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FSH treatment for normogonadotropic male infertility: a synergistic role for metformin?

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Abstract. – OBJECTIVE: The aim of this paper is to evaluate the effectiveness of follicle-stimulating hormone (FSH) administration in a cohort of insulin resistant (HOMA>2.5) patients with normogonadotropic idiopathic infertility.

PATIENTS AND METHODS: We subdivided patients in two clinical groups basing on the adopted therapeutic scheme: group A (n=44) received 150 units of FSH three times a week for three months (group A); group B (n=35) received 150 units of FSH three times a week for three months and 500 mg of slow-release metformin once a day for three months (group B). We evaluated the post-treatment sperm parameters, sperm parameters normalization rate, spontaneous pregnancy rate, and sperm DNA fragmentation normalization rate.

RESULTS: 40% of group A patients and 45% of group B patients became normozoospermic after the treatment, while 30% of group A patients and 32% of group B patients achieved a spontaneous pregnancy. B group patients also obtained higher sperm DNA fragmentation normalization rate (45% *vs.* 33%, $p = 0.03$). Compared to group A, group B showed a higher sperm concentration, progressive motility and morphology ($p < 0.0001$).

CONCLUSIONS: The results of this study suggest that the addition of the low-dose slow-release metformin in insulin-resistant patients with normogonadotropic infertility improves the efficacy of FSH therapy.

Key Words:

Follicle-stimulating hormone (FSH), Normogonadotropic male infertility, Metformin, Oligoasthenoteratozoospermia (OAT).

Introduction

Normogonadotropic male infertility is a medical condition characterized by an alteration of the conventional and/or biofunctional sperm parameters [oligoasthenoteratozoospermia (OAT)] in the presence of normal gonadotropins levels. This alteration is responsible for the failure of the establishment of clinical pregnancy in couples where there are no female factors of infertility[1]. In the clinical practice, the follicle-stimulating hormone (FSH) represent an important therapeutic option for normogonadotropic male infertility[2]. Accordingly, several studies have shown the positive effects of FSH on conventional (density, motility and morphology) and bio-functional sperm parameters[3]. In particular, evidence has been accumulated on the beneficial effects of FSH on sperm DNA fragmentation[4], a parameter closely associated not only with spontaneous pregnancy rates but also with assisted reproductive technology (ART) outcomes[5]. However, not all patients respond to FSH therapy and this heterogeneous response underlines the contrasting results obtained by clinical trials[6]. To date, there are no reliable predictors of response to treatment: the FSH receptor and FSHβ subunit polymorphisms could represent the most useful tool to predict the clinical response[4], but their use actually remains a prerogative of clinical research. Garolla et al[7] have highlighted the importance of the testicular cytology and of the presence of spermatids in the ejaculate, while no differences in the efficacy between recombinant human FSH and high purified FSH have been demonstrated[8]. Low interest has been attributed to the insulin resistance as a possible factor influencing the FSH efficacy so far. Despite the reproductive alterations in obese males and/or in insulin-resistant patients have been widely described, there is low evidence concerning the pharmacological use of metformin in addition to the FSH[9,10].

The aim of the present study is to evaluate the effects of FSH administered as a single treatment or combined with slow-release metformin in a cohort of normogonadotropic insulin-resistant patients with idiopathic infertility.

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Patients and Methods

We retrospectively evaluated the sperm parameters and the spontaneous pregnancy rate of patients referring to the Catania Andrology Centre for normogonadotropic male infertility who had been treated with FSH or FSH plus metformin, as follows:

- 44 patients with normal testicular volume (> 12 milliliters), normal serum levels of total testosterone (> 350 ng/dL), and HOMA index > 2.5 were treated with FSH (150 units three times a week) for at least three months (group A, used as a control group);
- 35 age-matched patients with similar endocrine features and HOMA index > 2.5 were treated with FSH (150 units three times a week) and slow-release metformin 500 mg once a day for at least three months (group B).

