The effect of Body Mass Index on the outcome of Controlled Ovarian Hyperstimulation and Intrauterine Insemination in Hospital Sultan Ismail, Johor Bahru

by

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~ *Noraida*

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LIST OF ABBREVIATIONS

AFS	American Fertility Society
ACOG	American Colleague Obstetric Gynaecology
BMI	Body Mass Index
CC	Clomiphene Citrate
COH	Controlled Ovarian Hyperstimulation
CPR	Clinical Pregnancy Rate
FSH	Follicular Stimulating Hormone
GnRH	Gonadotrophin Releasing Hormone
HCG	Human Chorionic Gonadotrophin
IGF-1	Insulin Growth Factor-1
IUI	Intrauterine Insemination
LH	Luteinizing Hormone
NSAID	Non-Steroidal Anti-Inflammatory Drug
PCOS	Polycystic Ovarian Syndrome
RCOG	Royal College of Gynaecology
SA	Seminal Analysis
TFI	Tubal Factor Infertility
TVS	Transvaginal Sonography
rFSH	Recombinant Follicular Stimulating Hormone
uFSH	Urinary Follicular Stimulating Hormone
WHO	World Health Organization

ABSTRAK

OBJEKTIF

Untuk menentukan kesan indeks jisim badan terhadap hasil permanian beradas dari hiperstimulasi ovari terkawal di Hospital Sultan Ismail Johor Bahru, seterusnya membandingkan kadar kehamilan dalam pesakit jisim badan normal dan berlebihan. Faktor yang berkaitan untuk hasil yang berjaya di kalangan jisim badan berlebihan telah ditentukan.

KAEDAH

Kajian retrospektif selama 5 tahun ini dilakukan di Hospital Sultan Ismail, Johor Bharu dari 2016 hingga 2020. Data dari 278 pesakit dengan 516 kitaran, yang menjalani hiperstimulasi ovari terkawal dan permanian beradas telah disemak. Pesakit telah dikategorikan kepada mereka yang mempunyai jisim badan yang normal dan berlebihan, yang mana hasil rawatan dibandingkan, menggunakan ujian chi kuasa dua. Pembolehubah seperti umur wanita, tempoh, punca dan jenis subfertility, dianalisis menggunakan ujian regresi untuk menentukan kitaran yang berjaya di kalangan pesakit jisim badan berlebihan. Nilai $p < 0.05$ telah diambil sebagai perbezaan ketara.

KEPUTUSAN

Sebanyak 516 kitaran permanian beradas telah dimasukkan dalam analisis. Tiga puluh lapan peratus daripada kitaran adalah dalam wanita berjisim biasa, manakala berat badan berlebihan dan wanita obesiti adalah 62%. Lapan puluh lima perpuluhan tiga peratus adalah etnik Melayu, 6.0% adalah Cina, 4.7% adalah India dan 4.0% yang lain, mencerminkan kepelbagaian etnik Malaysia. Purata dan sisihan piawai untuk jisim badan normal dan jisim badan berlebihan ialah 21.9 kg/m^2 dan 29.1 kg/m^2. Purata umur bagi semua subjek ialah 31 tahun. Tempoh purata subfertiliti dalam jisim

badan normal ialah 65 bulan (5.4 tahun) dan dalam jisim badan yang berlebihan ialah 62 bulan (5.2 tahun). Sebanyak 49 kehamilan (9.4%) telah dicapai dalam 516 kitaran. Daripada jumlah ini, 33 (67.3%) adalah daripada kumpulan jisim badan yang berlebihan dan 16 (32.7%) daripada kumpulan jisim badan biasa. Tiada perbezaan dalam kadar kehamilan klinikal antara jisim badan biasa dan wanita dengan jism badan berlebihan women (diselaraskan OR 1.4, 95% CI 0.66 – 3.11, p-value 0.369). Perbezaan dalam kehamilan ektopik dan keguguran juga tidak ketara. Korelasi negatif didapati antara umur wanita dan kejayaan yang menjalani hiperstimulasi ovari terkawal dan permanian beradas (p=0.815) di kalangan jisim badan berlebihan. Ketidaksuburan 'secondary' adalah lebih tinggi berbanding ketidaksuburan 'primary' (diselaraskan 2.8 (95% CI 0.97-7.92), p=0.058). Tempoh ketidaksuburan didapati tidak signifikan dalam hasil hiperstimulasi ovari terkawal dan permanian beradas. Dalam keputusan ini, semua punca ketidaksuburan berkorelasi negatif dengan hiperstimulasi ovari terkawal dan permanian beradas yang berjaya, mungkin disebabkan oleh bilangan pesakit yang kurang dalam kajian ini.

KESIMPULAN

Kesimpulannya, keputusan kami bercanggah dengan kepercayaan ramai bahawa jisim badan yang lebih tinggi mempunyai kadar kejayaan yang rendah untuk menjalani hiperstimulasi ovari terkawal dan permanian beradas. Hubungan antara jisim badan berlebihan dan hasil ketidaksuburan adalah kompleks dan boleh dipengaruhi oleh pelbagai faktor. Lebih banyak kajian dan sampel diperlukan untuk menilai hubungan jisim badan yang berlebihan dengan kejayaan hiperstimulasi ovari terkawal dan permanian beradas.

