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ORIGINAL ARTICLE

Prospective Study of Effect of Body Weight on *in vitro* Fertilization Outcome in Reproductive Age Group

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ABSTRACT

Background: Various prognostic factors in assisted reproduction procedures have been described and analyzed which includes woman's age, cause of infertility, ovarian response and uterine receptivity, the semen quality, and the body mass index (BMI). Optimal BMI is required for an optimal response. There is controversy among various reports, which is partly caused by the varying focus of investigators and differences in study designs, which led us to examine the relationship between BMI, *in vitro* fertilization (IVF) and intracytoplasmic sperm injection (ICSI) outcome in our unit.

Objective: To study impact of BMI on IVF outcome prospectively.

Materials and methods: It is a prospective study over a period of 1 year in the age group 25 to 35 years attending the IVF clinic was conducted at a tertiary infertility center in Bangalore, India between November 2010 and October 2011.

Results: There is a close association of increased BMI in particular when BMI is > 30 kg/m^2 and the reduced outcomes of IVF/ICSI treatment in the form of decreased clinical pregnancy and higher early pregnancy loss. Furthermore, increased BMI is related to higher dosage and duration of gonadotropins requirement increased risk of cancellation and fewer collected oocytes.

Conclusion: Obesity is associated with an increased risk of early pregnancy loss. Also need of high dose of gonadotropin, less number of collected oocytes is observed. Implantation rate, pregnancy rate and miscarriage rate was comparable but live births are high in normal weight and overweight as compared to extremes of BMI. So will be appropriate to recommend life style modifications including weight loss to achieve an appropriate BMI prior to IVF.

Keywords: Body mass index, Gonadotropin, IVF, Pregnancy.

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INTRODUCTION

Initiation and maintenance of reproductive functions are related to an optimal body weight in women. Underweight [body mass index (BMI) < 18.5 kg/m^2], as well as overweight (BMI ≥ 25 kg/m^2) and obesity (BMI ≥ 30 kg/m^2) are associated with an increased risk of anovulatory infertility.[1-3]

Reduced fecundity of underweight and overweight women is probably related to multiple endocrine and metabolic alterations, which include effects on steroid metabolism and altered secretion and action of insulin and other hormones, such as leptin, resistin, ghrelin or adiponectin.

These alterations can affect follicle growth, embryo development and implantation[4-6] and it is therefore of concern that being underweight or overweight may interfere with treatment of infertility with IVF and ICSI. Recently, a debate is started in literature whether or not restricting the access to fertility treatment on the ground of female BMI.[7-10]

In assisted reproduction, however, there are conflicting reports on the effect of obesity on oocyte quality, embryo development, lower number of mature oocytes, lower implantation and pregnancy rates.[11-15] Endometrium also seems to have a negative impact on reproductive outcome in studies based on oocyte donation model.[16-18]

Clinical observations on the effects of body weight during IVF and ICSI are conflicting changes in BMI has serious impact on the various aspects of health in particular the reproductive function of women.[19-22] There is controversy among various reports, which is partly caused by the varying focus of investigators and differences in study designs, led us to examine the relationship between BMI and IVF/ICSI outcome in our unit.

MATERIALS AND METHODS

Study Design

It is a prospective study over a period of 1 year in the age group 25 to 35 years (with 500 females subjects) attending the IVF clinic was conducted at a tertiary infertility center in Bangalore, India between November 2010 and October 2011. Approval was obtained from the Institutional Ethics Committee (IEC). An informed consent was taken from all patients.

Various inclusion and exclusion criteria were taken so as to avoid confounding variables as much as possible so as to get comparable results.

The patients had their BMI recorded at the initial consultation before starting the treatment cycle. Patients were divided into four groups: group A (BMI < 19 kg/m^2); group

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B (BMI between 19 and 24.9 kg/m^2); group C (BMI 25 to 29.9 kg/m^2) and group D (BMI > 30 kg/m^2).

Group A is underweight which comprised of 2% (10) patients, 41.6% (208) in group B (Normal BMI), 42.6% (213) in group C (overweight) and 13.8% (69) in group D (obese).

The primary end-point assessed was clinical pregnancy rate.

The secondary end-points included: cycle cancellation rate, the number of oocytes retrieved, mature oocytes, implantation rate and live birth rate.

Inclusion criteria: Fresh IVF/ICSI cycles, non donor cycles, age group 25 to 35 years.

