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Revisiting the predictability of follicular fluid leptin and related adiposity measures for live birth in women scheduled for ICSI cycles: a prospective cohort study

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Abstract

Background Our research question is could follicular fluid (FF) leptin solely or contemporaneously with other clinical, biochemical, and sonographic adiposity measures predict the probability of having a live birth during ICSI cycles? This is a prospective cohort study that enrolled infertile women without polycystic ovary syndrome scheduled for ICSI. At baseline, women had an assessment of obesity using different metrics: clinical, serum biochemical, and sonographic. Clinical measures encompassed waist circumference and body mass index. Biochemical evaluation comprised an assessment of the homeostasis model for insulin resistance, visceral adiposity index, and lipid accumulation product. Preperitoneal and subcutaneous abdominal fat were measured using ultrasound and body fat index was calculated. On the day of oocyte retrieval, pooled FF was sampled to assess FF leptin. Our primary outcome was live birth after one fresh embryo transfer cycle.

Results Out of 91 women analyzed in this study, 28 have a live birth (30.8%). No difference in FF leptin concentration was found between women with and without live birth (mean ± SD; 20336 ± 8006 vs 18493 ± 6655 pg/ml; $P = 0.2$). None of the assessed adiposity markers was a predictor for live birth. Substantially, follicular fluid leptin was positively correlated with insulin resistance in women with and without live birth ($r = 0.21$, $P = 0.04$). In logistic regression analysis, the outcome of the prior cycle, the ability to have cryopreserved embryos, and the oocyte maturation index were the predictors for live birth in our study.

Conclusions The present work could not find evidence that follicular fluid leptin, preperitoneal fat, and other evaluated adiposity measures could impact live births after ICSI cycles.

Keywords Body fat index, Central obesity, Follicular fluid leptin, ICSI, Insulin resistance, Preperitoneal fat

Background

In reproductive-aged women, obesity has been linked, in addition to the metabolic health risks, to ovulatory dysfunction, menstrual irregularities, and suboptimal outcomes of variant fertility treatments [19, 40].

Clinical metrics utilized as crucial descriptors for obesity include body mass index (BMI) and waist circumference (WC). However, body mass index considers total body weight and height without referring to body fat or

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central (abdominal) obesity. Also, waist circumference, although could accurately identify women with central obesity, cannot precisely reflect preperitoneal fat [39].

Hence, different imaging tools for measuring preperitoneal fat have been proposed in reproductive-aged women. Ultrasound has been determined as a valid tool for the assessment of intraabdominal and preperitoneal fat compared to computerized tomography scan and magnetic resonance imaging [41, 48, 49]. Moreover, ultrasound is more familiar in the clinicians' hands, inexpensive, and devoid of radiation hazards.

An earlier report demonstrated that preperitoneal fat thickness by ultrasound was superior to subcutaneous fat in predicting insulin resistance and other metabolic syndrome elements in the non-specific population [32].

At the level of the ovary, Ciavattini and colleagues found that increased preperitoneal fat as measured by ultrasound was associated with high reactive oxygen species in the follicular fluid and negatively correlated with oocyte and embryo quality [10]. Moreover, Obese anovulatory women with polycystic ovary syndrome (PCOS) who resume ovulation during a 6-month lifestyle program lost more visceral fat compared to the women who did not resume ovulation,despite similar subcutaneous fat loss in both groups [26].

Leptin is a product of the adipose tissue. It is a hormone that plays a key role in the regulation of the HPO axis to start puberty and maintain ovarian function [45].

In infertile women scheduled for assisted reproductive technology (ART), there are conflicting reports regarding the effect of follicular fluid (FF) leptin levels on IVF outcomes. Some suggested the deleterious effect of high leptin on embryo quality [38]. This controversy is evident in studies recruiting a non-PCOS infertile population. Heterogenous methodology and endpoints stand beyond these incongruent observations. These studies did not analyze or correlate the findings in relation to different adiposity measures such as the preperitoneal fat, body fat index, or visceral adiposity index. And, few of them reported a correlation with insulin resistance [27]. Neither body fat index (BFI) nor visceral adiposity index (VAI) have been assessed before in non-PCOS women for IVF.

The body fat index that compiled preperitoneal and subcutaneous fat in its calculation is a newly studied index in pregnant women as a predictability tool for gestational diabetes [6, 34, 46], while visceral adiposity index has been deemed to predict insulin resistance in PCOS women [37].

