Singapore Journal of Obstetrics and Gynaecology
July 2003; 34(2): 46-51

Establishment of an assisted reproductive technology unit in Pakistan, a developing country

Humaira Jamal Yousuf
Peter Baillie

ABSTRACT

Objective: To improve take home baby rates in the ART centre in Pakistan, a developing country.
Design: Assessment of changes in outcome with altered treatment protocols.
Setting: Patients enrolled in a university based assisted reproduction centre.
Patients: All patients submitted to IVF or ICSI at the ART unit of Baqai Medical University from 1996 to 2000 inclusive
Interventions: Introduction of: 1. Blastocyst transfer in 1999. 2. Correction of abnormalities prior to IVF of ICSI in 2000 according to reproductive biological concepts.
Main outcome measures: Total pregnancy rate per cycles started and the take home baby rate per cycle was assessed.
Results: Total pregnancy rate in 1998 was 27%; take home baby rate was 8%; abortions 22%. Total pregnancy rate in 1999 was 32% and take home baby rate was 18.6%; abortions 13.4%. Total pregnancy rate in 2000 was 39.4% and take home baby rate was 32.7%; abortions 6.7%.
Conclusions: A high success rate in terms of take home babies can be achieved in infertile couples from developing countries by the application of assisted reproductive techniques with improved conception rates rather than utilizing the conventional gynaecological approach.

Key words: IVF, ICSI, reproductive biology, PCOS, APA, implantation.

INTRODUCTION

Infertility, the inability to conceive after 1 or 2 years of regular unprotected sexual intercourse[1,2], is frequently a source of emotional trauma for infertile couples. The treatment of infertility received a boost in 1978 when Steptoe and Edwards[3] first reported the birth of a baby that was conceived through *in vitro* fertilisation and embryo transfer (IVF-ET). Since then, there has been considerable improvement in the original IVF methodology, and several procedures are now available to improve the chances of pregnancy for some couples who otherwise would have been irreversibly infertile. These methods, collectively called new reproductive technologies, include among others IVF-ET, gamete intrafallopian transfer (GIFT)[4,5], zygote intrafallopian transfer (ZIFT)[6] and intracytoplasmic sperm injection (ICSI)[7].

Despite some initial controversies[8,9], new reproductive technologies are now widely used by gynaecologists for the management of infertile couples in many developed countries of the world. By contrast, the situation in developing countries is presently unknown. However, developing countries have a large reservoir of infertility problems that are potentially treatable. In particular, IVF-ET was originally developed for the treatment of women with irreversible bilateral tubal

Baqai Institute of Reproduction and Developmental Sciences
Karachi,
Pakistan

Correspondence:
Dr. Humaira Jamal Yousuf
34-E Askari IV apartments
Gulistan e Jauhar
Karachi
Pakistan
E-mail: mhyousuf@cyber.net.pk

occlusion, and it is conceivable that the large number of women with tubal occlusion in developing countries would benefit from the procedure.

Another important area of concern in developing countries is whether a new reproductive technology programme can be carried out with the same degree of success and safety that has been established for the procedures in Western countries. To protect the public, some have expressed the view that the procedures should never be attempted unless they can be carried out with a high degree of proficiency, accuracy and safety. The successful performance of IVF-ET in developing countries suggests that these professionals have the required training and experience to carry out the technical aspects of the procedures. However, the proper organisation of reproductive technology programme involves more than technical expertise. For such a programme to be efficacious and sustainable it also requires the proper coordination of supporting services, a continuous supply of high quality consumables, such as culture media and the application of appropriate quality assurance procedures.

The safety of the procedures is another important issue to consider before a recommendation can be made for the use of these procedures. Reports from well established units in Western countries indicate that reproductive technologies are associated with increased incidence of multiple pregnancy, low birthweight babies, perinatal mortality, and ectopic pregnancy[10,11]. There are also increased risks for women associated with reproductive technologies, such as those linked with induction of superovulation by fertility drugs. These risks include ovarian hyperstimulation syndrome, cysts, coagulation abnormalities leading to thromboembolism, stroke and myocardial infarction, molar pregnancy and ovarian cancer[12,13]. These complications are particularly likely to occur in clinics with meagre facilities and with limited personnel experience to monitor accurately ovarian response. A related consideration is whether there would be resources available to treat these complications when they occur and whether the already over-burdened health care system can accommodate the additional strain. If the rate of complications is increased, it will tend to simultaneously increase the indirect costs and therefore reduce the cost-effectiveness of the procedure. To attain an acceptable standard, the programme will inevitably have to make provisions for the training, re-training and certification of physicians, possibly in institutions overseas.