ABSTRACT

OBJECTIVE

To determine the effect of BMI on COH with IUI outcome in Hospital Sultan Ismail Johor Bahru, comparing the clinical pregnancy rate in normal and excessive BMI patients. The associated factors for successful outcomes among excessive BMI were determined.

MATERIALS AND METHODS

This 5-year retrospective study was performed in Hospital Sultan Ismail, Johor Bharu from 2016 until 2020. The data of 278 patients with 516 cycles, who underwent COH with IUI cycles were reviewed. Patients were categorized into those with normal BMI and excessive BMI, for which the treatment outcome was compared, using a chi-square test. Variables such as female age, duration of subfertility, cause of subfertility, and types of subfertility (primary and secondary), were analysed using regression tests to determine their associations in successful cycles among excessive weight patients. A p-value of <0.05 was taken as a significant difference.

RESULTS

A total of 516 IUI cycles were included in the analysis. Thirthy-eight percent of the cycle were in the normal BMI women, while 62% from excessive BMI (overweight and obese). Eighty-five-point three percent were ethnic Malay, 6.0% were Chinese, 4.7% were Indians, and 4.0% were other ethnicities, reflecting the normal Malaysian population. The means and standard deviations for normal BMI and excessive BMI were 21.9 kg/m^2 and 29.1 kg/m^2 respectively. The mean age for all subjects was 31 years old. The mean duration of subfertility in normal BMI was 65 months (5.4 years) and in excessive BMI was 62 months (5.2 years). A total of 49 pregnancies (9.4%) were achieved over 516 cycles. Out of this, 33 (67.3%) are from the excessive BMI

group, and 16 (32.7%) are from the normal BMI group. There was no difference in the clinical pregnancy rate between normal BMI women and excessive BMI women women (adjusted OR 1.4, 95% CI 0.66 – 3.11, p-value 0.369). The difference in the ectopic pregnancy and miscarriage rates were also not significant. A negative correlation was found between female age and the success of COH IUI (p=0.815). Higher pregnancy rate was seen in secondary subfertility compared to primary subfertility (adjusted OR 2.8 (95% CI 0.97-7.92), p=0.058). The duration of infertility was found not significant in the outcome of COH IUI. In this result, all the causes of subfertility were negatively correlated with the successful COH IUI, possibly due to the small number involved.

CONCLUSION

In conclusion, our result contradicts the belief held by many that a higher BMI has a low success rate for COH IUI. The relationship between obesity and infertility outcomes is complex and can be influenced by multiple confounding factors. More studies and samples are necessary to evaluate the relationship between excessive BMI and successful of COH IUI.

CHAPTER 1: INTRODUCTION

Obesity is a global issue, and despite efforts to confront it, the worldwide incidence of obesity continues to escalate. The prevalence of obesity and overweight is increasing and has become an epidemic worldwide. Initially, its prevalence was relatively low among Asian populations, with 4% of Chinese and 0.5% of Indian women noted to be obese (Prentice AM,2006). However, in the last 20 years, its rate has tripled in developing countries. It was noted that 10% of children were overweight or obese.

The World Health Organization (WHO) has defined overweight and obesity as body mass index (BMI) ≥ 25kg/m^2 and more than 30 kg/m^2 and above, respectively. Obesity is associated with cardiovascular disease, diabetes, osteoarthritis, and malignancies such as colon and endometrial cancer. It is increasingly recognized that this current obesity epidemic has also contributed to fertility problems. The live birth rate decreases in obese women in both natural and assisted conceptions due to reduced pregnancy rates, increased miscarriage rates, and increased pregnancy complications (Zenyep Ozcan Dag and Berna Dilbaz,2015). However, many studies reported that the live birth rates were not affected by obesity (Rachel M. Whynott et al.,2021; Jennifer L. Eaton,2021; N. Yilmaz et al.,2009). Because of the conflicting data, we perform this study to evaluate the impact of BMI on the outcomes of Intrauterine Insemination (IUI) cycles in Hospital Sultan Ismail, Johor Bharu (HSIJB). We hypothesise that overweight and obese women would have a lower successful COH IUI outcome compared to women with normal BMI, and it may also affect the outcome of COH IUI cycles.

CHAPTER 2: LITERATURE REVIEW

One in every four couples in developing countries are affected by infertility. The burden remains high, as reported by a WHO study in 2012. From 1990 until 2010, the overall burden of infertility in women from 190 countries has remained high in levels and trends.

Infertility comprises primary and secondary infertility. Primary infertility defines a couple who has not become pregnant after at least one year of unprotected sexual intercourse. Secondary infertility refers to couples who have been pregnant previously, regardless of the outcome. Multiple studies have shown that couples with secondary infertility are more commonly associated with successful spontaneous pregnancy than with primary infertility.