In the current study, the researches targeted infertile women without PCOS to revisit the hypothesis of whether FF leptin as an adiposity biomarker could predict intracytoplasmic sperm injection (ICSI) outcomes in these women. Moreover, we integrated other adiposity measures in our evaluation (clinical, biochemical as well as sonographic) that have been proposed to be leptin-related. The reason for excluding PCOS women is to avoid the confounding effect of the pathophysiological mechanisms of PCOS on the study outcomes.

Materials and methods

Study design and setting

Our study is a prospective cohort study, conducted at the Assisted Conception Unit, Department of Obstetrics and Gynecology, Women's Health Hospital, Assiut University, Egypt. The study was registered (NCT03778684, www.clinicaltrials.gov). Recruitment was started in February 2019, and the study was completed on November 2022.

Study participants

Infertile women indicated for intracytoplasmic sperm injection (ICSI) were eligible for enrollment if they were non-PCOS, aged between 18 and 35 years, anticipated normal responders, and had normal uterine cavities by transvaginal ultrasound. Non-PCOS women scheduled for ICSI comprised women with anovulation, unexplained infertility, tubal disease, and male factor. Women with PCOS, those administering metformin, diabetic women, and poor responders based on Bologna criteria [16] were not included in the study. The Rotterdam European Society for Human Reproduction and Embryology (ESHRE)/American Society for Reproductive Medicine (ASRM) criteria were used to define PCOS [43]. Only one fresh ICSI transfer cycle for each participant was analyzed for the study outcomes.

Sample size calculation

Based on a study by Llaneza-Suarez and colleagues [27] who determined a mean FF leptin concentration of 16.8 ng/mL (SD ± 6.0 ng/ml), and 11.5 ng/ml (SD ± 4.6 ng/ml) in non-PCOS women without live birth and with live birth respectively, 50 women were required as a sample size at 85% study power, two-sided significance level of 0.05, and effect size of 0.88. Owing to the large effect size, we were willing to test the hypothesis at a modest effect size of 0.65 at the same power and significance level, so the concluded sample size was 88 non-PCO women. Sample size calculation was done using G-Power 3.1.9.2 software program.

Evaluated adiposity measures

Clinical (BMI and WC), biochemical (VAI, lipid accumulation product, insulin resistance, and follicular fluid leptin), and sonographic (BFI) obesity-related parameters were assessed.

Clinical measures

They were evaluated at baseline and comprised waist circumference and body mass index. Waist circumference was measured at the end of expiration by a tape applied to the skin of the participant at a plane perpendicular to the midline and passing through a point just above the top of the iliac crest [36].

For body mass index (BMI), weight and height were measured while the participant was standing and wearing neither more than one layer of light clothes nor shoes.

Biochemical measures in serum and follicular fluid

Before starting any ovarian stimulation, serum samples were taken following an overnight fast for assessment of serum glucose, serum insulin, and serum lipoproteins. Serum Glucose level was measured in mmol/L using ADVIA 1800 Chemistry Auto-Analyzer, Siemens Healthineers, USA. Serum fasting Insulin was measured in μIU/mL using the Bioscience Human Insulin ELISA Kit (Catalog number: 10801). Serum triglycerides and high-density lipoprotein cholesterol (HDL-C) levels were measured using ADVIA 1800 Chemistry Auto-Analyzer, Siemens Healthineers, USA.

On the day of oocyte retrieval, follicular fluid pooled from large follicles; 17 mm or more, containing cumulus-oocyte complex was selected for sampling. Fluids containing debris and blood were excluded. They were centrifugated at 1500 rpm for 5 min, then stored at − 80 °C until leptin measurement was performed using SinoGeneClon ELISA Kit (Catalog number: SG-10057). Follicular fluid leptin determination was by pg/ml. All biochemical tests were performed in the laboratory of Women's Health Hospital, Assiut University, Egypt.

Sonographic measures

We measured in this study the abdominal subcutaneous and preperitoneal fat utilizing the methodology validated in the literature using ultrasound [10, 20, 34, 41, 48, 49]. The maximum preperitoneal and the minimum subcutaneous fat were the target measurements. They were conducted by the same researcher (The third author: AAM) using a SONOACE R5 ultrasound machine with CN2-8 curved abdominal transducer (Samsung Medison Co., LTD). A 4-month duration of capacity building for the researcher was achieved before the study proposal submission to IRB through coupling with level 3 experience sonographer in order to ensure and maximize the quality of the scans.

