found that there was no significant difference in the incidence of antisperm antibodies between infertile males with normal seminal fluid analysis (10%) and those with abnormal seminal fluid analysis (15.8%) $p>0.05$, but most of infertile males with abnormal seminal fluid analysis (8 out of 10) their antibody titre was within high level (100- 115) U/ml, while 5 out of 6 infertile patients with normal seminal fluid analysis, their antibody titre was with low level (60- 75) U/ml. The high concentration of antisperm antibody was found in the age group (36-45) years in both groups (infertile patients with normal seminal fluid analysis and abnormal seminal fluid analysis) and most affected infertile males were those with duration of infertility of 6-10 years. There was significant association ($p<0.05$) between smoking habit and abnormal seminal fluid analysis, while there was no significant difference between smoker and non smoker infertile males in the incidence of antisperm antibody ($p>0.05$).

Keywords: seminal antisperm antibodies, infertile males, Kirkuk.

Introduction

Infertility defined as inability to conceive within 12 months of unprotected intercourse [1]. The incidence is more than 20% of all couples in reproductive age [2], and is classified as explained and unexplained infertility (couple with normal basic investigation: which include, seminal fluid analysis, ovulatory assessment, hysterosalpingeography, laproscopy and post coital test) [3,4].

The causes of infertility may be found in male or female partner, male infertility may be contributory in 30% to 40% of infertile couples [5], the causes may be in the form of defective spermatogenesis, antisperm antibodies , defective sperm transport which include, seminal pathway blockage due to obstruction in the epididymis or ejaculatory duct.

Antisperm antibodies are defined as immunoglobulines of IgG, IgA, and /or IgM isotypes that are directed to various aspects of spermatozoa including head, tail, midpiece or combination [6,7] and are claimed to cause explained and unexplained infertility.

In both men and women, antisperm antibodies may be found in the serum and /or reproductive tract secretion.

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The incidence of antisperm antibodies is 10% in the males and 5% in the females [8]. In the male the antisperm antibodies can be produced because of disruption of blood testis barrier due to vasectomy reversal, varicocel, and testicular torsion, congenital absence of vas deferens, testicular biopsy, cryptorchidism, testicular cancer, and infection [9-11]. The antisperm antibodies cause disordered spermatogenesis resulting in defects of sperm motility and concentration [12-14], interfere with migration through the female reproductive tract causing asthenospermia[15], cause autoaglutination of ejaculatory spermatozoa and disordered of sperm capacitation and also claimed to block sperm –ovum interaction.

In the clinical andrology, the detection of antisperm antibodies is regarded as one of the steps of infertility evaluation [16], the detection tests are quantitative and qualitative. The quantitative detection of antisperm antibodies detects immunoglobulines of IgG, IgA , IgM, etc levels that play a significant role in immune defense by using Enzyme - linked immunobsorbent (ELISA) technique [17], which is simple, rapid, and reliable test, not need fresh sample. Therefore, ELISA is an ideal assay method for the determination of antibodies directed against human spermatozoa. While qualitative detection of antisperm antibodies detects each immunoglobulin class separately such as complement dependent cytotoxicity which detects IgA and mixed agglutination reaction which detects IgG.

Antisperm antibodies can be treated medically by using steroidal and non steroidal drugs but the success rate is limited, while the application of variety of assisted reproductive technologies such as in vitro fertilization and intracytoplasmic sperm injection is the most effective treatment for antisperm antibodies [6, 7, 18].

Patients and Methods

The study included 123 infertile males were referred for infertility evaluation from private clinic from May 2004 to May 2005. After full history and clinical examination from male partner, seminal fluid was obtained by masturbation after 3-7 days of sexual abstinence, and examined according to WHO (world health organization) criteria of normal seminal fluid which includes: semen volume 1.5-3ml, normal sperm count $\geq$20 million /ml, actively motile sperm $\geq$ 50%, sperm morphology $\geq$30%.

123 infertile males were divided in to two groups:

A- Those with normal seminal fluid analysis. (60 patients)

B- Those with abnormal seminal fluid analysis. (63 patients)

Semen samples from all patients were collected for detection of antisperm antibodies in the seminal plasma by using ELISA kit. ELISA test utilize an antiglobulin to which an enzyme has been covalently linked. The antibody titres were measured colorimetrically, an absorbance at a wave length of 450nm greater than 60U/ml considered positive. The tittering of antisperm antibody levels of patient samples was obtained by the calibration curve of absorbance value for each sample and the corresponding concentration of antisperm antibody in U/ml.