Patients underwent weight and height measurement for the calculation of body mass index (BMI). A detailed medical history was collected. The testicular volume was measured by scrotal ultrasound (ultrasound evaluation: anteroposterior, transverse, and longitudinal diameters × 0.52). The semen analysis was performed in accordance with the 2010 World Health Organization manual (5th edition). The sperm DNA fragmentation was evaluated by the terminal deoxyuridine nick end labeling (TUNEL) assay. The FSH formulation used was Fostimon® (IBSA, Lodi, Italy). Glucophage Unidie 500 mg® (Bruno Farmaceutici SPA, Rome, Italy) was adopted as metformin formulation. The number of patients who normalized the conventional sperm parameter and sperm DNA fragmentation, and who reached a spontaneous pregnancy was recorded.

Statistical Analysis

Was performed by calculating the rate of patients who achieved these goals. The statistical comparison between the two groups was done using the Student's *t*-test. The difference considered statistically significant for *p*-values was < 0.05.

Results

The mean age of patients was 33.5±13.0 years; the mean BMI was 27.0±5.0 kg/m^2; the mean age of the female partner was 30.5 years. The BMI of group B was higher compared to group A patients (28.5 ± 2.0 *vs.* 24.0±2.0; $p < 0.05$). The age did not differ in the two groups (32.50 ± 4.0 *vs.* 34.0 ± 4.0; p0.72). The two groups did not differ for the HOMA-index.

Group A

After treatment 24 patients (40%) became normozoospermic, 18 patients (30%) achieved a spontaneous pregnancy, and 20 patients (33%) obtained the normalization of the sperm DNA fragmentation.

Group B

After treatment 27 patients (45%) became normozoospermic, 20 patients (33%) achieved a spontaneous pregnancy, and 28 patients (45%) obtained the normalization of the sperm DNA fragmentation.

No statistically significant differences in the sperm parameters normalization rate and spontaneous pregnancy rate have been demonstrated among the two groups. Regarding the sperm DNA fragmentation, the normalization rate was significantly higher in group B patients compared to group A patients ($p = 0.03$).

After treatment, group B showed a higher sperm concentration, progressive motility, and morphology compared to group A ($p < 0.0001$) (Figure 1).

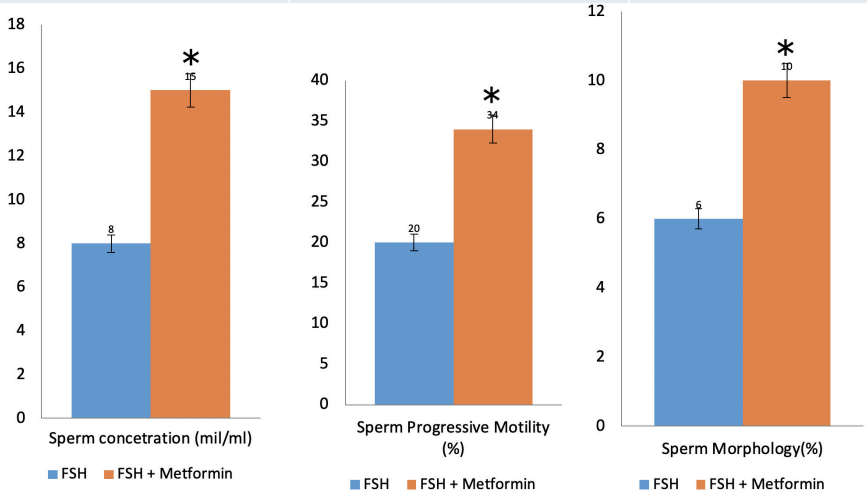
Discussion

The men with normogonadotropic infertility represent a heterogeneous group from a clinical point of view. The occurrence of insulin resistance is not usually considered before the treatment with FSH. The most adopted scheme in the treatment of infertile male with gonadotropins is the administration of FSH at a dose of 150 IU three times a week[8], but this scheme may not always be the most appropriate. If after three months of therapy the sperm parameters (conventional parameters and sperm DNA fragmentation) do not improve, it would be advisable to carefully evaluate in these patients the presence of other predictors of poor clinical response (i.e., FSH receptor and FSHβ subunit polymorphisms)[4].