Exclusion criteria: >35 years age, frozen embryo transfer, donor oocyte, gestational surrogacy, patients with an accompanying medical problem which may lead to abnormal BMI, such as diabetes mellitus, hyper or hypothyroidism.

Variables taken are: Clinical and embryological parameters stimulation protocol was decided based upon baseline endocrine profile, age and response in previous cycle if any, baseline hormonal levels, baseline vaginal ultrasound scan (for antral follicle count using 6.5 MHz vaginal probe and Prosound 4, Medison India Ltd. Ultrasound Machine) on Day 3, Anti Mullerian Hormone (AMH) values using DSL-ELISA kit and response in previous IUI/IVF cycles. Patients with good ovarian reserves were stimulated by long against down-regulation protocol for ovarian stimulation while women with poor ovarian reserves were stimulated by antagonist protocol. (Details of both the protocols are given vide infra).

Ovarian stimulation was achieved by human menopausal gonadotropin (Menopur, Ferring). As a rule, the starting daily FSH dose was 150 IU, Gonal F (Serono, Italy) or Recagon (Organon) with exception of women older than 35 years who received 225 IU and women with polycystic ovaries who received 75 to 150 IU.

The history and investigation reports were entered in a specially prepared performa. All patients undergoing conventional IVF-ET or ICSI were stimulated in accordance with the appropriate protocol decided for them and the treatment cycle was monitored using transvaginal ultrasound scans (TVS) and serum estradiol (E2), progesterone (P) and luteinising hormone (LH) levels, wherever required. Injection uhCG, 5000IU IM (Ovutrig HP, VHB Life Sciences, Mumbai, India, Profasi, Serono or Pregnyl, Organon) or recombinant hCG 250 mcg (Ovidrel, Serono, Inc.) used in some cases (poor responders, those at high risk of OHSS) was administered when the average diameter of lead follicle becomes more than 18 mm and at least two follicles become more than 16 mm in diameter. Oocytes were retrieved 34 to 36 hours after u hCG injection, by transvaginal ultrasound guided aspiration.

Standard laboratory protocols were followed, including ICSI, laser assisted hatching (LAH) for cleavage stage embryos and extended culture for blastocyst transfer, as clinically appropriate.

DISCUSSION

In our study, as per age distribution, 54.6% (273/500) were in age group >30 years and 45.4% (227/500) were <30 years age. Group A is underweight which comprised of 2% (10) patients, 41.6% (208) in group B (Normal BMI), 42.6% (213) in group C (overweight) and 13.8% (69) in group D (obese). Mean duration of infertility was 5.27 ± 2.49 years in group B and in group C, it was 5.63 ± 2.54 years (Fig. 1). Among the causes of infertility in all patients, most common was male factor in form of oligospermia, severe oligoasthenozoospermia, necrospermia, asthenozoospermia, azoospermia or sexual dysfunction in 60% (6) in group A, 58.7% (122) in group B, 62.9% (134) in group C and 65.2% (45) in group D followed by the tubal factor (20%), unexplained infertility (15%), ovulatory dysfunction and mixed causes in remaining patients (Table 1).

A number of frozen embryos were more in group B and C as compared to A and D though not statistically significant (p = 0.849). Endometrial thickness was 8 to 10 mm in 98 (47.1%) in group B, 106 (49.8%) in group C as compared to 4 (40%) and 32 (46.4%) in groups A and D with p = 0.988 (statistically non significant). Day 2/3 embryo transfer was done in 62% of patients and in remaining blastocysts were transferred in our clinic (p = 0.008).

Dosage of gonadotropins was high in overweight and obese group such that 41 to 60 ampules were used in 23% in group C compared to 14% in group B (Table 2).

The primary and secondary outcomes of our study are depicted in Table 3.

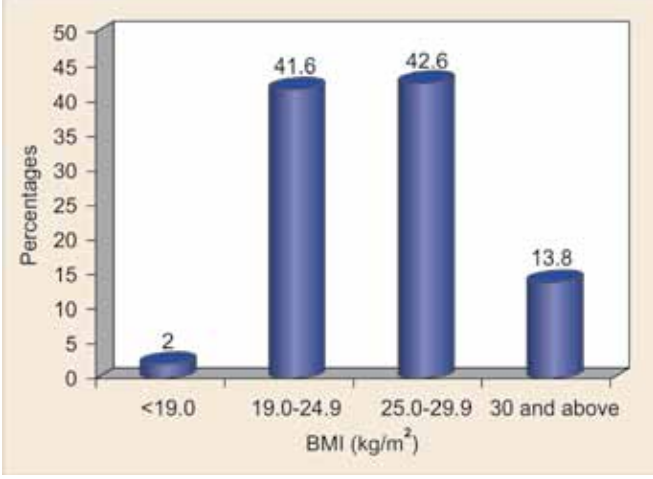

Fig. 1: Body mass index distribution

Days of stimulation were more in group C (11.43 ± 1.49 and group D (11.67 ± 1.79) with p = 0.037.

