DISSERTATION IN PARTIAL FULFILMENT OF MASTER OF MEDICINE IN OBSTETRICS AND GYNECOLOGY

UNIVERSITY OF NAIROBI

SUBMITTED BY : DR. FAUZIA BUTT

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OUTCOMES OF CONTROLLED OVARIAN HYPERSTIMULATION FOR ASSISTED REPRODUCTION

DECLARATION

This is to declare that this research is my original work and that it was done with the guidance of my supervisors.

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CERTIFICATION OF SUPERVISION:

This is to certify that this research was developed under my guidance

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Date 8/1/2010

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CERTIFICATE OF AUTHENTICITY.

This is to certify that this dissertation is the original work of Dr. Fauzia Butt, Mmed student registration number H58/8271/06 in the department of Obstetrics and Gynecology , University of Nairobi 2006 – 2010.

This research was carried out in the department of Obstetrics and Gynecology , Scool of Medicine , College of health Sciences. It has not been presented in any other university for award of a degree.

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Last but not the least, I would like to thank all those special people who may not have been mentioned but they were a motivation towards my work.

DEDICATION

I dedicate this work to all those couples who suffer from infertility and I hope that this piece of work will be valuable to all those clinicians practicing assisted reproduction in the third world countries.

TABLE OF CONTENTS

LIST OF ABBREVIATIONS:

ART – Assisted Reproductive Technology

AFC – Antral Follicle Count.

BMI – Body Mass Index.

COS – Controlled Ovarian Stimulation.

COH – Controlled Ovarian Hyperstimulation.

FSH – Follicle Stimulating Hormone.

GnRH –a – Gonadotrophin Releasing Hormone Agonist

hCG – Human Chorionic Gonadotrophin.

hMG – Human Menopausal Gonadotrophin.

IVF – In Vitro Fertilisation.

LH – lutenising Hormone.

ABSTRACT

Background

Infertility affects more than 80 million people worldwide with rates varying from less than 5% to more than 30% (1). The success of IVF treatment depends on adequate follicle recruitment which is achieved through controlled ovarian hyper stimulation. Overall, the incidence of poor response to assisted reproductive treatment is estimated to be 9 – 24 % (2). However the definition of poor response has been inconsistent using total number of oocytes retrieved, different numbers of M 2 oocytes ranging from <3 to 6 or minimal cumulative dose of gonadotrophins to define ovarian response (3,4).

Objective

To describe the outcomes of controlled ovarian hyperstimulation and factors associated with ovarian response.

Study design

A descriptive observational study.

Method

This was a descriptive observational study carried out at the IVF unit of The Aga khan university hospital from April 2008 to July 2009. Patients underwent controlled ovarian hyperstimulation using the GnRH agonist or GnRH antagonist protocol followed by transvaginal ultrasound guided aspiration of oocytes under sedation and oocyte quality was then determined by an embryologist. After obtaining ethical approval, recruitment of study participants who met the inclusion criteria was done. Data was obtained from patient records using a data collection form.

Data analysis

Data was analysed using SPSS version 15.0 and stata version 10 (stataCorp, texas)

Results

A total of 62 participants were studied, the mean age was 33.5 with a median of 34 years. Fifty one (79.7%) patients underwent GnRH agonist stimulation, the rest GnRH antagonist stimulation, whereby the mean dosage of exogenous gonadotrophins was 3012.8 i.u.

The mean number of oocytes retrieved was 10.4 with a median of 9, and good quality (M 2 quality) oocyte recovery rate was 59.6%. The mean oocyte count in women < 30 years was 15.3 as compared to 9.4 in women > 30 years which was significant with a p- value = 0.007.

The mean dosage requirement in women < 30 years was 2337.5 i.u and in those > 30 years, it was 3187.20 i.u .

Poor responders (< 6 oocytes) were older (mean age 35.21 vs. 31.19 years P=0.001) and received higher doses of exogenous gonadotrophins (mean dose 3559.84 i.u vs. 2391.35 i.u P<0.001) as compared to the normoresponders.

Conclusion

In this study we found that, outcomes of assisted reproduction in terms of quantity and quality of oocytes, as well as total exogenous gonadotrophins requirement varied with age of the female partner. From our findings, age plays a significant role in the eventual outcomes of controlled ovarian hyperstimulation. Much as controlled ovarian hyperstimulation may bypass the natural follicle recruitment , it is still limited to a certain extent by the quality of oocytes which are age dependant.

Recommendations

Work up for infertility should not be delayed for women in their thirties, as increasing age decreases the prognosis of assisted reproduction. Women should also be advised against delayed child bearing as the chances of success of assisted reproduction are also limited with increasing age .

INTRODUCTION

It is commonly accepted that infertility affects more than 80 million people worldwide. In general, one in ten couples experiences primary or secondary infertility, but infertility rates vary among countries from less than 5% to more than 30%. Most of those who suffer from infertility , live in developing countries , where infertility services in general and ART in particular are not available (1)

Ovarian stimulation is applied in a clinic to restore mono-ovulatory cycles in anovulatory women (ovulation-induction) or to induce the development of multiple dominant follicles for assisted reproduction. Assisted reproduction is the collective name for treatments designed to lead to conception by means other than sexual intercourse. It includes intrauterine insemination, in-vitro fertilization, intracytoplasmic sperm injection and donor insemination.

Despite improvements in ovarian stimulation protocols during the last two decades there are still women who fail to respond adequately to gonadotrophin stimulation (Tarlatzis 2003) and are classified under the broad term 'poor responders'. This study will describe the outcomes of patients undergoing controlled ovarian hyperstimulation.

One million oocytes are known to be present at birth, decreasing to between 300,000 and 500,000 at menarche. At all stages of the menstrual cycle a number of small antral follicles are recruited to undergo follicular maturation. The initial growth of follicles is influenced by FSH and maturation of the dominant follicle is influenced by LH in a complex interplay with hypothalamic pituitary axis and this hormonal interaction is utilized in artificial recruitment of multiple follicles in a controlled manner for the purpose of assisted reproduction.

Oocyte quality is established early during fetal life. The first produced oocytes (less susceptible to non-disjunction) ovulate first and 'poorer' oocytes ovulate later. There is also evidence to suggest that there is age dependent damage in oocytes due to gradual increase in intracellular oxidative stress that also leads to increased frequency of non disjunction. It is well documented that aneuploidy due to non-disjunction is related to increased miscarriage rate and reduced fecundity.

LITERATURE REVIEW

There are some factors that have been implicated as predictors of fertility, such as age of the female partner which has traditionally been considered the single most important determinant of reproductive success. Fecundability begins to decline in the early 30s and accelerates during the late 30s and early 40s. This decline in fertility is most probably due to diminishing quantity and quality of follicles in the ovary (4). It has been shown that the number of follicles leaving the pool of resting follicles to enter the growth phase towards the antral stages of development decreases with increasing age (5). Earlier epidemiological studies have also suggested that

there is a progressive reduction in the chance of conception with increasing duration of infertility, both in spontaneous and treatment related conceptions (Hull et al., 1985 Templeton et al ., 1996)(6) . Hull et al demonstrated that in an infertility clinic the chance of conception depends on the cause of infertility. They found conception rates of 96% after two years of amenorrhea, 72% for unexplained infertility; 11% for oligospermia; 27% in the context of abnormal postcoital test; 19% where tubal damage was present.

Various lifestyle factors have also been identified as affecting fertility. A recent review by Homan et al., 2007 concluded that there is definite evidence that smoking and obesity adversely affect fertility. Smoking has been shown to be associated with a two fold increased incidence of infertility (OR 1.6; 95% CI 1.34 to 1.91) Augwood et al., 1998.
Overweight status (BMI >25) and obesity are both associated with reduced chances of spontaneous or treatment – related conceptions. Time to conception doubles in overweight or obese women (RR 2.2; 95% CI 1.6 to 3.2 Homan et al., 2007). women with a BMI > 25 have a lower chance of pregnancy following IVF (OR 0.71; 95% CI 0.62 to 0.81) and require higher doses of gonadotrophins, but there is no evidence that these tests are useful in predicting outcomes of relevance to infertile couples (Broekmans et al 2006) (6).

The success of IVF treatment depends on adequate follicle recruitment. Controlled ovarian hyperstimulation is gonadotrophin induced stimulation of the ovaries for purposes of IVF treatment which specifically aims at inducing on going multiple follicle development rather than a single dominant follicle in ovulatory women (7). Protocols for controlled ovarian stimulation are based on this. Although Louise brown in 1978, was born following IVF – ET in a natural cycle , it soon became clear that the pregnancy rate was greatly improved if more

than one embryo was replaced in the uterus (8). However, the simultaneous risks of ovarian hyperstimulation syndrome and multiple pregnancies have led to the adoption of a compromise between pregnancy rates and multiple follicular development and restriction in the number of embryos transferred.

The definition of poor response to COS in IVF treatment varies widely based on one or a combination of some of the following factors (2) :

1. Variable numbers of mature follicles observed on ultrasound (ranging from less than 2 to less than 5).
2. Maximal Estradiol levels during COS (100 – 660 pg/ml).
3. Number of mature oocytes retrieved (less than 3 to less than 6). Minimal cumulative dose of gonadotrophins.
4. Days to gonadotrophins stimulation required in combination with factors known to be associated with poor response such as age > 40 years, early follicular phase FSH levels (6.5 – 15 miu/ml) (9).

