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Acknowledgments

Acknowledgments should only be made to funding institutions and organizations and, if to persons, only to those who have made substantial contributions to the study.

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(b) Chapter in book; Wilhelmsson L, Norstrom A, Tjugum 1, Hamberger L. Interaction between prostaglan dins and catecholamines on cervical collagen. In: Toppozada M., Bygdeman '. M., Hafez ESE, Eds. Prostaglandins and fertility regulation. Advances in reproductive health care. Lancaster, England, MTP Press Ltd., 1985 : 75 - 80.

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Letter from the Editor:

Dear esteemed colleagues,

Warm greetings

We welcome your comments as well as the scientific activity to be incorporated in the upcoming issues. We are pleased to invite you to the 25th Egyptian Fertility and Sterility conference, 28, 29 November 2019. Venue: Marriott hotel Zamalek Cairo. New treatments and developments in women reproductive health. Very important subjects are included in this issue. Importance of sperm DNA fragmentation in ICSI outcome. Relationship between maternal and neonatal serum leptin levels and preeclampsia. Quality of life after hysterectomy at tertiary care hospital in upper Egypt.

Best regards.

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Does Sperm DNA Fragmentation test in Cases of Male Factor Infertility Improve ICSI outcome?

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Abstract

Background: Sperm DNA fragmentation is an uprising factor affecting male fertility and some previous results have shown significant effect of SDF on ART techniques outcomes**.**

Objective: To study effect of DNA fragmentation testing and treatment on ICSI outcome among infertile couples with male factor infertility.

Patients and Method: After approval of ethics committee,prospective control randomized study was conducted among a total of 106 infertile couples arranged for intracytoplasmic sperm injection (ICSI) due to male factor. Males were divided by simple random allocation into 2 groups. **Group I** included 53 males that were tested for DNA fragmentation test (Halo test). **Group II** included 53 males that were directly offered ICSI cycle.Infertile couples were evaluated as regard to fertilization rate, day 5 embryo transfer, positive pregnancy test, clinical pregnancy evaluated by visible sac with viable fetus on ultrasonographic assessment and delivery of normal viable baby.

Results: Men with initially negative DNA fragmentation test and those with positive DNA fragmentation test who responded well to treatment were found to have higher fertilization ratio of M2 ova, day 3 grade I embryo, blastocyte, positive pregnancy test and positive clinical pregnancy compared to non responders to treatment. There was no statistically significant difference between testicular and ejaculated sperm ICSI outcome among males with persistent sperm DNA fragmentation test.

Conclusion: Sperm DNA fragmentation is an important factor that significantly affects ICSI outcome. None of testicular or ejaculated sperm is superior regarding ICSI outcome among males with DNA fragmentation.

Keywords: assisted reproduction, fertility, azospermia, oligospermia.

INTRODUCTION

Infertility is c common problem affecting about 15% of couples attempting to conceive **(1)**. Up to one third of infertile cases are in part related to male factors **(2)**. Functional sperm DNA is essential for normal embryo development as the genetic information passed on to the next generation depends on sperm DNA integrity **(3)**.

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Sperm DNA fragmentation (SDF) has emerged as potential biomarker in the assessment of male fertility. About 25% of infertile men have SDF levels higher than that found in fertile men **(4, 5)**. SDF has been also found to be associated with early poor pregnancy outcome (recurrent idiopathic miscarriage) **(6)**. Sperm DNA fragmentation may occur through either defective chromatin condensation that may occur during spermiogenesis; apoptosis that may occur during spermatogenesis; and oxidative stress may occur during the transit period through the male genital tract **(7, 8)**.

Outcome of assisted reproductive techniques (ART) has been found to be affected by presence of SDF **(9)**. Although sperm with fragmented DNA may fertilize an egg with apparently similar efficiency as sperm without DNA fragmentation, the negative impact of a damaged paternal chromatin to the integrity of embryonic genome is usually observed after implantation. This type of damage is often manifested by early pregnancy loss **(10)**.

The Practice Committee of the American Society for Reproductive Medicine (ASRM) has stated that sperm DNA fragmentation testing might be clinically informative for in vitro fertilisation/intra-cytoplasmic sperm injection (IVF/ICSI) outcomes **(11)**. The threshold of DNA fragmentation index (DFI) is quite important, as lower rates (<%30) have been strongly related to natural conception and success in intrauterine insemination (IUI), whereas higher rates (>30%) have been linked with decreased pregnancy odds in IVF **(12)**.

Several strategies have been tried in order to overcome the presence of SDF in couples undergoing ART including varicocele repair **(13)**, oral antioxidant therapy **(14)**, short ejaculatory abstinence periods **(15)** and recurrent ejaculations **(16)**, and laboratory sperm selection techniques such as magnetic cell sorting (MACS) **(17)**, physiological intracytoplasmic sperm injection (PICSI) **(18)**, and intracytoplasmic morphologically selected sperm injection (IMSI) **(19)**.

The use of testicular rather than ejaculated sperm – with either testicular sperm aspiration (TESA) or testicular sperm extraction (TESE) – for intracytoplasmic sperm injection (ICSI) among men with high SDF was found to be associated with better pregnancy outcome **(20,21)**.Current study was designed aiming to study effect of DNA fragmentation testing and treatment on ICSI outcome among infertile couples with male factor infertility.

Patientsand methods

After approval of ethics committee of Faculty of Medicine, Suez Canal University, the present prospective control randomized study was conducted among a total of 106 infertile couples arranged for intracytoplasmic sperm injection (ICSI) due to male factor. Couples age< 32 years and BMI 20 – 25 Kg/m2 were included into the study. Female factor was assessed through hysterosalpingogram (HSG), 3D vaginal ultrasound, routine office hysteroscopy, and laboratory assessment. Couples were included if HSG showed normal uterine cavity and patent tubes, 3D vaginal U/S showed normal cavity, normal office hysteroscopy, and normal T3, TSH, prolactine and normal FSH/LH ratio in 3rd day of cycle. Semen analysis of males showed severe oligospermia and asthenoteratospermia.

Studied 106 males were divided by simple random allocation into 2 groups.

Group I included 53 males that were tested for DNA fragmentation test (Halo test).

Group II included 53 males that were directly offered ICSI cycle.

DNA fragmentation test:

DNA fragmentation was tested via halo test using Halosperm test kit. This kit determines the degree of DNA damage of a human spermatozoon through a process called sperm chromatin dispersion (SCD), which is responsible for male infertility. This process "involves the denaturation and controlled lysis of the sample in an appropriate medium and can be used with both fresh and frozen samples. Spermatozoa with intact DNA produce a dispersion halo as a result of the chromatin released from proteins that can be easily analysed using fluorescence or bright field microscopy. In contrast, spermatozoa with fragmented DNA will not produce this halo. The technique is as easy as a routine leucocyte count" **(22)**. SDF level cut-off taken as high was SDF ≥ 30% using Fernandez protocol **(23)**.

Twenty five cases of group I subjected to DNA fragmentation test were found to have negative halo test and were offered ICSI cycle **(group Ix)**. Remain-

ing 28 cases of group I who showed positive halo test were offered 3 months of treatment receiving L-carnitine, vitamin E capsules daily and instructed to avoid smoking and exposure to toxins and then DNA fragmentation test was repeated after 3 months.

Post treatment DNA fragmentation test was negative among 16 of 28 treated cases **(Group Ia)** and remain positive among 12 of 28 cases **(Group Ib)**. Those cases with persistent positive halo test were offered Testicular sperm extraction by TESE or fine needle aspiration for sperm to avoid oxidative free radicals in epididymis that may cause DNA fragmentation. Eight cases have refused testicular extraction and offered ICSI with ejaculate **(Group Ib1)** while four cases have been subjected to testicular extraction **(Group Ib2).**

ICSI protocol:

All cases were finally arranged to ICSI cycle using long antagonist protocol with triptorelin (decapeptyl®) 0.1 subcutaneous from day 21 of the previous cycle and human menopausal gonadotropin (Fastimon ®) 300 IU at second day of the cycle. Ovarian response was evaluated by serial transvaginal folliculometry and serum E2. The dose of therapeutic regimen was adjusted according to the ovarian response.

When target follicular diameter of 3 or more follicles reached 18-21 mm. HCG 1000 IU was administered and ovum pick up was done after 34-36 hours. After embryo transfer, all cases were administered luteal phase progesterone support, folic acid supplement and low dose aspirin.

Outcome measures:

Infertile couples were evaluated as regard to fertilization rate, day 5 embryo transfer, positive pregnancy test, clinical pregnancy evaluated by visible sac with viable fetus on ultrasonographic assessment and delivery of normal viable baby.

Statistical analysis:

Gathered information was processed using SPSS version 25 (SPSS Inc., chiago, IL, USA.). Quantitative data was expressed as means ± SD while qualitative data was expressed as number and percentages (%). Unpaired t test was used to test significance of difference for quantitative variables and chi square was used to test significance of difference for qualitative variables. A probability value (p-value) <0.05 was considered statistically significant.

Results

There was no statistically significant difference between both groups of the study regarding women age, BMI, number of aspirated follicles and E2 level **(Table 1)**.

Men with initially negative DNA fragmentation test and those with positive DNA fragmentation test who responded well to treatment were found to have higher fertilization ratio of M2 ova, day 3 grade I embryo, blastocyte, positive pregnancy test and positive clinical pregnancy compared to non responders to treatment. Couples who didn't perform DNA fragmentation test have no statistically significant difference compared to men with initially negative DNA fragmentation test and those with positive DNA fragmentation test who responded well to treatment regarding positive pregnancy test and clinical pregnancy while take home baby was more evident among latter groups with statistically significant difference **(Table 2)**.

Table 1: Baseline characteristics among the studied patients:

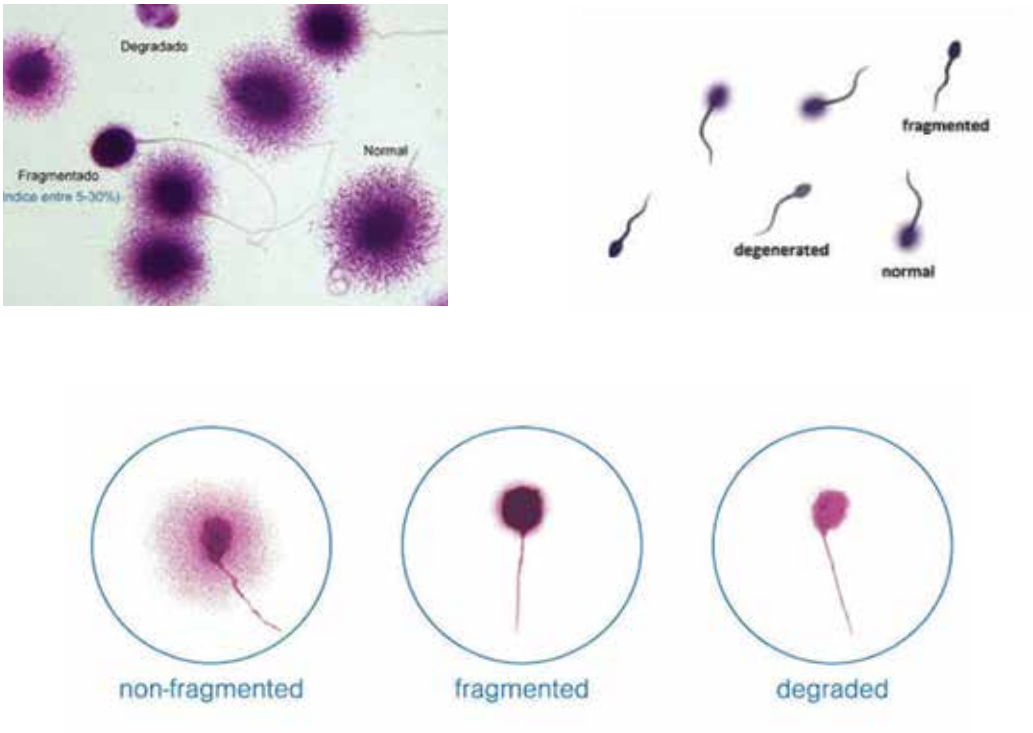
Type		Group I (n=53)	Group II (n=53)	p-value
Women Age	Mean ± SD	29.1 ± 2.7	28.9 ± 3.2	0.7 (NS)
	Range	25 – 32	25 – 32	
Women BMI	Mean ± SD	24.8 ± 3.1	24.9 ± 2.7	0.8 (NS)
	Range	20 – 25	21 – 25	
Number of aspirated follicles	Mean ± SD	9.3 ± 4.3	9.1 ± 4.8	0.8 (NS)
	Range	4 – 15	4 – 17	
E2 level	Mean ± SD	1800.9 ± 453.8	1850.2 ± 604.9	0.6 (NS)
		1100 – 2300	1000 - 2600	

Table 2: Outcome parameters:

	Group Ix (n=25)	Group Ia (n=16)	Group Ib (n=12)		Group II (n=53)	p-value
			Group Ib1 (n=8)	Group Ib2 (n=4)		
Fertilization ratio of M2 ova	20 (80%)a	14 (87.5%)a	2 (25%)b	3 (75%)ab	32 (60.4%)ab	0.001
Day 3 grade I embryo	5.2 ± 0.9a	5 ± 0.8a	2 ± 0.13b	4 ± 0.8c	5 ± 0.7a	0.001
Blastocyte	4 ± 1.06a	4.5 ± 0.9a	0.5 ± 0.08b	2 ± 0.7c	3 ± 0.9a	0.001
Positive pregnancy test	13 (52%)a	9 (56.25%)a	1 (12.5%)b	2 (50%)b	20 (37.7%)ab	0.001*
Positive clinical pregnancy	12 (48%)a	8 (50%)a	1 (12.5%)b	2 (50%)a	18 (33.9%)ab	0.001*
Take baby home	8 (32%)a	7 (43.75%)a	0 (0%)b	1 (25%)b	9 (16.9%)b	0.001*

GIx: men with initially negative DNA fragmentation test
GIa: Halo test became –ve after treatment
GIb: Halo test remain +ve after treatment
GIb1: refused TESE and offered ICSI with ejaculate
GIb2: subjected to testicular extraction
G2: offered ICSI directly with DNA fragmentation test
*statistically significant difference
a, b, c denote statistically significant within groups

Figure 1: Microscopic sperm morphology with DNA fragmentation



Discussion

The impact of DNA fragmentation as an important factor on male fertility and even pregnancy outcome is getting more attention recently. Studying DFI aims to add more prognostic information and planning to guide couples particularly those with repeated failure of ART attempts. The current study investigated burden of sperm DNA fragmentation on OCSI outcome and effectiveness of testicular versus ejaculated sperm on ICSI outcome among men with sperm DNA fragmentation.