Several studies have analysed the pregnancy predictors in IUI. Anabel et al (2020) review literatures to evaluate how patient and cycle specific factors affects IUI outcome specifically in clinical pregnancy rate, livebirth, spontaneous abortion rate and multiple pregnancy rate. The results including total motility sperm count >1 million associated with successful. In this analysis, advance maternal and paternal age has negatively impact pregnancy rates. Paternal obesity related to infertility and elevated maternal BMI increase in requirement of medication for COH without impacting pregnancy outcome. Furthermore, IUI mostly effective for women with ovulatory dysfunction and unexplained infertility but least effective for women with tubal factor and stage 3-4 endometriosis (Anabel et al.,2020). According to Philippe Merviel et al (2010), in the analysis of 1038 cycles in predictive factors for pregnancy after IUI, concluded that, couple with best probability of pregnancy are those in which women under the age of 30 years old (< 30 years old: 38.5% and >40 years old: 12.5%, P< 0.000001) and suffers from cervical infertility or anovulation (52.4% of ongoing

pregnancy). Apart from that, clinical pregnancy rate was 28.5% when total motility of sperm <5 million, compared to total motility sperm >5 million which was significantly higher, accounts for 44.3% (P <0.005) (Merviel, P.,2010).

2.1 Obesity and Reproductive functions

In the field of gynaecology and reproduction, obesity has been associated with menstrual disorder, hirsutism, infertility, miscarriage and obstetric complications. In addition, over a third to half of women with polycystic ovary syndrome (PCOS) are overweight or obese.

Several mechanisms are involved in the relationship between fertility and obesity. The insulin resistance and leptin levels are increased and hyperandrogenism occurs in obese women. Obesity may impair reproductive functions by affecting both the ovaries and endometrium (Bellver J,2007).

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Although many obese multiparous women can get pregnant despite their obesity, there is an increased prevalence of infertility in obese women. Vahrati and Smith

(2009), in their analysis, have found that a more significant portion of women who are seeking medical help to get pregnant are obese (Anjel Vahratian and Yolanda R. Smith,2009). A study by D C Gesink Law et al. (2007) among 7327 pregnant women demonstrated that the duration required to achieve a spontaneous pregnancy rate is increased, and pregnancy rates decreased in obese women, including regular ovulatory obese women (D C Gesink Law,2007). Obesity causes infertility through various pathways, including impaired ovarian follicular development, qualitative and quantitative development, and implantation.

Because of the obesity epidemic worldwide and its association with infertility, many overweight and obese women are treated using assisted reproductive techniques. Despite receiving advanced fertility treatment, the reproductive outcomes remained poor, especially in those with central obesity and PCOS.

The value of obesity as a predictor of infertility treatment is controversial. Lashen et al. (1999) stated that the extremes of body mass index do not adversely affect the outcome of IVF-embryo transfer treatment (H Lashen et al,1999). On the other hand, Dodson WC et al. (2006) concluded that obesity significantly affects treatment-related cycle fecundity (Dodson WC et al.,2006). A more extensive cohort study on 3,586 women receiving IVF, intracytoplasmic sperm injection, and gamete intrafallopian transfer, demonstrated a significant linear reduction in fecundity among obese women. Wang et al. in 2004 reported the same conclusion (Wang et al.,2004). In their earlier study involving 2,040 patients receiving IUI, Wang et al. (2000) reported that the fecundity rate increased among normal-weight to overweight and obese groups and even higher among the very obese group (Wang et al.,2000). They also stated that the possible mechanism is unclear and needs further exploration.

Ovarian function is dependent on weight. Obese women are more likely to have ovulatory dysfunction due to dysregulation of the hypothalamic-pituitary-ovarian axis. Rachel (2020) studied the effect of BMI on the IUI success rate. This study looked at 3,217 IUI treatment cycles in 1,306 patients. It showed that women with BMI 25 to 29.99 kg/m2 or ≥30kg/m2 were equal to having a live birth as women of normal BMI. Women with BMI ≥30 kg/m2 had a higher likelihood of biochemical pregnancy than those with normal BMI (Rachel M et al.,2021).

The endometrium may also affect by obesity. Bellver et al. investigated the potential role of the endometrium in the development of infertility in obesity in ovum donation cycles (Bellver et al.,2007). This study gives oocytes from healthy, young, and non-obese donors to recipients with different BMI. They found that the pregnancy rates per cycle initiated were significantly lower in obese women than in normal-weight women.

Souter I (2011) conducted a retrospective study examining the effect of BMI on patients undergoing COH with IUI. They concluded that gonadotrophins stimulation might overcome ovulatory dysfunction in obese, enabling them to have a success rate similar to normal weight patients (Souter I et al.,2011). Anabel et al. (2020) stated that maternal obesity increases the medication requirement for ovulation induction but does not affect the pregnancy outcome (Anabel et al.,2020).

There is evidence of an increased risk of miscarriage in obese women. Fedorcsk et al. (2000) noted that obesity was associated with an increased risk of early pregnancy loss before six weeks of gestation (Fedorcsk et al.,2000).