Calculations and benchmarking

Body mass index was calculated as weight (kg) divided by the square of height (m^2). According to the World Health Organization (WHO), BMI was categorized as normal (18.5–24. /m^2), overweight (25–29.9 kg/m^2), or obese (30 and above kg/m^2) [54].

To identify women with central obesity, a waist circumference equal to or more than 80 cm was used according to the International Diabetes Federation (IDF) and the report of WHO Expert Consultation on Obesity [1, 52, 53].

The homeostasis-model assessment for insulin resistance (HOMA-IR) was calculated using the equation: fasting insulin (μIU/mL) × glucose (mmol/L))/22.5. Participants were designated to be insulin resistant if HOMA-IR was equal to or more than 2.5 [7, 31].

Nassr et al [34] were the first to conclude and report on the body fat index formula. We utilized the same formula to calculate that new adiposity marker. It was calculated by multiplying pre-peritoneal fat (mm) and subcutaneous fat (mm), then dividing the product by height (cm) [34].

Sex-specific equations were employed to calculate the visceral adiposity index and lipid accumulation product [2, 51].

Cycle management, ovarian stimulation protocol, and embryo transfer

For each woman, personal and fertility data were reported, and the indication for ICSI was affirmed. Preparation of the patients and selection of the ovarian stimulation protocol followed the standardized protocols in ICSI practice and were individualized based on each patient's characteristics. The number of transferred embryos and the day of embryo transfer was not uniform for all enrolled patients; nevertheless, the same clinician conducted all transfers.

Study endpoints

The primary endpoint of the study was the probability of having a live birth (LB) per aspirated cycle as defined by the delivery of a live baby at 28 weeks of gestation or more. This is considered the standard definition in Egypt. We followed the standards of the Core Outcome Measure for Infertility Trials (COMMIT) initiative in reporting the primary and secondary endpoints [13]. The Time frame for reporting the outcomes was one fresh embryo transfer cycle. Enrolled women were contacted at the time of pregnancy test and estimated delivery date.

Statistical analysis

Statistical analysis was performed with the use of SPSS statistical package version 26.0 (IBM Corp, Armonk, NY, USA). The Kolmogorov-Smirnov test was used to determine data distribution. Normally distributed data are presented as mean (SD) however, abnormally distributed variables are presented as median (interquartile range

(IQR)). Comparisons were conducted between women with and without live birth. Also, comparisons were done between women with and without central obesity as well as among the common three indications of ICSI in our cohort; unexplained, male, and other factors in order to explore if there was any hidden effect of the indication of ICSI on the study variables and outcomes. Other factors encompass tubal disease, endometriosis, anovulation, and combined factors. Based on the comparisons, when appropriate, means were compared with the use of Student's *t* test or one-way ANOVA, and medians of non-parametric variables were compared utilizing the Mann-Whitney *U* test or Kruskal-Wallis test. Correlation analysis was conducted to determine the correlation between the adiposity measures and ICSI cycle variables and outcomes. Predictive models were constructed using regression and receiver operating characteristic (ROC) curve analyses to evaluate the predictability of adiposity measures for cycle outcomes. *P* value of < 0.05 was considered to be statistically significant.

Results

Ninety-one women were enrolled and completed the study. Their median (IQR) age was 30 (7) years with 54.9% of the cohort ($n = 50$) in their 30s. In our cohort, the common indications for ICSI were male and unexplained factors in 41.8% ($n = 38$) and 36.3% ($n = 33$) of women respectively. Other indications of ICSI included tubal (9.9%), anovulatory (4.4%), endometriosis (1.1%), and combined (6.6%) factors. History of ICSI was found in 25 (27.5%) women while the remainder were undergoing their first ICSI cycle. Antagonist protocol was chosen for about two-thirds of the participants ($n = 60$; 65.9%). Eleven women (12.1%) had normal BMI while 33 (36.3%) and 47 (51.6%) were overweight and obese respectively. Based on waist circumference 52 (57.1%) women had central obesity.

The total pregnancies were 31 (34.1%) ended in 28 live births: 25 term and 3 preterm births. There was one first-trimester miscarriage, one second-trimester miscarriage, and one ectopic pregnancy. Of live births, 11 cases were multiple pregnancies.

Based on the primary endpoint (live birth), comparisons between women with ($n = 28$) and without live birth ($n = 63$) are shown in Tables 1 and 2. Neither follicular fluid leptin concentration nor other adiposity measures were different between women with and without live birth.