Although many studies suggested that DNA fragmentation is an important indicator of successful fertilization **(2)**, others failed to find an effect for DNA fragmentation on fertilization rate claiming that fertilization is independent of the level of the DNA damage in spermatozoa **(24-26)**. The current study have shown higher fertilization rate, better embryo quality, higher rates of biochemical and clinical pregnancy and successful pregnancy outcome among couples with DNA fragmentation versus couples with DNA fragmentation. Successful treatment of DNA fragmentation has been found to significantly improve ICSI outcome among those couples.

Because the surgical extraction of sperm was adapted in ICSI, numerous studies have investigated the effect of sperm origin on ICSI outcomes. These studies have reported controversial results, with every study providing probable theories to support its findings. Some evidence suggested that ejaculated sperm should lead to a better outcome due to a crucial role of epididymis in the final steps of spermatogenesis, including epigenetic modification of genes **(27, 28)**, changes in the surface proteins of spermatozoa **(29)**, and maturation of sperm cells **(30)**. Some epigenetic remodelling processes is necessary for the stability of DNA and its resistance to damage and have been reported to play a crucial role in early embryogenesis **(31-35)**. Testicular sperm does not undergo these modification processes in the epididymis, and that was the explanation provided by many researchers shown higher normal fertilisation rates, implantation rates, and pregnancy rates **(36 – 38)**, and lower abortion rates in ICSI cycles using epididymal sperm than those using testicular sperm **(39, 40)**. However, some studies have shown no difference in outcomes of ICSI with testicular and epididymal sperm or with testicular and ejaculated sperm **(41, 42)** as well as current study. Some studies have even shown better fertility outcomes with testicular sperm than with ejaculated sperm **(27, 43, 44)**. These studies concluded that sperm selection during ICSI, significantly reduce the effect of epididymal transport on spermatogenesis and sperm quality **(42, 45)**. Damage to the sperm DNA along the genital tract might further explain why testicular sperm leads to better fertility outcomes than does ejaculated sperm. In the ICSI outcomes, studies have suggested that damage to sperm DNA may lead to impaired sperm decondensation, which reduces fertilisation rate, produces low-quality embryos, leads to implantation failure, and causes recurrent pregnancy loss **(46)**.

With persistent sperm DNA fragmentation despite treatment, current study has shown that there was no statistically significant difference between testicular versus ejaculated sperm regarding fertilization ratio, pregnancy rate and even successful pregnancy outcome and that only embryo quality was better with testicular sperm extraction. These findings are inconsistent with recent results by Arafa and colleagues **(1)**. Although they found that there was no difference in the fertilization rate using ejaculate and testicular spermatozoa, clinical pregnancy was significantly higher in TESA group compared to ejaculated group. Moreover, 17 live births were documented in TESA group, and only three live births were documented in ejaculate group **(1)**. Consistent with current findings; earlier studies showed no correlation between ICSI outcome and fertilization rate **(24, 26, 47)**.

Arafa and colleagues (1) ascribed their findings to thefact that spermatozoa obtained from the testis exhibit lower-DNA fragmentation and as such are more likely to undergo implantation **(48, 49)**. Furthermore, it can also be justified by results from other studies, which showed a negative correlation between clinical pregnancy and live-birth rate and high-sperm DNA damage **(49-51).** Inconsistent findings with current study could be explained by the fact that Arafa and co-wrokers haveinvestigated only men with high DNA fragmentation index that can cause to lower pregnancy outcome with ejaculated sperm.

Inconsistent with current findings, **Greco et al. (49)** havereported a higher percentage of clinical pregnancy in testicular spermatozoa compared to ejaculate (44.4% and 5.6% respectively). Similarly; **Esteves et al., (50)** recently examined ICSI outcomes using testicular versus ejaculate spermatozoa in patients with high % of SDF. They detected higher % SDF in ejaculate spermatozoa compared to testicular spermatozoa and a significant difference in the outcomes among the two groups. Reported results showed significantly higher clinical pregnancy and live-birth rates in the testicular ICSI group (52% and 47% respectively) in comparison with the ejaculate ICSI group (40% and 26% respectively).

Testicular sperm was found to be superior to ejaculated sperm in other many studies. **Pabuccu et al., (52)** found that ICSI using testicular spermatozoa obtained by TESA seems an effective option particularly for those with repeated ART failures in terms of clinical, ongoing pregnancies and miscarriages even though conventional sperm parameters are within normal range. Similarly; **Esteves et al., (53)** have shown that testicular sperm have lower levels of SDF than ejaculated sperm, with testicular sperm for high post-testicular SDF men improving ICSI outcomes compared with ejaculated sperm.

In their recent meta-analysis and systematic review; **Kang et al., (46)** showed that the risk ratios favour fresh testicular sperm for good quality embryo rate (1.17, 95% CI 1.05–1.30, P = 0.005), implantation rate (95% CI 1.02–2.26, P = 0.04), and pregnancy rate (RR = 1.74, 95% CI 1.20–2.52, P = 0.004). Inconsistently with current study, they concluded that the existing evidence suggests that testicular sperm is better than ejaculated sperm for ICSI in male with cryptozoospermia. Another systematic review supported our current findings and indicated that there is no difference between testicular sperm group and ejaculated sperm group with forest plots for pregnancy rate (OR = 0.53, 95% CI 0.19–1.42, P = 0.21, I2 = 67%) and fertilisation rate (OR = 0.91, 95% CI 0.78–1.06, P = 0.21, I2 = 73%) **(54).**

Limitation of our study: The main limitation of current study is relatively small sample size.

Conflicts of interest. None of authors have any conflict of interests

Conclusion and recommendations: Sperm DNA fragmentation is an important factor that significantly affects ICSI outcome and should be considered and adequately treated as well as it still requires further in depth investigation. Although current study has shown no significant difference between testicular and ejaculated sperm regarding ICSI outcome, divergent reports from previous studies showed that it is still controversial whether to obtain testicular sperm for better ICSI outcomes or not.

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Leptin Values in Maternal and Umbilical Cord Blood in Pregnant Women with Preeclampsia

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Abstract

Background: Leptin is a protein product of obesity gene and is synthesized mainly by adipose tissue.

Objective: The aim of this study was to determine maternal and neonatal serum leptin levels in preeclamptic and normal pregnancies.

Patient and Methods: This corss sectional study was performed on 45 preeclamptic and 45 normotensive pregnant women without other disease.Serum level of leptin was measured in all of pregnant mothers and after delivery,measured in umbilical cord of their neonates. This study was performed in Hospitals of Suez Canal University.

Results: In this study, when comparing the serum leptin levels between normal pregnancies and pregnancies complicated with both mild and sever preeclampsia there was a highly statistically significant deference in between ($p< 0.01$). plasmaleptin concentrations were significantly greater in women with pre-eclampsia than in normal controls.Umbilical cord leptin concentrations were significantly greater in women with pre-eclampsia than in normal controls. Cord blood leptin in the present study showed that there is significant positive correlation with severity of pre-eclampsia.

Conclusion: According to the results, we detected an increase in maternal plasma and umbilical cord leptin in preeclamticgroup.There was a highly statistically significant deference in between preeclamtic group and the matched control group

Keywords: Leptin, Pre-eclampsia, Umbilical Cord

Introduction

Leptin is a 16KD polypeptide hormone and the protein product of obesity gene,issynthesised and secreted mainly by adipose tissue (1). Under the supervision of the obesity gene,the adipocytes and placental trophoblasts secrete leptin which decreases the body weight by acting through its hypothalamic receptors and reducing the food intake.Leptin also act as ametabolic signal for the neuroendocrine and reproductive systems (2). Trophoblasts are responsible for the significantly increased plasma concentrations of leptin during the first two trimesters of normal pregnancy.This marked increase in synthesis of leptin is attributed to the prominent alterations of maternal weight,energy expenditure and hormonal status (3). The expression of leptin by the human trophoblasts significantly increased when cultured under hypoxic conditions.Placentalischaemia could therefor explain the rapid increase in leptin concentrations during late third trimester in preeclampsia(4). Processes associated with preeclampsia,includ-

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ing placental hypoxia lead to increased leptin levels for a compensatory mechanism to increase nutrient delivery to the fetus,by stimulating placental angiogenesis, aminoaciduptake and inhibiting apoptosis(5). Leptin levels are higher among women than men because women have abody mass index with relative higher content of fat and serum leptin level proportional to adiposity.Maternal serum leptin levels during pregnancy higher than those non pregnant women (6). Umbilical cord blood leptin is derived from fetal tissue.It is detectable by gestational week 18 but only increases to significant level after 34 weeks.Whereas much maternal leptinproduced by the placenta,most cord leptin is produced by fetal adipose tissue(7). Leptin levels in umbilical cord at term are correlated with birth weight(8).

Preeclampsia is a pregnancy-specific, multisystem disorder that is characterized by the development of hypertension more 140/90mmHg and proteiurea>300mg/day after 20 weeks of gestation in a woman with previously normal blood pressure(9). Preeclampsia is a common and serious complication of human pregnancy, affecting 5-7% of primigravid women and about 10-15% of all pregnant women(10). Preeclampsia can be subclassified into early onset, arrising before 34 weeks gestation, or late onset, arrising at or after 34 weeks gestation. Early onset preeclampsia carries a significantly greater risk of maternal and fetal morbidity and mortality due to larger and longer placental under-perfusion and prematurity (11). Preeclampsia may occur due to incomplete trophoblastic invasion and diffuse endothelial dysfunctions lead to hypoxic state and enhance leptin production (12). The exact aetiology of preeclampsia remains unknown, numerous biochemical, biophysical and clinical tests have been proposed for prediction, evaluation and assessement of aetiology, pathogenesis, courseand severity of this disease. However, it is unknown whether elevated leptin levels are the causes or consequences of preeclampsia (13).

Patient and Methods

This prospective case control study was carried out on 90 pregnant women in their third trimester of singleton pregnancies recruited from the obstetric department of Suez Canal University Hospital between April 2014 and April 2015. Divided into two equal groups. Consent was taken from each patient before investigation and management. 45 preeclamptic patients between 28-40weeks gestation subdivided into two groups, including 22 cases of mild preeclampsia and 23 cases of severe preeclampsia . 45 pregnant ladies with normal pregnancies without any symptoms or signs of preeclampsia.The inclusion criteria for the preeclampsia group were a blood pressure of 140/90mmHg or more and 300 mg or more protein in 24 hour urine sample ,or ++2 proteinuria dipstick finding in a random urine sample.preeclamptic patients wih these criteria who had not received any medications were included in the study.No medications was given to these patients before blood was taken for leptin levels and other routine tests.Patients with any of the following disorders were excluded:

1. Diabetes mellitus.
2. Essential hypertension.
3. Multifetal gestation.
4. Polyhydramnios.
5. Patient with any known medical diseases affecting the results as renal , cardiac and autoimmune diseases.
6. Body Mass Index > 27.

Sample preparation

Ten mL Blood were collected by sterile veniopuncture with 18 gauge needle from the patient before any treatment. 5 mL were transferred to test tube labelled with the name and number of the patient. The test tube was then centrifuged in cold centrifuge for 20 minutes. Serum was then separated and stored frozen at -70○C till the assay of leptin. The remaning 5 mL were similarly centrifuged, serum separated and transferred to assay of the remaning tests.5 ml blood were collected by sterile veniopuncture with 18 gauge needle from the umbilical cord after delivery and transferred to test tube labelled with the name and number of the patient to assay serum cord leptin.The maternal and umbilical cord blood leptin concentrations of preeclamptic group (n=45) were compared with those of normal pregnancies (n=45)who have similar BMI and gestational age matched.

Statistical Analysis

The clinical and laboratory data were recorded on a "investigative report form". The data were analyzed statistically using statistical software package

"SPSS", version 9.05, EchoSoft Corp., USA, 1998. The following stastical tests were used:

I. Descriptive statistics: Mean (X), standard deviation (SD) Minimum and maximum values (range)

II. Student "t" test: to compare between independent means of parametric data.

The results were evaluated at the 95% confidence interval, and $p<0.05$ significance level.

Results

In this study, 90 patients were analysed. Amoong which 45 were normotensive and 45 preeclampticpatients.Out of 45 preeclamptic, 22 were with mild preeclampsia and 23 were having sever preeclampsia.There was a significant statistical high level of maternalleptin in severe pre-eclampsia(48.96 ± 5.53ng/ml.) when compared to mild pre-eclampsia(28.05 ± 5.7ng/ml.) which in turn shows higher levels of leptin when compared to control group(26.18 ± 3.37ng/ml). Thus, the difference between the pre-eclamptic groups as a whole and the control normotensive group was statistically significant (table 1).There was a significant statistical high level of umbilical cord leptin in severe pre-eclampsia (6.65 ±1.11ng/ml.) when compared to mild pre-eclampsia (4.45 ± 0.86ng/ml.) which in turn shows higher levels of leptin when compared to control group (4.04 ±1.10ng/ml.). Thus, the difference between the pre-eclamptic groups as a whole and the control normotensive group was statistically significant(table 2).The age of mild pre-eclamptic group ranged from 21-40 years with a mean of 27.45±6.29 years.

The age of severe pre¬-eclamptic group ranged from 20-38 years with a mean of 27.13±5.91 years. The age of control group ranged from 20-40 years with a mean of 27.1±5.98 years.There was no statistical significant difference between the three studied groups regarding the age.

