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The Effect of L-carnitine supplement on seminal fluid parameters in males with infertility.

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Abstract :

Background: Male infertility is a significant problem affecting 7.5% of the male population. Approximately 60% of these cases are idiopathic and related to sperm dysfunctions such as oligo-asthenoteratozoospermia (OAT). Aim: The aim of this study was to verify the effect of L-carnitine on sperm parameters in patients who presented with infertility. Patients and Methods: The clinical trial study conducted on a total 142 men but 8 patients with varicocele were excluded. So this study was conducted on the remainder 134 patients presenting with infertility due to idiopathic oligo-astheno-zoospermia (mean age \pm SD: 30.6 \pm 6.07), who didn't use L-carnitine previously. Sample divided into 2 groupd (70 patients treated with L carnitine and 64 with placebo). L-carnitine was prescribed orally at a daily dose of 2 gm/day for 3 months. Before onset of drug intake and after the ending of the L-carnitine treatment, semen analysis was performed. Results: Seminal fluid analysis has been done before treatment and then 3 months after treatment. The mean values of sperm count, total motility and normal morphology of astheno-spermic and oligo-astheno-teratospermic were found significantly at ($p < 0.05$). There is a trend toward an improvement in seminal fluid parameters was noted when comparing baseline with 3 months points in both the placebo and the carnitine arm.

The results of the present study clearly demonstrated that L-carnitine at a daily dose of 2 gm/day for a treatment period of 3 months can improve sperm concentration, sperm count, percentage of actively motile sperm, and progressive motile sperm count among men with idiopathic oligo- and/or astheno-zoospermia. **Conclusion:** The present study concluded that L-carnitine at a daily dose of 2 gm/day for a treatment period of 3 months had positive effect and can be treat infertility.

المستخلص

مقدمة: عقم الرجال مشكلة واضحة تؤثر في 7.5% من الرجال. تقريبا 60% من الحالات اسبابها غير معروفة ولها علاقة بخلل عدد، شكل، حرك و عدد الحيامن. **الهدف:** تهدف الدراسة لتوضيح تأثير علاج ل_كارنتين على مؤشرات الحيامن للمرضى المصابين بالعقم. **لمرضى وطرق العمل:** دراسة تجريبية سريرية اجريت على 142 رجل عقيم تم استثناء 8 منهم كونهم مصابين بمرض دوالي الخصية. الدراسة طبقت على 134 مريض بالعقم لاسباب غير معروفة (معدل اعمارهم 30.6-/+6.07)

والذين لم يستخدموا علاج ل كارنتين مسبقا. قسمت العينة الى مجموعتين متكونة من 70 مريض عولج بال كارنتين و 64 مريض عولج ببلاسيبو، استخدم العلاج بأخذه عن طريق الفم بجرعة 2 ملغم يوميا لمدة 3 أشهر. اجري للمرضى اخبار السائل المنوي قبل البدء بالعلاج وبعد 3 أشهر من العلاج. **النتائج:** اجري فحص السائل المنوي قبل البدء بالعلاج وبعد 3 أشهر منه، وعند حساب معدل النطف والحركة الكلية والشكل الطبيعي للمرضى وجد ان الفرق واضح هناك

نزعة باتجاه تحسن مؤشرات فحص السائل المنوي لمجموعة ال كارنتين مقارنة بمستخدمي البلاسيبو. اظهرت النتائج بوضوح ان 2ملغم يوميا خلال 3 اشهر بامكانها ان تحسن تركيز النطف وعددها ونسبة النطف النشطة الحركة لمرضى العقم غير معروف الاسباب من الرجال. الاستنتاج: استنتجت الدراسة الحالية ان علاج ال كارنتين بجرعة 2 ملغم يوميا لثلاثة اشهر لها تأثير ايجابي ويمكنه علاج العقم.

Introduction

Infertility is a significant problem in humans. According to World Health Organization (WHO) it is defined as the inability of a sexually active, non-contracepting couple to achieve pregnancy in one year(1). Infertility affects fifteen percent of couples worldwide. Male and female factors coexist in about one third of cases, while one third of cases are secondary to male factors only (2).