Dosage of gonadotropins was more in overweight and obese patients (p = 0.003) compared to normal BMI and underweight groups.

A number of dominant follicles, number of mature (M2, Metaphase 2) oocytes, number of grade A (good quality) embryos formed and frozen were more in normal weight and overweight patients (groups B and C) as compared to extremes of BMI variations (underweight and obese) though

Table 1: Clinical characteristics of women according to body mass index (kg/m^2)

Clinical variables	*BMI (kg/m^2)*				*p-value*
	<19.0 (n =10)	*19.0-24.9 (n = 208)*	*25.0-29.9 (n = 213)*	*30 and above (n = 69)*	
Age in years	29.80 ± 2.20	29.98 ± 3.39	30.92 ± 3.19	32.36 ± 2.74	<0.001**
Duration of infertility	5.40 ± 2.67	5.27 ± 2.49	5.63 ± 2.54	6.21 ± 2.73	0.073*
Main causes of infertility					
• Mixed causes	0 (0%)	1 (0.5%)	1 (0.5%)	0 (0%)	0.946
• Tubal factor	1 (10%)	49 (23.6%)	47 (22.1%)	16 (23.2%)	0.786
• Anovulatory	1 (10%)	3 (1.4%)	2 (0.9%)	0 (0%)	0.055*
• Unexplained	2 (20%)	33 (15.9%)	29 (13.6%)	8 (11.6%)	0.762
• Male factor	6 (60%)	122 (58.7%)	134 (62.9%)	45 (65.2%)	0.676
Type of infertility					
• Primary	5 (50%)	148 (71.2%)	152 (71.4%)	42 (60.9%)	0.205
• Secondary	5 (50%)	60 (28.8%)	61 (28.6%)	27 (39.1%)	

*,**Statistically significant (more with inc in no. of stars)

Table 2: Clinical characteristics of women according to body mass index (kg/m^2)

Clinical variables	*BMI (kg/m^2)*				*p-value*
	<19.0 (n = 10)	*19.0-24.9 (n = 208)*	*25.0-29.9 (n = 213)*	*30 and above (n = 69)*	
Baseline FSH (Day 2/3) mIU/ml					
• <10	8 (80%)	184 (88.5%)	197 (92.5%)	67 (97.1%)	0.180
• 10-20	2 (20%)	21 (10.1%)	15 (7%)	1 (1.4%)	
• >20	0 (0%)	3 (1.4%)	1 (0.5%)	1 (1.4%)	
• Mean ± SD	7.52 ± 2.95	7.52 ± 3.66	6.94 ± 2.75	7.08 ± 4.37	
Number of frozen embryos					
• Nil	8 (80%)	172 (82.7%)	175 (82.2%)	56 (81.2%)	0.849
• 1-5	1 (10%)	27 (13%)	28 (13.1%)	10 (14.5%)	
• >5	1 (10%)	5 (2.4%)	6 (2.8%)	1 (1.4%)	
• Mean ± SD	7.00 ± 2.82	4.39 ± 2.16	4.16 ± 1.92	3.46 ± 2.02	
Endometrial thickness (mm)					
• 6-8	1 (10%)	18 (8.7%)	17 (8%)	5 (7.2%)	0.988
• 8-10	4 (40%)	98 (47.1%)	106 (49.8%)	32 (46.4%)	
• >10	2 (20%)	82 (39.4%)	81 (38%)	23 (33.3%)	
• Mean ± SD	9.61 ± 1.33	10.28 ± 1.48	10.22 ± 1.32	10.09 ± 1.52	
Day of embryo transfer					
• Day 2	0 (0%)	21 (10.1%)	34 (16%)	19 (27.5%)	0.008**
• Day 3	5 (50%)	109 (52.4%)	94 (44.1%)	28 (40.6%)	
• Day 5	3 (30%)	71 (34.1%)	75 (35.2%)	16 (23.2%)	
Protocol followed					
• Long	2 (20%)	44 (21.2%)	44 (20.7%)	14 (20.3%)	0.786
• Antagonist	7 (70%)	142 (68.3%)	152 (71.4%)	46 (66.7%)	
• Others	1 (10%)	21 (10.1%)	14 (6.6%)	9 (13%)	
Complications					
• No	10 (100%)	208 (100%)	213 (100%)	69 (100%)	1.000
• Yes	0	0	0	0	
Age in years					
• <25 years	1 (10%)	17 (8.2%)	8 (3.8%)	0 (0%)	0.005**
• 25-30 years	5 (50%)	95 (45.7%)	84 (39.4%)	17 (24.6%)	
• 30-35 years	4 (40%)	93 (44.7%)	116 (54.5%)	50 (72.5%)	
• 35-40 years	0 (0%)	3 (1.4%)	5 (2.3%)	2 (2.9%)	
No. of ampules of Gns					
• <20	0 (0%)	27 (13%)	17 (8%)	6 (8.7%)	0.012*
• 21-40	8 (80%)	147 (70.7%)	143 (67.1%)	43 (62.3%)	
• 41-60	1 (10%)	31 (14.9%)	49 (23%)	14 (20.3%)	
• >60	1 (10%)	3 (1.4%)	4 (1.9%)	6 (8.7%)	