For the statistical analysis, Chi square test was used.

Result

Among 60 patients with normal semen analysis, positive antisperm antibodies was observed in 6 (10%) patients with normal seminal fluid analysis compared with 10 (15.8%) patients with abnormal seminal fluid analysis. Although the incidence of antisperm antibodies in patients with abnormal seminal fluid analysis was higher than in those with normal seminal fluid analysis , the difference between the two groups was not statistically significant ($p>0.05$) table (1).

Table (2) shows the titre of antisperm antibodies detected by ELISA in infertile males, in patients with normal semen analysis, 5 cases from 6 positive cases the value was between the low titre (60-75)U/ml and only in one case the value was between (80-95)U/ml. While in patients with abnormal semen analysis most positive cases (8 out of 10) positive cases their value was in between (100-115) U/ml, and only in 2 remaining cases the value was between (80-95) U/ml.

It is indicated in the table (3) that high incidence of antisperm antibodies was among (36-45) years of age followed by (46-55) and (25-35) years respectively. Although there was no statistically significant difference between the groups in infertile males with normal and abnormal seminal fluid analysis, but the highest rate of incidence was among (36-45) years old in both groups (patients with normal and abnormal seminal fluid analysis) .

Regarding the duration of infertility, it was found that the most affected males were those with the duration of infertility of 6-10years, but no pattern of an increase in the percentage of their having antisperm antibodies was observed as the duration of infertility was prolonged as demonstrated in table (4).

The smoking rate in infertile males with abnormal semen analysis was higher (50.79%) than in patients with normal semen analysis (28.33%) and it was statistically significant ($p<0.05$) table (5).

Although the incidence of antisperm antibodies in smoker infertile males was higher (14.28%) than in those non smoker infertile males, but the difference was not statistically significant between the two groups ($p>0.05$) table (6).

Table (1) Distribution of antisperm antibodies among infertile males with normal and abnormal seminal fluid analysis.

S.F.A.	No. examined	No. positive	% positive
Normal	60	6	10
abnormal	63	10	15.8
Total	123	16	25.8

$\chi^2 = 6.468$ d.f = 1 ($p>0.05$)

Table (2) Titres of positive antisperm antibodies tests among infertile males

S.F.A.	No. positive	Titres (U/ml) of ASA		
		60- 75*	80-95	100-115
Normal	6	5	1	0
Abnormal	10	0	2	8

- >60 regarded positive

Table (3) Distribution of antisperm antibodies among infertile males according to the age

Age (years)	Patients with normal S.F.A.			Patients with abnormal S.F.A		
	No. examined	No. positive	% positive	No. examined	No. positive	% positive
25-35	34	0		24	1	4.16
36-45	20	5	25	34	8	23.25
46-55	6	1	16.66	15	1	6.66
Total	60	6	41.66	63	10	88.34

$\chi^2 = 4.02$ d.f = 2 ($p>0.05$)

Table (4) Distribution of antisperm antibodies among infertile males according to the duration of infertility.

Duration(years)	No. examined	No. positive	% positive
1-5	86	1	1.162
6-10	22	10	45.45
11-15	15	5	33.33

$\chi^2 = 36.6$ d.f = 2 ($p>0.05$)

Table (5) Smoking habit in relation with seminal analysis in infertile males.

S.F.A.	No. examined	No. smokers	% positive
normal	60	17	28.33
abnormal	63	32	50.79
Total	123	49	78.112

$\chi^2 = 6.468$ d.f = 1 ($p<0.05$)

Table (6) Distribution antisperm antibodies among smoker and non smoker infertile males.

	No. of smokers	No. positive for ASA	% positive
smokers	49	7	14.285
non smokers	74	9	12.162
Total	123	16	26.447

$\chi^2 = 0.117$ d.f = 1 ($p>0.05$)

Discussion

The role of antisperm antibodies in male infertility remains controversial, it may be regarded as a relative rather than absolute cause of infertility[5].

In our study, although the incidence of antisperm antibodies in infertile males with abnormal seminal fluid analysis was higher than those with normal seminal fluid analysis (15.8% Vs 10%), it was not statistically different , but most of patients with abnormal seminal fluid analysis (8 out of 10) positive case , their antisperm antibodies titre was within high level (100-115 U/ml) in contrast to the infertile males with normal seminal fluid analysis, (5 out of 6) of them was within low level (60- 75 U/ml).