In Receiver Operating characteristic curve (ROC) and logistic regression analyses, none of the evaluated adiposity measures (WC, BMI, BFI, VAI, LAP, HOMA-IR, and FF Leptin) in our study was a predictor for having a live birth. The area under the curve for WC, BMI, BFI, VAI, LAP, HOMA-IR, and Follicular fluid leptin was 0.57, 0.54, 0.55, 0.51, 0.53, 0.53, and 0.56, respectively.

Stepwise multivariable logistic regression analysis, showed that the outcome of the prior cycle, ability to have cryopreserved embryos, and the oocyte maturation index were the predictors for having live birth in our study (Table 3).

Investigating the relation among adiposity markers indicated that follicular fluid leptin was only correlated with HOMA-IR (Spearman's correlation coefficient $r = 0.21$, $P = 0.04$) (Fig. 1). This correlation is corroborated by the finding that HOMA-IR tends to be higher with increasing leptin tertile ($P = 0.047$) when categorizing leptin values of the studied cohort into 3 tertiles: the first tertile is < 15980.7 ($n = 31$ women), the second tertile is 15980.7 to 22130.9 ($n = 30$ women), and the third tertile is > 22130.9 ($n = 30$ women).

Loess regression with Epanechnikov kernel fitting was done to demonstrate the interaction of follicular fluid leptin and HOMA-IR on live birth (Fig. 2). The scatter plot indicates that leptin increases with increased insulin resistance both in women with and without live birth. However, surprisingly in both groups, this positive correlation was lost or even reversed when HOMA-IR approached 5 or more.

Comparing women with central obesity to their counterparts shows that they were obese and overweight, respectively (median (IQR) 32 (6.9) vs 27.1 (6); $P < 0.001$). Both groups were comparable regarding insulin resistance, follicular fluid leptin concentration, ICSI cycle characteristics, and outcomes. Miscarriage cases occurred in women without central obesity while all preterm deliveries were reported in central obesity women. However, indeed, we found central obesity women more likely to have higher BFI ($p < 0.001$), VAI ($p < 0.001$), and LAP ($p < 0.001$) (Supplemental Tables S1 and S2).

Unplanned subgroup post-hoc analysis according to the indication of ICSI showed that women in the male factor group had the lowest BMI ($p = 0.04$), and through borderline significance; the least preperitoneal fat thickness ($p = 0.05$), and the lowest fertilization rate ($p = 0.06$). All canceled transfers ($n = 4$) were also in the male factor group. Yet, follicular fluid leptin, the rest of the adiposity markers, and cycle parameters as well as outcomes did not differ among women in this subgroup analysis (Supplemental Tables S3 and S4).

Discussion

Main findings

Employing a prospective cohort study design, this study examined the follicular fluid leptin, along with a group of feasible clinical, biochemical, and sonographic

Table 1 Baseline characteristics and adiposity measures of women with and without LB

Variable	Women with LB ($n = 28$)	Women without LB ($n = 63$)	*P* value
Age (years) (median, IQR)	29.5 (9.25)	30 (7)	0.2
Age (years) (*n*,%)			0.6
18–< 25	7 (25%)	10 (15.9%)	
25–< 30	7 (25%)	17 (27%)	
30–35	14 (50%)	36 (57.1%)	
AMH (median, IQR)	2.2 (1.14)	2.2 (1.89)	0.9
Prior ICSI cycles (*n*, %)			**0.004**
Prior failed cycle	2 (7.1%)	12 (19 %)	
Prior successful cycle	8 (28.6%)	3 (4.8%)	
First cycle	18 (64.3%)	48 (76.2%)	
Causes of infertility (*n*,%)			0.9
Male factor	12 (42.9%)	26 (41.3%)	
Unexplained	10 (35.7%)	23 (36.5%)	
Tubal factor	2 (7.1%)	7 (11.1%)	
Anovulation	2 (7.1%)	2 (3.2%)	
Endometriosis	Zero	1 (1.6%)	
Combined	2 (7.1%)	4 (6.3%)	
BMI (kg/m^2) (median, IQR)	31 (6.9)	29 (8)	0.5
Class of body mass index (*n*, %)			0.4
Normal	2 (7.1%)	9 (14.3%)	
Overweight	9 (32.1%)	24 (38.1%)	
Obese	17 (60.7%)	30 (47.6%)	
Waist circumference in cm (median, IQR)	90 (22.8)	85 (22)	0.3
(range)	55–125	55–115	
% of women with central obesity (*n*,%)	15 (53.6%)	37 (58.7%)	0.6
% of insulin resistant women (≥ 2.5) (*n*,%)	14 (50%)	33 (52.4%)	0.8
HOMAIR (median, IQR)	2.6 (3.2)	3.3 (3.3)	0.6
Preperitoneal fat in mm (median, IQR)	10 (5.9)	11 (5.5)	0.5
(range)	(6–18.9)	(6.2–20)	
Subcutaneous fat in mm (median, IQR)	20.5 (8.7)	20.7 (14.7)	0.4
(range)	(10.4–33.7)	(6.90–42)	
Body fat index (median, IQR)	1.35 (1.5)	1.24 (1.84)	0.4
Visceral adiposity index (median, IQR)	1.44 (0.9)	1.42 (0.74)	0.9
Lipid accumulation product (median, IQR)	35.2 (23.24)	28.4 (29.84)	0.6
Follicular fluid leptin[a] in pg/ml (mean ± SD)	20336 ± 8006	18493 ± 6655	0.2