*,**Statistically significant (more with inc in no. of stars). SD: standard deviation

Table 3: Outcome of *in vitro*/intracytoplasmic sperm injection of women according to body mass index (kg/m^2)

Outcome	*BMI (kg/m^2)*				*p-value*
	< 19.0 (n = 10)	*19.0-24.9 (n = 208)*	*25.0-29.9 (n = 213)*	*30 and above (n = 69)*	
Days of stimulation	10.30 ± 0.67	11.32 ± 1.35	11.43 ± 1.49	11.67 ± 1.79	0.037*
Number of ampules of Gn	28.20 ± 13.92	30.75 ± 10.94	33.63 ± 11.37	43.97 ± 14.89	0.003**
Follicles > 14 mm	12.30 ± 6.15	11.32 ± 7.17	10.79 ± 6.73	10.24 ± 6.94	0.626
No. of M2 oocytes	5.10 ± 5.63	8.98 ± 4.81	7.62 ± 4.34	6.53 ± 4.96	0.654
Grade of embryo					
• A	6 (60%)	188 (90.4%)	201 (94.4%)	60 (87%)	0.593
• B	0 (0%)	12 (5.8%)	7 (3.3%)	3 (4.3%)	
No. of embryos transferred	2.37 ± 0.52	2.46 ± 0.68	2.49 ± 0.68	2.30 ± 0.66	0.284
ET (Embryo transfer)					
• Easy	8 (80%)	198 (95.2%)	187 (94.4%)	56 (81%)	0.045*
• Difficult	0 (0%)	3 (1.4%)	17 (8%)	9 (13%)	
• ET cancelled	0 (0%)	5 (2.3%)	9 (4.2%)	4 (5.8%)	
Cycle outcome					
• Cancelled	2 (20%)	10 (4.8%)	6 (2.8%)	6 (8.7%)	0.027*
• Negative	5 (50%)	123 (59.1%)	127 (59.6%)	41 (59.4%)	0.947
• Positive	3 (30%)	75 (36.1%)	80 (37.6%)	22 (31.9%)	0.828
Oocyte retrieval rate	89.29 ± 43.52	91.57 ± 17.55	106.29 ± 17.89	86.19 ± 17.02	0.012*
Fertilization rate	91.11 ± 26.67	91.67 ± 15.82	91.44 ± 15.61	93.41 ± 14.02	0.854
Cleavage rate	88.88 ± 33.33	97.22 ± 9.36	96.41 ± 10.42	98.90 ± 5.08	0.040*
Implantation rate	15.00 ± 33.74	13.26 ± 24.36	16.35 ± 27.35	16.87 ± 28.17	0.621
Outcome					
• Biochemical pregnancy	0 (0%)	16 (7.7%)	15 (7%)	2 (2.9%)	0.438
• Ectopic	0 (0%)	2 (1%)	1 (0.5%)	0 (0%)	0.817
• Missed abortion	0 (0%)	6 (2.9%)	9 (4.2%)	4 (5.8%)	0.630
• Single	1 (10%)	42 (20.2%)	44 (20.7%)	14 (20.3%)	0.879
• Twins	1 (10%)	7 (3.4%)	8 (3.8%)	3 (4.3%)	0.749
• Triplets	0 (0%)	1 (0.5%)	4 (1.9%)	0 (0%)	0.388
• Quadruplets	0 (0%)	0 (0%)	0 (0%)	1 (1.4%)	0.100

*,**Statistically significant (more with inc in no. of stars)

not statistically significant. Mean number (2.46 ± 0.68) of embryos transferred were comparable in four groups with p = 0.289.