Gonadotrophins for ovarian stimulation initially were extracted from human pituitary glands and urine, now they are produced from transformed cell lines. FSH, LH and hCG are now marketed as recombinant products. With the introduction of IVF and other ART procedures, the principles of ovarian stimulation changed and maximum yield of oocytes available for IVF was the aim, thus exogenous FSH was administered in larger amounts than before. From the mid – 1980s , the use of GnRH – analogues was introduced to achieve pituitary down regulation before stimulation with exogenous gonadotrophins (10).

However, the profound pituitary suppression with GnRH agonists made luteal support necessary since the output of LH remains blocked for at least ten days after cessation of the agonist (Broekmans 1992; Smitz 1988). Follicular recruitment however seemed unaffected and adequate follicle growth could be achieved even with pure FSH preparations lacking LH, like recombinant FSH. (Chapel 1991; Daya 2002, Out 1996).

The presumed redundancy of LH for ovarian hyperstimulation and the wish for more purified products drove the pharmaceutical industry to the conversion from hMG to r FSH which is completely devoid of LH. Studies have shown that GnRH antagonists can cause endogenous LH suppression to such an extent that follicle growth and pregnancy rates were adversely affected (Ganirelix study 1998). As a consequence the role of LH in ovarian stimulation became again a matter of debate (levy 2000)(11).

GnRH – agonists were introduced in ovarian stimulation for IVF to suppress premature surge of LH. They bind to pituitary receptors in the hypophysis and initially induce release of large amounts of FSH and LH (flare up effect) and an increase in the number of GnRH receptors (up regulation). However, prolonged use of the agonist is followed by a decrease in GnRH receptors (down regulation) . As a result, the pituitary becomes refractory to stimulation by GnRH leading to a decrease in circulating gonadotrophins (12).

Once the endogenous gonadotrophins have been suppressed, controlled ovarian hyperstimulation can then be achieved by using exogenous gonadotrophins under ultrasound monitoring of growth of ovarian follicles.

There are two protocols for using GnRH agonists :

- The long protocol - with this regimen , both pituitary and ovarian desensitization are induced by GnRH – agonist administration in the mid-luteal phase of the cycle preceding the planned IVF . Once densistization is obtained , ovarian stimulation with gonadotrophins is started and GnRH-a injections are continued until hCG is administered , 36 hours after which oocyte retrieval is done.
- The short protocol – This regimen takes advantage of the initial flare up of the serum gonadotrophins on follicular recruitment and of subsequent pituitary desensitization induced by daily agonist administration . Gonadotrophin administration is started in the early follicular phase (8).

Between August 1994 and March 1998, couples undergoing treatment with assisted reproduction technology at three Boston clinics were studied. Two of the clinics used Gnrh-agonist in a long protocol and one clinic used the short protocol. The short protocol involved use of leuprolide acetate 1mg subcutaneously from day 1 of the menstrual cycle, daily until administration of hCG. In the long protocol, leuprolide was administered from day 21 of the prior cycle.

It was found that, women who used the short protocol tended to have fewer follicles, (7.9 versus 9.6 p= 0.001) , and fewer oocytes retrieved (mean 8.7 versus 9.8 p= 0.006) , however their exogenous gonadotrophins requirement was approximately 3.5 ampoules less than the longer protocol. Using multiple regression analysis, age, year of treatment and use of short regimen were all signs predicting success , however with the addition of number of oocytes to

the model, use of the short protocol was no longer significant. This suggests that, the effect of the short protocol on the success of assisted reproduction technology is mediated largely by the lower number of oocytes retrieved (13).

A Cochrane intervention review was conducted to select randomized controlled trials comparing controlled ovarian hyperstimulation using r-FSH alone versus r-FSH with r-LH in IVF/ICSI . Comparison of the effect of co-administration of both gonadotrophins versus use of r-FSH alone was done in 11 trials. It was found that the amount of r-FSH was significantly lower in the combined group compared to r-FSH alone. (seven trials WMD – 192, 95% CI 244 TO 140). Eight trials reported on the amount of oocytes retrieved and significantly less oocytes were retrieved in the combination group .(7 trials WMD - 0.9 95% CI -1.07 TO -0.72). However, using the random effect model there was no difference in oocyte number in either of the protocols.

Although in this review no significant differences were found in clinical and on going pregnancy rates, it was pointed out that all pooled estimates indicate a beneficial effect of co-treatment of r-LH (Ferraretti 2004, De placido 2005, Barrasetxea 2006) . In conclusion, there was no evidence that co-administration of both gonadotrophins in GnRH agonist down regulated protocols women results in more live births (11).

A retrospective study was conducted in Bristol, involving women aged < 40 years who were undergoing their first IVF / ICSI cycles and were exposed to the long GnRH agonist protocol. All patients were banded into response groups according to their total FSH requirements.

In those patients whose ovarian response resulted in oocyte retrieval with total FSH requirement less than 3000 iu of r-FSH , pregnancy rates were found to be favourable (32.4 %) irrespective of the number of oocytes collected. Those patients in whom the r-FSH requirement exceeded 3000 iu, they had a significant overall reduction in pregnancy rate (21.9 % $p < 0.0005$). More so , it was seen that , the number of oocytes retrieved was not of significant importance when the total r- FSH requirement was less than 3000 iu, but when the requirement exceeded 3000 iu, then the pregnancy rates would decline if the oocytes retrieved were less than 4.

It was also seen that if the first cycle was cancelled at daily r- FSH requirement of less than 300 iu, then the chance of a subsequent cycle resulting in a pregnancy was good after the dosage was increased. However cancellation at dosage > 300 iu daily resulted in higher cancellation rates in the subsequent cycle (24.4% vs. 6.4% and 6.7% vs. 22.0%). These findings suggest that inadequate follicular development in response to 300 iu r-FSH daily identifies women whose reproductive performance is limited by the capacity of the ovaries to respond. Conversely an initial response to < 300 iu is unlikely to be detrimental and simply increasing the stimulation dose to 300 iu will result in a satisfactory response in most cases. The author also cautions against daily use of stimulation doses > 300 iu as cumulative pregnancy rate is <5 % despite more follicles being recruited. This gonadotropin requirement also appears to identify qualitative differences in oocytes produced(14).

A randomized controlled trial,in which 270 patients were recruited, was performed in those patients who had one or more previous failed IVF cycles in which 5 or less oocytes were retrieved despite using >300 iu of gonadotrophins / day. Patients were randomized to receive either Gnrh-agonist or Gnrh antagonist protocol.

Patients in the agonist group received 0.05mg of triprorelin daily from cycle day 2 until and including the day of hCG administration, whereas patients in the antagonist group received 0.25mg of Ganirelix , initiated in the presence of a follicle whose mean diameter was 14mm at ultrasound. Both groups then received 400 iu of gonadotrophins .

A significantly higher on going pregnancy rate was observed with the use of the flexible antagonist protocol (12.2% versus 4.4 % $p = 0.048$). The total units of FSH administered during stimulation as well as the number of oocytes retrieved were similar in the two groups. It was not clear as to what was the source of the difference in pregnancy rates but it may be related to the fact that the flexible antagonist protocol creates a hormonal environment which resembles the natural cycle more closely avoiding the high levels of LH in the early follicular phase caused by the agonist protocol (15).

RATIONALE

Assisted reproduction is a rapidly changing field and its success rates vary widely. A lot of research is on going especially in the developed countries which then create protocols and standards of management but suited to their population.

Kenya is one of the countries where assisted reproduction technology is in its infancy, and a lot of research is required to find out about the need in the local population, patient characteristics and how best assisted reproduction can help improve fertility rates by creating protocols and standards of practice tailored to our population.

This descriptive observational study aims to determine local population characteristics and outcomes of the various stimulation protocols used. This information can be compared with similar research conducted elsewhere in order to improve management of subsequent patients.

RESEARCH QUESTION

What is the outcome of controlled ovarian hyperstimulation in patients requiring assisted reproduction.

BROAD OBJECTIVE

Describe the outcomes of controlled ovarian hyperstimulation and the factors associated with poor response.

SPECIFIC OBJECTIVES

Among patients undergoing ART in AKU-H:

1. Describe the characteristics of patients undergoing controlled ovarian hyperstimulation for assisted reproduction
2. Describe the outcomes of controlled ovarian hyperstimulation
3. Compare patients with favourable outcomes with those with unfavourable outcomes. Patients with favourable outcomes are those with more than 6 oocytes at retrieval after controlled ovarian hyperstimulation, with unfavourable outcomes as less than 6 oocytes.

METHODOLOGY :

Study Area

The study was conducted at the IVF centre of The Aga Khan university teaching hospital situated in Nairobi. It is the main teaching hospital for the college of health sciences Aga Khan University and it caters for patients from within Nairobi and its environs. This unit caters for patients recruited from within the hospital or from other hospitals. Numbers ?

