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Significance of Some Trace Elements in Semen of Infertile Men

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Abstract

Objectives: The biological significance of trace elements in male infertility was studied. Levels of Zinc, Copper, Iron and Magnesium in the seminal plasma of infertile men and controls were measured. The relationships between these elements, seminal characteristics, and serum reproductive male hormones were explored. **Patients and Methods:** Seventy two infertile men on no treatment were selected from the central infertility center, Al Shifa hospital, Gaza, Palestine were studied and 72 known fertile males were used as controls. Semen samples were analyzed according to WHO criteria and seminal plasma trace elements were analyzed by atomic absorption spectrophotometry. Serum hormonal levels were measured by conventional methods. **Results:** The mean values of Zinc and Magnesium were significantly lower in infertile men than controls (69 vs. 122 mg/L and 67 vs 120 mg/L) respectively. All studied seminal parameters (sperm count, forward motility, weak motile and non-motile) were significantly lower in the infertile group than in controls. Serum luteinizing hormone and testosterone were significantly lower in the infertile group than in controls ($p<0.05$). No significant difference in serum follicle stimulating hormone levels was detected between groups. Within the infertile group, seminal plasma Zinc and Magnesium levels correlated directly to the sperm count ($r=0.376$, $P=0.001$ and $r=0.293$, $P=0.013$ respectively), and testosterone ($r=0.293$, $P=0.012$ and $r=0.324$, $P=0.003$ respectively). Zinc and Magnesium were inversely related to the seminal volume ($r=-0.251$, $P=0.034$ and $r=-0.369$ $P=0.001$ respectively). **Conclusion:** Our findings supports a possible role for Zinc and Magnesium in spermatogenesis and steroidogenesis.

Key words: Trace elements, Male infertility, Semen composition

Introduction

Male factor infertility plays a role in approximately 30-55

% of infertile couples (1,2). A significant subset of these subfertile men are classified as having unexplained male infertility (UMI). The commonest single defined cause of male infertility is sperm dysfunction (2). This problem has been worsening due to the deterioration of the human semen quality by as much as 3% per year suggesting that male reproductive problems may be increasing over time (3). Despite the difficulty in assessing the prevalence of infertility in developing countries, it is estimated that 8-12% of couples around the world have difficulty conceiving a child at some point in their lives thus affecting about 50-80 million people (3). In Palestine, infertility has both psychological and financial burden on the local population (4).

In 90% of infertile men, sperm count is low (5). The role of trace elements in the quality of human semen has received much interest recently (6). Although the significance of trace elements in male fertility has been realized, the biological role of these elements is not fully understood (7,8). Zinc was found to be in high levels in semen from mammals, and Zinc was thought to be critical to spermatogenesis. Deficiency of Zinc is associated with hypogonadism and insufficient development of secondary sex characteristics in humans (9). Also, deficiency of Zinc may cause failure in spermatogenesis due to atrophy of the seminiferous tubules in rats (10). However, high concentrations of Zinc have been reported to depress oxygen uptake in the sperm cells (11), head-tail attachment/detachment and nuclear chromatin condensation decondensation is also influenced by seminal Zinc (12). Also sperm motility has been suggested to be affected by Zinc levels (13). The generation of oxidants, also described as reactive oxygen species (ROS), in the male reproductive tract has been a real concern because of their potential toxic effects, at high levels, on sperm quality and function (14,15). A number of reports indicated the significance of trace elements in male infertility and its effect on the level of antioxidants (16). ROS are needed for the regulation of normal sperm functions, such as sperm capacitation, the acrosome reaction, and sperm-oocyte fusion (17). Increased levels of metal ions in semen (19) or blood plasma (18) appear to be significantly and positively correlated with male infertility (20). On the other hand, spermatogenesis in mammals requires the action of a number of peptide and steroid hormones. Sex hormones are critical for both regulation of male germ cell development and proliferation and function of the somatic cell types required for proper development of the testis (21-23). We wished to explore the biological significance of trace elements in male infertility. Levels of Zinc, Copper, Iron and Magnesium in the seminal plasma were compared in infertile men and controls. The relationship between these elements and both seminal characteristics, serum reproductive male hormones were examined.