The use of insulin-sensitizing drugs in the therapy of male infertility has been described. We have previously documented the improvements in the sperm mitochondrial function obtained by myo-inositol which is a weak insulin-sensitizing agent[11]. The insulin-resistant males, through the

Metformin improves the efficacy of FSH therapy in infertile men with insulin resistance

	FSH (n=44)	FSH + Metformin (n=35)
Age	42	44
BMI	27	28
WC	104	105
HOMA-IR	3.5	3.7



*p< 0.0001

Slow release Metformin: 500 mg daily; uFSH: 150 IU three times a week for three months

Figure 1. Semen parameters after treatment in the two groups.

reduction of the levels of the Sex Hormone Binding Globulin have an increase of free estrogens serum concentration which is also favored by the excessive aromatization of testosterone that occurs in visceral adipose tissue. Estrogens through the negative modulation of Kisspeptin determine a down-regulation of GnRH secretion. At the same time, the elevated levels of leptin contribute to reduce the GnRH pulsatility and, moreover, a condition of leptin resistance is applied to Leydig cells[12]. Insulin has direct effects on the spermatozoa that are rich in glucose transporters and, therefore, are considered a responsive element to glycemic fluctuations and the peripheral biological action of insulin. In particular, within the Sertoli cell, insulin positively modulates the expression of the specific lactate transporters within the tubular fluid to prepare it for an active transport into the germ cell mediated by other transporters. This functional aspect is of fundamental importance for the production of energy within the spermatozoa[13]. In the animal model, a diabetic disease causes a reduction of the intratesticular testosterone levels (which is necessary for the post-meiotic phase of spermatogenesis). Furthermore, the antioxidant defense systems in the epididymis are altered. The use of metformin significantly improves these aspects and in particular the expression of mRNA of the enzymes involved in steroidogenesis[14]. Also, in the animal model, which was experimentally rendered obese after a diet rich in fats, the pharmacological intervention with metformin reduces the sperm apoptosis and determines an increase in the testicular volume[15]. On the clinical level, the positive effects of metformin on the quality of sperm chromatin compaction have been described, particularly when associated with antioxidant agents[16]. Morgante et al[17] showed the improvement in the sperm parameters of patients with oligo-astheno-teratozoospermia with metabolic syndrome. They suggested a mechanism associated with an improvement in the hormonal pattern of hypogonadism after six months[17]. In eight-day-old

rat Sertoli cell cultures, the action of metformin counteracts the proliferative effect of FSH which is a feature of the neonatal phase. This would suggest a different action of metformin in addition to FSH in the prepubertal phase[18]. Finally, there are no clinical data on the efficacy of the combined treatment of FSH and metformin in adult normogonadotopic male infertility, therefore, the present study represents the first clinical evidence concerning this aspect.

Taken all this into account, the data of the present study suggest that metformin improved the efficacy of FSH therapy in normogonadotropic idiopathic-infertile patients with insulin resistance. The insulin receptor (IR) has been demonstrated to be involved in the Sertoli cell proliferation[19,20]. Also, the role of the insulin-like growth factor 1 receptor (IGF1R) in the FSH signaling in Sertoli cells has been demonstrated[21]. Since the IGF1R and the IR belong to the same tyrosine kinase receptor family, the possible interplay between the IR and the FSH signaling should be deepened. In particular, the IR substrate 1 (IRS1), which is involved in the FSH signaling[22], is known to be dysregulated in the insulin resistance[23]. Thus, when dysregulated, the IRS1 might also influence the FSH responsiveness. The pharmacological correction of the insulin resistance, by acting on the IRS1, may indirectly promote FSH signaling, improving the responsiveness to exogenous FSH and explaining out our findings. However, this hypothesis needs to be investigated.

Conclusions

This is the first study demonstrating the influence of the insulin resistance on the effects of FSH therapy and the benefits of its correction on FSH efficacy. Before FSH administration, the infertile patients should be assessed for the insulin resistance and, if present, it should be treated.

Conflict of Interest

The Authors declare that they have no conflict of interests.

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