Study design

A descriptive observational study in which patients undergoing controlled ovarian hyperstimulation at the IVF unit of Aga khan hospital for assisted reproduction were recruited between April 2008 to July 2009.

Study population

Study population was drawn from women with primary or secondary infertility, requiring assisted reproductive technology procedures undergoing controlled ovarian hyperstimulation. At the IVF unit, all patients had the following lab investigations done as a routine and not a pre requisite for this study : Glucose tolerance test, Thyroid function tests , lipid profile, Serum testosterone levels, and Serum Fsh and Estradiol levels during the course of ovarian stimulation, semen analysis for all male partners . Radiological procedures were : pelvic ultrasound , Hysterosalpingogram , diagnostic laparoscopy where radiological findings were inconclusive.

Sample size

In order to describe a hypothesized proportion of poor response to ART of 24% plus or minus 5% with 95% confidence, the sample size was calculated as shown below :

$$n = \frac{Z^2_{1-\alpha/2} P_1 (1-P_1)}{\delta^2}$$

Where
n = sample size required
P_1= hypothesized proportion of poor response to ART (24%)
$Z_{1-\alpha/2}$ =Normal deviate corresponding to a 95% confidence interval in a two tailed test (=1.96)
δ =error margin of 5%

$$n = \frac{(1.96)^2 \text{ X } 0.24(1-0.24)}{(0.05)^2}$$

n = 280.28 participants

Since the estimated sample size is more that 5% of the finite population of 80 women who visited the AKU-H infertility clinic for a period of one year the sample size was corrected using the formula below;

n finite population correction = n/(1+n/N)
= 280/(1+280/80)
= 62

Where;
N = Population of women who have attended the infertility clinic (80)
n = sample size required

Inclusion criteria

1. Presence of both ovaries.
2. No evidence of endocrine disorders.
3. less than 45 years of age.

Exclusion criteria

1. Failure to retrieve oocytes due to technical difficulties.
2. Patients who have undergone radiotherapy or chemotherapy.

Ethical approval / Ethical considerations

Approval to carry out the study was sought from the Ethics and research committee of Kenyatta National Hospital as well as The Aga Khan university teaching hospital.

Total confidentiality of patient identification was ensured. A standard data collection form was used and patients were identified by use of a number. This study did not require any blood samples from patients other than those taken as routine prior to assisted reproduction, or those tests done to monitor follicle growth, and it did not involve a large number of personnel, hence breach in confidentiality was minimised. This study was presented to the department of obs/gyn at the proposal level and results and was approved. The study aims to promote the quality of services offered by fertility experts in this country, and provide a ground for further research in assisted reproduction in this country.

PROCEDURE

After obtaining ethical approval, all patients who met the inclusion criteria were recruited in the study. Data was obtained from patient records and a data collection form was used to obtain the information relevant to the study. Data collection forms were coded and none of the patients were identified by their names in order to maintain confidentiality.

Protocol for controlled ovarian hyperstimulation :

Long Gnrh – agonist protocol – In this method, patients received 0.5 mg of leupride subcutaneously from day 21 of the previous cycle till day 2 of the next cycle. From cycle day 2 until the day of hCG administration patients received 0.25mg of leupride . 225 iu r-FSH and 75 iu of r-LH were administered from day 2 till day 5, a Transvaginal ultrasound was done to determine the size of the follicles. Subsequently, from day 5 onwards, the dosage of gonadotrophins was adjusted depending on the growth of follicles . On cycle day 9 or 10 , majority of the patients had attained follicle diameter of 18 mm , hCG was administered on that day, 36 hours after which oocyte retrieval was done.

Gnrh – antagonist protocol – This protocol involved administration of 225 iu of r-FSH and 75 iu of r- LH until follicles attained diameter of 12 – 14 mm when cetrotide was given to suppress the endogenous LH surge . Cetrotide was also given until and including the day of hCG administration along with gonadotrophins which were adjusted according to the growth of follicles.

For both protocols, stimulation was monitored by Transvaginal ultrasound which was done on cycle day 2 of stimulation, day 5 , 7, 8 and 9 or even up to day 10 depending on the growth of the follicles. The aim was to achieve follicle diameter of 18 mm for hCG to be given for oocyte maturation.

For both cases, final oocyte maturation was achieved with 6500 UI of hCG when 3 or more follicles reached a diameter of 18mm.

Quality control of Data collection.

This pertains to data which was available in the patient records. While carrying out data collection it was ensured that quality of data is not compromised. Data collection was done by the principal investigator.

GnrH agonist and GnrH antagonist – All patients received medications belonging to the same manufacturer , hence we don't expect any differences in our findings attributable to the difference in quality.

Ultrasound – All patients were scanned by the same sonographer through out the study period, using the same ultrasound machine, this ensured consistency in reporting of findings.

Oocyte quantity and quality – Ovum retrieval was done under sedation , under guidance of Transvaginal ultrasound scan. As soon as oocytes were retrieved the quantity and quality was recorded by the embryologist. The microscope used by the embryologist is connected to an LCD monitor which was viewed by principal investigator as the quantity and quality were being assessed.

The quality of oocytes used by this IVF unit is divided into 3 types :

M 2 – The oocyte has undergone first meiotic division , and is in metaphase of second meiotic division which would be completed at fertilisation just as in a natural cycle. An oocyte is called so when it demonstrates a polar body, and the nucleus or nucleolus is not visible.

M1 – This is an intermediate phase , the oocyte is less mature than M2 , and it is still in the first meiotic division . Upon examination, it does not demonstrate a polar body , but spindle formation is seen to be occurring.

M3 – This is an immature oocyte, it is in the initial phases of the yet to begin cell division. It demonstrates a nucleus and a nucleolus which are typical features of a cell when it is in its initial stages of cell division.

Therefore , M2 is the best quality of oocyte , M1 is intermediate and Germinal vesicle is a poor quality oocyte.

Data collection instrument

A comprehensive data collection form that was closed and pre-coded was used to collect data pertaining to the patients involved. It covered all the necessary information required for purpose of this study.

Data analysis

Data summarization for categorical values used percentages while data summarization for quantitative variables used means and standard deviation for measures of central tendency and dispersion respectively. Associations were determined using P – values.

STUDY LIMITATIONS

1. Since this was a retrospective study, some of the data was found to be missing at the time of data collection.
2. The outcomes of ART in terms of pregnancy rates and live birth rates could not be used in this study since time was a limiting factor.
3. A Larger sample size would have given more elaborate results which was not possible because ART is a fairly new concept in this country and the study site was still new.
4. ART being a sensitive issue to the partners affected by infertility, it was not possible to interview patients and analyse better the patient characteristics and their sociodemographic profiles.

RESULTS

PATIENT CHARACTERISTICS

With reference to table 1 below, majority of the patients were Africans 36 (57.1%) followed by Asians 21 (33.3%), few of them were Caucasians 3 (4.8%) and of mixed race 3 (4.8%).

Out of 62 patients 29 (46.8%) had never conceived before and only 15 (24.2%) had carried their pregnancy to term while 7 (11.3%) had spontaneous abortions, 5 (8.1%) had elective terminations and 3 (4.8%) had ectopics. Of patients who had had a term pregnancy, 11(19.3%) had delivered vaginally and 4 (7%) had undergone a caesarian section. Three patients (4.8%) had hypertension and 26 (40.6%) had had previous reproductive tract surgery. All patients were tested for diabetes and thyroid disease but none were found to have these medical illnesses. Only 17 (26.6%) patients had received ART prior to the current treatment.

Table 1: Patient characteristics

Patient characteristics	**N (%)**
Race	
African	36 (57.1)
Asian	21 (33.3)
Caucasian	3 (4.8)
Mixed	3 (4.8)
Obstetric History	
No pregnancy	29(46.8)
PREVIOUS PREGNANCIES :	
Abortions	12(19.4)
Ectopic pregnancy	3(4.8)
Pre term deliveries	3(4.8)
Term pregnancy	15(24.2)
Delivery mode	
Normal	11 (19.3)
Caesarean Section	4 (7.0)
Presence of any medical conditions	
Hypertension	3(4.8)
Reproductive tract surgery	
	26 (40.6)
Previous ART	
	17 (26.6)

AGE DISTRIBUTION OF STUDY PARTICIPANTS .