Semen analysis is routinely used to evaluate the male partner in infertile couples and provides useful information for diagnosing male infertility. Each of the sperm measurements helps to distinguish between fertile and infertile men however none is a powerful discriminator (3,4). In male infertile patients semen analysis reveals a decreased number of spermatozoa (oligozoospermia), decreased motility (asthenozoospermia) and many abnormal forms on morphological examination (teratozoospermia). These abnormalities usually occur together and are described as the oligoasthenoteratozoospermia (5). Free L-carnitine (beta-hydroxy-gamma N trimethyl aminobutyric acid) is biologically active amino acid that was first isolated from beef muscle in 1905 (3). Meat and milk are the most significant dietary sources of exogenous carnitine for humans (6). Approximately 75% of the body stores of L carnitine are derived from the diet, whereas only 25% are synthesized de novo from lysine and methionine (7). It has long been assumed that carnitine is not an essential

component of diet as humans have the ability to synthesize this compound(8). L-carnitine is concentrated in high energy demanding tissues such as skeletal and cardiac muscles and in a specialized reproductive tract organ, the epididymis (9).

In 1973, Casillas (10) demonstrated that spermatozoa accumulate carnitine in mammalian epididymis, which is closely related with the development of fertilizing capacity by spermatozoa. The concentration of L-carnitine in epididymal plasma and spermatozoa varies from 2 to 100 mmole, which is nearly 2000 fold greater than circulating levels (10-50 mole) (11,12). In epididymis, free L-carnitine is taken up from the blood plasma and is transported into the epididymal fluid. It is then passively diffused into the spermatozoa, where it accumulates as both free and acetylated L-carnitine (13,14). The initiation of sperm motility occurs in parallel with the increase in concentration of free L-carnitine in the epididymal lumen (6).

By providing readily available energy for use by spermatozoa thus positively affecting sperm motility, maturation and the spermatogenic process (12,13,14) a key role in sperm metabolism is strongly suggested by the high levels of LC found in epididymal fluid due to an active secretory mechanism (9) and there is also evidence that the initiation of sperm motility is related to an increase of LC in the epididymal lumen and LAC in sperm cells (15,16). Based on these fundamental roles, numerous clinical trials have attempted to demonstrate

beneficial therapeutic effect of LC and/or LAC when administered to infertile men with various forms of sperm dysfunction. This study was conducted to evaluate the effectiveness of carnitines in male infertility.

Patients and methods

Ethical consideration: Permission was taken from patients regarding their information and drug intake and its side effect then they agreed to participated in a study.

This clinical trial study was conducted between first of October 2011 to the end of March 2014, a total of 142 infertile male patients (aged 17-52 years) referred to the infertility clinic in Tikrit Teaching Hospital but 8 cases excluded because they had varicocele; therefore the drug and placebo applied on 134 patients.

Study groups chosen by convenience sampling method, this sample (134) patients divided into two groups (70 treated with L-carnitine 2 mg/day and 64 given placebo). Semen analysis done before starting treatment and after 3 months of treatment.

Exclusion criteria were: history of smoking habit, alcohol consumption, and occupational chemical exposure; history of major renal and hepatic disorders and myopathy; treatment with other drugs within the 3 months before enrolment in this study; history or presence of primary testicular disease (cryptorchidism, orchitis, varicocele) or testicular volume ≤ 12 ml; infected semen; elevated (>10 mIU/ml) serum FSH concentrations or other abnormal hormonal assay; and abnormal sonography.

Infertility diagnosed according to (WHO) criteria, male patients, with

infertility >1 year, having regular sexual intercourse with a gynecologically normal partner who has no apparent factors of female factor infertility were chosen.

The diagnosis was made after medical assessment which included: history; clinical examination especially for varicocele detection and testicular volume evaluation; semen analysis; Follicular stimulating Hormones (FSH), Leutinizing Hormone (LH), testosterone, Prostaglandins (E2), and prolactin assay; sonography of genitalia.

The seminal analysis, including sperm concentration, total and forward sperm motility (percentage at one hour after ejaculation) and sperm morphology (percentage of atypical forms), according to WHO standard procedures.

Data presented with simple tables and analyzed statistically to test significance at p value <0.05 .

Results

This study considered 134 patients with unexplained astheno-spermia followed up after treatment with L-carnitine. The mean age was 30.6 ± 6.07 years. The mean duration of infertility was 4.7 ± 5.56 (1 to 8 years) with 70 patients on L carnitine for three months and 64 patients in the placebo.

The median seminal fluid volume, total motile sperm count and percent motility of the treatment group before treatment were 2.46 ml, 19.4 million and 27.0% respectively, which were not statistically different from the placebo group: 20.9 million sperm and 30.3% - 3.2%. respectively ($P < 0.05$).