The poor productive performance in obese women, both in natural and assisted conception cycles, maybe a result of a combination of lower implantation and

pregnancy rates, higher preclinical and clinical miscarriage rates, and increased complications in pregnancy for both mother and foetus. These have been related to various endocrine and metabolic disturbances, such as effects on steroid metabolism and alterations in the secretion and action of insulin and other hormones, such as leptin, resistin, ghrelin, and adiponectin, which may affect follicle growth, corpus luteum function, early embryo development, trophoblast function, and endometrial receptivity.

2.2 Causes of infertility

The aetiology of infertility is a contributing factor that influences the IUI outcome (Y W Azantee,2011). Unexplained factors have a better prognosis in clinical pregnancy than other etiological factors (Cohlen B,2005). However, Costello MF et al. (2004) noted that moderate male factors and anovulation have the highest clinical pregnancy in IUI (Costello MF et al.,2004).

2.2.1 Ovulatory disorders

Ovulation disorders cause infertility in around 25% of couples with difficulty conceiving. The WHO categorizes ovulation disorders into three groups:

Group I ovulation disorders (hypogonadotropic hypogonadal anovulation) are caused by hypothalamic-pituitary failure. This category includes conditions such as hypothalamic amenorrhea and hypogonadotropic hypogonadism. Typically, women present with amenorrhoea (primary or secondary), characterized by low gonadotrophins and oestrogen deficiency. Approximately 10% of women with ovulation disorders have a group I ovulation disorder.

Group II ovulation disorders (normal-gonadotropic, normo-oestrogenic anovulation) are dysfunctions of the hypothalamic-pituitary-ovarian axis. This category includes conditions such as PCOS and hyperprolactinemic amenorrhoea. Around 85% of women with ovulation disorders have a group II ovulation disorder.

Group III ovulation disorders (hyper-gonadotropic, hypoestrogenic anovulation) are caused by ovarian failure. Around 5% of women with ovulation disorders have a group III ovulation disorder.

PCOS is an example of group ll ovulation disorder. It is the commonest endocrine disorder in women of reproductive age, occurring in approximately one in seven women. Of these women, approximately two-thirds will not ovulate regularly and consequently may seek treatment for ovulation induction. Approximately 5-7% of obese women with infertility and signs or symptoms associated with androgen excess, are affected by PCOS. PCOS is diagnosed by two out of three Rotterdam Criteria (2003), including hyperandrogenism (clinically or biochemically), ovulatory dysfunction (oligo/ anovulation) and polycystic ovaries (12 or more antral follicles measuring 2 to 9 mm in diameter or ovary that has a volume of greater than 10 mL on ultrasound) in ultrasound. Over a third to half of PCOS subjects are overweight or obese. Evidence shows that even normal-weight PCOS women present with the abdominal phenotype of fat distribution. It is increasingly recognized that in the majority of women with PCOS, insulin resistance and compensatory hyperinsulinemia play an essential role. Essentially, insulin resistance has a complex genetic and environmental aetiology. Insulin resistance appears to be selective with impaired glucose uptake, while other intracellular actions of insulin are preserved. At the ovarian level, insulin synergizes luteinizing hormone action by acting primarily through its receptor and IGF-1 receptor and stimulates ovarian steroidogenesis in thecal and granulosa cells. Mahnaz et al. (2013) reported that in 1348 IUI cycles, Group II

ovulation disorders have a higher clinical pregnancy rate of 13.8% (Mahnaz et al.,2013).

2.2.2 Endometriosis

Endometriosis is a benign gynaecological oestrogen-dependent inflammatory condition defined by the presence of endometrial-like tissue in extrauterine locations. It affects 10-15% of women of reproductive age. Endometriosis is classified using the American Fertility Society (AFS) system of Classification, and the management would depend on the stages of endometriosis.

The pathophysiology of endometriosis causing infertility, including dysmenorrhoea, resulting in excessive use of non-steroidal anti-inflammatory drugs (NSAIDs) as analgesia, might interfere with the implantation. The formation of endometrioma (accumulation of chocolate cysts in ovaries) and ectopic endometrium lining lead to poor oocyte quality and quantity. Furthermore, tubal adhesion or blockage causes external pelvic adhesion and disrupts cilial movement at the fallopian tubes. The hostile environment to sperms and oocytes results in an imbalance of the inflammatory mediators, subsequently preventing fertilization. Lastly, dyspareunia leads to less frequent and less effective sexual intercourse.

According to Matoras et al. (2002), medical treatment does not enhance pregnancy rates in minimal to mild endometriosis. Stimulated cycles slightly improved the outcome (Matoras et al.,2002). Surgical ablation does not improve the fertility rate. In moderate to severe endometriosis, a surgical approach by normalizing pelvic anatomic distortion and by adhesiolysis may enhance fertility may be beneficial.

