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Treatment regimens in ovulation induction and ovarian hyperstimulation

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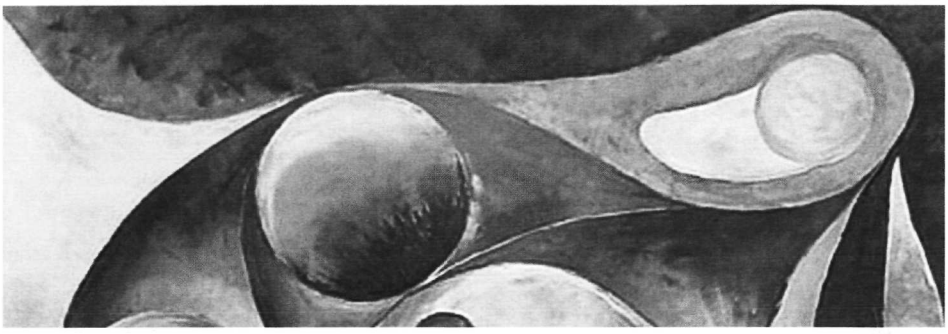
Chapter 9



Summary and implications

The aim of this thesis was to make a contribution to finding optimal treatment regimens for ovulation induction and ovarian hyperstimulation.

In women with polycystic ovary syndrome (PCOS) ovulation induction with clomiphene citrate (CC) is still the treatment of first choice. Women not responding to CC present a clinical challenge. In these women both ovulation induction with recombinant FSH (rFSH) in a chronic low dose step-up regimen and laparoscopic electrocautery of the ovaries are standard treatments.

Ovulation induction with FSH is associated with a higher chance for multiple pregnancies, caused by multiple follicular development and is expensive. Laparoscopic electrocautery is therefore an interesting alternative treatment modality for PCOS patients. Arguments in favour of the laparoscopic approach include the minimal morbidity associated with a laparoscopic procedure, the eliminated need for cycle monitoring and the low risks of multiple pregnancies. Furthermore, it has been shown that some CC-resistant women respond once again to CC after laparoscopic cauterisation. Disadvantages are that it is a surgical procedure under general anaesthesia, and that possible long-term effects on ovarian function are unknown.

To find out the best way to treat these women we designed a randomised clinical trial comparing a treatment strategy starting with laparoscopic electrocautery of the ovaries followed by CC and rFSH if anovulation persisted versus ovulation induction with rFSH. In the evaluation of these treatments we focussed on clinical effectiveness, side effects, costs and the identification of predictors for success or failure with these respective procedures in specific subgroups of women.

For women with PCOS not responding to ovulation induction or with other co-existing fertility problems IVF or ICSI and thus ovarian hyperstimulation is considered a last resort treatment. Recombinant FSH nowadays is most commonly being used for ovarian hyperstimulation in down-regulated normogonadotrophic ovulating women as well as in women with PCOS although its superiority has not been proven. Therefore, whether a pure FSH preparation or a mixture of LH and FSH should be preferred for ovarian hyperstimulation in GnRHa down-regulated women was unclear.

Furthermore, FSH is not just a single entity but exists as 20 FSH isoforms. These isoforms differ in their isoelectric points - the lower the isoelectric point, the more acidic the FSH isoform will be - and in the complexity of the oligosaccharides. The FSH isoform profile of commercially available gonadotrophin preparations has not been in focus when evaluating treatment outcome in connection with ART.

The first part of the thesis describes four studies on ovulation induction in women with CC-resistant PCOS. In a randomised trial a laparoscopic electrocautery strategy was

compared to ovulation induction with rFSH with respect to clinical outcome and costs. Using prognostic modelling it was further studied whether specific predictors can be identified for treatment failure following laparoscopic electrocautery and for treatment success following ovulation induction with rFSH.

The second part of the thesis describes three studies on ovarian hyperstimulation. In a retrospective study the effectiveness of hMG, uFSH and rFSH for ovarian hyperstimulation in IVF and ICSI cycles in women with PCOS was compared. A meta-analysis was performed to compare the effectiveness of hMG and rFSH for ovarian hyperstimulation in IVF and ICSI cycles in normogonadotrophic ovulating women. Finally, a review was performed to study whether the FSH isoform profile of gonadotrophin preparations is of clinical significance.

The aim of this thesis as outlined in chapter 1 was to answer six specific questions.

Chapters 2 and 3 addresses the first question

How does a laparoscopic electrocautery strategy compare with ovulation induction with rFSH in CC-resistant women with PCOS in terms of clinical outcome and costs?