Many studies indicated the association between antisperm antibodies and abnormal semen parameters. Kipersztok [19], Devine [16],and Brandy [20] found that positive semen samples had a significant abnormal sperm count, motility, morphology, volume, liquefaction time and white blood cell count, while this association was have not been noticed in other studies such as Munuce [21] and Carizza [22] who could not found association between antisperm antibodies and semen parameters.

This controversy in finding may be due to sample of infertile males who were selected for the study and to different methods used for detection of antisperm antibodies.

Regarding the titre of antisperm antibodies, Handersleman [23] found that the high titres of antisperm antibodies associated with reduced normal semen parameters, and Hojori [24] recorded that the low level of antisperm antibodies do little harm in male infertility, many previous studies also found that the high titres of antisperm antibodies very rarely occur except in patients with fertility problems [25,26].

The highest incidence of antisperm antibodies was observed in infertile males with age group (36-45) years and those with duration of infertility (6-10) years. This may be due to that this test is not routinely used for evaluation of infertile patients which cause delay in the detection and giving the appropriate treatment.

Although the smoking rate in infertile males with abnormal semen parameters was significantly higher than in those with normal semen parameters, but no statistically significant difference was observed in the incidence of antisperm antibodies between infertile smokers and non smokers, this may be attributed to the smoking association with nitrous oxide level in the semen which affect adversely on normal semen parameters and fertility [27,28], while the treatment of antisperm antibodies in infertile males associated with the lowering of nitrous oxide level in

semen and improvement of semen quality [29]. This lead to prognosis the possible relation between antisperm antibodies and smoking habit in infertile males.

Antisperm antibodies interfere with human reproductive events, and once they are bound to spermatic surface , they affect both the transport of spermatozoa and interaction between gametes. The formation of antisperm antibodies has still not been completely explained and the antigen map of spermatic surface has not been yet established, we have many diagnostic procedures but actually very few therapeutic options [30].

But in recent years owing to the improvement and spreading of IFV (in-vitro fertilization) techniques, it has been possible to demonstrate the effect of antisperm antibodies at the level of in-vitro gamete interaction. The literature demonstrates that the various previously used treatments for immunological infertility, i.e. medical therapy, intrauterine insemination with husband's spermatozoa and IVF, usually have poor success. The primary choice of treatment in infertility, especially in the most severs cases and when the sperm head is involved is ICSI (intra- cytoplasmic sperm injection). Antisperm antibodies evaluation in all couples who undergo the various techniques of insemination or IVF is imperative [31].

Conclusions

1. Most of infertile males with abnormal seminal fluid analysis have antibody titres with high levels(100-115 U/ml), while infertile males with normal seminal fluid analysis have antibody titres with low levels (60-75 U/ml).

2-There was significant association between smoking habit and abnormal seminal fluid analysis.

References

1. WHO laboratory manual for the examination of human semen and semen cervical – mucous interaction. *Cambridge Uiversity Pess.* **2001**.
2. Lomardo, F.; Gandini, L.; Dondaro F.; et al. Antisperm antibodies in natural and assisted reproduction . *Hum. reprod.Update.* **2001**; 7(5):450-456.
3. Price GA.; Cornelissen CN.; and Hobbs MM. Immunogenicity of gonococcal transferring binding proteins during maternal infection. *Infect.Immun* ..**2004**; 72(1):277-38.
4. Westerlaken L. ; Helmerhorst F.; Dieben S. ; et al. Intradytoplasmic sperm injection as a treatment for unexplained total fertilization failure or low fertilization after conventional in vitro fertilization. *Fertility Sterility.* **2005**; 83(3):612-617.
5. American society for reproductive medicine. *Formely the American fertility society.* 4/**2001**.
6. Bouvent BR. ; Brufman AS.; and Paparella CV. ; et al. Macrofagos en semen humano. *Arch.Esp.Urol.* **2003**; 56(9):983-987.
7. Chamley L.W.C. ; and Chiu W.W.C. Use of antisperm antibodies in differential display Western blotting to identify sperm proteins important in fertility. *Human reproduction.* **2002**; 17(4):984-989.
8. Bohring C.; and Krause W. Immune infertility: Toward a better understanding of sperm (auto) - immunity. The value of proteomic analysis. *Human reproduction.* **2003**; 18(5):915-924.
9. Borges JE. ; Rossi FL; Posqalotto FF.; et al. Different interval between vasectomy &sperm retrieval interfere in reproductive capacity from vasectomized men . *J. Assist Reprod.Genet.* **2003**; 20(1): 33-37.
10. Munce M.; Bregni C.; Carizza C.; et al. Semen culture, leuckospermia and the presence of sperm antibodies in seminal hyperviscosity. *Arch. Androl.* **1999**; 42(1):21-8.
11. Kay DJ. ;Hosken B. ; Boettcher B. ; et al. Antispermatozoal antibodies in three men with infertility due to congenital aplasia of the vas deferentia. Am. J.Reprod. Immunol. Microbiol.**1988**; 17(2):48-52.
12. Taylor C. Bilijan M.; Kingslad C. ; et al. Inhibition of human spermatozoa- oocyte interaction in vitro by monoclonal antibodies to CD46. *Human reproduction.* **1994**; 9:907-911.
13. Salma R.; Eustache F.; Duot B.; et al. The pregnancy &semen parameters: a cross sectional study among fertile couples from four European cities. *Human reproduction.* **2002**; 17(2):503-515.
14. Shaup P; and Folstad I. Dose immunity regulate quality and fertility in human? *Behavioral Ecology.* **2005**; 16(2):410-416.
15. Mc;achlan RI. Basis, diagnosis and treatment of immunological infertility in men. *J. Reprod. Immunol.* **2002**; 57(1-2)35-45.
16. Kastner C.; Jakse G. Measurment of immunoglobulins in seminal fluid with modified nephelometry- an alternative diagnostic tool for chronic prostitis. *Prostate. Cancer. Prostatic. Dis.* **2002**; 68-89.