2.2.3 Uterine factor

The uterus plays many significant roles in conception, implantation, and pregnancy. Uterine abnormalities can be either congenital or acquired. Uterine septum, bicornuate uterus, and uterine didelphys are common congenital uterine abnormalities. Acquired anomalies include leiomyomas, polyps, and Asherman syndrome. Leiomyomas account for 15% of couples with infertility and are the commonest benign tumours in the female reproductive tract. Even though their role in infertility is still questionable, evidence to date suggests that the anatomic location may be related to reproductive outcomes. Several possible mechanisms have been reported on how leiomyomas may affect fertility, such as anatomical distortion of the endometrial cavity, abnormal uterine contractility, reduced blood supply to the endometrium, and altered endometrial receptivity (Louise M Hafner,2015).

Decreased embryonic implantation potential and early pregnancy loss were both reported in the presence of endometrial polyps. They are associated with a decreased mid-secretory concentration of IGFBP-1, TNF-alpha, and osteopontin, as markers of implantation, which were shown to be reserved following surgical polypectomy.

2.2.4 Tubal factor

Tubal factor infertility (TFI) contributes to 30-35% of female infertility. It is caused by obstructions, damage, scarring, congenital malformations, or other factors which impede the descent of a fertilized or unfertilized ovum into the uterus through the Fallopian tubes and prevent a normal pregnancy and full-term birth.

The most common cause of TFI is occlusion of the fallopian tubes due to an infection by a sexually transmitted disease, namely *Chlamydia trachomatis* or *Neisseria*

gonorrhoea. Clinical observations and experimental data have indicated a role for antibodies against *Chlamydia trachomatis* proteins, such as the 60-kDa heat shock protein 60 (cHSP60), in the immunopathogenesis of TFI. When released from infected cells, cHp60 can induce proinflammatory immune responses that may functionally impair the FTs leading to fibrosis and luminal occlusion. Chlamydial pathogenesis of irreversible and permanent tubal damage results from innate and adaptive host immune responses to ongoing or repeated infections (Louise M Hafner,2015). The tubal factor is associated with the lowest clinical pregnancy rate after IUI. A study by Sahakyan et al. (1999) found that patients only conceived within the first two IUI cycles (Sahakyan et al.,1999). The authors suggest that these patients with tubal factor move onto IVF treatment after two unsuccessful cycles, given these low pregnancy rates.

2.2.5 Male factor

According to RCOG, male infertility affects 7% of all men; evaluating the male partner is essential (RCOG,2020). Male infertility is attributed to several causes, including anatomical (congenital or acquired), endocrine, iatrogenic, behavioural, lifestyle factors, and adverse environmental exposure. Causes of male infertility are classified into pre-testicular, testicular, and post-testicular.

Conventional semen analysis (SA) is essential in evaluating male fertility, and it remains the initial laboratory evaluation for infertile men. SA should be performed by an accredited andrology laboratory and to the standard described by WHO for the examination.

WHO reference limits (the lower fifth centile of the fertile population) describe minimal standards of adequacy for semen characteristics (Table 2.1).

RCOG suggested that a single SA is usually sufficient to determine the most appropriate management pathway. A repeat should be considered if the initial SA shows one or more abnormal parameters.

Table 2.1: WHO reference limits and 95% confidence intervals for semen analysis

Parameter	Reference limit	95% confidence interval
Semen volume (ml)	1.5	1.4-1.7
Sperm concentration (10^6/ml)	15.0	12-16
Total number (10^6/ejaculate)	39.0	33-46
Total motility (%)	40.0	38-42
Progressive motility (%)	32.0	31-34
Normal form (%)	4.0	3.0-4.0
Vitality (%)	58.0	55-63

Source from an update on the management of male infertility RCOG, 30 September 2020

2.3 Other associated factors for infertility

2.3.1 Age

According to Azantee et al. (2011), female age is one of the significant predictors of successful infertility treatment (Azantee et al.,2011). Age-related fertility problems increase after 35 years old and dramatically after 40 years. For these patients, an earlier fertility evaluation is made six months after trying (American Society for Reproductive Medicine, 2012). Female fecundity declines with age, reflecting, in part, the progressive loss of oocytes that occurs with age. Aging oocytes also accumulate meiotic defects and DNA damage, causing deterioration of gamete quality and increased risk of aneuploidy embryos and miscarriage (Crawford NM, Steirer AZ,2015).

Kimberly Liu et al. (2011) emphasized that women need to be informed the risk of spontaneous pregnancy loss and chromosomal abnormalities increases with age. They should be counseled about and offered appropriate prenatal screening once pregnancy is established (Kimberly Liu et al.,2011).

Macizo Soria et al. (1999), in their analysis of 3,012 cycles from 1,201 couples, evaluated the impact of maternal age on IUI outcomes after ovulation induction with gonadotrophin or clomiphene citrate. They demonstrated diminished pregnancy rates in ≥40 years old, with rates of 4.1% to 7% per cycle compared to 13.7% to 17% in women <40 years old (Macizo S et al.,1999).