Improvement in conventional methods of treatment of infertility should also form part of the overall control efforts in order to increase the number of infertile couples who can benefit from simple treatment methods. The development of the new reproductive methods in the country should be a long term endeavour and should only be undertaken after careful consideration of their efficacy, cost effectiveness, the occurrence of multiple pregnancies are other associated hazards, and socio-cultural, legal and ethical issues.

Based on these considerations, IVF-ET and ICSI are in great demand in developing countries including Pakistan despite considerable difficulties[14]. In order to achieve equivalent results to that of developed countries as outlined above, appropriate measures need to be adopted to maintain high quality procedures with adequately trained personnel. In this report, we describe the evolution of a university based assisted reproductive technology (ART) centre in Karachi.

PATIENTS AND METHODS

A total of 514 patients were recruited for IVF and ICSI procedures from 1995 until December 2000. Gynaecological abnormalities were corrected prior to ovulation induction. Down regulation with inhaled GnRH was carried out followed by a high and low dose stimulation protocol depending on age, FSH and LH estimation and the presence of polycystic ovaries. When the leading follicle was ≥ 18 mm diameter, HCG 5000 i.u. was administered and vaginal ultrasonically guided ovum retrieval was carried out 36 hours later.

Since 1999, all embryo transfers were carried out with the two best being selected on day 5, the remainder being frozen.

Post-transfer luteal support was carried out by vaginal pessaries daily of 200mg micronized progesterone as well as HCG 5000i.u. intramuscularly twice weekly if overstimulation was not a possibility.

Serum βHCG, progesterone and estradiol were estimated on day 12 after ovum retrieval. Further early pregnancy hormonal support was determined by the rate of change of these hormones integrated with ultrasonically determined trophoblastic volume.

At 32 to 34 weeks gestation, patients were returned to their practitioners for delivery.

From January 2000, the protocol was altered to target ovum quality improvement prior to ovulation induction, the effects of an inflammatory environment on implantation and insulin resistance with polycystic ovaries. The changes included:

(i) counselling aimed at stress alleviation

(ii) normalization of weight

(iii) the recognition and treatment of underlying disease by full blood count, erythrocyte sedimentation rate (ESR), C-reative protein (CRP) and anti-phospholipid antibodies (APA). Thyroid evaluation including microsomal antibodies were carried out when indicated clinically. Mantoux tests were carried out for unexplained ESR and if >12 mm, a laparoscopy and appropriate biopsy was undertaken.

(iv) Anti-phospholipid antibodies were eliminated (<5) prior to ovulation induction with 75mg Dispirin initially and occasionally corticosteroids.

(v) Aspirin 75mg was administered daily following ovum retrieval and luteal support was increased if spinbarkheit was present at ovum transfer.

(vi) If polycystic ovaries were detected ultrasonically, fasting sugar, insulin and triglycerides were estimated. Insulin resistance[15] was diagnosed if the fasting insulin was >12.2 or >10 with triglycerides >150mg%. Metformin 1500mg daily was started and ovulation induction delayed until the serum insulin was <8 with no polycystic ovaries detected ultrasonically. This usually required more than 3 months therapy.

(vii) A similar approach was carried out in male patients with suboptimal semen analysis.

RESULTS AND DISCUSSION

The primary message in this preliminary communication is to demonstrate that both IVF-ET and ICSI procedures can be performed in reproductive centers in developing countries. Data from Table 1 indicate that the total pregnancy rate in 1998 was 27%; in 1999 it increased to 32% and in the year 2000 the pregnancy rate was 39.4%. Similarly, an increased success rate occurred with reference to the take home baby rate between 1998-2000 being 8% in 1998, 18.6% in 1999 and a much better outcome of 32.7% in 2000.

A steady increase in successful outcome was noted since start-up culminating in a 33% take home baby rate achieved per cycle initiated in 2000 despite the steadily increasing age[16,17] of our patients. This is also reflected in the increasing number of cycles abandoned, a course of action that is avoided if at all possible in developing countries.