ڕێژەی دژە سپێرم لە نەخۆشی نێر ینەكە نەزۆكی سەرەتای یان هەیە لە شاری كەركوك

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پوختە

ڕێژەی دژە سپێرم لە پلازمای تۆماو لە (١٢٣) نێزی نەزۆك كرا بۆ (٦٠) نێر شی كردنەوەی تۆماویان نورماڵ بوو (٦٣) سستی سپێرمیان هەبوو. بە بەكارهێنانی ئامێری Eliza . ئەنجامەكانی توێژینەوەكە پیشانی دا كەوا جیاوازیەكی ئەو تۆنی لە نێوانی هەردوو گروپەكەدا بەلام (٨ لە ١٠) نێر كەشی كردنی تۆماویان سستی سپێرمی هەیە ڕێژەی سپێرم زۆر بەرزبوو (١٠٠-١١٥ یەكە/ ١ سی سی) بەلام ئەو گروپەی تۆماویان نۆرماڵ بوو ڕێژەكە (٦٠-٧٥ یەكە/ ١ سی سی) هەروەها توێژینە-وەكە پیشانی دا كەوا ڕێژەی دژە سپێرم لە نێوان جگەرە كێشان و جگەرە نە كێشان هیچ جیاوازیەكەی نی یە.

دراسة مستقبلية حول نسبة حدوث أضداد الحيامن لبلازما السائل المنوي لدى الذكور العقيمين ذو العقم الأولي في مدينة كركوك.

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الخلاصة

تم تقدير نسبة حدوث أضداد الحيامن في بلازما السائل المنوي ل١٢٣ ذكر عقيم (٦٠ ذكر ذو معايير السائل المنوي السوية و٦٤ ذكر ذو معايير السائل المنوي الغير السوية) باستخدام اللوازم الخاص بالامتصاص المناعي المرتبط أنزيميا". أظهرت نتائج الدراسة بان ليس هنالك اختلاف ملموس إحصائيا" في نسبة حدوث أضداد الحيامن بين الذكور العقيمين ذو معايير السائل المنوي السوية والذكور العقيمين ذو معايير السائل المنوي الغير السوية. ولكن اغلب الرجال العقيمين (٨ من ١٠) ذو معايير السائل المنوي الغير السوية لديهم معايير أضداد الحيامن ضمن المستويات العالي (١٠٠-١١٥ وحدة/مل) أما الذكور العقيمين ذو معايير السائل المنوي السوية كانت لديهم معايير أضداد الحيامن ضمن المستويات الواطئة (٦٠ -٧٥ وحدة / مل). وكذلك أظهرت الدراسة ان أكثر نسب حدوث أضداد الحيامن ظهرت في مجاميع الأعمار (٣٦-٤٥سنة) للذكور العقيمين ذو معايير الحيامن السوية والغير السوية واغلب هؤلاء الذكور كانت لديهم فترة العقم (٦-١٠ سنوات). كما لم تظهر الدراسة وجود اختلاف إحصائي ملموس في نسبة حدوث أضداد الحيامن في الرجال العقيمين المدخنين والغير المدخنين.

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وەرگرتن لە ٢٠٠٦/٣/٢٥دا و پەسەندكردن لە ٢٠٠٧/٢/٦دا.