2.3.2 Duration of infertility

According to Hakan E. Duran et al. (2002), the duration of infertility becomes one of the predictors of the success of infertility treatment apart from female age (Hakan E. Duran et al.,2000). However, there are differences in the outcome of whether the duration of infertility successfully affects the outcome of IUI. Nuoja-Hutturen et al. (1999) studied 811 IUI cycles with clomiphene citrate/hMG and concluded a significant decrease in pregnancy rate with increasing duration of infertility. A more extensive population study by Ashrafi et al. (2013) on 1,348 IUI cycles arrived at the same conclusion (Ashrafi et al.,2013). Conversely, studies were done by Zainul et al. (2006) and Tay et al. (2007) did not find to have any significant association with the duration of infertility (Zainul et al.,2006; Tay et al.,2007).

2.3.3 Smoking

Cigarette smoke contains many substances, including nicotine, carbon monoxide, and recognized carcinogens and mutagens such as radioactive polonium,

benzopyrene, dimethyl benzantracene, naphthalene, and methylnaphtelene (Tulay Irez et al.,2013). Smoking has a detrimental effect on various parameters of semen analysis. A cross-sectional analysis of 254 healthy men from 1987 to 2004 by Ramlau-Hansen et al. found that on semen analysis, cigarette smokers had lower semen volumes, sperm counts, and percentage of motile sperm compared to men who did not smoke (Ramlau-Hansen et al.,2007). Jacob et al. (2009) evaluated the influence of smoking on the outcome of COH and IUI in subfertile couples. The study involved 885 patients, comparing COH IUI outcomes on 679 non-smokers with 206 smokers. They concluded that smokers who underwent COH with IUI required significantly more gonadotrophins ampoules than non-smokers to achieve a comparable pregnancy rate (Jacob et al.,2009). Furthermore, smokers gained a thinner endometrium of the day hCG administration than a non-smoker. However, this study did not state on primary or secondary smoker effect in the selected patients. Tulay's (2013) study investigated the effect of cigarette smoking on IUI outcome in men with normal sperm concentration, and the outcome is higher progressive sperm motility, chromatin condensation, and pregnancy ratio have been found in non-smokers (Tulay et al.,2013).

2.4 COH with IUI

Clomiphene Citrate (CC) is a selective oestrogen receptor modulator with both oestrogenic and anti-oestrogenic properties. It was first approved for use in women with anovulation in 1967 and has been used as a first-line ovulation induction agent for over 40 years. Acting as an antioestrogen, CC competitively inhibits the binding of oestradiol to its receptors in the hypothalamus and pituitary, which in turn blocks the negative feedback effect on endogenous oestrogen, including oestradiol. This results in an increased secretion of pulsatile gonadotrophin-releasing hormone (GnRH) from the hypothalamus, leading to an increase in follicle-stimulating hormone (FSH) and

luteinising hormone (LH) production and secretion from the pituitary gland. This increase in FSH secretion stimulates follicular growth and oestradiol production, thereby inducing a mid-cycle LH surge and subsequent ovulation.

Kousta et al. (1997) reported that CC had an ovulation rate of 60-85% and a pregnancy rate of 30-50% after six ovulatory cycles (Kousta et al.,1997). Hart and Norman (2006) commended that this apparent discrepancy between good ovulation and lower pregnancy rates has been attributed to the anti-oestrogenic effect of CC on the endometrium and cervical mucous (Hart and Norman,2006).

The risk of multiple pregnancies in case of pregnancy was 6% higher when two follicles were present than monofollicular growth. Multifollicular growth is associated with increased pregnancy rates in IUI with COH. According to Rumste et al. (2008), since in cycles with three or four follicles, the multiple pregnancy rate increased without a substantial gain in overall pregnancy rate, IUI with COH should not aim for more than two follicles (Rumste et al.,2008).

IUI is often suggested for infertile couples in which the women have at least one patent fallopian tube, and the partner has partially modified sperm or low sperm quality. Other indications for IUI include unexplained infertility, cervical factors, ovulatory dysfunction, and stage I/II endometriosis. Patients with ovarian failure, significant male factor infertility, significant tubal adhesions, tubal dysfunction, and significant uterine abnormality contraindicate IUI.

2.4.1 Ovarian stimulation and follicle rupture timing

In HSIJB, most female patients in this hospital were stimulated using CC alone, ranging from 50mg to 150mg OD (from day 2 to day 6 of menses) are used. A serial

transvaginal scan (TVS) was performed by general specialist and medical officers, to monitor the growth of the follicles. hCG was administrated to the patient when the size of follicles reached ≥18mm. IUI was performed 36-40 hours post hCG injection.

2.4.2 Sperm preparation and IUI

A semen sample from the husband was collected by masturbation following an abstinence period of 3 days and left to liquefy for less than 1 hour. Pure sperm media was used for sperm preparation. In the gradient method, 45% and 90% solutions were prepared and then topped layered with the semen sample. The sample was centrifuged at 1200rpm for over 20 minutes, leaving the pallet to sink to the bottom, and the supernatant was discarded. The pellet was then washed with 2ml of media wash and centrifuged again at 2000rpm for 10 minutes.

IUI was carried out using a soft IUI catheter, with the patient in dorsal position. The procedure was done in a sterile environment. Two weeks after the IUI procedure, if the patient remained amenorrhoeic, a urine pregnancy test was done, and if it was positive, a TVS was performed to observe for gestational sac.