[a] Results of follicular fluid samples for 6 cases (4 in women with LB group, and 2 in their counterparts) showed minimally than detected levels that were attributed to an error in sampling or storage. So, leptin values for these 6 cases were computed through the linear interpolation method for missed data calculation. *HOMA-IR* Homeostasis-model assessment for insulin resistance, *BMI* body mass index, *AMH* Anti-mullerian hormone

adiposity markers as predictors of ICSI cycle outcomes in infertile non-PCOS women. Follicular fluid leptin was the basis for the power analysis of the sample size. According to BMI, only 12.1% of women had normal BMI while the remainder were overweight and obese. None of the tested parameters (including follicular fluid leptin levels) could predict a live birth. Insulin resistance was the only adiposity marker that has been positively correlated to follicular fluid leptin.

Strengths

Only FF leptin and insulin resistance were tested before in women without polycystic ovary syndrome. In the present work, we tested multiple markers that never were evaluated before in such a patient cohort particularly; preperitoneal fat, body fat index, and visceral adiposity index. Multiplicity of the evaluated markers was to seek explanations, detect superiority, and build combined predictors, in case of significance. The concept of testing

Table 2 ICSI Cycle characteristics and outcomes in women with and without LB

Variable	Women with LB ($n = 28$)	Women without LB ($n = 63$)	*P* value
Antral follicular count (median, IQR)	12 (6)	12 (6)	0.9
Toral gonadotropins dose (median, IQR)	3450 (1425)	3600 (1500)	0.6
Total days of stimulation (median, IQR)	11 (1)	12 (2)	0.5
Peak E2 (pg/ml)* (median, IQR)	2618 (1636)	2738 (1125)	0.9
Day of trigger progesterone (ng/ml) ** (median, IQR)	0.94 (0.57)	0.95 (0.44)	0.8
Day of trigger endometrial thickness (mm) (median, IQR)	10 (2)	10 (2)	0.5
(range)	(8–13)	(7–14)	
Oocyte retrieved (median, IQR)	11 (9.5)	12 (7)	0.6
Metaphase II oocytes (median, IQR)	9.5 (8.5)	8 (9)	0.1
Embryo transfer (*n*, %)			0.1
One embryo	0	4 (6.3%)	
Two embryos	12 (42.9%)	25 (39.7%)	
Three embryos	16 (57.1%)	25 (39.7%)	
Four embryos	0	5 (7.9%)	
Cancelled transfer	0	4 (6.3%)	
Maturation index in %	**84.7%**	**76.4%**	**0.02**
Fertilization rate in %	75%	66.7%	0.1
Women with good quality embryo transfer (at least one) (*n*,%)	**27 (96.4%)**	**46 (73%)**	**0.03**
Women who cryopreserved (*n*, %)	**19 (67.9%)**	**18 (30.5%)**	**0.001**

Peak E2 *, and Progesterone ** were analyzed for 77 and 65 cases, respectively due to missing data

Table 3 Predictability of cycle parameters for having a live birth

ICSI cycle parameter	*P* value	OR	95% CI
Prior cycle result	**0.028**	2	1.07–3.72
The ability to yield good-quality embryos	0.279	0.55	0.18–1.63
The ability to have cryopreserved embryos	**< 0.001**	5.28	2.01–13.83
Oocyte maturation index	**0.044**	22.16	1.09–451.24

multiple variants of predictors in overweight and obese women comes from observations that the sole utility of leptin is not adequate to construct the predictive model. Leptin to body mass index ratio has been reported to be superior to leptin alone as an IVF outcome predictor [8].