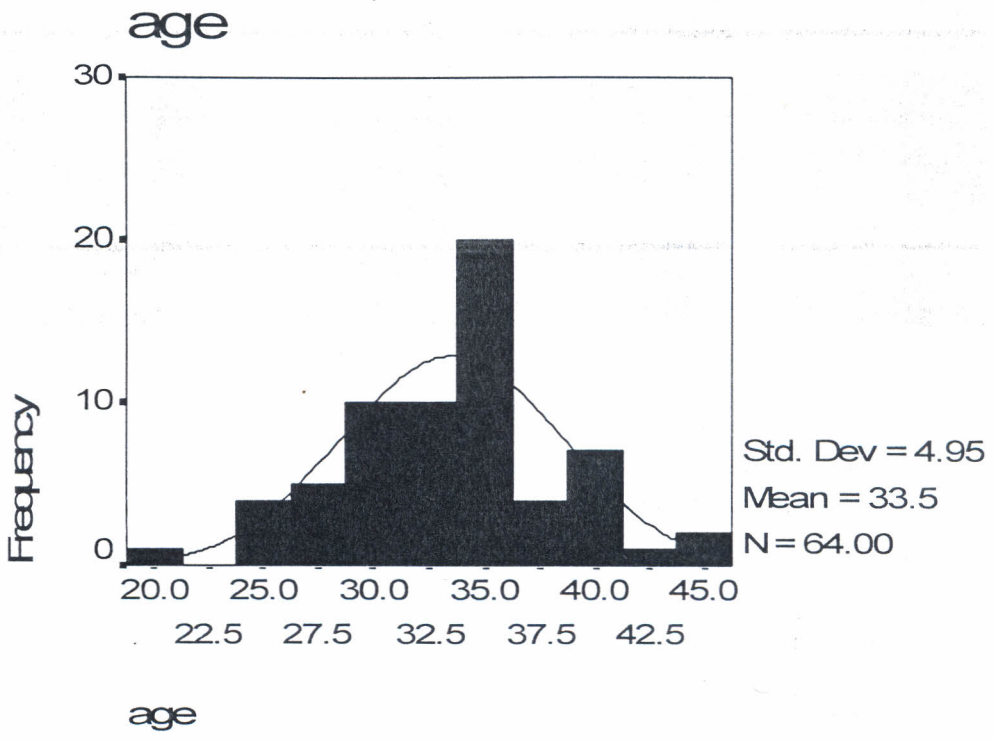


Figure 1.

With reference to figure 1 above, majority of women were in the second half of reproductive age with a median of 34 years and mean of 33.5 years (S.D = 4.95).

TYPES OF INFERTILITY BY AGE GROUP

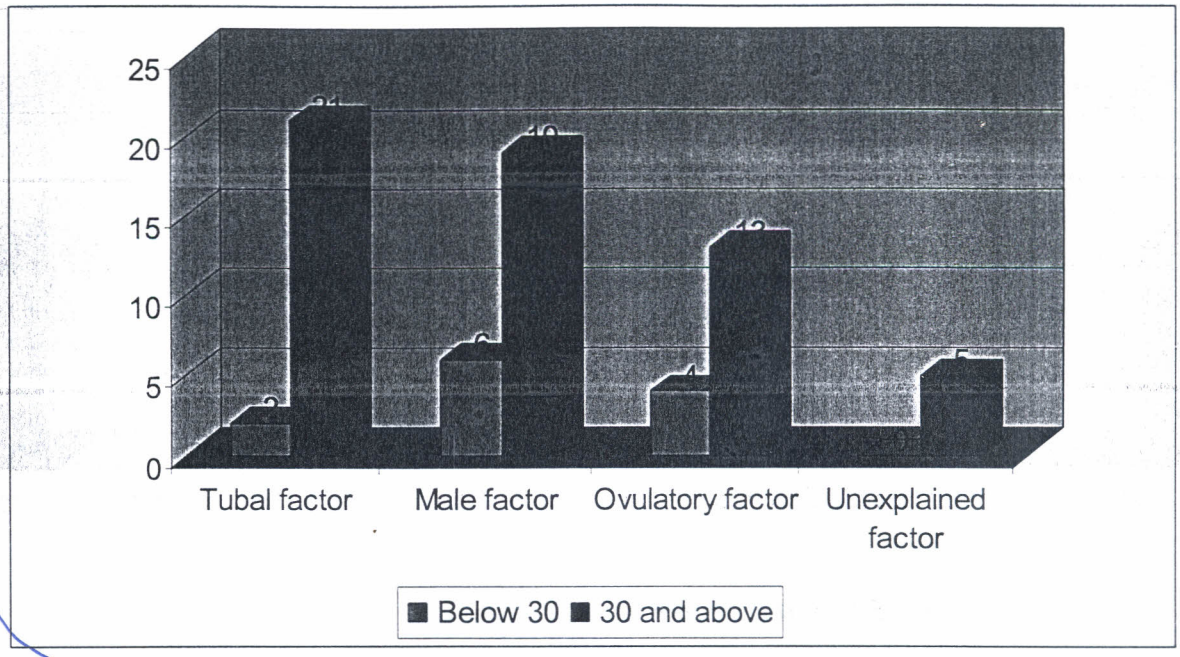


Figure 2.

With reference to figure 2 above,women who were below 30 years of age were mainly affected by male factor infertility and that was their main reason for requiring assisted reproduction. Whereas women above 30 years of age required assisted reproduction due to tubal factor infertility.

DURATION OF INFERTILITY BY AGE. (p – value)

	Below 30 years	30 years and above	P - value
Mean	2.78	5.14	0.005
Median	3.00	5.00	
Std. Deviation	1.481	2.349	
Minimum	1	1	
Maximum	5	12	

Table 2.

As expected,women below 30 years of age had an average duration of infertility of 2.78 years(SD = 1.48) whereas women above 30 years had an average duration of infertility of 5.14

years (SD = 5.14) as shown in table 2 above, which is statistically significant with a p- value = 0.005.

TYPE OF INFERTILITY

Male factor was the main contributor of infertility in this population (39.7%), followed by tubal factor infertility (37.4%) and ovulatory factors (27.4%) while only (8.2%) patients had unexplained infertility . Patients who had primary infertility were, 29 (46.8%) .

CATEGORY OF STIMULATION PROTOCOL

51 patients (79.7%), which comprise majority of the total population , underwent the GnRH agonist stimulation protocol while the rest underwent the GnRH antagonist stimulation protocol.

TOTAL DOSAGE OF GONADOTROPHINS

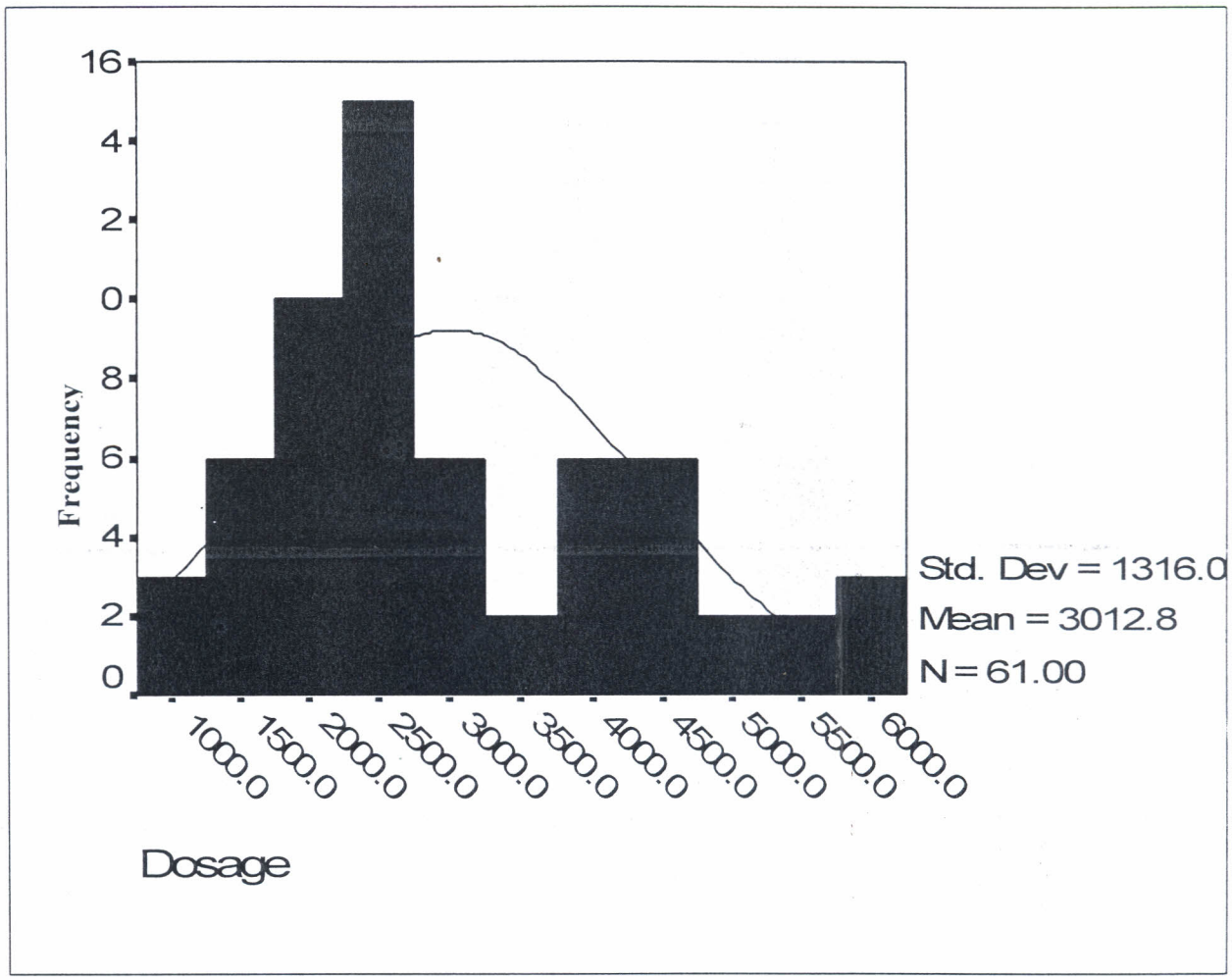


Figure 3

With reference to figure 3 above, when dosages in both the protocols were combined, it was found that the minimum dosage of exogenous gonadotrophins used was 1050 i.u and the maximum dosage was 6225 i.u with a median of 2575 i.u. The average dosage of gonadotrophins used was 3012.8 i.u (SD= 1316.05).