Materials and Methods

Subjects

All subjects provided informed written consent before the study. The case group consisted of 72 infertile men aged 20-50 years with oligospermia or/and asthenospermia. Controls consisted of 72 fertile men aged 20-50 years whose wives were pregnant or had delivery of a child within the previous 12 months. The number of individuals was chosen to be able to carry out Pearson test analysis. The case group included men whose wives were not able to conceive at least for 12 months of unprotected sex having established no causes of infertility in the female partners. All men were non-smokers, did not consume alcohol and had had normal dietary pattern of life.

Table 1. Comparison between seminal parameters (mean±standard deviation) of case group of 72 cases and 72 controls.

Semen parameters	Cases	Controls	P value
Semen volume (ml)	3.0±1.1	3.3±0.9	NS
Sperm count (million)	23.9±22	63.1±16.7	<0.001
Forward motility	22.8±14	51.3±5.36	<0.001
Weak motile	21.3±10.6	18.5±5.08	<0.05
Non-motile percentage	55.9±18.6	30.3±6.27	<0.001

Methods

Seminal fluid collection and analysis were carried out in strict compliance with the WHO guidelines. Semen specimens were best collected by masturbation into a sterile container. Serum hormonal profiles of LH, FSH and testosterone. Seminal plasma trace elements (Zinc, Copper, Iron and Magnesium) using standard laboratory methods.

Results

Sperm characteristics

Sperm parameters excluding the volume of the patients were below the WHO reference range (Table 1). Predictably, the mean sperm count was markedly lower in cases than in controls (P<0.001). Forward progression percentage was lower in patients than in controls (P<0.001). Weak-motile and non-motile percentages were lower in the patient group than in the controls.

Hormonal parameters

The mean serum FSH, LH and testosterone levels were normal in both groups. However, LH level was significantly lower in the patients than in the controls (P<0.05) and mean

Table 2. Comparison between select relevant endocrine parameters (mean±standard deviation) of the case group of 72 infertile men with the control group of 72 fertile men

Endocrine parameters	Cases	Controls	P value
Serum LH (IU/L)	4.52±1.45	5.05±1.23	<0.05
Serum FSH (IU/L)	5.33± 0.6	5.37±0.65	NS
Serum Testosterone (nmol/l)	5.09±1.03	5.4±0.9	<0.05

Table 3. Comparison between concentrations of seminal plasma trace elements in the case group of 72 infertile men with the 72 healthy (fertile) controls.

	Cases	Controls	P value
Zinc (mg/L)	68.9±37.7	122±26.1	<0.001
Magnesium (mg/L)	67.1±36.4	120±28.2	<0.001

Table 4. The linear regression analysis between Zinc and Magnesium seminal plasma level and seminal parameters in case group n = 72

Seminal parameters	Slope		r		P value	
	Zn	Mg	Zn	Mg	Zn	Mg
Semen volume	-8.482	-12.04	- 0.251	-0.369	0.034	0.001
Sperm count	0.646	0.484	0.376	0.293	0.001	0.013
Forward motility	-0.488	-0.290	- 0.182	-0.112	NS	NS
Weak motile	0.702	0.268	0.197	0.078	NS	NS
Non-motile	0.025	7.86	5.075	0.040	NS	NS
Serum LH	5.472	9.577	0.211	0.383	NS	0.001
Serum FSH	4.532	6.55	0.074	0.110	NS	NS
Testosterone	10.694	11.37	0.293	0.324	0.012	0.006

Zn: Zinc; Mg: Mangnesium; L: Leutinizing hormone, FSH = Follicle stimulating hormone.