There was statistically significant difference between the three studied groups regarding the gestational age, blood pressure, blood picture, renal function, liver function and proteinuria. The results are summarized in table (3).

Table (1): Maternalleptin among the studied groups.

	Maternal leptin			ANOVA	
	Range	Mean ±	SD	f	P-value
Group A1 (Mild P.)*	22 - 35	28.05 ±	5.7	48.9	<0.01
Group A2 (severe P.)	38 - 60	48.96 ±	5.53		
Group B (control)	20 - 33	26.18 ±	3.37		
Tukey's test					
A1 & A2	**A1 & B**			**A2 & B**	
<0.01	<0.05			<0.01	

*P : preeclampsia

Table (2): Umbilical Cord leptin among the studied groups.

	Cord leptin			ANOVA	
	Range	Mean ±	SD	f	P-value
Group A1 (Mild P.)*	3 - 8	4.45 ±	0.86	48.3	<0.01
Group A2 (severe P.)	5 - 9	6.65 ±	1.11		
Group B (control)	3 - 7	4.04 ±	1.10		
Tukey's test					
A1 & A2	**A1 & B**			**A2 & B**	
<0.01	<0.05			<0.01	

Table (3): Statistical comparison of studied parameters between studied groups.

parameters	Mild Preeclampsia N=22 Mean±SD	Severe Preeclampsia N=23 Mean±SD	Control N=45 Mean±SD	Anova(F) test	Anova(F) test
Gestational age	35.78±1.73	31.83±2.64	37.05±1.86	41.4	<0.01
Systolic Bl.Pr.	145.45±5.10	186.96±17.95	114.70±5.05	198.3	<0.01
Diastolic Bl.Pr.	94.55±5.10	111.30±7.57	74.90±5.06	112.9	<0.05
Haemoglobin	10.35±0.61	9.85±0.55	10.9±0.62	3.9	<0.05
Platelet Count	194.32±33.00	181.22±21.04	224.42±49.35	10.1	<0.01
Urea	25.09±5.29	30.91±7.58	22.84±3.37	18.4	<0.01
Creatinine	1.05±0.13	1.12±0.12	1.00±1.00	16.3	<0.01
Uric Acid	4.15±0.54	4.76±0.89	3.72±0.60	18.3	<0.01
SGPT	18.23±2.09	38.17±7.19	16.11±2.55	22.6	<0.01
SGOT	19.42±3.02	37.92±6.99	17.2±1.99	26.4	<0.01
Protinuria	429.0±65.3	2707.2±485.7	<300	10.99	<0.01
Prothrombine	90.59±3.80	87.30±3.90	92.71±4.02	12.4	<0.01

There was positive correlation between maternal leptin and umbilical cord leptin, systolic and diastolic blood pressure, urea, uric acid,creatinine, SGOT, SGPT and protinuria (table 4).Also,there was positive correlation between cord leptin and maternal leptin, systolic and diastolic blood pressure, urea, uric acid, creatinine, SGOT, SGPT and protinuria (table 5). Cut off between Group A (pre-eclampsia) and Group B (control) as regard maternal leptin = 39 by sensitivity = 88.6, specifity =100.0, positive predictive value =100.0, negative predictive value =71.4 by accuracy = 0.957.

Cut off between Group A (pre-eclampsia) and Group B (control) as regard cord leptin = 5.5 by sensitivity = 87, specifity =60.0, positive predictive value =88.6, negative predictive value =60.0 by accuracy = 0.730

Table (4): Correlation between maternal leptin and studied variable

	Maternal leptin	
	r	**P.value**
Age	-0.090	>0.05
Gestational age	-0.536	<0.01
Systolic blpr	0.741	<0.01
Diastolic blPr	0.641	<0.01
Hg	-0.332	<0.05
Platelets	-0.222	>0.05
Sgpt	0.88	<0.01
Sgot	0.87	<0.01
Urea	0.262	>0.05
Creatinine	0.391	<0.01
Uric acid	0.294	0.05
Prthrombin activity	-0.427	<0.01
Umbilical cord leptin	0.602	<0.01
protinuria	0.739	<0.01

Table (5): Correlation between cord leptin and studied variable

	Umbilical cordleptin	
	r	**P.value**
Age	-0.113	>0.05
Gestational age	-0.511	<0.01
Systolic blpr	0.66	<0.01
Diastolic blPr	0.639	<0.01
Hg	-0.052	>0.05
Platelets	-0.318	<0.01
Sgpt	0.7	<0.01
Sgot	0.69	<0.01
Urea	0.482	<0.01
Creatinine	0.291	<0.01
Uric acid	0.455	<0.01
Prthrombin activity	-0.397	<0.01
Maternal leptin	0.602	<0.01
protinuria	0.739	<0.01

Discussion

Preeclampsia may be mild or severe depending on the degree of blood pressure elevation, degree of proteinuria, extent of edema and the presence of signs and symptoms, including epigastric pain, severe headache and blurred vision. However severe pre-eclampsia can result in bleeding disorders and death (14).

The present study demonstrated that maternal plasma leptin is increased in women with pre-eclampsia compared to normal pregnant woman. Furthermore, maternal plasma leptin concentration in pre-eclampticspositivily correlates with fetal cord concentrations. This is only present in pre-eclamptic woman but not in healthy pregnant woman.

In a longitudinal study done by Kolyingit et al. (15) showed that increased plasma leptin level with severity of pre-eclampsia could be taken as a marker of placental hypoxia in severe pre-eclampsia.

In the current study, the mean value of plasma leptin level in control group (normotensive pregnant women) was 26.18 ± 3.37ng/ml, in mild preeclamptic group it was 28.05 ± 5.7ng/ml and in sever preeclamptic group it was 48.96 ± 5.53ng/ml.

In the current study, according to Receiver Operating Characteristic (ROC) curve of serum leptin and preeclampsia, the best cutoff value was 39ng/ml which gives a sensitivity of 88% and specificity of 100 %.

In this study, when comparing the serum leptin levels between controls and pregnancies complicated with both mild and sever preeclampsia there was a highly statistically significant deference in between ($p< 0.01$). This in correspondence with the study by Anim-Nyame et al. (16)in whichCirculating concentrationsof leptin were measured (19 women with pre-eclampsia and 13 normal pregnantcontrols) In this study, plasma leptin concentrationswere significantly greater in women with pre-eclampsia thanin normal controls. This study confirmed thatplasma leptin concentrations are increased in established pre-eclampsia.

Also, the results of the study is in agreements with another study done by Donghong et al. (17)which was carried out to determine the changes in serum levels of adiponectin, leptin and soluble leptin receptor, and in the free leptin index in women with pre-eclampsia.

The result of the present work showed that maternal plasma leptin concentration reflects accurately the severity of hypertension in pre-eclamptic women. This is evidence by the significant positive correlation between leptin values and blood pressure measurements.This finding is consistent with finding of Donghong et al. (17)who found that maternal plasma leptin levels reflect the severity of hypertension in woman suffering from EPH Gestosis as they reported that there was a significant difference in leptin levels between those with severe EPH Gestosis as compared to mild or control and there was a significant positive correlation between leptin values and values of blood pressure.

Cord blood leptin in the present study showed

that there is significant positive correlation with severity of pre-eclampsia. the mean value of cord leptin level in control group (normotensive pregnant women) was 4.04 ± 1.10ng/ml, in mild preeclamptic group was 4.45 ± 0.86ng/ml and in sever preeclamptic group was 6.65 ± 1.11ng/ml. This is consistent with the study of Salvatores et al. (18), Acromite et al. (19),Kocyigit et al. (20) and Akerman et al. (21)who's study included 40 normotensive pregnant women and 80 women with pre-eclampsia measuring the concentration of leptin in the umblical cord and reported that the level of cord leptin increased with the severity of pre-eclampsia.

Many studies measured the concentration of leptin in umbilical cord and reported that the level of cord leptin varies between 2.5-10 ng/ml (19,22) and this correlates with the present study which ranged from 3-9 ng/ml.

Conclusion

Maternal serum leptin is increased in preeclamptic women and more in sever than mild preeclampsia. From previous data, it coud be concluded that serum levels of leptin may be used as a marker of severity of preeclampsia.Umbilical cord leptinincreased inpreeclamptic women than normal pregnancies.

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Quality of life after hysterectomy at tertiary care hospital in upper Egypt

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Abstract

Background: Hysterectomy is the most common major gynaecological surgery often performed for benign lesions.Many studies have reported adverse psychosocial outcomes post-hysterectomy. This study is to evaluate psychological wellbeing, and quality of life in patients undergoing hysterectomy for non-malignant conditions, in comparison with patientsundergoing surgery other than hysterectomy.

Methods: A comparativestudy was conducted on 90consecutive out-patients who underwent hysterectomy for non-malignant indications at least 6 months ago. The comparison group comprised of 45 consecutive out-patients whounderwent gynaecological surgery other than hysterectomy at least 6 months ago formed the comparison group. Thestudy participants were evaluated on , Psychological General Wellbeing Index (PGWBI), and Women's Quality of Life Questionnaire (WOMQOL).

Results: No statistically significant different on socio-demographic characteristics such as patient age, marital status, social and education levels .There were no significant differences in the study groups on scores of PGWBI, and WOMQOL . Both the study groups had good marital adjustment andmajority reported no depression and anxiety.

Conclusion: There is no major psychiatric morbidity, decline in marital adjustment and quality of life afterhysterectomy for benign conditions among women at Aswan university hospital in upper Egypt.

Keywords: Hysterectomy, complications, Psychological well-being and Quality of life.

INTRODUCTION

Hysterectomy is the removal of uterus and it is the commonest major surgical procedure performed in gynaecology.(1)

It is the second most common operative procedure performed on women in world after lower segment caesarean section(LSCS). Its incidence varies between 6.1 to 8.6 per 1000 female. (2)

Hysterectomy for benigngynaecological lesions are usuallyundertaken toimprove the quality of life (QOL) of affected females. (3)(4)

The rateof hysterectomy vary with geographic area, patientexpectations, training and practice patterns of localgynaecological surgeons.(5)

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Although hysterectomy is the definitive management formany conditions, it is not risk free. It is associated withrisk of iatrogenic premature menopause, surgical andanaestheticrisks. (5)

Hysterectomy is theorized to cause depression because of the perceived loss of feminine self-image, strength, and self-esteem, as well as feelings of deformation, mutilation, and the mourning of the loss of fertility.(6)

The uterus symbolizesfemininity, fertility, sexuality, strength,vitality, youth, attractiveness, competency, regulation of body processes, and control of the life rhythm.(7)

Anticipatory guidance regardingdecreased libido, physical changes, loss and griefreactions, and the possible complications of surgery can affect sexual functioning, marital adjustment, andconsequently, QOL after removal of uterus.(8)

The positiveoutcomes of hysterectomy include decreases in chronic pelvic pain (CPP) and pain during intercourse, as well as the elimination of dysmenorrhea and dysfunctional uterinebleeding (DUB).(9)

However, hysterectomy may sometimes result in new symptoms related to pain, sexual dysfunction, and psychological distress, as well as in long-term adverse effects related to ovarian failure(OF).(7)

Some researchers have reported adverse sequelae of hysterectomy such as depression, psychosis, anxiety andpsychosomatic disturbances.(10)(11)

On the contrary, fewprospective studies concluded that hysterectomy does not lead to psychiatric disorders. Hence the need for this study.

METHODS

This comparative studywas conducted in the Departments of Obstetrics and Gynaecology,Aswan University hospital, Aswan, EgyptbetweenAugust 2017and August 2018a written informed consent was obtained from all the studyparticipants. The study group comprised of 90 consecutive adult out-patients who underwent abdominalor vaginal hysterectomy at least 6 months ago.

The comparison group consisted of 45 out-patients who have undergone gynaecological surgeryexcept hysterectomy6 months ago.

The socio-demographic and clinical data such as age,marital status, domicile, education level, nature ofgynaecologicaldisorder, indication for hysterectomy andother surgery was recorded on a specific proformadesigned for the study. The following tool were used.

The Women's Quality of Life questionnaire(WOMQOL): Quality of life is a multi-dimensionalconstruct and defined subjectively. The WOMQOL wasdeveloped as part of a community-based study of women's health, including mental health through themenstrual cycle with no known pathology.(12)

A generic conceptualization of QOL was used in theconstruction of the measure that weighted health and no health factors to ensure the representation of the lifeexperiences of a broad range of women in thecommunity-based Women Wellness study. Theparticipants were asked to answer "yes", "no" or "notapplicable" to the 40 questions in the WOMQOL based on how they have felt in the last week of their life.

The Psychological General Well-being Index (PGWI)contains 22 questions, covering the six subscales anxiety,depressed mood, positive well-being, general health, vitality and self-control (13)

The validity and reliability of this instrument are welldocumented, and it has previously been used to comparepatient groups and to determine the effect of anintervention on the patient's sense of subjectivewellbeing.(14)

Results

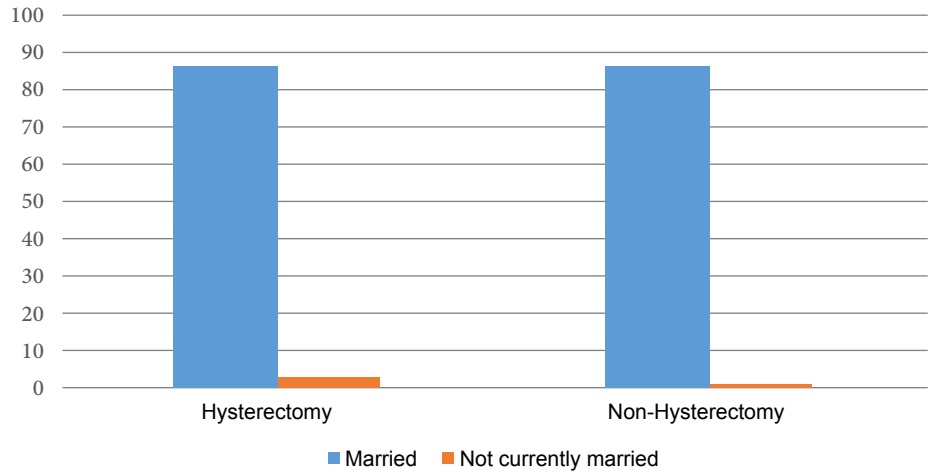
In our results the hysterectomy group and non-hysterectomy comparison group were not statistically significant different on socio-demographic characteristics such as patient age, marital status, social and education levels

Table 1: Patient age

Item	Hysterectomy	Non-hysterectomy
Mean±SD (years)	45.5±11.5	44±9
<35 years	6(6.66%)	9(25.71%)
35-49 years	76(84.44%)	24(68.57%)
≥50 years	8 (8.88%)	2(5.71%)

Table 2: Patient marital status

Item	Hysterectomy	Incidence	Non-hysterectomy	Incidence
Married	87	(96.66%)	44	(97.77%)
Not currently married	3	(3.33%)	1	(2.22%)



This table and chart show that the patient marital status among hysterectomy group were 87 patients married with incidence 96.66% and 3 patients not currently marriedwith incidence 3.33%. But non-hysterectomy group were 44 patients married with incidence97.77% and 1 patients not currently married with incidence2.22%.