CHAPTER 3: STUDY JUSTIFICATION AND RESEARCH QUESTIONS

Overweight and obese women seeking to conceive naturally experience a longer time to conceive, with higher rates of infertility and miscarriage and lower pregnancy rates. However, the impact of high-BMI women on the outcome of fertility treatments is debatable.

This study was done to evaluate the impact of BMI on COH IUI outcomes. In HSIJB, there is an increasing trend of overweight and obese patients seeking infertility treatment. This group often requires a higher dose of COH and cycle cancelation. Hence, it is essential to understand the impact of high BMI on various infertility interventions to counsel better patients considering COH IUI.

In HSIJB, most of the stimulation induction agents are CC ranging from 50mg to 150mg OD (from day 2 to day 6 of menses), and only small numbers use other types. For that reason, only COH IUI using CC as the stimulating agent was recruited.

3.1 Research Questions

1. What is the relationship of excessive BMI towards the outcome of COH IUI?
2. What is the proportion of different classes of BMI patients receiving COH IUI in HSIJB?
3. What are the differences clinical pregnancy rate of COH IUI between the two groups of BMI (normal BMI vs excessive BMI)?
4. What are the associated factors (age, duration of infertility, type of infertility, causes) that affect the outcome of COH IUI among excessive BMI patients?

CHAPTER 4: STUDY OBJECTIVES

4.1 General objective

To study the effect of BMI on COH IUI outcome in HSIJB.

4.2 Specific objectives

1. To determine the proportion of different classes of BMI patients receiving COH IUI in HSIJB.
2. To compare the clinical pregnancy rate of COH IUI between the two groups of BMI (normal BMI vs excessive BMI).
3. To determine the associated factors (age, duration of infertility, type of infertility, causes) that affect the outcome of COH IUI among excessive BMI patients.

CHAPTER 5: METHODOLOGY

5.1 Study design, venue, and duration

5.1.1 Study design

This study was a cross-sectional study based on retrospective data. All cases of COH IUI from 2016 until 2020 were obtained from the medical record system.

5.1.2 Study period

This study was carried out over one year, from 1 March 2021 until 31 January 2022. The 5- years data, from 1 January 2016 until 31 December 2020, were collected from the Record Office of HSIJB.

5.1.3 Study location

This study was conducted in the Department of Obstetrics and Gynaecology Clinic, HSIJB.

5.2 Study population

5.2.1 Reference population

All infertility patients who received COH IUI cycles in HSI, and who fulfilled the inclusion and exclusion criteria, were included in this study.

5.2.2 Source population

All infertility patients who received COH IUI cycles.

5.2.3 Study participants

All infertility patients who received COH IUI using Clomiphene Citrate ranging from 50mg to 150mg OD (from day 2 to day 6 of menses).

5.3 Sample size calculation

The sample size for this study was calculated according to the study objectives, as shown in Table 5.1 below.

Table 5.1: Sample size calculation

Specific objective	Formula	Sample size calculation
Specific objective (1): To determine the proportion of different classes of BMI patients receiving COH+IUI in HSIJB	Single population proportion: $X = Z_{\alpha/2}^2 * \frac{p*(1-p)}{d^2}$ $Z_{\alpha/2}^2$: 1.96	**Normal BMI** (Veronique Viardot et al, 2014): P=0.093 d^2=0.05 Sample size: 130 Add 20% drop out; sample size= **163** **Excessive BMI** (Veronique Viardot et al, 2014): P=0.907 d^2=0.05 Sample size: 130 Add 20% dropout; sample size= **163** Final sample size: **163**

Specific objective number 2: To compare the clinical pregnancy rate of COH IUI between the two groups of BMI; normal BMI vs excessive BMI)	Two proportion formula was used: Reference: (Huijuan Gan, 2021) $n = (Z_{\alpha/2}+Z_{\beta})^2 * (p_1(1-p_1) + p_2(1-p_2)) / (p_1-p_2)^2$ p_1: clinical pregnancy rate in normal BMI undergoing COH IUI (0.31) p_0: clinical pregnancy rate in excessive BMI undergoing COH IUI (0.143) $Z_{\alpha/2:}$ 1.96, type 1 error at 5% (2-sided) $Z_{\beta:}$ 0.84, type 2 error at 20% (power 80%) Drop out: 20%	Sample size: 123 per arm Total sample size= **246** <u>Final sample size:</u> **<u>246</u>**
Specific objective (3): To determine the associated factors that affect (age, duration of infertility, type of infertility, causes) the outcome of COH IUI among excessive BMI patients	Two proportion formula was used: $n = (Z_{\alpha/2}+Z_{\beta})^2 * (p_1(1-p_1) + p_2(1-p_2)) / (p_1-p_2)^2$ p_0: successful clinical pregnancy rate (with associated factors) in excessive BMI undergoing COH IUI p_1: non successful clinical pregnancy rate (with associated factors) in excessive BMI undergoing COH IUI	**Age** (Reference: Arzu Yavuz et al, 2013) P_0: 0.07 P_1: 0.22 Sample size: 86 Add 20% drop out; sample size= 108 **Duration of infertility** (Reference: Arzu Yavuz et al, 2013) p_1: 0.06 p_0: 0.21 Sample size: 81

