

Serum Protamine 1 Level in Oligozoospermia Patients in Najaf Province and its Correlation with Some of Seminal Fluid Parameters

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Abstract

Objective :The study was performed to estimate the concentration of Protamine 1 (PRM 1) level in serum of oilgozoospermia patients (40 person) that attended to fertility center in al Sader medical city in Najaf Province and comparing it with normal fertile men(20 person),also correlation was done between PRM 1 and some seminal fluid parameters.

Methods: Seminal fluid parameters were estimated including sperm concentrations, sperm progressive motility percent and Sperm normal morphology percent. PRM 1 serum level assessment in all sample as a biomarker.

Result : There is significant increase (**p <0.05) in protamine 1 level were observed in oligozoospermia patients (413.2 ± 21.94) by comparing with control group (326.2 ± 12.70), also there are negative correlation between PRM 1 and seminal fluid parameters, a significant increase in PRM 1 concentration may be cause a defect in male fertility.

Conclusion : From current study conclude that The biomarker PRM1 may be good predictive indicator of defect in male fertility espically in Oilgozoospermia Patients, and there are negative correlation between PRM1 and some of seminal fluid parameters.

Keywords: Protamine 1, Oilgozoospermia Patients, Fertility, Seminal Fluid Parameters

INTRODUCTION

There is little attention is placed on male infertility in developing countries because of the widely erroneous belief that infertility is a female problem [1]. Infertility is a typical illness of the reproductive system, incapacity to have healthy birth following one year of effectively endeavors of unprotected free intercourses [2]. Infertility is seen as a social and public issue across virtually all cultures and all societies and social orders. It influences 13%–15% of the reproductive aged couples around the world. The pervasiveness of infertility shifts broadly from region to region, being higher in developing nations, where there is an absence of assets for examination and treatment, than in developed nations [3]. The male contribution to infertility among couples worldwide has been estimated to be about 33%, the male partners' contribution to sub fertility is estimated to be about 54% based on semen analysis alone [4]. Moreover, approximately 15% of patients with male factor infertility have a normal semen analysis [5]. Whereas in 8% of men with normal sperm parameters, different forms of sperm DNA damage are found[6]. Spermatogenesis is a tightly regulated and complex biological process of cellular differentiation that results in the production of haploid male germ cells. During spermatogenesis, a complex and dynamic process of proliferation and differentiation occur as spermatogonia are transformed into mature spermatozoa. This unique process involves a series of meioses and mitoses, changes in cytoplasmic architecture, replacement of somatic cell-like histones with transition proteins, and the final addition of protamines, leading to a highly packaged chromatin. The presence of DNA damage in the male germ line has been linked with a variety of adverse outcomes such as low fertilization rates, decrease in embryo implantation, miscarriage, cancer and other diseases in the offspring[7]. The organization of sperm nuclear DNA takes place in the haploid stage of spermatogenesis is called spermiogenesis. In the testicular phase, histones are replaced at first by lysine-rich transition proteins (TPs) and then by protamines[8]. Protamine has an important role in the condensation of spermatozoa chromatin. It shows that protamine is required in the designing and capacity of spermatozoa [9].

There are two classes of protamines (*PRM1* and *PRM2*) are available in human sperm, where *PRM1* contains 50 amino acids rich in arginine and cysteine and *PRM2* is a group of 3 proteins derived from a single gene. The analogical gene for protamines of 28.6kb is located in the p arm of chromosome 16 DNase I-sensitive mass [10].

In the human, it has been known for many years that the chromatin of the mature sperm nucleus can be abnormally packaged. In addition, abnormal chromatin packaging and nuclear DNA damage appear to be linked [11]. There is also a strong association between the presence of nuclear DNA damage in the mature spermatozoa of men and poor semen parameters [12].