TOTAL NUMBER OF OOCYTES

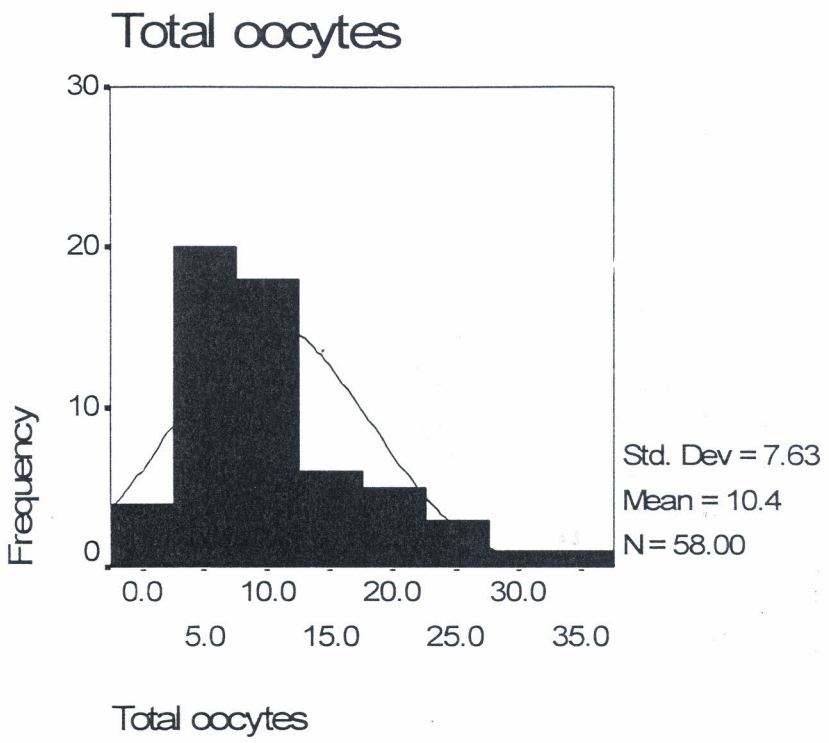


Figure 4

The mean number of oocytes was 10.4 (SD = 7.63) and median was 9 with a range of 0 to 37 as from figure 4 above.

Table 3. OOCYTE QUALITY

Quality of oocytes	Mean (SD)	Range	% of total Oocyte
M1(Intermediate quality)	1.56 (1.6)	(0,8)	14.5 %
M2(High Quality)	6.28 (4.6)	(0,24)	59.6 %
M3(Poor Quality)	2.78 (3.4)	(0,14)	25.9 %
Total	10.2 (7.6)	(0, 37)	100 %

With reference to table 3 above, in this study , quality of oocytes was divided into three main groups based on their level of maturity. M 2 quality of oocytes which are considered best consisted the majority at 59.6% , whereas 40.4% were of M 1 intermediate quality and M 3

quality contributing to 64 % of them which are of the poorest quality also known as germinal vesicle. Majority of the patients therefore were stimulated to produce good quality of oocytes, M 2 and M 1.

TOTAL OOCYTE COUNT BY AGE GROUP.

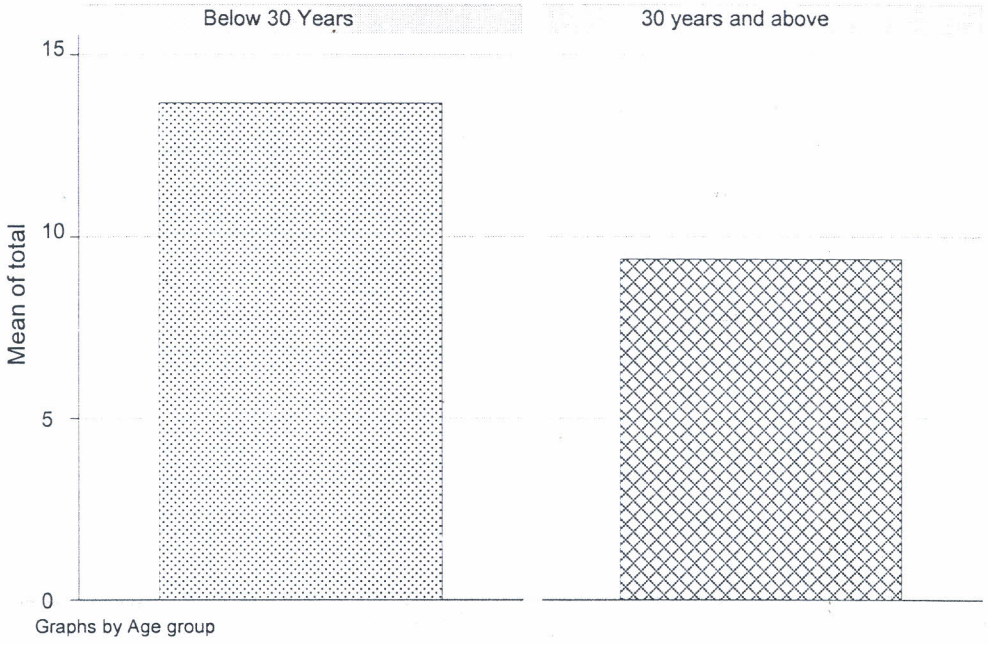


Figure 5

Total Oocyte characteristics by age group . Table 4.

	Below 30	=>30	P - value
Mean	16.1	9.4	0.0538
Median	15	8	
Std. Deviation	8.0	7.2	
Minimum	5	0	
Maximum	30	37	

??
not clear

The median number of oocytes retrieved from women below 30 years of age was 15 as compared to women above 30 years whose median oocyte count at retrieval was 8 . In both categories, the oocyte count was within acceptable limits as can be seen from figure 5 and table 4 above.

OOCYTE QUALITY BY AGE GROUP .

Table 5.

Oocyte quality	Below 30	30 and above	P - value
	Mean (SD)	Mean (SD)	
M 1 (Intermediate Quality)	1.8 (1.35)	1.51 (1.77)	0.161
M 2 (High quality)	8.36 (4.8)	5.82 (4.49)	0.062
M 3 (Poor quality)	5.4 (4.6)	2.24 (2.93)	0.012
Total	15.3 (7.9)	9.4 (7.24)	0.007

Women < 30 years of age have a greater total number of oocytes compared to women > 30 years ($p = 0.007$) which is a statistically siginificant difference, as shown in the table 5 above. In all the specific categories , women < 30 years have a trend towards a higher number of oocytes in all the three categories although they did not reach a statistical significance due to smaller population size.

DOSAGE OF EXOGENOUS GONADOTROPHINS BY AGE GROUP

	MEAN DOSAGE	S.D
AGE < 30 YRS	2337.50 I.U	897.6 I.U
AGE > 30 YRS	3187.20 I.U	1355.8 I.U
P - value	0.0738	

Table 6.

The mean dosage of gonadotrophins ,as seen in table 6 above, in women below 30 years of age was 2337.50 i.u which is much less than that of women above 30 years whose mean dosage of gonadotrophins was 3187.20 i.u. However, this is a trend not statistically significant (p – value 0.0738) and this is because the study did not have power to detect this difference. It has been reported in literature severally that older women tend to have a lower ovarian reserve hence a higher dosage is likely to be used for ovulation induction in order to achieve multifollicular development.

OOCYTE QUALITY BY PROTOCOL

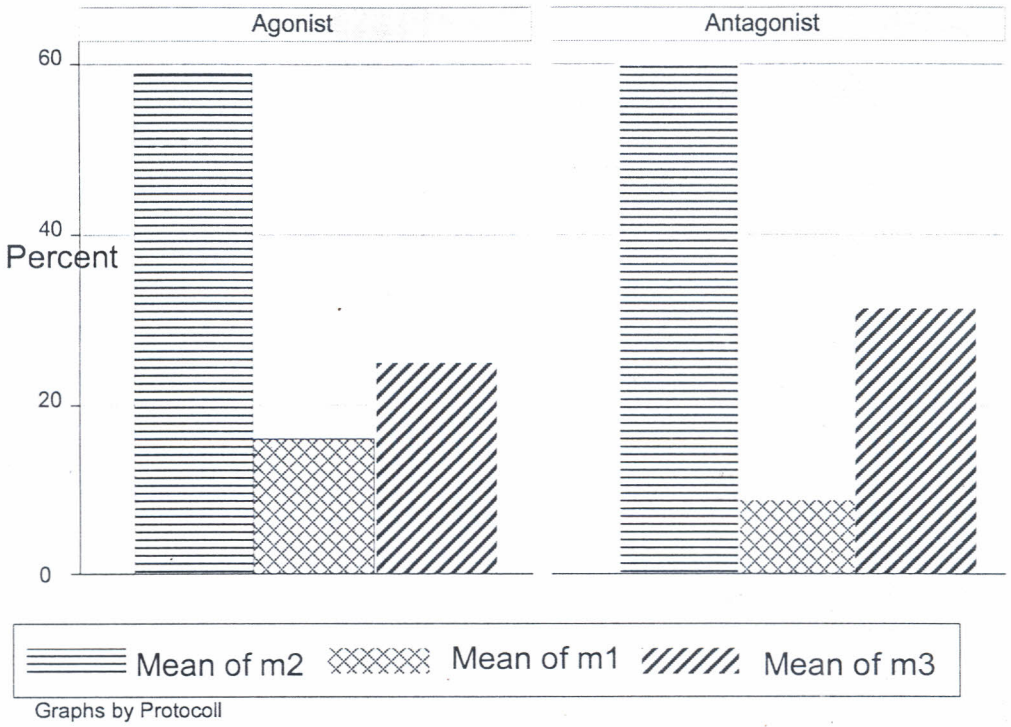


Figure 6.