Table 3:Patientdomicile

Item	Hysterectomy	Incidence	Non-hysterectomy	Incidence
Urban	68	(75.55%)	33	(73.33%)
Rural	22	(24.44%)	12	(26.66%)

Table 4:Patienteducation

Item	Hysterectomy	Incidence	Non-hysterectomy	Incidence
Primary-Highschool	73	(81.11%)	37	(82.22%)
College	17	(18.88%)	8	(17.77%)

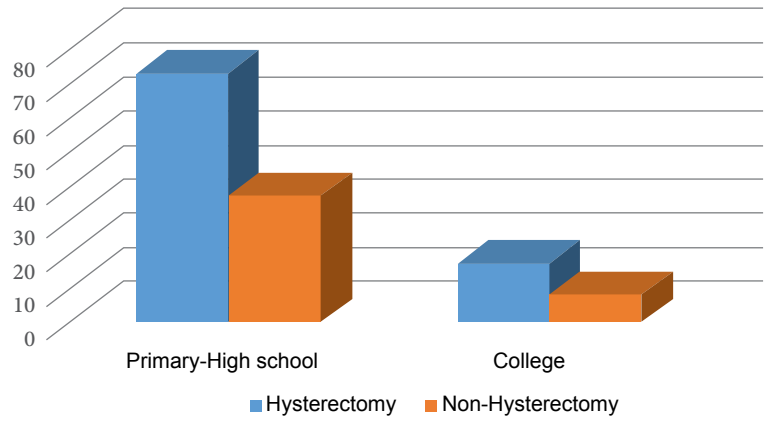


Table 5: Mean scores of the two groups on study measures.

Item	Group	N	Mean	Std. Deviation
WOMQOL	Hysterectomy	90	34.44	5.538
	Non-hysterectomy	45	34.70	5.335

Table 6: Mean scores of the two groups on study measures

Item	Group	N	Mean	Std. Deviation
PGWBI	Hysterectomy	90	84.79	14.224
	Non-hysterectomy	45	85.72	10.264

As regard The Women's Quality of Life questionnaire (WOMQOL) inhysterectomy group the mean was 34.44 and Std. Deviation was 5.538 , but in the non-hysterectomy group the mean was 34.70 and Std. Deviation was 5.335.

As regard The Psychological General Well-being Index (PGWI) inhysterectomy group the mean was 84.79 and Std. Deviation was14.224 . but in the non-hysterectomy group the mean was 85.72 and Std. Deviation was 10.264.

The mean scores of the study groups on measures of (WOMQOL) and (PGWI) there were no significant differences in the study groups on these measures.

DISCUSSION

Hysterectomy for benigngynaecological lesion are usually undertaken toimprove the quality of life of affected females. (3)(4)

The rateof hysterectomy vary with geographic area, patientexpectations, and training and practice patterns of localgynaecological surgeons. (5)

The positive outcomes of hysterectomy include decreases in chronic pelvic pain (CPP) and dyspareunia, as well as the elimination of dysmenorrhea and dysfunctional uterine bleeding (DUB).(9)

However, hysterectomy may sometimes result in new symptoms related to pain, sexual dysfunction, and psychological distress, as well as in long-term adverse effects related to ovarian failure(OF).(7)

In the current study weaimed to evaluate prevalence of, psychological well-being and QOL in women post-hysterectomy in comparison to women undergoing gynaecological surgeryother than hysterectomy at a tertiary care centre.

Earlier studies have reported higher prevalence ofpsychiatric morbidity post-hysterectomy.(15)(16)(17)(18)

In the current study The a minority ofpatients after hysterectomy have depression and anxiety, butSnaith reported prevalence of depressionfollowing hysterectomy to be around 20 %.(19) andAckner found that Psychiatriccomplaints were found in 30% of women post hysterectomy.(8)

The findings of the presentstudy are in agreement with previous researchers whofound no significant increase in depressive disorders afterhysterectomy.(20)

Present study found most women having good quality of life which isconsistent with the observations of earlier studies. (21)(22)

Hysterectomy is effective in reducingdyspareunia and pelvic pain which may translate intobetter sexual functioning, and consequently better marital-adjustment and quality of life.

Recent prospective studies have determined that nonegative effects resulted from hysterectomy overall, andsome authors have even found positive effects ofhysterectomy on the psychosocial and sexual well-beingof women.(23)

CONCLUSION

This study suggests that there is no major psychiatricmorbidity, decline in marital adjustment and quality of lifeafterhysterectomy for benign conditions among women at Aswanuniversity hospital.

RECOMENDATION

Future multi-centric research on the peculiarsocio-cultural implications, use of larger sample size and effect of hysterectomy will be a significant addition to theavailable evidence in Egypt.

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Use of Metformin versus Chromium Picolinate in the Management of Polycystic Ovarian Syndrome: A Randomized Controlled Clinical Trial

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Abstract

Objective: To compare the effects of metformin and chromium picolinate on females with polycystic ovarian syndrome.

Background: Polycystic ovarian syndrome is the most common endocrinopathy of reproductive-aged women. It presented with excessive hair growth(hyperandrogenism), menstrual irregularities (anovulation), and polycystic ovaries.They are commonly accompanied by obesity, insulin resistance, and infertility.

Methods: Sixty female patients with polycystic ovarian syndrome were enrolled for our study at Suez Canal University Hospitals, from January 2016 to December 2016. They were randomly assigned into 2 groups. FSH, LH, testosterone, TSH, prolactin, fasting blood sugar, fasting insulin, QUICKI, HOMA-IR were measured for all patients. The first group received metformin 500 mg twice daily while the other group received chromium picolinate 200 μg once daily for 3 months.

Results: Fifty-four patients completed the study, we compared the effect of metformin and chromium in patients with PCOs, it showed significant difference regarding free testosterone, serum prolactin (p=0.01), FSH level, FBS, Fasting insulin and QUIKI between the two groups (p=0.001).

In Group I, there was significant difference regarding the values of BMI, free testosterone, TSH, fasting blood sugar, fasting insulin, HOMA-IR, QUICKI, ovarian volume(p <0.001)prolactin (p=0.01), and hirsutism score (0.02) before and after treatment with metformin. In Group II, there were significant differences regarding the values of testosterone (p=0.01), BMI LH, FSH, TSH, fasting blood sugar, fasting insulin, HOMA-IR, QUICKI, ovarian volume and hirsutism score before and after treatment with chromium picolinate(p =0.001).

After treatment, twenty-two (81.48%) patients had normal menstruation in Group I compared to 24 (88.89%) patients in Group II (p=0.35) meanwhile, 11(40.7%) patients had normal ovulation in Group I compared to 12 (44.4%) patients in Group II (p=0.78). Pregnancy occurred in 6 (22.2%) patients in group I and 5 (18.5%) patients in Group II with no significant difference (p=0.73). After 3 months of treatment, there were no significant differences between the groups regarding the side effects except abdominal discomfort which was more significant with metformin (p= 0.018).

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Conclusion: Chromium picolinate was better tolerated than metformin due to lower side effects; nevertheless, no significant differences were observed between the two groups regarding ovulation and pregnancy rates. Therefore, metformin could be replaced by chromium in some PCOS patients.

Key words: Polycystic Ovarian Syndrome – metformin – Chromium Picolinate

Introduction

Polycystic ovarian syndrome (PCOS) is the most common endocrinopathy of reproductive-aged women, affecting 6–8% of this population (1). The major clinical features are excessive hair growth (hyperandrogenism), menstrual irregularities (anovulation), and polycystic ovaries. This triad of symptoms is commonly accompanied by obesity, insulin resistance, and infertility (2).

The biguanide, metformin (dimethylbiguanide), was introduced in 1957 as an oral glucose-lowering agent to treat non-insulin dependent diabetes mellitus (3). For women who remain anovulatory with such simple measures, metformin therapy was found to be as effective as treatment with gonadotrophins but without the associated risk factors of multiple pregnancy and Ovarian Hyperstimulation Syndrome (4).

Chromium picolinate, consists of trivalent chromium, is an extremely safe and highly tolerable trace mineral which is present in normal diet and is combined with picolinate acid in order to enhance gut absorption (5). It effectively reduces insulin resistance and treats hyperinsulinemiaas well as hyperandrogenemia but it did not significantly affect the hormonal changes (6). In the women with PCOS, chromium picolinate (200 µg daily) improved the glucose tolerance but did not improve ovulation or hormonal profiles (7).

Amooee et al. (2013) compared the effect of combination of clomiphene + metformin and clomiphene + chromium picolinate on ovulation induction and pregnancy rate in clomiphene citrate-resistant patients with PCOS. They concluded that chromium picolinate was better tolerated compared to metformin (8). So, we aimed to compare the effects of metformin and chromium on patients with polycystic ovarian syndrome.

PATIENTS AND METHODS

Amooee et al. (2013) compared the effect of combination of clomiphene + metformin and clomiphene + chromium picolinate on ovulation induction and pregnancy rate in clomiphene citrate-resistant patients with PCOS. They concluded that chromium picolinate was better tolerated compared to metformin (8). So, we aimed to compare the effects of metformin and chromium on patients with polycystic ovarian syndrome.

This prospective randomized controlled clinical trial was conducted at Gynecology outpatient clinic, Suez Canal University Hospitals. The study was approved by faculty of medicine, Suez Canal University and an informed written consent was obtained from all participants. The study included 60 female patients with polycystic ovarian syndrome in the period from January 2016 to December 2016.Six patients were lost during follow-up, so they were excluded from the study (Fig.1).

The inclusion criteria included patients with age ranging from 18 to 30 years and fulfilling two out of three of the criteria in diagnosis of PCOS according to the revised Rotterdam Consensus Workshop Criteria (9); Chronic anovulation, Clinical and/or biochemical evidence of androgen excess and Polycystic-appearing ovaries on transvaginal ultrasound. Patients were excluded if they had kidney disorders (metformin and chromium picolinate excreted via the kidney), diabetes, adrenal tumors, Cushing's syndrome, Thyroid gland dysfunction, Hyperprolactinemia, Lung diseases, Liver diseases or Heart failure. Patients received oral contraceptives in the past 2 months were excluded from the study.Semen analysis for the husband was done to exclude medical problem for the husband.

Thorough examination was done to all patients, hirsutism was assessed according to the modified Ferriman-Gallwey score (9 body locations) (10) and Body mass index (BMI) was calculated.

On the third day of a spontaneous or induced/ cycle and after 8 hours of overnight fasting (at 9 am in the morning), Serum FSH, LH, total testosterone, free testosterone, prolactin, fasting blood sugar (FBS) and fasting Insulin were done. Oral Glucose Tolerance test was performed. Insulin sensitivity was assessed through quantitative insulin sensitivity check index (QUICKI) defined as 1/ [log (fasting insulin) + log (fasting glucose)] while insulin resistance was assessed by Homeostasis model assessment (HO-

MA-IR) defined as [fasting glucose - fasting insulin] / 22.5 (11). All the hormonal assays were performed twice at baseline and after 3 months of treatment.

Baseline transvaginal ultrasound was done,ultrasonographic criteria used for the diagnosis of PCOS were the presence of 12 or more 2–9-mm ovarian follicles, a peripheral distribution of ovarian follicles (necklace appearance), the increased echogenity or surface of ovarian stroma on a cross-sectional cut, anovulation and endometrial thickness and an ovarian volume of more than 10 cm3.Sixty patientswere randomly (using a computer-based random digit generator) assigned to one of the two groups.Fifty-four completed the study, Group I included 27 patients received metformin 500 mg twice daily for 3 months in the form of Cidophage Tablets (Manufactured by CID Company) and group II included 27 patients received chromium picolinate 200 μg once daily for 3 months in the form of Chromium Capsules (Manufactured by MepacoMedifood Company). Women were evaluated for possible side effects of therapy including abdominal discomfort, vomiting, diarrhea, indigestion, headache, nausea, and loss of appetite. β-HCG was checked for detection of pregnancy 1 week after the missed period. If β-HCG level was >25 (by the vidas method), the patient was considered pregnant and medications were discontinued.

Statistical analysis

All data were collected, tabulated and statistically analyzed using SPSS 15.0 (SPSS Inc., Chicago, IL, USA). Continuous Quantitative variables were expressed as mean ± SD and range while Categorical Qualitative variables were expressed as number and percentage.Two sample t-test(paired and unpaired) were used to test the significance of difference for quantitative variables while the Chi-square test was used to test the significance for qualitative variables. A probability value (p-value) < 0.05 was considered statistically significant.

RESULTS

Our study included fifty-four patients with PCOS, the first group received metformin and the second group received Chromium picolinate for 3 months.

Demographically, the mean age was 27.1 ± 3.3 and 27.9 ± 3.0years ingroup I and Group II respectively, with no statistically significant difference (p=0.33). The mean BMI was 30.8 ± 2.5 kg/m3 in Group I and 30.7 ± 2.3 kg/m3 in Group II. There were no significant differences between both groups regarding body mass index (p=0.87). Nulliparous women were 62.96% in group I and 55.56% in Group II, there was no significant difference in parity between the two groups (p=0.6) or duration of infertility (p=1.0).Regarding menstrual pattern among the studied groups, only 11.1 % had normal menstrual cyclein group I and 7.5%in group II. The difference was not statistically significant (p=0.94). (Table 1).