These results are comparable to those from the developed nations in the world[18] despite the difficulties we encountered in initiating the ART programme. With limited resources, microdeletions[19] and such factors as lead toxicity[20,21] were inadequately evaluated. However, 86% of patients undergoing ART were observed to be insulin resistant (Table 2).

It is noteworthy that the improvement in 1999 coincided with the introduction of blastocyst culture and emphasizes the importance of a high quality laboratory, something that is often deficient in a developing world situation and emphasizes the importance of a university based centre in these circumstances.

In 2000 we altered our approach to deal with ovum quality prior to stimulation and implantation problems – the two major factors limiting succes rates in ART[22–24].

As initial cohort selection occurs up to 200 days prior to ovulation[25] and is non gonadotrophin dependent, attention to improvement of ovum quality by such interfering factors as weight[26], stress[27–29], inflammatory conditions[28] and polycystic ovaries require considerable pre-treatment, and although not a bio-equivalence study, we feel that our improved results justify continuation and further evaluation of this approach[30].

It is noteworthy that insulin resistance is very common in our patients with polycystic ovaries as has been reported from South India[31]. This is possibly due to a combination of genetic factors[32,33] and the Barker hypothesis[34,35]. Metformin was used liberally and has been reported to be associated with increased fertility[36,37], decreased abortion[38,39] and virtual elimination of gestational diabetes[40] – we only had one patient with gestational diabetes. Metformin does not alter placental transfer of glucose and although a small molecule, transplacental passage may be beneficial[41]. No increase in fetal abnormalities have been noted recently[42]. Nestler does not even measure insulin resistance in polycystic ovaries prior to administering metformin[43].

Our decison to minimize inflammation as deleterious to implantation has considerable justification in world literature – aspirin significantly improved outcome in an Argentinian study[44], as did pre-treatment of hydrosalpinges[45]. Estrogen dominance, common in ART, is also inflammatory[46]. Hence we used aspirin in all patients and eliminated inflammatory markers in patients with fibroids and endometriosis prior to stimulation. It is also noteworthy that we did not achieve a single take home baby in any patient with a non anaemic raised ESR. More controversial is our hypothesis that low levels of anti-phospholipid antibodies may subtly influence implantation[28,47–50.] Elimination of these antibodies was associated with a similar outcome to patients without antibodies.

Asbortion rates also decreased but this may be due to many factors and the small numbers examined preclude statistical analysis.

In conclusion, highly satisfactory results are achievable in ART in developing countries. Attention to laboratory quality is mandatory and it is possible that a reproductive biological approach as opposed to gynaecological methods will compensate for a lack of resources and is worth pursuing in developing countries.

TABLE 1
Outcome of ART procedure following the embryo transfer programme (1996–2000)

Year	Cycles	Average	>35 years	Single	Multiple	Abortions*
2000	104	33.9	30	27	7	7†
1999	134	33.4	28	22	3‡	18§
1998	100	30.8	20	5	3	22
1997	56	30.7		3	0	16
1996	20			0	0	4

* Prior to 2000, abortions also included biochemical pregnancies
† Four deliveries at 16-24 weeks gestation
‡ One triplet
§ One delivery at 16-24 weeks gestation

CYCLES ABANDONED: 2000 – 11
(Included in results) 1999 – 03
1998 – 04

Only three of these patients were under 40 years of age.

TABLE 2
Clomid resistant ultrasonic polycystic ovaries, insulin resistance and outcome for 1999-2000

	Number	Deliveries	Abortions
Treated insulin resistance	56	14	03
No insulin resistance	15	06	02
Insulin not measured	78		
Inadequately treated	34		

90 out of 105 (86%) investigated patients were insulin resistant (1999-2000)

TABLE 3
Treated autoimmune disease and outcome 1999-2000

	Number	Deliveries	Abortions
Treated APA*	60	16	01
No APA abnormality	29	08	01
APA not estimated	110		
APA inadequately treated	21		

* Four patients required steroids for elimination of APA, 2 with positive ANA
APA = Anti-phospholipid antibodies

No patient with an unexplained ESR of more than 20 mm/hour became pregnant.

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