A dearth of literature reported live birth as a primary endpoint for the impact of FF leptin on ICSI. We followed our participants till delivery to reflect on live births concurring with the published standards for reporting infertility trials [13, 21].

Limitations

There are limitations that should be pointed out. First, the underrepresentation of normal BMI women in the enrolled cohort, which could affect the generalizability of conclusions and mitigate the discriminative threshold of the evaluated indices.

Second, follicular fluid sampling like other studies presents a limitation [24]. We used pooled follicular fluid which is still better than sampling just the first follicle. The ideal is to sample fluid from each follicle to conduct follicle-to-embryo tracking and sibling oocyte cohort analysis. However, it is difficult from the implementation point of view. Third, the study was powered for the detection of leptin predictability but not for other tested parameters. The lack of comparable studies with similar study designs presents a challenge in proposing assumptions during sample size calculation. Thus, the conclusions for the other parameters should be taken with caution. Fourth, although excluding PCOS women was justifiable, the performance of these indices to predict cycle outcomes in PCOS women remains elusive. Lastly, if we followed the outcomes after 2 or more embryo transfers, a difference in the outcomes would be disclosed.

Comment on the study adiposity measures

Leptin is a neuroendocrinal protein that exists in excess in obese women. Integrating follicular fluid leptin in our study is rationalized by the following: first, leptin is a reflection of the oocyte microenvironment; second, leptin receptors and m-RNA are expressed in granulosa cells [28], oocytes [4, 11] and pre-implantation embryos [4],third, leptin is depicted to regulate ovarian

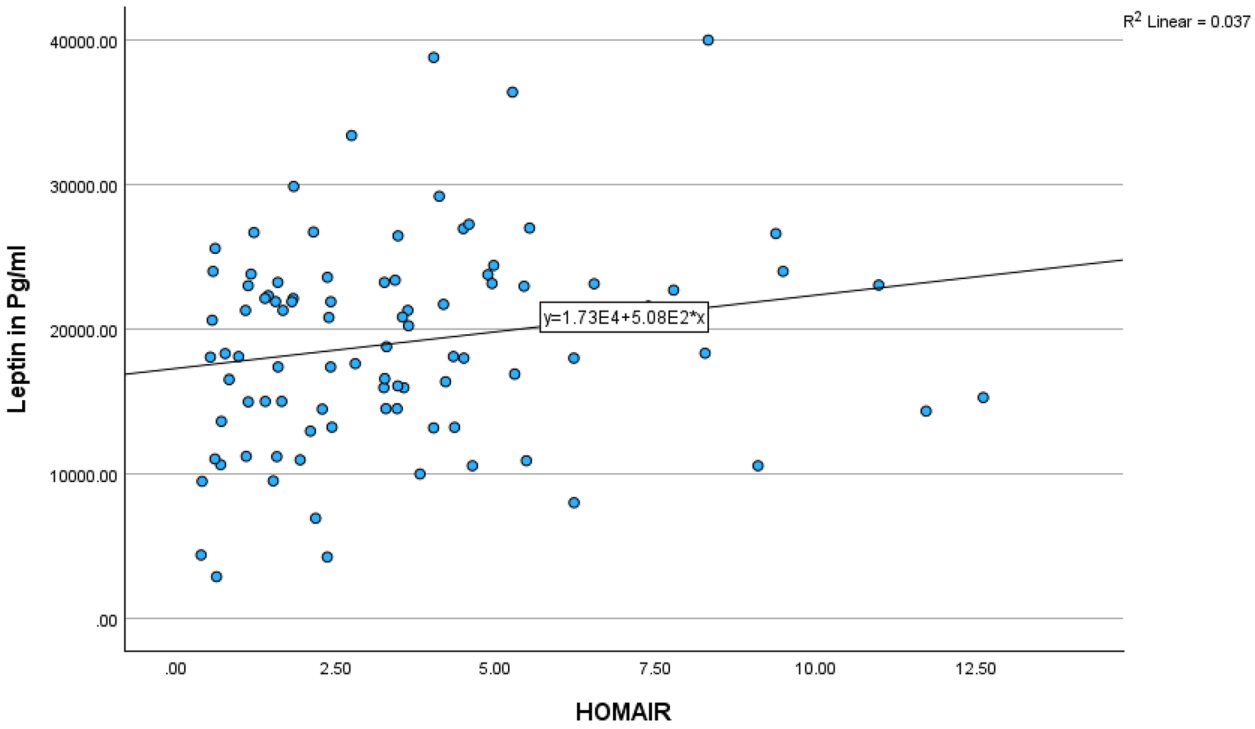

Fig. 1 Scatter plot showing positive correlation between HOMA-IR and follicular fluid leptin ($r = 0.21$, $P = 0.04$). HOMA-IR: homeostasis-model assessment for insulin resistance