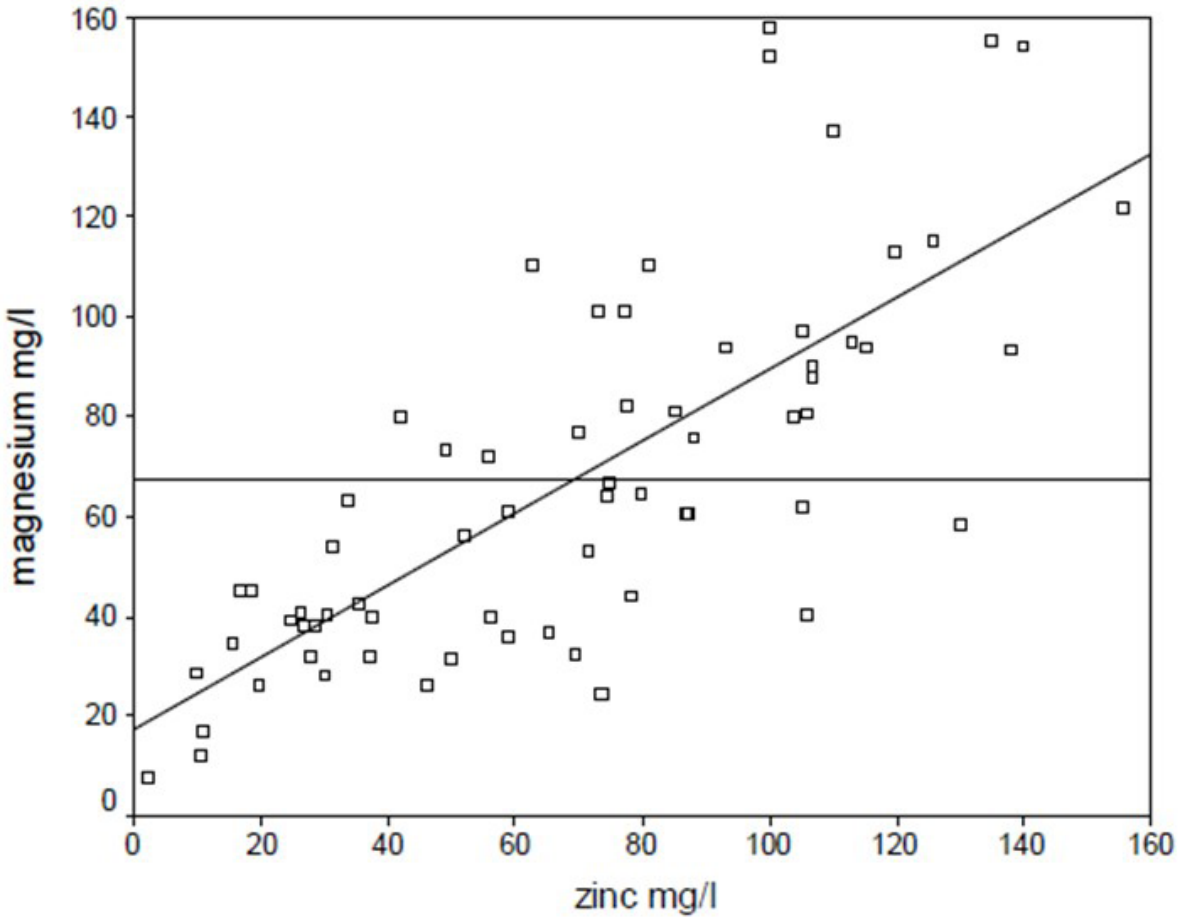

Figure 1. Relationship between seminal plasma Zinc and seminal plasma Magnesium in patients' group n = 72.

testosterone level was significantly lower in the patients than in the controls (P<0.05). The serum FSH levels were not significantly different between groups (Table 2).

Trace elements levels in seminal plasma

The seminal plasma trace elements Zinc, Copper and Magnesium levels were significantly lower in the patients than in controls (Table 3). The differences were particularly seen in Zinc concentrations (P<0.001). In the patient group, Zinc correlated inversely with semen volume (P<0.05), correlated directly with sperm concentration (P<0.001) but and not with the other seminal plasma parameters (P>0.05). Seminal plasma Zinc correlated positively with testosterone levels in the patient group ($p < 0.05$). The Magnesium and Zinc levels in seminal plasma of patients showed a strong direct correlation (Table 4 and Figure 1). Magnesium seminal plasma of infertile men showed an inverse correlation with semen volume (P<0.001), direct correlation with sperm count (P<0.01) but not with other semen parameters (Table 4). Seminal plasma Magnesium in patients correlated directly serum LH and serum testosterone but not with serum FSH level (Table 4).

Discussion

Abnormalities associated with trace elements can be due to specific efficiency from dietary inadequacies and imbalances, or abnormality secondary to other diseases. Both kinds of abnormality can be diagnosed by analysis of trace elements in body fluids or other tissues. However secondary changes, which occur as a result of diseases, are not exactly understood. Our study suggests that Zinc is an essential trace element for male infertility. This supports early studies indicating that Zinc is an essential trace element required for normal spermatogenesis and steroidogenesis and that Zinc deficiency being one of the factors responsible for decreased testicular function in

infertile males (25).