There were no significant differences between groups inthe hirsutism score (p=0.17), free testosterone (p= 0.43), luteinizing hormone (p= 0.36), thyroid stimulating hormone (p= 0.14), serum prolactin (p= 0.39), fasting blood sugar (p= 0.49), fasting insulin (p= 0.59), QUICKI (p= 0.36), HOMA-IR (p= 0.64) and ovarian volume (p= 0.32) before treatment (Table 2).

Table (3) compared the effect of metformin and chromium in patients with PCOs, it showed significant difference regarding free testosterone, serum prolactin (p=0.01), FSH level, FBS, Fasting insulin and QUIKI between the two groups (p=0.001).

In Group I, there was significant difference regarding the values of BMI, free testosterone, TSH, fasting blood sugar, fasting insulin, HOMA-IR, QUICKI, ovarian volume(p <0.001)prolactin (p=0.01), and hirsutism score (0.02) before and after treatment with metformin (table 4), while no difference was found in the values of LH and FSH. In Group II, there were significant differences regarding the values of testosterone (p=0.01), BMI LH, FSH, TSH, fasting blood sugar, fasting insulin, HOMA-IR, QUICKI, ovarian volume and hirsutism score before and after treatment with chromium picolinate(p =0.001). There was no difference regarding the values of prolactin (Table 5).

After treatment, twenty-two (81.48%) patients had normal menstruation in Group I compared to 24 (88.89%) patients in Group II with no significant difference (p=0.35). 11(40.7%) patients had normal ovulation after treatment in Group I compared to 12 (44.4%) patients in Group II with no significant difference (p=0.78). Pregnancy occurred in 6 (22.2%) patients in group I and 5 (18.5%) patients in Group II with no significant difference (p=0.73) (Table 6). After 3 months of treatment, there were no significant differences between the groups regarding side effects as nausea, indigestion, vomiting, diarrhea, loss of appetite or headacheonly, abdominal discomfort was more significant with metformin (p= 0.018).

Fig. 1 the flowchart of the studied population.

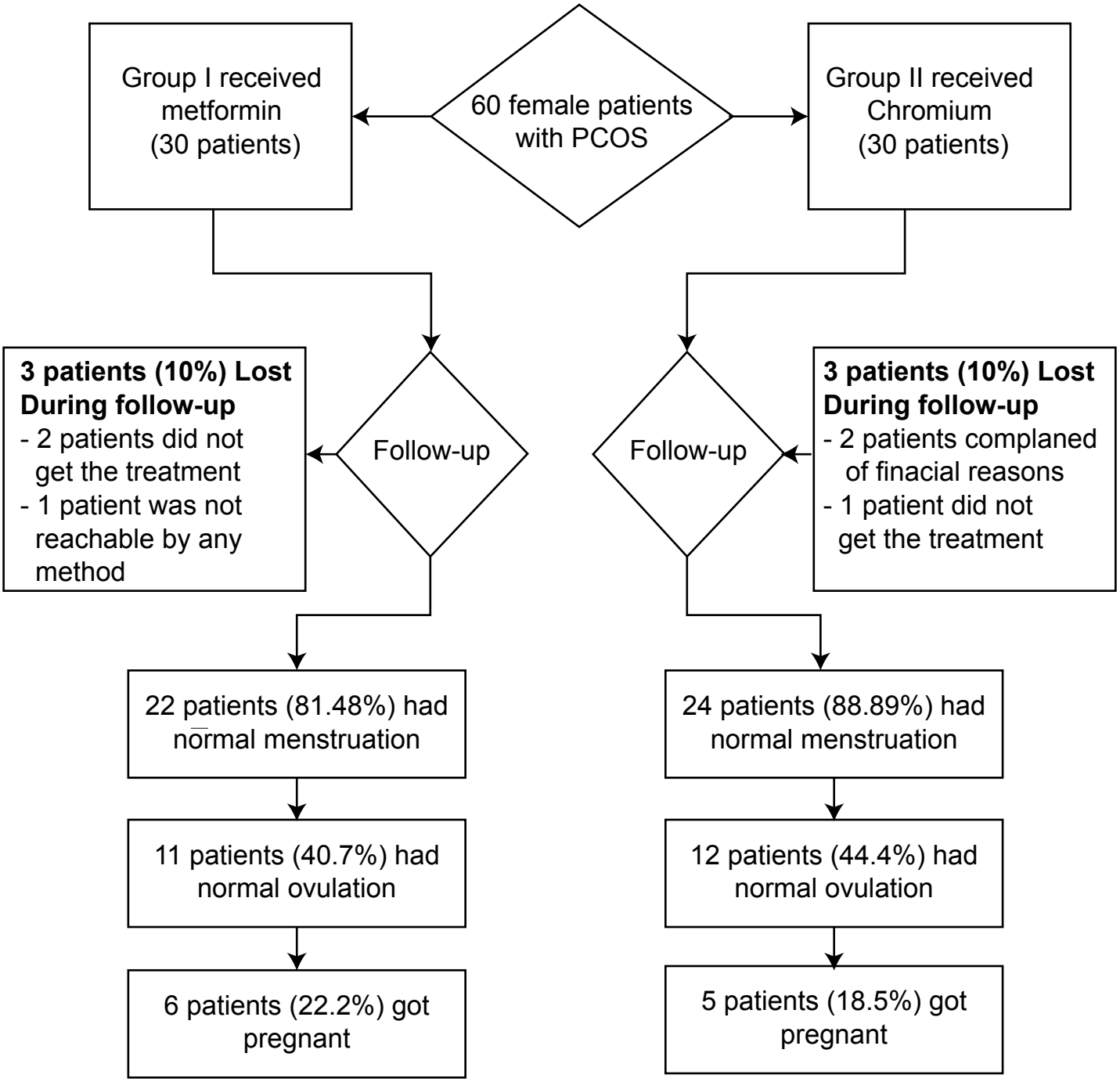

Table (1): Comparison between metformin and chromium picolinate groups regarding demographic characters.

Title		Metformin (n=27)		Chromium (n=27)		P value
		No.	%	No.	%	
Parity	Nulipara	**17**	**62.96**	**15**	**5.56**	**0.6NS**
	Para 1	**6**	**22.22**	**5**	**18.52**	
	Para ≥2	**4**	**14.81**	**7**	**25.93**	
Age (years)	22-26 Y	**13**	**48.15**	**10**	**37.04**	**0.69NS**
	27-31 Y	**11**	**40.74**	**14**	**51.85**	
	32-35 Y	**3**	**11.11**	**3**	**11.11**	
	Mean±SD	**27.56±3.36**		**27.93±3.16**		**0.33 NS**
BMI (kg/m2)	Mean ±SD	**30.83±2.52**		**30.73±2.30**		**0.87 NS**
Infertility duration	Mean±SD	**2.7±1.1**		**2.76±1.3**		**1.0 NS**
Menstrual pattern	Normal N (%) Oligomenorrhea Amenorrhea Irregular	**2 (7.41)** **8 (29.63)** **6 (22.22)** **11 (40.74)**		**3 (11.11)** **7 (25.93)** **5 (18.52)** **12 (44.44)**		**0.94 NS**

NS: Not statistically significant difference

Table (2): Comparison between metformin and chromium picolinate groups regarding baseline hormonal profile, hirsutism, FBS, fasting insulin, and ovarian volume.

Title	Metformin (n=27)		Chromium (n=27)		P value
	Mean	SD	Mean	SD	
Hirsutism Score (0-36)	22.74	7.52	19.63	8.91	0.17NS
F.Testosterone (ng/dl)	2.67	0.51	2.57	0.41	0.43 NS
LH (mIU/ml)	9.10	1.43	8.70	1.73	0.36 NS
FSH (mIU/ml)	6.60	0.82	7.07	1.43	0.14 NS
TSH (µg/dl)	3.55	0.61	3.24	0.71	0.09 NS
Prolactin (ng/dl)	8.15	2.87	8.81	2.74	0.39 NS
FBS (mg/dl)	115.89	10.02	114.00	9.77	0.49 NS
Fasting insulin (µU/ml)	16.30	3.16	15.80	3.57	0.59 NS
Homa-IR	4.43	0.52	4.37	0.44	0.64 NS
QUICKI	0.31	0.01	0.31	0.01	0.36 NS
Ovarian Volume (cm3)	11.70	1.83	12.10	0.92	0.32 NS

NS: Not statistically significant difference

Table (3): Comparison between metformin and chromium picolinate groups after treatment regarding hormonal profile, hirsutism, FBS, fasting insulinand ovarian volume.

Title	Metformin (n=27)		Chromium (n=27)		P value#
	Mean	SD	Mean	SD	
BMI (kg/m2)	28.51	2.44	28.60	2.35	0.89 NS
Hirsutism Score (0-36)	7.07	4.90	5.52	3.26	0.18 NS
F. Testosterone (ng/dl)	2.10	0.31	2.50	0.38	0.01*
LH(mIU/ml)	9.08	1.22	8.60	1.53	0.21NS
FSH(mIU/ml)	6.50	0.51	4.64	1.17	0.001*
TSH(µg/dl)	3.20	0.31	3.05	0.41	0.13 NS
Prolactin(ng/dl)	7.37	1.87	8.63	1.61	0.01*
FBS(mg/dl)	95.22	7.36	79.81	5.98	0.001*
Fasting insulin (µU/ml)	12.70	2.01	9.10	2.24	0.001*
Homa-IR	3.67	0.32	3.14	0.28	0.001*
QUICKI	0.32	0.01	0.35	0.01	0.001*
Ovarian volume (cm3)	8.37	2.96	8.09	2.28	0.71 NS

* Statistically significant difference
NS: Not statistically significant difference

Table (4): Comparison of metformin group before and after treatment regarding hormonal profile, infertility, hirsutism, FBS, fasting insulin, and ovarian volume.

Title	Before treatment (n=27)		After treatment (n=27)		P value#
	Mean	SD	Mean	SD	
BMI (kg/m2)	30.83	2.52	28.51	2.44	0.001*
Hirsutism Score (0-36)	22.74	7.52	7.07	4.90	0.02*
F. Testosterone (ng/dl)	2.67	0.51	2.10	0.31	0.001*
LH(mIU/ml)	9.10	1.43	9.08	1.22	0.92 NS
FSH(mIU/ml)	6.60	0.82	6.50	0.51	0.33 NS
TSH(µg/dl)	3.55	0.61	3.20	0.31	0.001*
Prolactin(ng/dl)	8.15	2.87	7.37	1.87	0.01*
FBS(mg/dl)	115.89	10.02	95.22	7.36	0.001*
Fasting insulin (µU/ml)	16.30	3.16	12.70	2.01	0.001*
Homa-IR	4.43	0.52	3.67	0.32	0.001*
QUICKI	0.31	0.01	0.32	0.01	0.001*
Ovarian volume (cm3)	11.70	1.83	8.37	2.96	0.001*

*Statistically significant difference
NS: Not statistically significant difference

Table (5): Comparison of chromium picolinate before and after treatment regarding hormonal profile, infertility, hirsutism, FBS, fasting insulinand ovarian volume.

Title	Before treatment (n=27)		After treatment (n=27)		P value#
	Mean	SD	Mean	SD	
BMI (kg/m2)	30.73	2.30	28.60	2.35	0.001*
Hirsutism Score (0-36)	19.63	8.91	5.52	3.26	0.001*
F. Testosterone (ng/dl)	2.57	0.41	2.50	0.38	0.01*
LH(mIU/ml)	8.70	1.73	8.60	1.53	0.60 NS
FSH(mIU/ml)	7.07	1.43	4.64	1.17	0.001*
TSH(µg/dl)	3.24	0.71	3.05	0.41	0.10 NS
Prolactin(ng/dl)	8.81	2.74	8.63	1.61	0.61NS
FBS(mg/dl)	114.00	9.77	79.81	5.98	0.001*
Fasting insulin (µU/ml)	15.80	3.57	9.10	2.24	0.001*
Homa-IR	4.37	0.44	3.14	0.28	0.001*
QUICKI	0.31	0.01	0.35	0.01	0.001*
Ovarian volume (cm3)	12.10	0.92	8.09	2.28	0.001*

*Statistically significant difference
NS: Not statistically significant difference

Table (6): Comparison between metformin and chromium picolinate groups regarding ovulation and pregnancy rates.

Title		Group I	Group II	p value
Menstrual pattern	Regular Irregular	22(81.49%) 5 (18.5%)	24 (88.89%) 3 (11.1%)	0.35NS
Ovulation Rate	Yes	11 (40.7%)	12 (44.4%)	0.78NS
	No	16 (59.3%)	15 (55.6%)	
Pregnancy Rate	Yes	6 (22.2%)	5 (18.5%)	0.74NS
	No	21 (77.8%)	22 (81.5%)	

NS: Not statistically significant difference

DISCUSSION

Polycystic ovary syndrome (PCOS) is the usual cause of anovulatory infertility. Induction of ovulation with clomiphene citrate (CC) was reported in the 1960s with ovulation rate of 75-80% and a cumulative pregnancy rate of 70-75% after 6-9 cycles of treatment (12).

The use of metformin in the treatment of PCOS was started in 1998 by Nestler and colleagues with some skepticism (13) but it is now accepted to be a valuable and inexpensive therapeutic modality (14). Lord and colleagues have indicated that metformin is highly effective in inducing ovulation and increasing pregnancy rates (15).

As data regarding the use of chromium picolinate are not well established so this current research was done in order to compare the effect of metformin and chromium picolinate on ovulation induction and pregnancy rate in patients with PCOS.

In the study, age ranged from 22 to 35 years (27.1 ± 3.3 years in Group I and 27.9 ± 3 years in Group II). There was no considerable variation ($p > 0.05$).