Ninety percent of embryo transfers done were easy. Difficult embryos transfers were more in group C (8%) and group D (13%) compared to group A (none, 0%) and group B (1.4%).

Embryo transfer was cancelled in 9 (4.2%) patients in group and 4 (5.8%) in group D as compared to 2.3% in group B and none in group A (p = 0.045) (Fig. 2).

Oocyte retrieval rate was high in group B (91.57 ± 17.55) and group C (106.29 ± 17.89) as compared to group A and D. Fertilization rate was comparable. Cleavage rate was high in groups B, C and D as compared to group A (p = 0.040). Implantation rate was almost similar in all four groups (16-17%) with p = 0.621. Pregnancy rate was 38% in group C (maximum) followed by 36% in group B, 31% in group D and 30% in group A (p = 0.828), so it was comparatively statistically nonsignificant.

A number of miscarriages were maximum in overweight and obese patients but comparable to normal BMI group with p = 0.630 (Fig. 3).

In our study, out of 500 patients recruited, 171 were pregnant so overall pregnancy rate was 34% (171/500) with clinical pregnancy rate of 31% and 28% in overweight and normal BMI patients as compared to 17% and 20% in obese and underweight patients in comparable age group (p = 0.163) (Table 4 and Fig. 4).

While live birth rate was upto 26% in group C and D as compared to 20% and 24% in groups A and B (p = 0.929).

Changes in BMI have serious impact on the various aspects of health in particular the reproductive function of