	$Z_{\alpha/2}$: 1.96, type 1 error at 5% (2-sided) Z_{β}: 0.84, type 2 error at 20% (power 80%) Drop out: 20%	Add 20% drop out; sample size= 102 **Types of infertility** **-primary** (Reference: Arzu Yavuz et al, 2013) p_1: 0.04 p_0: 0.19 Sample size: 70 Add 20% drop out; sample size= 88 **Smoking** (Reference: Huijuan et al, 2021) p_1: 0.03 p_0: 0.18 Sample size: 65 Add 20% drop out; sample size= 82 Largest sample will be taken: 108 per arm Sample size for 2 arms: 216 Final sample size: **216**

The largest calculated sample size was 246 per arm. Therefore, the minimum total sample size taken for this study was 492.

5.4 Ethical approval

Ethical approval obtained from the Medical Research Ethic Committee Ministry of Health Malaysia dated 10th May 2021 (NMRR initial approval: NMRR-21-645-59078 (IIR)) (Appendix 2) and the Human Medical Research and Ethics Committee of USM dated 5th August 2021 (Study protocol code: USM/JEPeM /21040308) (Appendix 3).

5.5 Inclusion and Exclusion criteria

5.5.1 Inclusion criteria

1. Couple who tried to conceive with unprotected intercourse for at least 12 months (female age < 35 years) and 6 months (female age 35 years and above)
2. Couple who had undergone COH IUI cycle in HSIJB, stimulated with clomiphene citrate ranging from 50mg to 150mg OD (from day 2 to day 6 of menses)

5.5.2 Exclusion criteria

1. Couple who underwent COH monitoring in other hospitals
2. Severe abnormality in sperm parameters (low count <5 million, asthenospermia, severe teratospermia and aazospemia)
3. Had intracervical insemination rather than intrauterine insemination
4. Multiple intrauterine insemination failure >4 cycles

5.6 Research tool

Data was collected using the Data collection sheet as shown in Appendix 1.

5.7 Data collection method

Before starting the research, ethical clearance was obtained from the National Medical Research Centre (NMRC) and the Human Research Ethical Committee of USM (JEPeM). After obtaining permission to conduct the study from the Director of HSIJB, the list of infertility who received COH IUI from the Record Office was recorded.

One week prior to or during stimulation, the patient's BMI, together with detail of the patient, was taken and documented in the data collection form. The data were analysed and processed to obtain results.

5.8 Statistical analysis

To analyse the data, IBM SPSS Statistics Version 24.0 was used. Selected variables were analysed using descriptive statistics. The categorical data were presented as frequencies and percentages, means, and standard deviations for the numerical data if they were normally distributed or as medians and interquartile ranges if they were not.

The Chi-square test was used to compare the clinical pregnancy rates between the two studied groups. The p-value of <0.05 was taken as the significant difference. Regression logistic tests were used to determine the associated factors for successful COH IUI among excessive BMI patients.

5.9 Definition of operational term

1. Infertility: A disease of the reproductive system defined by the failure to achieve a clinical pregnancy after 12 months or more of regular unprotected sexual intercourse.

2. Controlled ovarian hyperstimulation: Technique used in assisted reproduction involving the use of fertility medication to induce ovulation by multiple ovarian follicles.
3. Obesity and overweight: Body mass index >30 kg/m^2 and 25.0 to 29.9 kg/m^2 respectively
4. Intrauterine insemination: Artificial reproductive technique whereby a placement of sperm that have been washed of seminal fluid directly into the uterus bypass the cervix.
5. Duration of subfertility: The duration that any form of reduce fertility with prolonged time of unwanted non-conception.
6. Primary infertility: When a woman is unable to ever bear a child, either due to the inability to become pregnant or the inability to carry a pregnancy to a live birth.
7. Secondary Infertility: When a woman is unable to ever bear a child, either due to the inability to become pregnant or the inability to carry a pregnancy to a live birth following either a previous pregnancy or a previous ability to carry a pregnancy to a live birth.
8. Number of stimulated follicles: Number of stimulated follicles in the ovary after COH to ensure that multiple oocytes are available within a given IUI cycle.
9. Endometrial thickness: The measurement of the thickest echogenic area from one basal endometrium interface across the endometrial canal to the other basal surface by ultrasound.
10. Pregnancy confirmation: Defined as positive urine or serum beta hCG test.
11. Fecundity: Probability of achieving at least one pregnancy throughout the course of IUI treatment, which usually consists of 2-4 insemination cycles.
12. Clinical pregnancy: A pregnancy that is confirmed by both a high level of hCG and ultrasound confirmation of a gestational sac or heartbeat/ fetal heartbeat.
13. Clinical pregnancy Rate: Presence of a fetal heartbeat at 6-7 weeks of pregnancy