MATERIALS AND METHODS

Sample Preparation

Semen and serum specimens were collected from infertile oligozoospermia patients and control group (Fertile Normozoospermia) that attended to fertility center in al Sader medical city in Najaf Province from October 2017 until February 2018. The average age of infertile patients was (30.45 ± 54) years, the samples were collected are 100 and tested sample are 60, the sample which obtained from control group (fertile) was 20 samples (Normozoospermia), and 40 samples from Oligozoospermia on the day they delivered semen sample semen analysis was done according to WHO guidelines[13]. and an abstinence of 3-5 days was considered.

Biochemical Test

A biochemical test was performed on (60) samples had been measured protamine 1 protein by immunological method (Enzyme-Linked-Immuno-Sorbent- Assay) by using ELISA reader (Huma Germany origin). All specimens and reagents must be allowed to come to room temperature before use. All reagents must be mixed softly without foaming. Once the procedure has started, all steps must be completed without interruption, and biochemical tests were conducted in the laboratories of Biology Department/ Faculty of Sciences/ University of Kufa. The ELISA kits used in this study was human protamine 1, (E-EL-H5684) Elabscience company china in Origin.

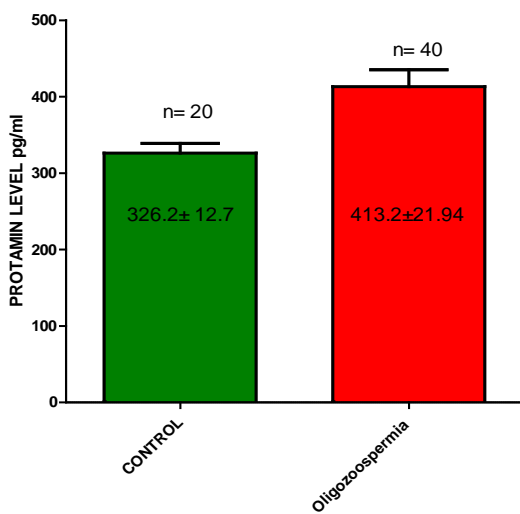
Statistical analysis

Protamine -1- level in serum of oligozoospermia patients presented as mean and standard error of mean (SEM) sample groups were compared using the Student's *t* test. Statistical analysis was performed with GraphPad Prism software version 5.03 (GraphPad Prism software ,Inc. San Diego, California, USA). *p* values <0.05 were considered to be statistically significant. Correlation coefficients were calculated to estimate the correlation between protamine-1- and parameters, the descriptive statistics and correlation coefficients were performed by using mega stat (Version v 10.12) for excel 2010 [14].

RESULTS

Protamine -1- level in oligozoospermia patients serum

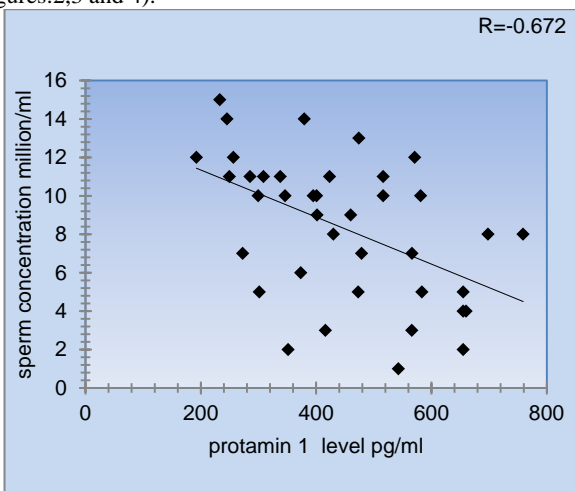
Results show significant increase (***p*<0.05>) in protamine 1 level were observed in oligozoospermia patients (413.2 ± 21.94) by comparing with control group (326.2 ± 12.70), as shown in Figure 1.