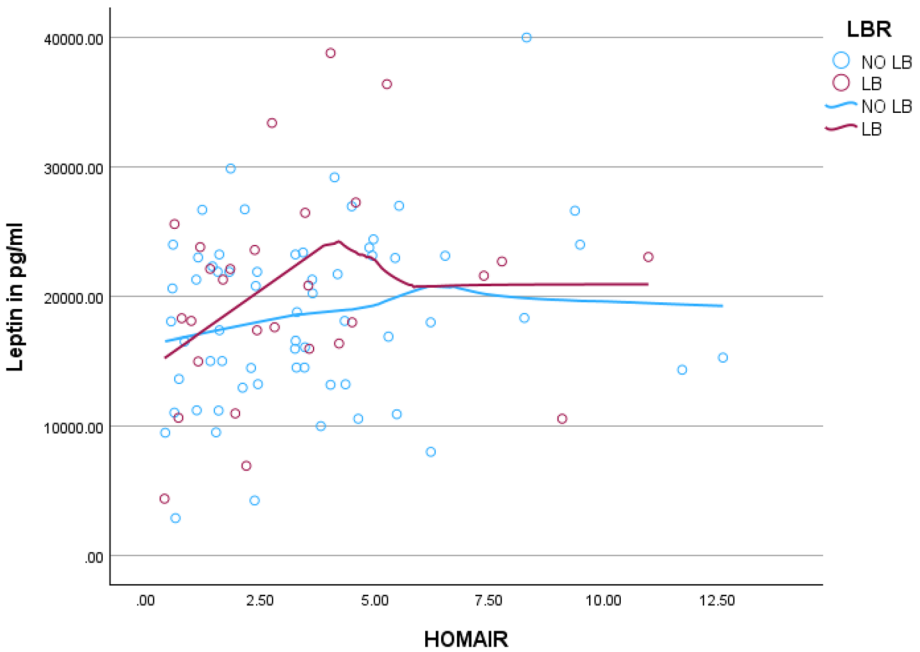

Fig. 2 Loess fitting scatter plot showing the interaction of HOMA-IR and follicular fluid leptin on LB. HOMA-IR: homeostasis-model assessment for insulin resistance

steroidogenesis, follicular growth and apoptosis, and oocyte maturation [12, 47], Fourth: controversial reports exist for the impact of FF leptin on cycle outcomes in the studied infertile cohort [3, 24, 30].

Our results concur with the results of a recent metanalysis evaluating [24] 11 observational studies (266 pregnant and 552 non-pregnant cases). This metanalysis indicated that pregnancy was unrelated to leptin levels in the follicular fluid. Highlighting the dilemma of methodological heterogenicity in follicular fluid sampling and cycle outcome reporting, different groups of researchers conveyed favorable cycle outcomes with low follicular fluid leptin concentrations. Mantzoros et al. [30] sampled fluid from the first aspiration of the dominant follicle and demonstrated low FF leptin in pregnant women (11.9 vs 17 ng/ml) within three ART cycles. However, this could bias their conclusions as comparable follicular fluid microenvironment among all follicles cannot be guaranteed. Another study by Anifandis et al. [3] did not explore the sampling details of follicular fluid and found women with peak estradiol range 1001–2000 ($n = 53$) yielded the highest pregnancy rate (35.8%) and had the lowest follicular fluid concentration (52 ng/ml). However, they did not test the predictability of follicular fluid leptin for clinical pregnancy after adjustment of peak serum estradiol. A study reporting live birth as an outcome found FF leptin sampled from the first dominant follicle predicted live birth at a concentration of 16 ng/ml with sensitivity and specificity of 78.3% and 54.2%, respectively [27]. Another group has addressed higher sensitivity and specificity of similar leptin concertation for the oocyte maturation when follicular fluid was pooled from the largest 3 follicles [23].