From the figure 6 above , both the protocols resulted in a high proportion of M 2 quality of oocytes (60%) implying that both protocols were effective in achieving the desired goal of obtaining a greater proportion of the best M 2 quality of oocytes. About 25% of total oocytes in the agonist protocol and 30% in the antagonist protocol were of M 3 quality. M 1 oocytes comprised the least of them all, resulting probably from majority of oocytes entering the second mitotic division implying better oocyte maturity

RELATIONSHIP BETWEEN GONADOTROPIN STIMULATION AND QUANTITY OF OOCYTES RETRIEVED

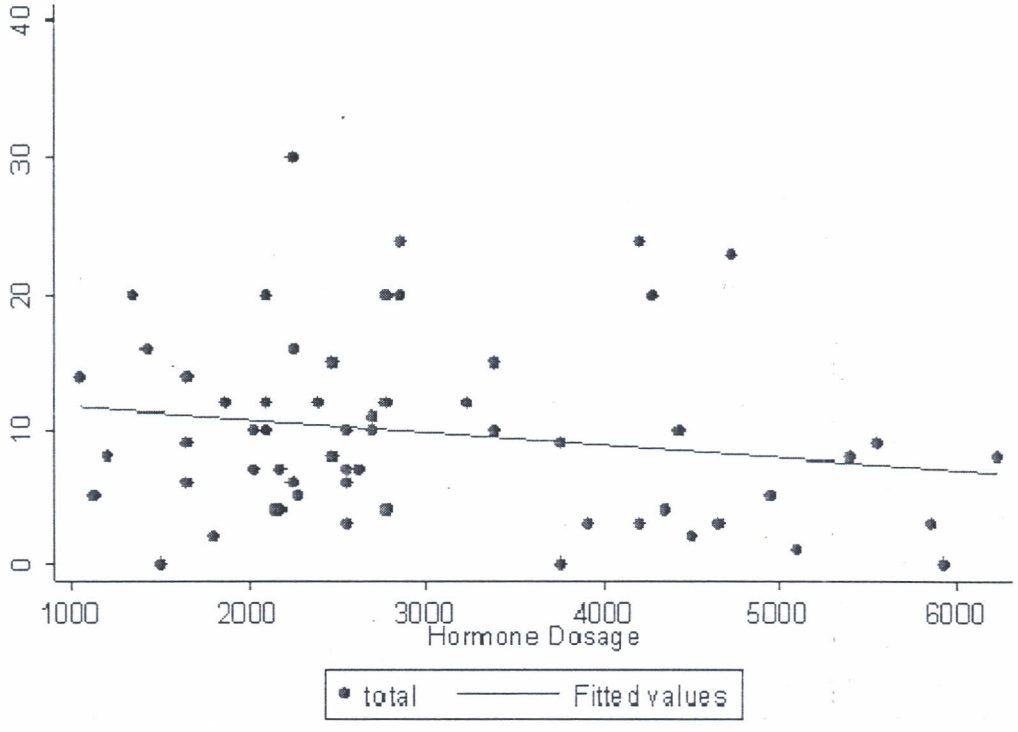


Figure 7.

correlation= -0.1755

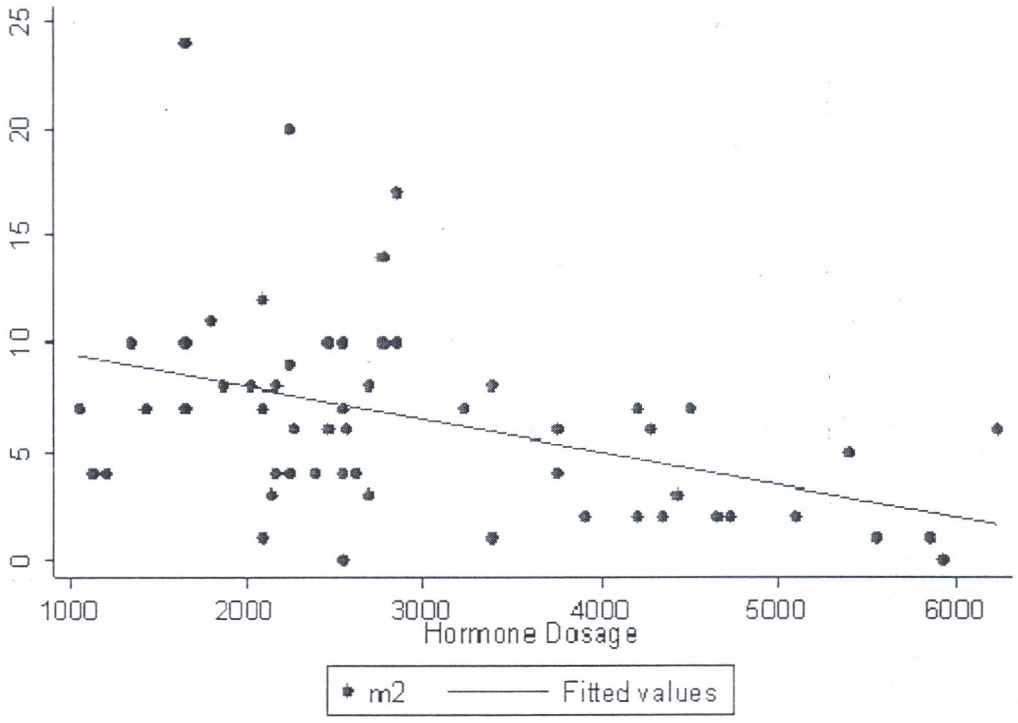
p =0.1838

The Spearman rank correlation from figure 7 above , demonstrated a marginal correlation between the total exogenous gonadotrophin use and the total number of oocytes retrieved r = -0.1755. Women whose ovaries have fewer oocytes or whose ovarian reserve is low usually require a higher dosage of gonadotrophins. Their response to ovarian stimulation is poor .

Is this how you study

RELATIONSHIP BETWEEN DOSAGE OF EXOGENOUS GONADOTROPHIN AND M 2 QUALITY OF OOCYTES

As the dosage of gonadotrophins is increased, the quantity of M 2 , the best quality of oocytes , is also decreased as shown in figure 8. There is a stronger correlation between dosage of exogenous gonadotrophins and the decline in M 2 quality, this shows that, even though the quantity of oocytes appears to increase with the dosage up to a certain point , this however does not appear to increase the quality of oocytes.



Correlation = -0.4466
P = 0.0005

Figure 8.

COMPARISON OF PATIENTS WITH FAVOURABLE OUTCOMES WITH THOSE WITH UNFAVOURABLE OUTCOMES.

M2 Category		Dosage	age	infertility duration
< 6	Mean	**3559.84**	**35.21**	**4.77**
	Std. Deviation	**1423.687**	**4.804**	**2.232**
>6	Mean	**2391.35**	**31.19**	**4.60**
	Std. Deviation	**817.831**	**4.243**	**2.746**
	P – Value	P < 0.001	P =0.001	P =0.693

Table 7.

Using Mean M 2 oocyte count of > 6 as favourable and < 6 oocytes as unfavourable, the average age, duration of infertility and exogenous gonadotrophins dosage were compared in table 7 above.

Women whose total oocyte count at retrieval was 6 and above were found to be relatively younger (31yrs) and their exogenous gonadotrophins requirement (2391.35 i.u) was much less than women in whom the total oocyte count was less than 6. In the two groups however, the duration of infertility was quite similar.

Student t test demonstrated that there was a statistically significant difference in the mean dosage that was administered in the two M 2 categories (P < 0.001) and in the mean age between the two groups (p = 0.001) however there was no statistically significant difference in the duration of infertility between the two groups.

DISCUSSION

Provision of Assisted reproductive technology to overcome both female and male factor infertility is in line with the reproductive rights agenda developed at the international conference on population and development in Cairo, 5 years ago. Infertility is a highly prevalent global reproductive health problem affecting at least 15% of reproductive age couples worldwide (16)

The scope and gravity of the infertility problem is much more severe in some regions of the world especially in Sub–Saharan Africa owing largely to untreated reproductive tract infections.