These results are comparable to those reported by Roy et al. (2009) who reported that, the mean age in first group was 28.2±0.7 compared to 28.8±0.9 years in second group with statistically non-significant difference between both groups (16). Amooee et al., (2013) found that the mean age of the patients was 26.9±5.1 (range 18-38) years (8).

Due to the random distribution of the patients into the 2 groups in this work, there were no significant differences between both groups regarding baseline characteristics, menstrual pattern, hirsutism score and laboratory investigations

In the current study, 22 (81.48%) patients had normal menstruation in Group I and 24 (88.89%) patients in Group II after 3 months of treatment, with no significant difference. Haas et al., (2003) concluded that some (but not all) women with PCOS had improvements in their menstrual cycles while on metformin (17). The effect of metformin on regulation of the menstrual period has also been reported by Kazerooni and Dehghan-Kooshkghazi, (2003) (18). Lucidi et al., (2005) revealed regulation in menstruation after administration of 200 microgram chromium (7).

In our study, ovulation occurred in 11(40.7%) patients in Group I and 12(44.4%) patients in Group II. Metformin has nearly the same effect as chromium on the pregnancy rate. In the study done by Amooee et al., (2013), among those who received chromium picolinate, 22(47.8%) patients ovulated during the study period and 9(19.6%) patients conceived. Also, 20 (43.5%) patients of the metformin group ovulated and 10 (21.7%) conceived during the study period. They concluded that metformin had the same effect as chromium on the pregnancy rate (8). Aruna et al., (2004) stated that using metformin improved ovulation and the pregnancy rate (19) while Lucidi et al., (2005) suggested that regulation in menstruation could be a sign of ovulation (7).

In our study, a significant decrease was observed in BMI in Group I ($p < 0.001$). In the study by Kazerooni and Dehghan-Kooshkghazi, (2003), 500 mg metformin was used three times a day. The decrease in BMI was completely overt (18). Also, Aruna et al., (2004) also conducted a study using 500 mg metformin two times a day in 50 patients and reported a decrease in BMI (19) In contrast, no decrease in BMI was observed in PCOS patients in the study by Genazzani et al., (2004) which used 500 mg metformin two times a day for 6 months (20). In Group II, the difference was also significant between pre and post treatment with chromium for 3 months ($p < 0.001$). In contrast to the present study,Lydic et al., (2006), 1000 µg chromium was used daily in PCOS patients for 2 months, but no significant change was found in BMI (5). In Anderson (1998), no changes in body composition after receiving chromium were encountered (21).

In the current study, there were significant differences in the values of free testosterone, TSH, prolactin, fasting blood sugar and fasting insulin in Group I before and after treatment with metformin. In Group II, the difference was significant in free testosterone, LH, FSH, TSH, fasting blood sugar and fasting insulin.

In Amooee et al., (2013) study, the serum levels of free testosterone decreased by 0.2 and 1.1 in chromium and metformin groups, respectively and the difference was statistically significant (8). The decreasing effect of metformin on testosterone was reported by Kolodziejczyk et al., (2000) (22). No changes were observed in testosterone level in the study by Aruna et al., (2004) (19). Similar results were also obtained by Genazzani et al., (2004) who had conducted their study on non-obese patients (20). In the chromium group, no change was found in free levels of testosterone by Lucidi et al., (2005) and Lydic et al., (2006) (5,7).

Metformin is known to decrease FBS, fasting insulin and QUICKI index and similar results were also achieved in our study. On the contrary, these findings were not reported by Genazzani et al., (2004) which could be due to the low body weight of their study patients (20). Also,Aruna et al., (2004), found no decrease in FBS, fasting insulin and QUICKI index (19), while Kazerooni and Dehghan-Kooshkghazi, (2003) showed using metformin to be effective in decreasing FBS (18). Cabezas et al., (2012) observed an improvement in HOMA-IP in metformin group as in our study (23).

Hummel et al., (2007) have confirmed the effectiveness of chromium in decreasing FBS, HbA1C, fasting and 2 hours insulin in the patients and insulin sensitivity. They stated that Chromium picolinate improved insulin sensitivity as measured by

QUICKI index at the insulin receptor level and, at the elevated level of intake, was devoid of adverse effects in human studies (24). In our study, insulin resistance as measured by HOMA-IR index was improved after chromium administration. Also, Lydic et al. (2006) have shown some improvement in insulin resistance following the use of chromium (5) while Ali et al., (2011) did not observe any change in HOMA-IR after 6 months of chromium when they compared it with placebo (25).

Badawy and Elnashar, (2011) observed that chromium picolinate effectively reduced insulin resistance and HOMA-IR index and treated hyperinsulinemia as well as hyperandrogenemia. They concluded that chromium picolinate (200 μg daily) improved the glucose tolerance in women with PCOS but did not improve ovulation or hormonal profiles (6).

There were significant reductions in ovarian volumes in both groups. In Group I, it was 11.7 ± 1.8 ml before treatment and 8.37 ± 2.91 ml after treatment ($p < 0.001$). In Group II, the ovarian volume was 10.9±1.5 ml before treatment and 9.7 ± 1.24 ml after treatment ($p < 0.001$). Sanoee et al., (2011) investigated the possible effects of metformin administration in women with PCOS on the ovarian volume. The mean ovarian volume was 11.2 ± 4.31 ml before treatment. After three months of treatment the mean ovarian volume declined to 8.17 ± 3.71 ml (p 0.001). They concluded that metformin therapy, even in a relatively short time such as three months, in patients with PCOS may cause a decrease in the ovarian volume by decreasing intraovarian stromal androgens (26). In Amr and Abdel-Rahim, (2015) study, the mean ovarian volume was >10 cm3 in 19 (54%) patients before treatment and in 12 (34%) patients after treatment with chromium. Ovarian volume decreased to 10 cm3 or less in (10/19, 53%) patients who originally had ovaries >10 cm3. The change in mean ovarian volume with treatment was highly significant ($p < 0.001$). They concluded that the observed effects of chromium on ovarian morphology might be due to the effect of chromium on insulin sensitivity, or due to an otherwise unknown mechanism, or it may also be due to rectified pre-existing chromium deficiency (27).

There were significant reductions in hirsutism score in both groups. In Group I, it was 22.7 ± 7.5 before treatment and 7.07 ± 4.9 after treatment ($p < 0.001$). In Group II, it was 19.61 ± 8.9 before treatment and 5.52 ± 3.26 after treatment ($p < 0.001$). The aim of Harborne et al., (2003) study was to elucidate whether metformin does have an effect on hirsutism in women with PCOS. The beneficial effects do not appear to be mediated by suppression of circulating androgens, which makes it possible that hyperinsulinemia or related metabolic pathways may be important determinants of end-organ responses at the level of the hair follicle. They found that the FG score was significantly reduced after treatment with metformin and the results of this prospective, randomized, controlled study showed that metformin is an effective treatment for moderate to severe hirsutism in women with PCOS (28). In Amr and Abdel-Rahim, (2015) study, no significant improvement in hirsutim was noted with chromium. This was most likely explained by the fact that the life cycle of the hair follicles is relatively long so it could improve with longer period of follow-up. The reduction of free testosterone levels is expected to slow the growth of terminal hair and reduce new hair growth (27).

In our study, there were no significant differences between chromium picolinate group and metformin group regarding side effects, except for abdominal discomfort which exhibited statistically a significant difference between the two groups. In Albarracin et al., (2008), chromium had very low side effects (29). In the study done by Amooee et al., (2013), the patients who received metformin experienced more side effects compared to those receiving chromium picolinate. Moreover, metformin administration was accompanied by higher incidence of abdominal discomfort, nausea, vomiting, diarrhea, and indigestion, while chromium picolinate was accompanied by loss of appetite and headache (8).

CONCLUSION

Chromium picolinate was better tolerated than metformin due to lower side effects; nevertheless, no significant differences were observed between the two groups regarding ovulation and pregnancy rates. Therefore, metformin could be replaced by chromium in some PCOS patients.

Conflict of interest

The authors declare that they have no conflict of interest.

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Serum Concentration of Cancer Antigen 125 in Normal and Preeclamptic Pregnancies

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Abstract

Aim: To measure the serum concentration of CA125 in pregnancies complicated by preeclampsia and low risk pregnancies with the possibility it being considered to be used in early diagnosis of severe preeclampsia so, it may be a marker of the severity of the disease.

Methods: One hundred and eighty-nine pregnant women were participated in the study, it was carried out at department of obstetrics and gynecology, Suez Canal University Hospitals. They were divided into 3 groups mild, severe preeclampsia and normal study group. Urine analysis, complete blood count (CBC), serum AST and ALT, uric acid, urea, creatinine and serum CA125 were assayed for all participants.

Results: The mean serum concentration of CA-125 was (32.59 ± 1.63), (39.70± 1.19) and (52.92 ± 2.88) in control, mild and severe preeclampsia respectively, the difference was statistically significant (p=0.001).It wasfoundthatCA125positivelycorrelatedwithsystolic blood pressure, diastolic bloodpressure, proteinuria, hemoglobin level,ALT, AST, serum uric acid,serum creatinineand urea,meanwhile, the plateletcountshowed negative correlation withCA125($p<0.05$).Receiver operating characteristics (ROC) curve was used to find out the best cut off value of CA 125 in mild and sever preeclampsia, it was 39 in mild preeclampsiawith sensitivity of 80.9%, specificity 99 %, PPV 99.4% and NPV 84%.The best cut off value was 49.5 in severe preeclampsia, the sensitivity, specificity, PPV and NPV were 82.5 %, 99 %, 99 % and 87.2 respectively.

Conclusion: SerumCA-125leveliselevatedsignificantly inmild and severepreeclampsia,itcorrelated with the severity of preeclampsia,soserum concentration ofCA125 maybeusedas a marker ofsevarityof preeclampsia.

Key words: Cancer Antigen 125 Concentration, Preeclampsia
Maternal serum ferritin may be a useful test in the prediction of asymmetric IUGR.

Introduction

Preeclampsiaisasyndromespecificto pregnancythataffectsalmostevery organ[1]. Hypertensive disorders of pregnancy affect8–10% of primigravidafemalesand aretheleading causeofmaternalandfetal morbidity andmortality worldwide. Preeclampsia isa hypertensive syndrome occurring during pregnancy thatis clinically diagnosed by hypertension (bloodpressure >140/90mmHg), proteinuria(>300 mg/24 h)and varying degreesof ischemic endorgandamage, whichist-

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houghtto betheresultof abnormal trophoblastic invasion and diffuseendothelialdysfunction-likeinthebrain, eye, kidneyand also in placenta[2].

Theexactpathophysiology of preeclampsiaisunknown;however, the mechanismmay beduetoabnormal trophoblastic invasionof uterinevessels, abnormalnitric oxide and lipid metabolism, immunologicintolerancebetweenfeto-placental and maternal tissue, geneticabnormalities,inadaptability to inflammation and cardiovascularchanges, andmetabolicandnutritionalfactors[3].

Cancerantigen125(CA-125) isa highmolecular weight heterogeneously structuredglycoprotein.It hasbeenused indiagnosis;followup treatmentandrecurrence ofepithelial ovariancancer. CA-125isa valuable marker of other gynecologicalconditions including pleuralandperitoneal involvement,the sourceof CA-125during pregnancy isthefetalchorionamnioticfluid, and maternaldecidua[4].**Schrocksnadel et al (2000)** who was the first to compare the plasma CA-125 level in healthy non-pregnant women, pregnant patients with hypertensive disorders and healthy women with singleton pregnancies at term and found no significant difference(5).On the other hand,Cebesoyand Dikensoy (2009)confirmed the relationship between the cancer antigen-125 (CA-125) with preeclampsia. They stated that CA-125 level was higher significantly in severe preeclampsia(6), soon the merits of thisdebate,weaimed to measuretheserumconcentrationofCA125inpregnancies complicatedbypreeclampsiaandlowriskpregnancieswiththepossibilityitbeing consideredtobeusedinthe earlydiagnosisof severepreeclampsiaso it may be amarkeroftheseverityof the disease.

Patients and methods

A prospectivecase-controlstudywas carried out at emergency ward, obstetrics and gynecology department at Suez Canal University Hospitals, from April 2014 to April 2016. The study was approved by the ethical committee of faculty of medicine, Suez Canal University and an informed written consent was obtained from all participants. One hundred and eighty-nine pregnant women were recruited for this study, they were divided into 3 groups 63 each;mild preeclampsia group(blood pressure ≥ 140/90 mmHg and<160/110 with proteinuria, 300mg/24hr urine collection, or +1 dipstick in urine sample).Severe preeclampsia group;pregnant women had one or more of the following criteria systolic B P ≥ 160mmHg, diastolic BP ≥ 110mmHg, proteinuria≥5gm in 24hrs urine collection or +3 dipstick in urine sample, oliguria (<500ml/d), cerebral or visual disturbance, pulmonary edema, epigastric or right upper quadrant pain, impaired liver function, thrombocytopenia (<100000) or fetal growth destruction(1) and healthy pregnant women control group. Inclusion criteria were age between 18 and 35 years, primigravida, gestational age from 28 to 40 weeks and Singleton pregnancy.Exclusion criteria were history of chronic hypertension, diabetes mellitus, renal disease, cardiovascular disease or autoimmune disease.

Complete urine analysis, complete blood count, serum AST and ALT, kidney function tests (urea and creatinine) and serum CA125 were done for all participants.Venous blood samples were withdrawn and the serum was separatedwith exclusion of grossly hemolytic, lipemic and turbid samples. Specimens were stored -20oCtill measurement of serum CA-125concentration.CA125 (Human) ELISA Kits were purched for Abnova Company Taiwan (BIOTEC Alex). CA125 was measured using the ELISA method at wave length 450nm.

Statistical analysis

The data collected were tabulated & analyzed by SPSS (statistical package for the social science software) statistical package version 11 on IBM compatible computer. Quantitative data were expressed as mean& standard deviation (SD) and analyzed by ANOVA-test for comparison of three groups followed by post hoc test. Qualitative data were expressed as number and percentage (N & %) and analyzed by applying Chi-square test. **Spearman's correlation** test was done to study correlation between one qualitative variable and one quantitative variable or two quantitative variables of not normally distributed data.**Roc curve (Receiver operating characteristic curve)**: was done to detect cut level of any tested variable where at this level there is the bestsensitivity and specificity.