Body mass index has been the benchmarking for decades in IVF/ICSI daily practice in pre-IVF/ICSI preparation of obese/overweight infertile women to achieve the optimum outcomes for the service stakeholders [15, 17]. The contradictory conclusions about the effect of body mass index and weight reduction on IVF/ICSI outcomes [14, 25, 33, 42, 44, 50] should encourage the clinicians to test other or new feasible adiposity predictors for the cycle outcomes such as visceral adiposity and body fat indices.

Obesity and abdominal preperitoneal fat alter the metabolic milieu of follicular fluid in IVF women by increasing lipid peroxides and reducing total antioxidant capacity [5, 10, 22, 35]. Reflection of the preperitoneal fat on FF leptin in not been addressed before. In our results, neither preperitoneal fat nor the new body fat index improved the predictability of FF leptin for cycle outcomes. Body fat index has been evaluated before in pregnant women and revealed to predict gestational diabetes at cut-offs of 0.5 and 0.88 [6, 34, 46].

The concept of preperitoneal fat measurement is warranted by evidence that it is a source of multiple inflammatory mediators contributing to insulin resistance and reproductive dysfunction [18, 29, 55]. The correlation between insulin resistance and leptin found in our study is observed in other studies [27] and is supported by the fact that insulin stimulates leptin secretion [9]. This correlation was unrelated to live birth in our women. Moreover, surprisingly, the increase in leptin with insulin resistance was lost at higher insulin resistance levels denoting that a phenomenon of desensitization could be exhibited by leptin receptors at high insulin resistance levels. Further research is needed to elucidate this finding.

According to the reference 'The Vienna consensus: report of an expert meeting on the development of art laboratory performance indicators' the benchmarking range for maturation index is 75–90%. Although statistically significant between women with and without live birth, the maturation index of both groups is still reaching the benchmarking range (84.7% in women with LB and 76.4% in women without LB; as shown in Table 2). Conducting the comparisons based on central obesity and ICSI indication, still all subgroups achieved a maturation index within the benchmarking range (as shown in Supplemental Tables S2 and S4)

For the fertilization index based on the Vienna consensus, the competency value should be ≥ 65, and the benchmark value should be ≥ 80. In the current study, the fertilization index is 75% and 66.7% in women with and without LB, respectively. We are above the competency value, however, we did not reach the benchmark value. The lowering of the overall fertilization rate could be attributed to including the male factor in the study that seemed to be of poor prognosis and achieved the lowest fertilization rate among the groups (63.6%) (below the competency value) (Supplemental Table S4).

Conclusion

The current work limited the inference to only non-PCOS women and could not find evidence that a correlation exists between follicular fluid leptin and live birth after ICSI cycles. Furthermore, prediction for live birth after IVF, could not be demonstrated when any of the tested parameters (clinical, biochemical, preperitoneal fat, and BFI) was used. Further research should be warranted to evaluate the plausibility of these indices in all BMI categories of women without PCOS. Further research that includes another arm of PCOS in the evaluation of variant adiposity markers in ART women is needed.

Supplementary Information

The online version contains supplementary material available at https://doi.org/10.1186/s43043-024-00164-y.

Additional file 1: Supplementary Table S1. Baseline characteristics and adiposity measures of the study groups based on waist circumference. **Supplementary Table S2.** Cycle characteristics and outcomes of the studied groups based on waist circumference. **Supplementary Table S3.** Characteristics and adiposity measures of women based on ICSI indication. **Supplementary Table S4.** ICSI cycle parameters and outcomes of women based on ICSI indication.

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Authors' contributions
(1) The conception and design of the study or acquisition of data, or analysis and interpretation of data: Ahmed Abdelmagied, Mohammed K. Ali, Safwat Abdel-Rady, Ahmed A. Abdel-Alleem, Alaa Makhlouf, Azza Abo Elfadl. (2) Drafting the article or revising it critically for important intellectual content: Ahmed Abdelmagied, Mohammed K. Ali. (3) Final approval of the version to be submitted: Ahmed Abdelmagied, Alaa Makhlouf, Ahmed A. Abdel-Aleem, Safwat A. Mohamed, Azza Abo Elfadl, Mohammed K. Ali.

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Availability of data and materials
The datasets used and/or analyzed during the current study are available from the corresponding author upon reasonable request.

Declarations

Ethics approval and consent to participate
The Institutional Review Board (IRB) of the Faculty of Medicine, Assiut University approved the study on January 22, 2019 (IRB approval number: 17200286). Every patient was informed about the steps of the study and written informed consent was obtained from each patient.

Consent for publication
Not applicable.

Competing interests
The authors declare that they have no competing interests.

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