Tubal factor infertility is the main reason for infertility in resource poor countries, and sexually transmitted reproductive tract infections are also a major cause of male infertility.

The diagnosis of tubal factor infertility is reported to be as high as 85% in sub-Saharan Africa (17). This high incidence of tubal factor disease can be explained by a high rate of unprotected intercourse with infected partners at a very young age, unsafe abortions, and postpartum pelvic infections.

In our study, it was found that male and tubal factors were the main contributors to infertility . Male factor affected 25 (39.7%) which was only slightly higher than tubal factor 23(37.1%), and ovulatory factor was found in only 17 (27.4%) patients.

Local data from The Nairobi IVF centre , included 362 IVF cycles from August 2005 to July 2008 also reported tubal factor infertility to be a major cause of infertility in patients requiring ART . Blocked tubes were a contributory factor in 123 (36.4%) patients whereas anovulation was a factor in 68(20%) patients, uterine synechiae were found in only 3 patients.

Female factor infertility was found in 58% of the patients and male sub fertility in 31.4% of patients (18). This was very similar to what we found in our study.

Male factor infertility is a neglected reproductive tract health problem yet it contributes to at least half of all the cases of sub fertility worldwide. This was also evident from the high rate of ICSI , 58.4% that was performed at The Nairobi IVF centre mainly due to male factor infertility as compared to 41.6% of patients who underwent IVF.

It is now realised that a significant percentage of male infertility cases , particularly those that are severe are due to genetic abnormalities (19). Male factor infertility is quite concerning in our set of patients and it seems to have affected a significant proportion of couples. Majority of the patients were found to have low or abnormal sperm counts .

Relative or absolute decline in fertility with advancing age is a well know and widely studied phenomenon. It is partially explained by the decline in number of ovarian follicles as the age increases and this occurs on a monthly basis when there is recruitment of a cohort of follicles from which only one follicle will be mature enough to ovulate while the rest will become atretic. Follicular atresia, decreasing endometrial receptivity and decline in oocyte quality have been implicated in decline in fertility(20) .

In our descriptive observational study, ovulatory factor was found in 17 (27.4%) of patients which was mainly found in women above 30 years of age. This correlates well with the scientific background where numerous studies have attributed and proved decline of fertility to the increasing age of a woman.

Fecundability begins to decline in the early 30s and accelerates during the late 30s and early 40s (4). It has been shown that the number of follicles leaving the pool of resting follicles to enter the growth phase decreases with increasing age (5). In our study we found that the median age of patients presenting with infertility was 34 years , with a range of 19 – 44 years. This is quite similar to what has been reported by Dr. Noreh (18) whose patients had a mean age of 35 years and a range of 20 – 50 years. From our study it was found that women below 30 years of age were mainly requiring assisted reproduction due to male partner infertility. The effectiveness of ART has been reported to depend on age and conception rates start declining when the woman is aged > 30 years (Federation CECOS et al 1982, Van Noord Zaadstra et al) .

However , our study population is much younger than that found in the developed parts of the world whereby majority of the women actually start child bearing at a much later age , around the mid thirties, hence their age at infertility is usually higher.

It can be appreciated that there is a significant decline in oocyte number and quality at a cut – off of 30 years, which supports what has been reported by previous authors that there is a steep decline in fertility from the early 30s. In our study , we found that there was a significant difference in the total oocyte count in women < 30 years which was 15.3 and 9.4 in those > 30 years (p = 0.007) . The mean of M 2 count according to age was also found to be greater in women below 30 years as 8.36 and 5.82 in those above 30 years. There is a higher trend of oocytes in younger women even though it did not reach statistical significance for the individual qualities which may have occurred due to a smaller sample size.

A Brazilian retrospective study conducted on women below 35 years, found that there was a significantly greater exogenous gonadotrophins requirement in the poor responder group, and

they chose to define poor responders as having < 4 oocytes at retrieval. Their total oocyte retrieval rate was also lower but the M 2 oocyte rate was similar among poor and normal responders. Poor responders had an M 2 oocyte recovery rate of 66.8 % and normal responders had a recovery rate of 73.3 % (p = 0.2471)(21) . However both types of their responders belonged to the same age category , below 35 years . Furthermore, in our study there was a statistically significant difference in the ages of poor responders, 35.2 years and good responders 31.19 years (p = 0.001) . The Brazilian study found equal rates of fertilization, high quality embryos and similar implantation rates for both types of responders and they concluded that for women below 35 years quality of oocyte is not as important as quantity of oocytes retrieved.

From our study population, it is seen that there is a decline in quantity and quality of oocytes in women above 30 years with an increase in exogenous gonadotrophin requirement although this significant difference is yet to be correlated with pregnancy and live birth rates .

The mean cumulative dosage of exogenous gonadotrophins from our study was 3012 i.u . From the Bristol study (14), 3000 iu was used as a cut-off for determining favourability of outcome of ART. This study was assessing ovarian response in a standard IVF programme using a Long GnRH agonist protocol in women below 40 years. They reported that pregnancy rates were much more favourable at dosage of less than or equal to 3000 i.u (32.4%) as compared to > 3000 i.u, furthermore they found that oocyte quality became significantly important when more than 3000 iu were used. The pregnancy rates with use of > 3000 i.u and > 4 oocytes were poor, but the rate declined even further at the same dosage but with oocyte count being < 4.

In our study , we used a cut – off of 6 oocytes to define favourable outcomes (2) and it was found that the difference in the mean dosage of exogenous gonadotrophins used between those with less than or more than 6 oocytes was statistically significant. Patients with more than 6 oocytes required a mean dosage of 2391.35 i.u and those with less than 6 oocytes had required a higher dosage of 3559.8 i.u with p value < 0.001. Although the Bristol study looked at all patients below 40 years, we looked at patients who were below and above 30 years . Furthermore , from the analysis it was found that the mean age of women who produced more than 6 oocytes was 31 years and those women who produced less than 6 oocytes had a mean age of 35 years and this difference was found to be statistically significant $p = 0.007$.

In our study we looked at patients who were undergoing controlled ovarian hyperstimulation using either GnRH agonist or GnRH antagonist stimulation. Majority of the patients 51 (79.7%) underwent GnRH agonist stimulation. From our analysis , it was found that both stimulation protocols resulted in an M 2 oocyte recovery rate of 60% hence both protocols were fairly effective in achieving the desired outcome of attaining as many oocytes and that too of a good quality. In this study we did not look at other outcomes such as live birth rates to compare the two groups due to time limitation .

Meta analyses by Ludwig et al suggested that clinical pregnancy rate and on going pregnancy rate were not statistically significant between the GnRH agonists and antagonists . In 2006, a meta analysis based on 22 fully published RCTs used live birth rates as an outcome measure and no statistically significant difference was found between the two analogues (12) (Gnrh agonists and antagonists.)

Our findings were limited to the oocyte quality obtained but it demonstrated that both analogues in our study were equally effective in achieving a recommended quantity of the best quality of oocytes .

A Spearman rank analysis was conducted to determine a correlation between the total dosage of exogenous gonadotrophins used for the combined protocols and the quantity and quality of M 2 oocytes. It was found that there was a marginal correlation between the total exogenous gonadotrophins dosage and the total number of oocytes, r = - 0.1849.

However , this correlation demonstrated that the quantity of oocytes reduced as the exogenous gonadotrophins requirement increased . This effect can be explained by the fact that , as the ovaries become depleted of oocytes and fewer oocytes are left , the so called reduced ovarian reserve, then a higher dosage of gonadotrophins is required for ovarian stimulation. The ovaries are said to have become relatively resistant to stimulation .

The correlation between dosage of exogenous gonadotrophins and M 2 quality of oocytes was stronger (r = - 0.4233) as compared to total oocyte number (r = - 0.01849) , implying that women especially of the older age group will only be able to produce a limited number of M 2 oocytes even if a higher dosage of exogenous gonadotrophins is used. The quantity of oocytes may be increased up to a certain point by increasing the stimulation dose, but this will not however have a marked effect in increasing the quality as well.

SUMMARY AND RECOMMENDATIONS

With a mean age of 33.5 years and a median of 34 years, majority of the patients were affected by male and tubal factor infertility which has been the trend in the Sub-Saharan Africa. Age plays a significant role in determining fertility potential of the female partner in terms of quantity and quality of oocytes retrieved after controlled ovarian hyperstimulation.

There was a statistically significant difference in the mean age and mean dosage of exogenous gonadotrophins used between the poor responders (< 6 M 2 oocytes) and the good responders (> 6 M 2 oocytes). As the minimal cumulative dose of exogenous gonadotrophins requirement increases, both the quantity and quality of oocytes decline , implying a decline in ovarian reserve of good quality of oocytes hence even if a higher stimulation dose is used , and the ovaries do not respond linearly.

Clinicians and infertility experts need to modify stimulation protocols in order to achieve a satisfactory number of oocytes that comprises the good quality of oocytes and not just an excessive number of oocytes .