To answer this question we performed a multicenter randomised controlled trial between 1998 and 2001. in 186 clomiphene citrate resistant women with PCOS. These women were allocated either to an electrocautery strategy entailing laparoscopic electrocautery of the ovaries followed by CC and rFSH if anovulation persisted, or ovulation induction with rFSH.

After randomisation the ovaries of women allocated electrocautery strategy were cauterised immediately. The cauterisation of the ovaries was performed with an Erbotom ICC 350 Unit using a bipolar insulated needle-electrode. Depending on the size of the ovary, 5-10 punctures were created on each ovary, distributed randomly over the surface. Clomiphene citrate was given when anovulation persisted within eight weeks following electrocautery or if anovulation reoccurred during follow-up. Women who did not ovulate on 150 mg CC received rFSH in a chronic low-dose step-up regimen. Women randomised to rFSH were treated in the same step-up protocol. In both study arms women were treated until six subsequent cycles were achieved within 12 months. The primary end point was ongoing pregnancy within 12 months.

Within a time span of 12 months, 56 of the 83 women (67%) in the electrocautery strategy group and 57 of the 85 women (67%) in the rFSH group reached an ongoing

pregnancy (RR 1.01, 95% CI: 0.81 to 1.24). In the electrocautery strategy arm 61% (228 of 375) of the cycles were ovulatory versus 69% (188 of 272) of the cycles in the rFSH arm.

No cases of ovarian hyperstimulation syndrome were observed and miscarriage rates were comparable in both treatment arms. However, multiple pregnancies only occurred after rFSH treatment, and as a result significantly less multiple pregnancies were found in the electrocautery strategy arm (n=1) than in the rFSH arm (n=9) (RR 0.11, 95% CI: 0.01 to 0.88).

An economic evaluation was set up alongside the multicenter randomised clinical trial. Data on resources used for treatment and productivity loss were collected prospectively up to an eventual ongoing pregnancy with a time horizon of 12 months. Mean total costs until an ongoing pregnancy per woman were € 5308 for the electrocautery strategy and € 5925 for treatment with rFSH, resulting in a mean difference of € 617 (95% CI € minus 382 to € 1614). A scenario-analysis without a diagnostic laparoscopy preceding rFSH treatment led to almost identical costs of the two treatment regimes. In that case the mean total costs in the rFSH group would be € 5371 with a resulting costs difference of only € 63 (95% CI minus € 945 to € 1070). As multiple pregnancies occurred only after rFSH treatment we subsequently estimated delivery costs of singleton and higher order pregnancies on basis of the literature. The estimated direct medical costs of treatment including the delivery costs in the electrocautery strategy group were 22% lower than in the rFSH treatment group (€ 11301 versus €14489).

In summary, with this study we have shown that both the electrocautery strategy and ovulation induction with rFSH are effective and safe treatment strategies with comparable cumulative ongoing pregnancy rates after 12 months at comparable costs. The major difference between the two strategies is that multiple pregnancies can largely be prevented by treating women with electrocautery and clomiphene citrate prior to rFSH.

Chapter 4 relates to the second question

What are predictors of ovarian response after laparoscopic electrocautery of the ovaries and of treatment outcome after laparoscopic electrocautery of the ovaries followed by clomiphene citrate?

To answer this question we performed two prognostic studies in the 83 CC-resistant women with PCOS that had been allocated to the electrocautery strategy as described in Chapter 2. We studied whether clinical, ultrasonographic and endocrine characteristics during initial screening of CC-resistant PCOS patients can predict ovarian response after laparoscopic

electrocautery of the ovaries. Following the results of our randomised clinical trial as described in Chapter 2, laparoscopic electrocautery has been put forward as treatment of choice in women with CC-resistant PCOS in the guidelines of the National Institute of Clinical Excellence of the British National Health Service. However, it is possible that certain women do not respond favourably to laparoscopic electrocautery. Therefore, it would be helpful if we could identify women with PCOS with a high probability of treatment failure following electrocautery of the ovaries. Treatment failure following laparoscopic electrocautery of the ovaries was defined as failure to ovulate within eight weeks after treatment.

From the set of clinical, ultrasonographic and endocrine characteristics the LH/FSH ratio came out as the most predictive variable for ovarian response after electrocautery. Women with a LH/FSH ratio below 2 were more likely to have persistent anovulation while women with a higher LH/FSH ratio were more likely to ovulate. Year of menarche was the second predictive variable for ovarian response, i.e. women who had their menarche before their 13th birthday were more likely have persistent anovulation than women who had their menarche later. Furthermore, chances to remain anovulatory appeared to decrease as fasting glucose levels increased. Although age at menarche and glucose level improved the model, their association with ovarian response was not significant.