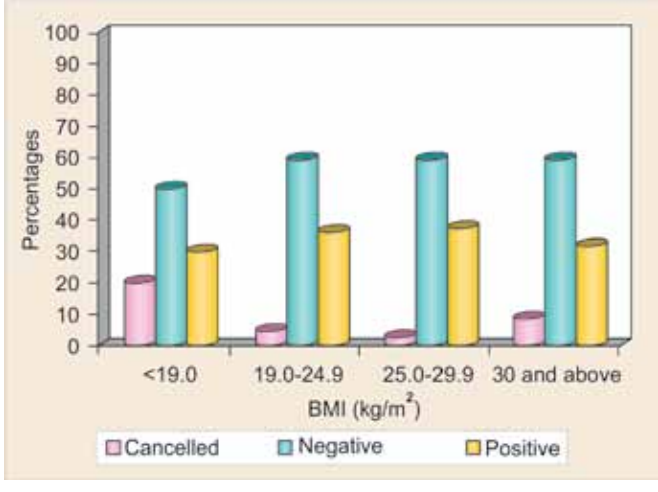

Fig. 2: IVF/ICSI cycle outcome

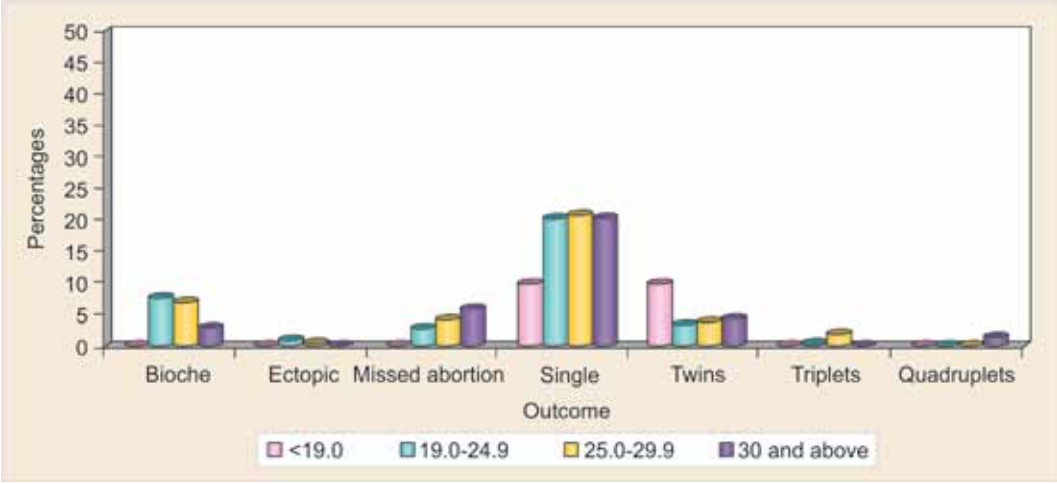

Fig. 3: Pregnancy outcome

women.[23] The reduced fecundity of underweight and overweight women is probably related to multiple alterations in their metabolic and endocrine functions. Previous studies have suggested that women with extremes of BMI had lower numbers of oocytes,[24] lower fertilisation rate[25] and lower implantation rate[26,27] and it is therefore of concern that being underweight or overweight may interfere with treatment of infertility with IVF and ICSI.

In overweight or obese compared with normal weight women, increased FSH requirement during ovarian stimulation, frequent cycle cancellations, low pregnancy rate and higher miscarriage rate have been observed.[28-30]

In summary, this study suggests that obesity is associated with lower clinical pregnancy and live birth rate and the need for higher and longer period of FSH stimulation due to impaired ovarian response but underweight was not related to an impaired outcome.

STRENGTH OF STUDY

The study has some significant strength. First of all, the major strength was its prospective nature. In the literature, most of the data about this topic have been based on retrospective studies or pooled data, thus allowing potential for observer bias.

Another advantage was, this study included only first assisted reproductive techniques (ART) cycle. There are many studies including more than one cycle of the same participants. In this situation, undiagnosed bad prognostic factors of an individual may cumulatively affect on results.

Lastly, we calculated BMI of the participants at the initiation of ovarian stimulation.

CONCLUSION

Body mass index has a role in counselling couples before initiation of ART.

Optimal BMI should be a pre-requisite before recruiting the patients for the controlled ovarian stimulation (COS). Obese patients should be strongly encouraged to loose weight before starting ART.

Obesity is associated with reduced pregnancy rates and increased requirement gonadotropin for ovulation induction.

Obesity is associated with an increased risk of early pregnancy loss occurring before 6 weeks gestation.

Implantation rate, pregnancy rate and miscarriage rate was comparable but live births are high in normal weight and overweight as compared to extremes of BMI due to

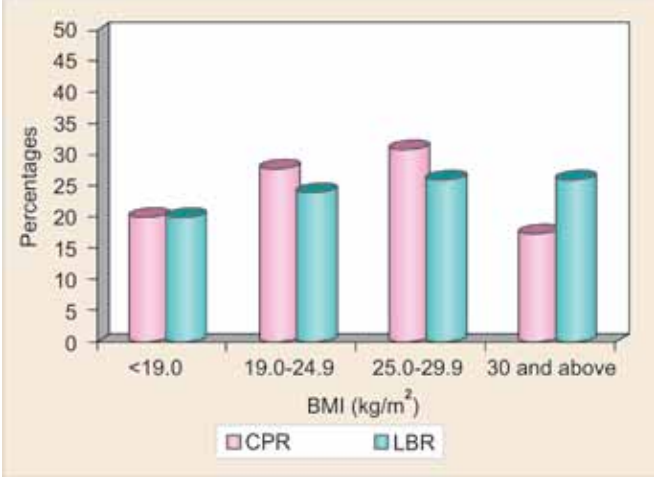

Fig. 4: Clinical pregnancy rate and live birth rate correlation with BMI

Table 4: Outcome of *in vitro* fertilization/intracytoplasmic sperm injection of women according to body mass index (kg/m^2)

Outcome	*BMI (kg/m^2)*				*p-value*
	< 19.0 (n = 10)	*19.0-24.9 (n = 208)*	*25.0-29.9 (n = 213)*	*30 and above (n = 69)*	
CPR	2 (20.0%)	58 (27.9%)	66 (30.9%)	12 (17.4%)	0.163
LBR	2 (20.0%)	50 (24.1%)	56 (26.3%)	18 (26.1%)	0.929

high number of miscarriages in extremes of BMI though not statistically significant.

Though, it appears appropriate to recommend weight loss prior to IVF in patients especially in young patients while in older patients, a more immediate and aggressive approach to ART may be warranted.

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