With the availability of ART services in the country , women should be advised to seek medical care at an earlier stage rather than wait for many years as age plays a significant role in determining the outcomes of ART and it seems that age remains a limiting factor even with the application of assisted reproduction.

Research needs to be done with regards to male factor infertility, looking at the risk factors involved and trends in male factor infertility so that preventive measures can be applied and clinicians can then be able to advise couples accordingly based on local population studies and intervene where possible.

CONFLICT OF INTEREST

This study was a descriptive observational study that was done in partial fulfilment of the MMED degree in Obs/gyn and it was not funded by any organization or the IVF unit from which the data was collected.

APPENDIX

DATA COLLECTION FORM

Date of data collection .....

Participant number .....

Age .....

Body weight .....

Race : African Asian Caucasian

Date of last pregnancy	Gestational age	Outcome of Pregnancy	Mode of delivery	Any antenatal complications

Level of Education : Pry / Secondary/ College/ University

(Encircle where applicable)

Past medical history of : yes = 1 , no = 0 , not indicated = 9

(1) Hypertension .....

(2) Diabetes.....

(3) Autoimmune disorder .....

(4) Any surgeries done on the reproductive tract.....

.....

Past obstetric history of : yes =1 , no = 0 , not indicated = 9.

(1) First trimester miscarriages.....

Dates 1. .....

2. .....

3. .....

(2) Blighted ovum .....

Dates 1. .....

2. .....

3. .....

Congenital malformations : yes = 1, no = 0, not indicated = 9.

.....

Infertility : (encircle the number applicable)

Cause of infertility : yes = 1 , no = 0 , not indicated = 9.

Ovulatory

Tubal factor infertility

Male factor infertility

Unexplained cause

Others

.....

Duration of infertility .....

Previous ART treatment yes =1, no = 0 , not indicated = 9.

Number of previous attempts .....

Serum hormonal levels and date of collection of samples :

FSH level .....miu/ml Date of collection __ __ / __ __ / __ __ __

Estradiol levels .....pg/ml Date of collection __ __ / __ __ / __ __ __

Exogenous gonadotrophins used :

Total number of vials / Dosage (i.u) used .....

Oocytes retrieved

Quality of oocytes	Number of oocytes
M 1	
M 2	
M 3	
TOTAL	

APPENDIX II

KENYATTA NATIONAL HOSPITAL
Hospital Rd. along, Ngong Rd.
P.O. Box 20723, Nairobi.
Tel: 726300-9
Fax: 725272
Telegrams: MEDSUP", Nairobi.
Email: KNHplan@Ken.Healthnet.org

Ref: KNH/UON-ERC/ A/190

8th April 2009

Dr. Fauzia Butt
Dept. of Obs/Gynae
School of Medicine
University of Nairobi

Dear Dr. Butt

Research proposal: "Association of Basal Serum FSH levels with Oocyte Quantity and Quality after Controlled ovarian Hyperstimulation: A Descriptive – Observational study" (P24/1/2009)

This is to inform you that the Kenyatta National Hospital Ethics and Research Committee has reviewed and **approved** your above revised research proposal for the period 8th April 2009 –7th April 2010.

You will be required to request for a renewal of the approval if you intend to continue with the study beyond the deadline given. Clearance for export of biological specimen must also be obtained from KNH-ERC for each batch.

On behalf of the Committee, I wish you fruitful research and look forward to receiving a summary of the research findings upon completion of the study.

This information will form part of database that will be consulted in future when processing related research study so as to minimize chances of study duplication.

Yours sincerely

PROF. A N GUANTAI
SECRETARY, KNH/UON-ERC

c.c. The Chairperson, KNH/UON-ERC
The Deputy Director CS, KNH
The Dean, School of Medicine, UON
The Chairman, Dept. of Obs/Gynae, UON
Supervisors: Dr.James Kiarie, Dept.of Obs/Gynae, UON
Dr. Samson Wanjala, Dept.of Obs/Gynae, UON

ETHICS AND RESEARCH COMMITTEE,
KENYATTA NATIONAL HOSPITAL,
Nairobi – Kenya.

Dear Sir,

RE : Association of Basal FSH with quantity and quality of oocytes retrieved after controlled ovarian hyper stimulation (P24/1/2006)

Data collection for the above study is ongoing. During interim analysis it was realised that information on basal FSH was missing for most participants. We have therefore decided to conduct analysis on the outcomes of controlled ovarian hyperstimulation in relation with the stimulation protocols. I am therefore requesting for approval of the revised proposal.

This analysis I believe has greater scientific merit and will involve the following changes in the proposal :

- Title changed to outcomes of controlled ovarian hyperstimulation.
- Time frame from which patients will be eligible extended from April 2008 to July 2009.
- The sample size has been increased from 61 to 62 participants.

There will be no changes in the data collection procedures

I look forward to hearing from you soon.

Yours faithfully ,

Dr. Fauzia Butt
MMED , Part II
OBS/GYN , UON.

KENYATTA NATIONAL HOSPITAL
Hospital Rd. along, Ngong Rd.
P.O. Box 20723, Nairobi.
Tel: 726300-9
Fax: 725272
Telegrams: MEDSUP", Nairobi.
Email: KNHplan@Ken.Healthnet.org

Ref: KNH-ERC/ MOD/405

26th August 2009

Dr Fauzia Butt
Dept. of Obs/Gynae
School of Medicine
University of Nairobi

Dear Dr. Butt

Re: Request for modification - study titled "Association of Basal FSH with quantity and quality of oocytes retrieved after controlled ovarian hyper stimulation" (P24/1/2006)

Refer your communication to the KNH/UON-ERC on the above study.

This is to inform you that the revised protocol has been approved according to the changes below:

1. Study title
2. Time frame
3. Sample size

Yours sincerely

PROF C. S. KIGONDU
AG SECRETARY, KNH/UON-ERC

c.c. Prof. K.M.Bhatt, Chairperson, KNH-ERC
The Deputy Director CS, KNH

THE AGA KHAN UNIVERSITY

Faculty of Health Sciences

Post Graduate Medical Education

15th May 2009

Dr. Fauzia Butt
Kenyatta National Hospital
P.O Box 30270 – 00100
Nairobi

Dear Dr Butt,

Re: Association of basal serum FSH levels with oocyte quantity and quality after controlled ovarian hyper-stimulation

---

It is my pleasure to inform you that the Aga Khan University Research Committee (EA) has approved your application for seed funding for your research and has referred your proposal for Ethical review. This approval has been given subject to the following conditions.

1. Approval is given for a specified period as stated in your protocol. If the project takes longer than the specified period to complete, a request for an extension of the ethics clearance should be sought.
2. Any alterations to the approved protocol must be submitted to the Research Committee for approval prior to alterations being effected.
3. Progress reports must be submitted to the Research Support Unit within the first six months and subsequently on a quarterly basis during the period of the project.
4. Research could be audited by the Research Ethics Committee to ensure compliance with guidelines.
5. The Research Support Unit must be informed when a project is curtailed, terminated or completed.
6. A copy of the research project final report must be submitted to the Research Support Unit upon completion of the project.

Note that as the Principal Investigator, you have the full administrative, scientific, and ethical responsibility for the management of the research project in accordance with the University policies and guidelines.

Best regards,

Best regards,

Prof. William Stones
Chair, AKU (EA) Research Committee

Cc. Dr. Ahmed Mushtaq
Associate Dean (Medical Education-EA)

Jawaid Bachlani
Finance Officer

Prof. William Stones
Chair, AKU (EA) Research Committee
Aga University Teaching Hospital
Nairobi – Kenya.

Dear Sir,

RE : Association of Basal FSH with quantity and quality of oocytes retrieved after controlled ovarian hyper stimulation (P24/1/2009)

Data collection for the above study is ongoing. During interim analysis it was realised that information on basal FSH was missing for most participants. We have therefore decided to conduct analysis on the outcomes of controlled ovarian hyperstimulation in relation with the stimulation protocols. I am therefore requesting for approval of the revised proposal.

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- The sample size has been increased from 61 to 62 participants.

There will be no changes in the data collection procedures

I look forward to hearing from you soon.

Yours faithfully ,

Dr. Fauzia Butt
MMED , Part II
OBS/GYN , UON.

THE AGA KHAN UNIVERSITY

Faculty of Health Sciences

Post Graduate Medical Education

21st October 2009

Dr. Fauzia Butt
University of Nairobi
Dept of Obstetrics & Gynaecology -Kenyatta National Hospital
P.O. Box 30197 – 00100
Nairobi

Dear Dr Butt,

Re: Association of Basal FSH with quantity and quality of oocytes retrieved after controlled ovarian hyper stimulation (P24/1/2009)

Thank you for informing the Aga Khan University Research Committee about the changes made to the above mentioned study. The minor changes have been noted and approved by Research Committee Chair's action. These changes are;

1. Change of study title to "Outcomes of controlled ovarian hyper stimulation"
2. Revision of study time frame
3. Revision of sample size

You will be required to submit a final copy of your dissertation to the Aga Khan University Research Office.

Best regards,

Prof. William Stones
Chair, AKU (EA) Research Committee

Cc. Mr. John Arudo
Chair, Aga Khan University Research Ethics Committee

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