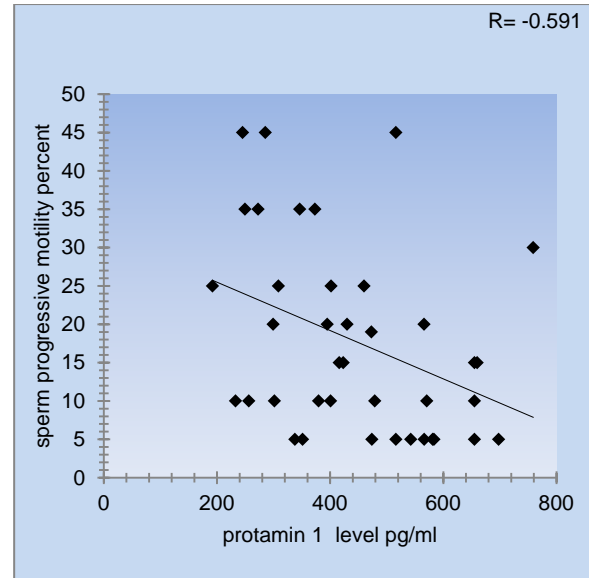
Figure(1): comparison between protamine 1 concentration in oligozoospermia patients and control group.

Correlation between protamine 1 with seminal fluid parameters.

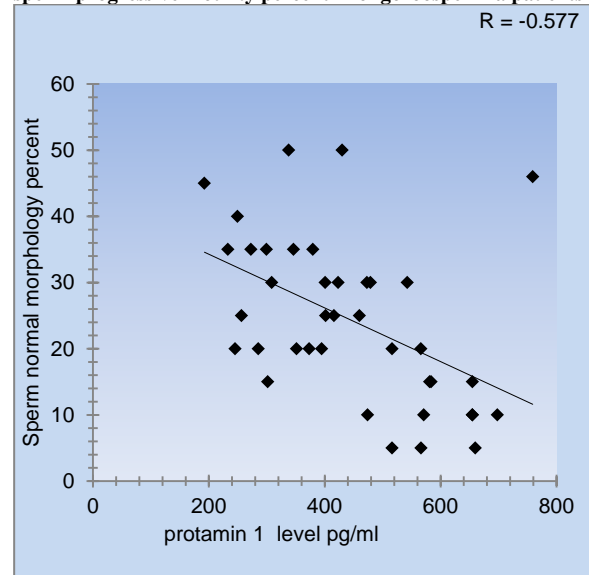
The result showed there are negative correlation between PRM1, sperm concentrations, sperm progressive motility percent and Sperm normal morphology percent of seminal fluid analysis of semen specimens in oligozoospermia patients as shown in (Figures:2,3 and 4).



Figure(2): The correlation between PRM1 concentration (pg/ml) and sperm concentration in oligozoospermia patients .



Figure(3): The correlation between PRM1 concentration (pg/ml) and sperm progressive motility percent in oligozoospermia patients .



Figure(4): The correlation between PRM1 concentration (pg/ml) and sperm normal morphology percent in oligozoospermia patients .

DISCUSSION

The present study showed a significant increase (***p*<0.05) in the level of serum PRM1 in oligozoospermia patients by comparing with control group. Current study agreed with study done by [15]. Also our study agreed with study done by [16]. The explanation of that due to deregulation of *PRM1* in spermatozoa of infertility patients, less than deregulation of *PRM2* , based on the cause that Protamine2 deregulation is responsible for the majority of cases including an aberrant protamine ratio [17]. Studies of protamine evolution have revealed that the P2 gene is more recently derived than P1 and highly variable within the mammalian genera [18]. The present study also shows that protamine 1 expression in spermatozoa had a negative correlation with sperms concentration, Sperm Progressive motility and sperm normal morphology in oligozoospermia. Our result agree with several studies that show negative correlation found between protamine protein, with sperms concentration, Sperm Progressive motility and sperm normal morphology [19], this may occur due to protein synthesis that effected by accompanying with spermatogenesis caused by increase free radicals which in turn damage spermatozoa

and subsequently the parameters[20]. Furthermore, many studies have indicated a significant correlation between DNA damage may be protamine 1 degradation and high levels of ROS in infertile patients[21].

CONCLUSION

From current study conclude that The biomarker PRM1 may be good predictive indicator of defect in male fertility, and there are negative correlation between PRM1 and some of seminal fluid parameters .

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