Table 1: Effects of L-carnitine treatment on the mean \pm SD of seminal fluid volume, sperm concentration, and sperm count in idiopathic infertile patients(n=70).

Sample Groups	Volume(ml)	Million/ml	Million/ejaculate
Patients before treatment	2.46 \pm 0.23	19.4 \pm 1.6	142.47 \pm 4.41
3 Month after treatment	2.81 \pm 0.21	31.83 \pm 2.59 *	79.41 \pm 7.87 *

*p<0.05 represents significant difference between before treatment and after treatment.

Table 2: Effects of L-carnitine treatment on the mean \pm SD of sperm motility (represented as percent of total) in idiopathic infertile patients(n=70)

Sample Groups	Active motile	Sluggish	Immotile
Patients before treatment	27.03 \pm 2.52	33.44 \pm 1.85	36.44 \pm 3.51
3 Month after treatment	35.77 \pm 2.70 *	34.77 \pm 1.55	29.44 \pm 3.30 *

*p<0.05 represents significant difference between before treatment and after treatment.

The above results had demonstrated that L-carnitine at a daily dose of 2 gm/day for a treatment period of 3 months can improve sperm concentration, sperm count, percentage

of actively motile sperm, and progressive motile sperm count among men with idiopathic oligo- and/or astheno-zoospermia.

Table 3: placebo Effects on mean \pm SD of seminal fluid volume, sperm concentration, and sperm count in idiopathic infertile patients(n=64)

Sample Groups	Volume (ml)	Million /ml	Million /ejaculate
Patients before treatment	2.43 \pm 0.17	20.37 \pm 1.11	20.9 \pm 3.61
3 Month after treatment	2.50 \pm 0.11	20.72 \pm 1.70 *	21.17 \pm 5.29

*p<0.05 represents significant difference between before treatment and after treatment.

Table 4: placebo Effects on mean \pm SD of sperm motility (represented as percent of total) in idiopathic infertile patients(n=64)

Sample Groups	Active motile	A sluggish	Immotile sperm
Patients before treatment	46.06 \pm 1.47	34.24 \pm 1.39	19.68 \pm 1.65
3 Month after treatment	46.89 \pm 1.72	33.44 \pm 1.21	19.65 \pm 1.71

*p<0.05 represents significant difference between before treatment and after treatment.

Discussion

Asthenozoospermia is a relevant issue in male infertility management. The efficiency of sperm motility, required for fertilization capacity (19). Other similar studies have suggested an improvement in semen parameters of men with low sperm motility following treatment with oral carnitine (20, 21, 22). These observations suggest a role of carnitine or acetylcarnitine as empiric therapy for idiopathic asthenospermia (23). The significant effect of L-carnitine on increasing sperm concentration, sperm count, and percentage of actively motile sperm in this study could be due to or explained by that L-carnitine is an essential cofactor that could accelerate lipid metabolism and has a pivotal role in mitochondrial β -oxidation of long-chain fatty acids for cellular energy production (4,5). There is also an unknown effect of L-carnitine in Sertoli cell-spermatogenic line interaction, an action on the postmeiotic phases of spermatogenesis (for example, on the chromatin stability or mitochondrial function of spermatocytes or spermatids), or an improvement in the quality of the epididymal microenvironment, reducing gamete phagocytosis at this level while increasing ejaculated spermatozoa (17).

Carnitines may be also responsible for removing excess intracellular toxic acetyl-CoA, which protects spermatozoa from oxidative damage.(33) Although some evidence suggests a key role of carnitine for sperm motility, its real effective role still remains an interesting open question. In addition, carnitine administration increases prostaglandin E2 concentration(34), which affects sperm count (35). Moreover, carnitine protects cell membrane and DNA against damage induced by free oxygen

radicals. It also prevents protein oxidation and lactate oxidative damage (36). Hence, it acts as an “anti-aging” substance, protecting against damage induced by free oxygen radicals (37).

Although pregnancy was not a principal end point in many studies (15, 14), including this study, as it is difficult to avoid the many confounding variables acting on naturally induced fertilization and subsequent pregnancy, three pregnancy was recorded in group L-carnitine patients after 2 months of treatment.

Conclusion: The present study had demonstrated that L-carnitine at a daily dose of 2 gm/day for a treatment period of 3 months can improve sperm concentration, sperm count, percentage of actively motile sperm, and progressive motile sperm count among men with idiopathic oligo- and/or asthenozoospermia.

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