The predictive model for persistent anovulation after electrocautery had an area under the Receiver Operating Characteristic curve of 0.74, expressing a modest discriminative performance. The model allowed a distinction between women with a poor chance to ovulate and women with a high chance to ovulate. Yet, the clinical value of the model is limited, as the next logical treatment in women with persistent anovulation is CC. Therefore, a model that predicts treatment failure after electrocautery followed by CC would be more relevant for clinical practice than predicting ovarian response after electrocautery. However, we were unable to identify a subgroup of women with a poor chance to reach an ongoing pregnancy after electrocautery followed by CC.

As laparoscopic electrocautery is now well established as the treatment of first choice for CC-resistant women with PCOS, our findings imply that no single woman should a priori be excluded from this treatment.

Chapter 5 focuses on the third question
Which patient characteristics can predict treatment outcome following ovulation induction with rFSH

To answer this question we performed a prognostic study in the 85 CC-resistant women with PCOS that had been allocated to ovulation-induction with rFSH as described in Chapter 2. We studied whether clinical, ultrasonographic and endocrine characteristics during initial screening of cc-resistant PCOS patients can predict treatment success after rFSH treatment. Treatment success after rFSH was defined as the occurrence of an ongoing pregnancy within one year.

The rationale behind this study was that although laparoscopic electrocautery is the preferable treatment, it is a surgical procedure under general anaesthesiology for which patients and/or gynaecologists might not opt. The counselling of these women would benefit from knowledge about the chances of success with rFSH in specific patients groups.

Oligomenorrhea, short duration of infertility and a low free androgen index (FAI) were favourable predictors for ongoing pregnancy, resulting in a predictive model with an area under the Receiver Operating Characteristic curve of 0.72, expressing a modest discriminative performance. Furthermore, the model allowed distinction between women with a poor prognosis and women with a good prognosis. Women with a probability below 5% to reach an ongoing pregnancy were those who had amenorrhea, an infertility duration over 3 years and a FAI above 9. Women with a probability above 25% to reach an ongoing pregnancy rate typically had oligomenorrhea, an infertility duration of less than 2 years and a FAI below 9.

Chapter 6 relates to the fourth question
What is the effectiveness of hMG, uFSH and rFSH for ovarian hyperstimulation in IVF and ICSI cycles in women with PCOS?

To compare the effectiveness of hMG, uFSH and rFSH in women with PCOS a historical cohort study was performed at the AMC. All women with PCOS treated by ovarian hyperstimulation for IVF or ICSI between 1993 and 2003 were included in the cohort.

Thirty-six women with PCOS underwent 87 attempts with hMG, 11 women underwent 22 attempts with uFSH and 38 women underwent 93 attempts with rFSH. Data were analysed with multivariable logistic regression models correcting for clinical and treatment differences between the three groups. There were no significant differences in live birth (OR

2.15; 95% CI: 0.77 to 5.90). The hMG group however, had a significantly higher probability of conception than the rFSH group (OR 2.81; 95% CI 1.11 to 5.81).

To further control for the bias inevitably introduced by the retrospective design of this study a reference population was formulated existing of 425 IVF attempts in 170 women treated just before and just after each first cycle of each woman in the historical cohort.

Usage of the three gonadotrophins was represented by different time periods. In the PCOS and the reference population hMG was used until 1997, uFSH was used from 1997 until 2000 and rFSH was used from 1997 onwards. In the reference population more live births occurred in the rFSH group than in the hMG group, while in women with PCOS live birth rates were lower in the rFSH group compared to the hMG group although this difference did not reach statistical significance. Numbers were too small to determine whether there was a significant difference in live births or conceptions in women treated with uFSH compared to hMG and rFSH.

In summary, this retrospective cohort study suggests that ovarian hyperstimulation with hMG is as effective as rFSH in terms of live birth in women with PCOS undergoing IVF or ICSI cycles.

Chapter 7 focuses on the fifth question

How do hMG and rFSH compare in terms of clinical outcome when used for ovarian hyperstimulation in IVF and ICSI cycles in normogonadotrophic ovulating women ?

To answer this question we performed a systematic review and meta-analysis of randomised clinical trials comparing hMG and rFSH for controlled ovarian hyperstimulation in down-regulated women undergoing IVF or ICSI. This review relied on the search strategy developed for the Cochrane Menstrual Disorders and Subfertility Group and described in the Cochrane database. To identify relevant trials we searched the Cochrane Menstrual Disorders and Subfertility Group trials register, PubMed, MEDLINE and Web of Science. In addition, cross-references from all identified articles were checked.