Result

Onehundredandeighty-nine pregnant women were enrolled in the study, 63 patients with mild preeclampsia, 63 patients with severe preeclampsia and 63 low risk controls. There was no statistically significant difference between the three studied groups as regards the mean of maternal age and parity (P>0.05), meanwhile, the mean gestational age was statisticallysignificant, it was (38.9±1.2), (38.1±1.5) and (36±2.5) in the control, mild and severe preeclampsia groups respectively with p-value < 0.001(table 1).

In the current study the mean serum concentration of CA125 was (32.59 ±1.63), (39.70± 1.19), (52.92 ± 2.88) in control mild and severe preeclampsia respectively, the difference was statistically significant (p=0.001), which indicates that serum CA125 level increase with the severity of preeclampsia. There was statistically difference between the studied groups regarding BP, HB, platelet count, uric acid, creatinine, urea, ALT, AST (p<0.05) (table 2).

Post-hoc analysis was performed for each variable the difference between control and severe preeclampsia groups was statistically significant for all variables and also, the difference between mild and severe preeclampsia groups was significant, meanwhile, the difference between control and mild preeclampsia groups was significant only for systolic and diastolic blood pressure, serum uric acid, creatinine and CA125 (P<0.05).

The relations between cancer antigen 125 (CA125) and each of: systolic blood pressure, diastolic blood pressure, proteinuria, hemoglobin level, platelet count urea and AST and ALT were assessed by spearman's correlation coefficient, all correlations were positive except for platelet count which shows negative correlation with CA 125. All correlations were statistically significant (p<0.05) (table3).

Receiver operating characteristics (ROC) curve was used to find out the best cut off value of CA 125 in mild preeclampsia, 39 was chosen as the best cutoff value (figure I) with sensitivity of 80.9%, specificity 99 %, PPV 99.4% and NPV 84%(table 4). In severe preeclampsia, the best cutoff value was 49.5(figure II), the sensitivity, specificity, PPV and NPV were 82.5 %, 99 %, 99 % and 87.2% respectively (table 5).

Table (1): Comparison of socio-demographic characteristics of the studied population

Title		Control (n=63)		Mild (n=63)		Sever (n=63)		P-Value#
		No.	%	No.	%	No.	%	
Age	17-	25	39.68	19	31.16	22	34.92	0.485
	23-	25	39.68	23	36.51	26	41.27	
	29-34	13	20.63	21	33.33	15	23.81	
		Mean	**SD**	**Mean**	**SD**	**Mean**	**SD**	
		24.52	4.53	25.37	4.55	24.54	4.36	
Parity	PG	63	100.0	63	100.0	63	100.0	
Gestational Age	28 to <33	0	0.00	0	0.00	9	14.29	0.000*
	33 to <38	9	14.29	15	14.29	37	58.73	
	38-41	54	85.71	48	85.71	17	26.98	
		Mean	**SD**	**Mean**	**SD**	**Mean**	**SD**	
		38.98	1.19	38.1	1.51	36.00	2.51	

ANOVA Test* statistically significant difference

Table (2): Comparison of clinical and laboratory measures between the studied groups

Title	Control (n=63)		Mild (n=63)		Severe (n=63)		P-Value#	LSD p-value
	Mean	**SD**	**Mean**	**SD**	**Mean**	**SD**		
CA125	32.59	1.63	39.70	1.19	52.92	2.88	0.001*	A:0.01* B:0.00* C:0.01*
SBP	107.06	11.35	144.29	3.09	171.27	9.16	0.001*	A:0.01* B:0.00* C:0.01*
DBP	63.73	6.09	95.79	3.83	112.78	4.38	0.001*	A:0.00* B:0.00* C:0.00*
HB	10.20	1.01	10.21	1.01	10.87	0.69	0.002*	A:0.915 B:0.00* C:0.01*
PLT ($x10^3$)	268.86	59.78	278.48	65.35	212.44	51.51	0.001*	A:0.363 B:0.00* C:0.01*
Serum uric acid	2.93	0.30	4.53	0.28	5.67	0.67	0.000*	A:0.02* B:0.00* C:0.01*
Serum Creatinine	0.58	0.10	0.77	0.15	0.93	0.15	0.001*	A:0.01* B:0.00* C:0.01*
Serum Urea	33.27	2.74	33.27	2.74	35.13	4.15	0.014*	A:1.00 B:0.00* C:0.00*
AST	26.32	4.49	26.32	4.49	39.38	5.30	0.001*	A:1.00 B:0.00* C:0.00*
ALT	27.16	4.21	27.16	4.21	38.78	5.34	0.002*	A:1.00 B:0.00* C:0.00*

SBP: systolic blood pressureDBP: diastolic blood pressure.
Kruskal Wallis Test * statistically significant at 95% level of confidence.
(LSD) least significant difference was determined:A: between control and mild.
B: between control and severeC: between mild and severe.

Table (3): Spearman's correlation coefficient (r) between CA 125, blood pressure and laboratory investigations.

	CA 125	
	R	**p-value**
Systolic blood pressure	0.903	0.000*
Diastolic blood pressure	0.908	0.000*
Proteinuria	0.936	0.001*
Hemoglobin level	0.235	0.001*
Platelet count	-0.364	0.002*
Uric acid	0.888	0.000*
AST	0.620	0.001*
ALT	0.614	0.001*
Creatinine	0.675	0.002*
Urea	0.174	0.017*

*Statistically significant at 95% level of confidence.

Table (4): Best cut-off value of CA 125 in mild preeclampsia from controls

	Mild preeclampsia No (%)	**Control No (%)**
≥ 39	51	0
< 39	12	63
Sensitivity	80.9%	
Specificity	99 %	
PPV	99.4%	
NPV	84%	
Area under the curve	0.905(0.845-0.964)	
p-value	0.001*	

PPV: positive predictive valueNPV: negative predictive value
*Statistically significant at 95% level of confidence.

Table (5): CA 125 best cutoff value in severe preeclampsia.

	Severe Preeclampsia No (%)	**Control and mild preeclampsia No (%)**
≥ 49.5	52	0
< 49.5	11	75
Sensitivity	82.5 %	
Specificity	99 %	
PPV	99 %	
NPV	87.2 %	
Area under the curve	0.965(0.94-0.989)	
p-value	0.000*	

PPV: positive predictive value.NPV: negative predictive value.
*Statistically significant at 95% level of confidence.

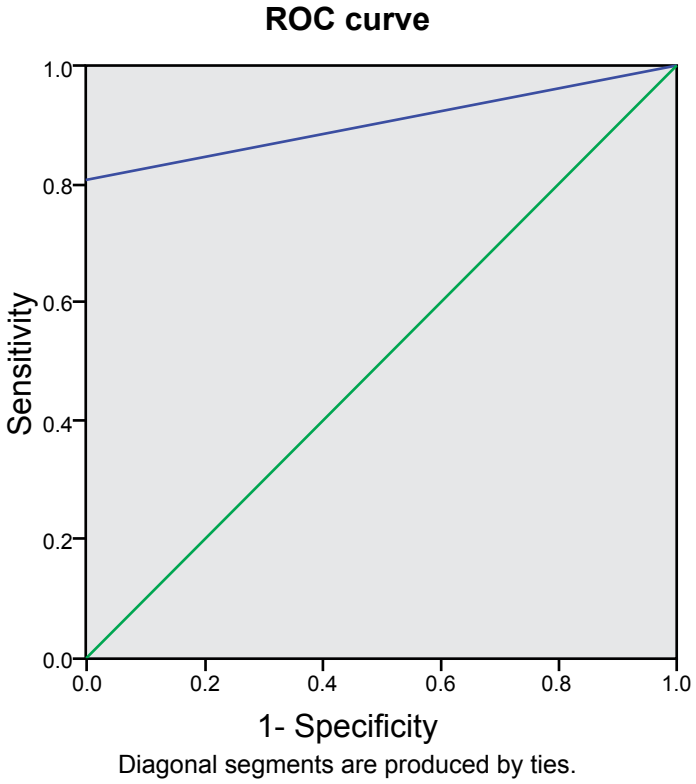

Fig (I): ROC curve for CA 125 values in mild preeclampsia.

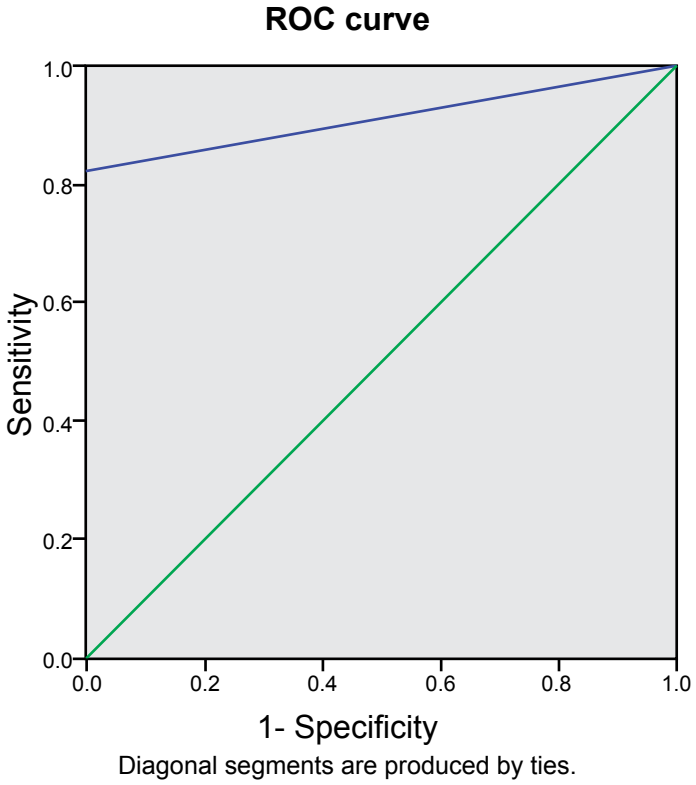

Fig (II): ROC curve for CA 125 cut off values in severe preeclampsia.

Dissussion

Preeclampsia is a hypertensive disorder of pregnancy which may cause morbidity and even mortality for both the mother and the fetus. Blood pressure elevation is the most visible sign of preeclampsia, but this disease may cause generalized damage to the maternal endothelium,kidneys and liver through the release of vasoconstrictive substances[4].CA-125 is a glycoprotein antigen which is located on cell surface; it is a molecule of 200-kDa glycoprotein initially identified on the surface of the OVCA433 ovarian carcinoma cell line[7]. CA-125 is widely distributed on the surface of both healthy and malignant cells of mesothelial origin, including pleural, pericardial, peritoneal, and endometrial cells, as well as in the normal genital tract and amniotic membrane. Fetal chorion and maternal decidua have been indicated as the potential sources of high serum CA-125 levels which are detected during the first trimester of pregnancy and postpartum period [8].

In the present study, the demographic data of the patients enrolled in the study, there were no statistically significant difference between the three studied groups control, mild and severe preeclamptic groups regarding the maternal age but there were statistically significant difference between them regarding the mean of gestational age, these findings were confirmed by **Danisman and Rousso(2011)**[9] who studied CA125 concentration in 242 patients with singleton pregnancies and primigravida divided into three groups control, mild and severe preeclampsia and found no statistically significant differences between the mean maternal age groups, while statistically significant differences between them regarding gestational age due to preterm delivery of the fetuses with preeclampsia with increasing severity of the disease.

In the current study the **mean serum** concentration of CA125 was (32.59 ± 1.63), (39.70± 1.19), (52.92 ± 2.88) in control mild and severe preeclampsia respectively which indicates that serum CA125 level increase with the severity of preeclampsia. This may be explained as serum CA125 level related to impaired placentation which causes intermittent disruption of placental perfusion, ischemia-reperfusion type injury, oxidative stress and systemic inflammatory response.This was in agreement with **Karamanand Ark (2013)** [10],

who studied 93 patients who were primigravida, they found that the mean serum concentration of CA125 in control group was (34.25 ± 3.34), it was (39.70±8.72) in mild preeclampsia and (56.11± 4.28) in severe group, an underlying inflammatory process may play a role which worsens with the severity of preeclampsia in patients. Maternal serum CA-125 level can be high during early pregnancy and the postpartum period. The potential source for this elevation is the fetal chorion, amniotic fluid and maternal decidua, these increased serum levels of CA-125 during early pregnancy and immediately afterbirth indicate disintegration of maternal decidua as a potential source, that is, extension of decidual destruction and separation of trophoblasts from the decidua are proposed as the mechanism underlying the increased CA-125 levels[10].

It is assumed that preeclampsia is related to reduce trophoblastic migration into the maternal decidua, which leads to chronic inflammation within the placenta. This process may lead to increased expression of CA-125. Thus, it can be hypothesized that maternal serum CA-125 levels will be higher in females with severe preeclampsia than in other patients due to the increase in inflammatory process. It may be assumed that the extension of decidual destruction and failure of trophoblastic invasion in preeclampsia may induce the secretion of CA-125 within placenta [11].

In the current study maternal serum concentration ofCA125 were positively correlated with proteinuria (r =0.936 p <0.001), this finding was in agreement with **Han and Karaman (2013)** who found the same correlation (r =0.789 p<0.001).Elevation of the maternal serum concentration of CA-125 level in severe preeclampsia more than mild preeclampsia and control group[12], also, it was in accordance with the findingofCebesoy and **Dikensoy (2009)**[6] who reported that the serum concentration of CA-125 was significantly higher in women with preeclampsia in comparison to healthy pregnant women and added that serum CA-125 in severe preeclampsia was significantly higher than mild preeclampsia. There was a positive correlation between CA-125, albumin level and mean arterial pressure (MAP).Gungor andYenicesu (2011) [13] compared CA-125 values of healthy and preeclampticwomen throughout a given time interval (from the middle of the second trimester to term) and documented that serum concentration of CA-125 did not differ with respect to either pregnancy outcome or gestational age. However, there was a trend toward an elevation in CA-125 concentration for pregnancies that are destined to develop preeclampsia.