Our results also indicate that both Zinc and Magnesium co-exist either intracellulary or extracellularly as shown by the positive correlation between them. Since Magnesium is the second major intracellular cation to Potassium, the positive correlation found with Zinc indicates that Zinc is important inside the reproductive cells. Previous work has shown that increase in cAMP due to hormonal changes enhances that outlet of Magnesium and its increase extracellularly. It is well known that FSH and LH act intracellularly by increasing the levels of cAMP. Therefore it is expected that FSH and LH promotes the export of Magnesium, potassium and consequently Zinc (26). Our study indicates that increase in seminal plasma Magnesium or Zinc reduces seminal plasma volume. Since deficiency in Magnesium is considered a deficiency in potassium, it is expected that the outlet of either Magnesium or Zinc will lead to reduction in Potassium therefore to loss of water in the seminal plasma therefore leading to reduction in the seminal plasma volume. We concur with the previous reports which found inverse relationship between Zinc concentration in seminal plasma and semen volume more than 5ml and a positive correlation with hypoviscosity of semen and conclude that Zinc and calcium and physical analysis of ejaculate is clinically useful for evaluating the secretary activity of the seminal vesicles and prostate; abnormal coagulation, liquefaction, volume, viscosity and pH strongly suggest gland dysfunction (27).

Our results support the hypothesis that Zinc is essential for spermatogenesis and steroidogenesis, as shown by the positive correlation between the Zinc concentration and the semen count and serum testosterone. Our results agrees with other studies which have found a positive association between seminal plasma Zinc concentration with sperm count and with sperm motility, positive association with serum testosterone levels of the case group (28). Our study is concordant with others that shown that seminal plasma Zinc concentration was significantly correlated with sperm density (7,11,12). Zinc is a trace element essential for normal functioning of the male reproductive system. Numerous biochemical mechanisms are Zinc dependent, including more than 200 enzymes in the body (29).

Zinc deficiency is associated with decreased testosterone levels and sperm count, an adequate amount of Zinc ensures proper semen motility and production. Zinc levels are generally lower in infertile men with diminished sperm count, and several studies found supplemental Zinc may prove helpful in treating male infertility (30). The effect of Zinc supplementation on testosterone, dihydrotestosterone and sperm count was studied in 37 patients with idiopathic subfertility of more than 5 years duration and diminished sperm count received 24 milligrams of elemental Zinc from Zinc sulfate for forty five to fifty days (31). The results were dramatic in the twenty two subjects with initially low testosterone levels; a significant increase in testosterone levels and sperm count (from 8-20 millions/ml) was noted, along with 9 resulting pregnancies (31). LH increases seminal Magnesium levels. The lower levels of Magnesium in infertile population indicate that Magnesium might play a role in male fertility. Despite the lack of direct analytical studies on the role of Magnesium in infertility, a number of studies have shown that Magnesium is essential for energy requiring cells such as muscle and heart cells (32). It binds to ATP in the mitochondria in great amounts. However when hormones such as adrenaline binds to the cells it increases the levels of cAMP by activating Adenylate cyclase. The increase of cAMP will encourage the release of ATP-Mg from the mitochondria (33). This will disassociate leading to free of ATP and exporting Magnesium outside the cells. LH, which acts on leydig cells during spermatogenesis, has been shown to increase the levels of cAMP (26). Therefore we assume that this will lead the export of Magnesium into the seminal fluid. This could explain the direct relation between the increase in LH levels and Magnesium levels, which our data has demonstrated. Therefore the increase in Magnesium in seminal plasma improves fertility. It is noteworthy that the increase in seminal plasma means free ATP in spermatocytes and good fertility. Since Magnesium being an intracellular component means that it moves against water. Since the increase in LH increases Magnesium in the seminal plasma, this will lead to lower amount of water in the seminal plasma. This explains the inverse relations between the levels of both Magnesium and LH and seminal volume in our studied population. In conclusion, our findings indicate a possible role for Zinc and Magnesium in spermatogenesis and steroid genesis in man and supports the notion of the need of further research in this area.

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