Six trials were included entailing 1030 women. In each of the six trials, the treatment direction for clinical pregnancy per woman was in favour of hMG. Pooling the five trials that used a long GnRHa protocol, resulted in a higher clinical pregnancy rate for hMG compared to rFSH (RR: 1.22; 95% CI: 1.03 to 1.44). Excluding one quasi-randomised trial, the pooled RR for clinical pregnancy was 1.19 (95% CI: 1.00 to 1.42) for hMG versus rFSH. However, there was no evidence of a difference in ongoing pregnancy or live birth per woman between hMG and rFSH (RR: 1.20; 95% CI: 0.99 to 1.45). No differences were found in gonadotrophin dose used, oocytes retrieved, miscarriage and multiple pregnancy.

In summary, use of hMG resulted in higher clinical pregnancy rates than did use of rFSH in IVF/ICSI cycles after GnRH agonist down-regulation in a long protocol.

Chapter 8 addresses the sixth question
Is the isoform profile of FSH preparations of clinical significance?

A review was performed of information on the FSH molecule and its isoforms, the FSH isoform profile of gonadotrophin preparations and observed differences in bioactivity of FSH isoforms in FSH preparations. The FSH molecule is not just a single entity but exists as a number of different isoforms. In humans 20 different FSH isoforms have been identified. The more acidic isoforms contain high number of sialic acid residues reflecting a more complex branching pattern, whereas less-acidic isoforms have fewer sialic acid residues often reflecting lack of branching of the carbohydrates moieties. The difference in acidity can be used to separate isoforms of FSH according to their electric charge.

Most studies suggest that the more acidic isoforms possess a reduced *in vitro* bioactivity compared to the less-acidic isoforms. The amount of FSH isoforms with an isoelectric point below 4, i.e. the more acidic isoforms, is usually higher in urine derived preparations as compared to the recombinant products. The isoform distribution of rFSH is more basic and resembles uFSH more closely than highly purified uFSH, which is more acidic.

Each FSH preparation is characterised by a specific isoform profile that relates to the charge heterogeneity, the complexity of the oligosaccharides attached to the peptide back-bone, and the *in vitro* bioactivity. To further explore whether the FSH isoform composition may influence the clinical effectiveness of the preparation we pooled the data of randomised clinical trials that compared urinary-derived highly purified FSH with a highly acidic isoform profile (Metrodin-HP) and rFSH rich in less-acidic isoforms (Gonal F) in a meta-analysis, with ongoing pregnancy as primary outcome. A literature survey in PubMed, MEDLINE and EMBASE (all searched 1985 to March 15 2004), identified 5 studies that specifically compared Metrodin-HP with Gonal F for ovarian hyperstimulation in IVF or ICSI cycles in women that were down-regulated using a standard GnRHa protocol. The results of these five trials could be pooled.

A difference in ongoing pregnancy per woman could not be proven but the direction of the effect was in favour of rFSH (OR 1.27, 95% CI 0.98 to 1.65). In the rFSH group a significantly lower amount of gonadotrophin was used (WMD: minus 3.0 ampoules, 95% CI minus 3.8 to minus 2.3) and the duration of treatment was shorter (WMD: minus 1.2 days, 95% CI minus 1.4 to minus 0.9) as compared to the highly purified uFSH group. Furthermore, in the rFSH group a significant increased number of follicles (>10 mm on the

day of hCG) developed (WMD: 2.3 , 95% CI 1.6 to 2.9) and an increased number of oocytes was retrieved (WMD: 2.3 , 95% CI 1.6 to 2.9) as compared to the highly purified uFSH group.

The lower gonadotrophin dose used, shorter duration of treatment and increased number of follicles and oocytes in the rFSH group compared to highly purified uFSH point to a higher in vivo bioactivity of rFSH. However, a difference in effectiveness in terms of an ongoing pregnancy per woman could not be proven.