The present study disagreed withSchrocksnadel et al (2000)who was the first to compare the plasma CA-125 levels of 50 healthy non-pregnant women, 50 pregnant patients with hypertensive disorders and 50 healthy women with singleton pregnancies at term, they reported that there were no statistically significant differences or an increasing trend could be noted for CA-125[5].Previous study compared serum CA-125 concentration of 120 women with pathological outcome of pregnancy (spontaneous abortion, fetal death, intrauterine growth retardation, chromosomal and structural abnormalities, and preeclampsia/eclampsia) to those of 350 women with normal outcome of pregnancy. They confirmed that maternal CA-125 serum concentrations were significantly higher in the first and the third trimesters of pregnancy when compared to those in the second trimester, but not significantly different from those obtained in pathological pregnancies [14].

Danismanet al (2011) reported in their study that CA-125 is a biochemical marker which reflects the severity of the underlying inflammatory process in preeclampsia; it may be assumed that the extension of decidual destruction and failure of trophoblastic invasionin preeclampsia may induce the secretion of CA125 within placenta(9).Another explanation for elevation of maternal serum CA125 in females with severe preeclampsia may be due to the formation of ascites resulting from the decreased albumin level. The albumin level in females with severe preeclampsia was significantly lower than that in females with mild preeclampsia and normal pregnancies. The presence of ascites may lead to peritoneal irritation and increased CA-125 levels [10].

The current study revealed the blood pressure and all laboratory results were statistically significantbetween the control and studied groups, meanwhile, the difference between control and mild preeclampsia groups was significant only for systolic and diastolic blood pressure, serum uric acid,

creatinine and CA125, this finding agreed with that of (Danisman et al. 2011)[9].

Serum uric acid levels showed statistically significant differences between control, mild preeclampsia and severe preeclampsia groups in the present study. Karumanchi and Naljayan (2013) documented a correlation betweenhyperuricemia and the severity of the disease[15]. Many et al (1996)stated that decreased renal tubular excretion may be responsible for the rise in serum uric acid levels in preeclampsia[16], More recently, increased oxidative stress and formation of reactive oxygen species have been proposed as another contributing source of the hyperuricemia noted in preeclampsia.

In current study there was a strong correlation between CA125 with systolic blood pressure, diastolic blood pressure, proteinuria and serum uric acid, and it was moderately correlated with AST and serum creatinine and also shows a weak correlation with hemoglobin level, platelet count, ALT and urea. Ozat et al (2011) [17]found that serum CA-125 concentrations were correlated positively with systolic blood pressure, diastolic blood pressure, serum uric acid level and proteinuria. Moreover, they concluded that CA-125 is a biochemical marker of the severity of the underlying inflammatory process during preeclampsia. Thus, CA-125 seems to be a promising marker of preeclampsia.In the current study, the best cutoff value of CA-125 in severe preeclampsia was > 49.5, with a sensitivity of 82.5%, a specificity of 99%, a positive predictive value of 99%, a negative predictive value of 87.2% and area under the curve of 0.965% (0.94 – 0.989) as a marker of severe preeclampsia, this agreed with Ozat et al (2011)who found that the CA125 cutoff value of>50, serum CA-125 had a sensitivity of 97.2%, a specificity of 96% [17].

Conclusion:

SerumCA-125leveliselevatedsignificantly inmild and severepreeclampsia,itcorrelated with the severity of preeclampsia,soserum concentration ofCA125 maybeusedas a marker ofseverityof preeclampsia.

Conflict of interest

The authors declare that they have no conflict of interest.

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Ovarian doppler changes following tubal salpingectomy for hydrosalpinx

Waleed F. Gharib MD

Abstract

Background: Hydrosalpinx is a frequent tubal pathology encountered among infertile female. Hydrosalpinx impairs the expression of factors essential for differentiation of the endometrium impairing endometrial receptivity; reducing the in vitro fertilization success rate. Removal of a hydrosalpinx can increase the implantation rate of in vitro fertilization. However, salpingectomy could potentially affect the ovarian vascularity and may impose a potential change in ovarian Doppler indices. .

Objective: to assess the changes in ovarian vascularity following laparoscopic tubal salpingectomy in terms of change in ovarian Doppler indices.

Setting: Department of Obstetrics and Gynecology, Suez Canal University Hospitals, Ismailia, Egypt.

Patients and Methods: This prospective cohort study included 25 patients with unilateral communicating hydrosalpinx treated with laparoscopic tubal salpingectomy and ultrasound ovarian Doppler indices were assessed before and after surgery.

Main outcome measures: Tubal salpingectomy showed increased local ovarian vascular resistance with significant ovarian Doppler indices changes.

Results: Both pulsatility index and resistive index increased significantly 3 months after surgery denoting increased vascular resistance (p value 0.03 and 0.01 respectively).

Conclusion: laparoscopic tubal salpingectomy for treatment of hydrosalpinx can potentially affect ovarian Doppler indices with significant increase in ovarian local vascular resistance.

Keywords: Ovarian Doppler, Laparoscopic salpingectomy.

Introduction

Tubal disease is responsible for 25% to 35% of cases of female infertility (1).Hydrosalpinx is known to affect one third of women with tubal pathology. It is a pathologic tubal disorder in which distally obstructed fallopian tubes of various pathologies become filled with fluid, forming a saccular structure (2).

The expression of factors essential for differentiation of the endometrium is impaired by hydrosalpinx consequently deteriorating endometrial receptivity; reducing the IVF success rate, implantation rate,

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and pregnancy rate by 50% and doubles the rate of spontaneous abortion(3).

It is generally recognized that removal of a hydrosalpinx can increase the implantation rate of in vitro fertilization. However, whether salpingectomy affects ovarian reserve is uncertain, with some studies suggesting that salpingectomy deceases ovarian reserve (4), and other studies indicating that it has no effect on ovarian reserve (5).

The close anatomical association of the vascular and nervous supply to the fallopian tubes and ovaries constitutes the rationale for the risk of impaired ovarian function after surgery (6,7).

The purpose of this work was to document the changes in ovarian Doppler indices following laparoscopic tubal salpingectomy for hydrosalpinx.

Patients and methods

Patients

This is an observational prospective cohort study which was performed at the department of Obstetrics and Gynecology, Suez Canal University hospital. This study was approved by the faculty ethical committee; and all patients gave an informed consent before inclusion in the study. This prospective cohort study included 25 patients with unilateral communicating hydrosalpinx more than 3 cm long by ultrasound with the age between 20 and 40 years.

All patients had an initial assessment to diagnose hydrosalpinx. Hydrosalpinxwas diagnosed when an elongated tubular mass with echogenic wall and linear echoes in the lumen was observed by ultrasound and confirmed by Hysterosalpingography; Hydrosalpinx appear enlarged and irregular, with absent rugae, and most often with failure of contrast medium spilling from the tubes into the pelvis.

Methods

After obtaining informed consent all the patients in the study were subjected to detailed history taking, General , and local examinations.

Ultrasound indices of ovarian function:

Assessment of ultrasound parameters of ovarian function were done by measurement of antral follicular count (AFC), ovarian volume and ovarian stromal blood flow.

AFC is defined as the number of all small follicles (between 2and 9 mm) counted in the ovary. Ovarian volume calculated by multiplying the 3 dimensions of the ovary, then by 0.5. i.e. 0.5233 x Dl (length) x D2 (width) x D3 (breadth)(Cm3).

Ovarian stromal blood flow indices included pulsatility index (PI) resistivity index (RI) and systolic diastolic ratio S/D ratio

All ultrasonographic examinations were done at the ipsilateral ovaries before and 3 months after laparoscopic surgery between day one and day 4 of their cycles using a Philips HD11 XE Transvaginal ultrasonography with a 7.5-mHz probe. All examinations were conducted by the same investigator to remove interobserver bias, and the parameters were measured at least three times and the mean value was recorded.

Surgical intervention:

Laparoscopic salpingectomy was performed using bipolar cautery and scissor. Adhesiolysis was performed if necessary. The mesosalpinx was transected just below the fallopian tube to minimize any compromise to the Collateral blood supply of the ipsilateral ovary. The fallopian tube was transected 1–1.5 cm from the cornual region.

Results

The demographic and clinical characteristics of the study population are presented in table (1).

There was statistically significant difference between the ovarian doppler indices before and after surgery ,with a statistically significant increase in resistive index (from 0.65±0.19 to 0.76±0.16 and p value =0.01) and a statistically significant increase in pulsatility index (from 1.2±0.95 to 1.78±0.9 and p value =0.03) table (2).

Discussion

This prospective cohort study was performed on infertile women below 40 years of age with unilateral communicating hydrosalpinx to detect the effect of laparoscopic tubal salpingectomy on ovarian Dopplerindices. 25 females with unilateral communicating hydrosalpinx were recruited in the study.They were all subjected to treatment

with laparoscopic salpingectomy, and the ovarian Doppler indices were recorded before and three months after surgery.

In our patients the follow-up evaluation has been done 3 months after surgery. Nevertheless, a lot of studies demonstrate that choosing the third postoperative month to test the effect of surgery on ovarian functions is enough to assess the extent of recovery after acute ovarian damage (14).

Moreover, ovarian transplantation studies have clarified that the formation of small preantral and antral follicles from quiescent primordial follicles requires at least 3 months (19),a specific time point that we choose as follow-up in our study.

The results of the present study showed higher values of pulsatility (PI) index, resistivity index (RI) and S/D ratio in the post-operative follow up compared to the preoperative values. There was a statistically significant increase in resistive index from 0.65±0.19 to 0.76±0.16 and p value =0.01, and statistically significant increase in pulsatility index from 1.2±0.95 to 1.78±0.9 and p value =0.03.

Salpingectomy has been postulated to decrease ovarian reserve through several mechanisms. One common argument is that ovarian blood flow may be compromised during salpingectomy. Because the tubal branch of the uterine artery originates at the same point as the ovarian branch of the uterine artery, damage to ovarian branch as it traverses the mesosalpinx, by surgical disruption or thermal spread may decrease blood flow to the ovary. (15)

Strandell et al in 2001 examined 26 women who underwent salpingectomy because of hydrosalpinx and acted as their own controls before and after surgery. The study did not describe any signs of compromised ovarian function after surgery. Two other similar studies reached the same conclusion (17, 18).Also, hemodynamic studies using Doppler ultrasonography in patients with hydrosalpinx have revealed that the blood flow to the endometrium and ovary may be impaired at baseline. Thus, a decrease in blood flow to ovary may occur after salpingectomy for hydrosalpinx compared to tubal ectopic pregnancies.(8,9)

Some surgeons have also argued that disruption of blood flow may occur only with poor surgical technique .The use of either a 5-mm Harmonic® scalpel or LigaSure device in cauterization of the mesosalpinx as close as possible to the fallopian tube can markedly minimize damage to ovarian blood supply, as these instruments minimize lateral thermal spread compared to monopolar or conventional bipolar electrocautery, thereby reducing unintended tissue damage.

However, whether these postsalpingectmy ovarian vascular changes affect the ovarian reserve or not,it is still in debate . In a systematic review to investigate the impact of salpingectomy on ovarian reserve, the overall analysis as well as subgroup analysis based on laterality, age and AMH(antimullerian hormone) kits revealed no short-term changes in serum AMH concentrations after salpingectomy (10). These results are surprising given the expected post-salpingectomy damage of ovarian reserve as a result of impairment of ovarian blood supply as shown in several previous studies (11,12,13)

Possible explanation for these results is that the postulated post-salpingectomy decline of ovarian reserve may be a chronic process that could take a long time to occur. In other words, the possible post-salpingectomy impairment of blood supply may lead to chronic ovarian ischemia that could take a relatively long time to cause reduction in the number of the small antral follicles with subsequent fall in circulating AMH, which is exclusively secreted from these follicles. This hypothesis, however, requires validation through further long-term follow-up studies on the changes of circulating AMH after salpingectomy. (16)

In contrast to their result, our measures were ovarian stromal blood flow in the ovarian stroma at a maximum distance from the ovarian capsule not the ovarian artery which may explain the difference.

The limitations of this study are its small sample size and lack of long-term follow up. We do not know whether the differences in ovarian volume and AFC would begin to appear as the participants get further out from surgery.

Conclusion

The results of the present study showed higher values of pulsatility (PI) index, resistivity index (RI) and S/D ratio in the post-operative follow up compared to the preoperative values .Which postulates that laparoscopic salpingectomy in-

creases local ovarian vascular resistance which could affect ovarian reserves over longer duration. The revert to other surgical modalities rather than salpingectomy in the management of hydrosalpinx in infertile females with already embarrassed ovarian reserve seems to be a wiser option.

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Appendix

Table (1): Demographic and clinical characteristics of the study group.

Age (years)	**Mean ±SD** **Range**	**26.4±4.5** **20-39**
Smoking	Smoker Non-smoker	0 25(100%)
BMI	Mean ±SD Range	24.6±3.93 20-34
Infertility	Primary Secondary	6(24%) 19(76%)
Years of infertility	3.4±1.2	
Parity	0 1 2	6(24%) 13(52%) 6(24%)
Abortion	0 1 2	6(24%) 16(64%) 3(12%)
Ectopic	0 1 2	16(64%) 8(32%) 1(4%)
Pelvic surgery	Yes No	17(68%) 8(32%)

Table (2) : Ovarian doppler indices before and 3 months following salpingectomy

Parameter	**Pre-salpingectomy**	**Post-salpingectomy**	**t-test**	**p-value**
Ovarian volume(cm3)	5.2 ± 1.55	4.85 ± 2.2	1.4	0.18(NS)
Antral follicular count	8.3±2.4	9.6±1.4	1.39	0.14(NS)
Pulsatility index	1.2±0.95	1.78±0.9	2.08	0.03*
Resistivity index	0.65±0.19	0.76±0.16	2.45	0.01*
S/D ratio	3.6±2.88	4.95±4.3	1.21	0.23(NS)

(NS) Statistically non-significant difference between both groups (P value > 0.05)

* Statistically significant difference between both groups (P value $\leq$ 0.05)