Conclusions

1. The laparoscopic electrocautery strategy and ovulation induction with rFSH are equivalent in terms of ongoing pregnancy and costs within a time horizon of one year. However, the electrocautery strategy leads to significantly less multiple pregnancies and should therefore be the treatment of choice.
2. Persistence of anovulation after electrocautery can be predicted and women with a high risk of persisting anovulation could be distinguished. However, parameters that predict failure to reach an ongoing pregnancy after laparoscopic electrocautery followed by clomiphene citrate cannot be identified and hence, a subgroup of women with a poor chance to reach an ongoing pregnancy after electrocautery followed by CC cannot be identified. As laparoscopic electrocautery is now well established as the treatment of first choice for CC-resistant women with PCOS, our findings imply that no single woman should a priori be excluded from this treatment.
3. Treatment success after ovulation induction with rFSH and ovarian response after treatment with laparoscopic electrocautery can be predicted. A model consisting of oligo/amenorrhea, duration of infertility and FAI level allows distinction between women with a poor prognosis, and women with a good prognosis to reach an ongoing pregnancy.
4. There is no evidence for the superiority of rFSH for ovarian hyperstimulation in women with PCOS.
5. Ovarian hyperstimulation with hMG leads to more clinical pregnancies than rFSH in women undergoing IVF or ICSI with GnRHa down-regulation in a long protocol, although differences are small.
6. A difference in ongoing pregnancy per woman between a FSH product with a highly acidic isoform composition (Metrodin-HP) compared to a less-acidic isoform composition (Gonal-F) could not be proven in women undergoing IVF or ICSI with GnRHa down-regulation in a long protocol. However, the lower gonadotrophin dose used, shorter duration of treatment and increased number of follicles and oocytes in the Gonal-F group compared to Metrodin-HP group do point to a higher *in vivo* bioactivity of Gonal-F.

Implications for future research

Clomiphene citrate is still the treatment of first choice in women with PCOS and CC-resistance is a relevant policy problem. Therefore it would be sensible to direct our research efforts to preventing CC-resistance. During the last years it has become common practice to use metformin, generally as co-treatment, for ovulation induction in women with polycystic ovary syndrome. The insulin-sensitiser metformin appears to enhance successful ovulation induction with CC.[1] However, definite evidence for its effectiveness is still lacking. Whether standard treatment with CC is truly enhanced by concurrent administration of metformin and whether this addition can prevent clomiphene-resistance in women with PCOS is currently being studied in large randomised placebo controlled trials.

Prediction models for success and failure after clomiphene citrate are available although their validity still has to be evaluated.[2,3] It would be interesting to make a prediction model for failure after CC in combination with metformin when more evidence becomes available of the effectiveness of that strategy.

Based on results described in this thesis laparoscopic electrocautery has been put forward as treatment of first choice in women with CC-resistant PCOS. It would be of interest to know whether metformin can also enhance the effectiveness of electrocautery. Therefore a, preferably placebo-controlled, trial with adequate power should be performed comparing electrocautery with and without metfomin.

At present, most western countries use rFSH for IVF and ICSI in down-regulated normogonadotrophic women. This thesis contains evidence that rFSH may not be the most effective gonadotrophin. Do women benefit from exogenous LH after all?

Studies that looked at the added value of recombinant LH (rLH) to rFSH stimulated cycles have produced contradicting results. Age may have been a confounding factor in these studies. There are indications that additional rLH results in higher implantation rates in women aged 35 years or older while younger women may not benefit from rLH.[4,5] Furthermore rLH appears to be beneficial in women with poor response to rFSH.[6]

How can the addition of exogenous LH lead to higher implantation and pregnancy rates in these subpopulations of women? One possible explanation could be an intrinsic effect of LH on the endometrium. However, exogenous LH also had an impact on implantation rate in an oocyte donation program.[7] A more likely explanation is the effect on the oocyte- and embryo competence. LH is known to play a role in oocyte growth and maturation and subsequently may play a relevant role in optimising fertilisation and embryo competence. When added in the right amounts LH has been reported to increase the number of good morphology cleaving embryos[8] or to increase the blastocyst rate.[9-11]

It will be interesting to examine whether the addition of rLH to standard treatment with rFSH results in more competent embryos than treatment with rFSH only for ovarian hyperstimulation in women below and above 35 years of age.

Currently most ovarian hyperstimulation regimens employ just a single FSH preparation throughout the entire period of stimulation. With this approach a woman will only be exposed to the FSH isoform profile of the specific product used. In contrast, the isoform profile of the FSH that granulosa cells experience during the follicular phase of a natural cycle changes considerably, being most acidic in the early follicular phase when follicles are recruited and less-acidic as ovulation approaches and selection of the dominant follicle has taken place. By first administering a more acidic FSH preparation during the early follicular phase followed by less-acidic preparations at the mid-follicular phase we may be ably to mimic the natural follicular phase more closely. It would be of interest to study this approach in a randomised clinical trial.

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