# Women's experience of pain and pain relief in assisted reproductive technology

Ann-Louise Gejervall


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Department of Obstetrics and Gynecology
Institute of Clinical Sciences
The Sahlgrenska Academy, Göteborg University
Göteborg, Sweden

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# CONTENTS

# ABSTRACT

The overall aim of this thesis was to evaluate and compare different methods for pain relief during oocyte retrieval and to study women's expectations and experiences of pain during oocyte retrieval in conjunction with in vitro fertilization.
Paper I, an open prospective randomized controlled trial, including 160 women had the primary aims of comparing the pain relieving effects of electro-acupuncture and conventional analgesia, comprising opiates, in conjunction with oocyte retrieval, and to compare post-operative well-being between groups. For measurements of pain the Visual Analogue Scale (VAS) was used and post-operative well-being was assessed using the State Trait Anxiety Inventory test. Our findings showed that women who used electro-acupuncture had significantly more pain during surgery than women who received conventional analgesia. Well-being between groups was comparable.
In paper II, a prospective single blinded randomized multicentre study with a total of 183 women two techniques for local anesthesia were compared; pre-ovarian block and paracervical block, in combination with conscious sedation. VAS and the McGill Pain Questionnaire were used for pain ratings. No significant difference between paracervical block and pre-ovarian block was found in terms of pain relieving effects. No differences in fertilization and embryo development were observed.
Study III, an observational two-centre study of 124 women, evaluated women's expectations regarding pain before oocyte retrieval and whether their experienced pain was in accordance with the expected pain. VAS and multiple choice questions of our own construction were used for measurements. It was found that women experienced significantly less pain during oocyte retrieval than they expected before surgery.
Study IV was a retrospective study evaluating the effects of analgesic drugs used at oocyte retrieval, in particular different doses of alfentanil, on fertilization rate and/or embryo quality. A total of 663 women were included. Data was collected from the clinical database at Reproductive Medicine, Sahlgrenska University Hospital and from the women's hospital records. No differences in fertilization rate or embryo quality were observed in relation to the amount of drug used for analgesia.

*In conclusion,* the results of these studies showed that electro-acupuncture cannot generally be recommended as a general pain relief method for oocyte retrieval, but might be used as an alternative for women desiring a non-pharmacological method (Paper I). One advantage of electro-acupuncture was significantly less tiredness and confusion compared with conventional analgesia.
Both pre-ovarian block and paracervical block offered good pain relief and were considered safe methods with rapid recovery and ease of administration and monitoring (Paper II).
Since women experienced significantly less pain in conjunction with oocyte retrieval than they expected before surgery, this is important information for women who are about to start IVF. It might reduce their apprehension about pain levels during the procedure (Paper III). High doses of alfentanil compared to low doses were not associated with any adverse effects on fertilization rate, embryo development or clinical pregnancy rate, assuring that women can be offered adequate pain relief (Paper IV).

*Keywords: oocyte retrieval, pain relief, randomized controlled trial, pain, fertilization rate, embryo development, Visual Analogue Scale*

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# LIST OF PUBLICATIONS

**I. Electro-acupuncture versus conventional analgesia; a comparison of pain levels during oocyte aspiration and patient's experiences of well-being after surgery.**
Gejervall A-L, Stener-Victorin E, Möller A, Janson PO, Werner C and Bergh C.
*Human Reproduction 2005;20;3:728-735.*

**II. Pre-ovarian block versus paracervical block for oocyte retrieval.**
Cerne A, Bergh C, Borg K, Ek I, Gejervall A-L, Hillensjö T, Olofsson J I, Stener-Victorin E, Wood M and Westlander G.
*Human Reproduction 2006;21;11:2916-2921.*

**III. Pain aspects in oocyte aspiration for IVF.**
Gejervall A-L, Stener-Victorin E, Cerne A, Borg K and Bergh C.
*Reproductive BioMedicineOnline 2007;14;2:184-190.*

**IV. Effects of alfentanil dosage during oocyte retrieval on fertilization and embryo quality.**
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# ABBREVATIONS AND DEFINITIONS

| | |
|---|---|
| **ART** | Assisted reproductive technology |
| **CA** | Conventional analgesia |
| **EA** | Electro-acupuncture |
| **FSH** | Follicle stimulating hormone |
| **GABA** | γ-amino-butyric acid |
| **HPA** | Hypothalamus-pituitary-adrenal axis |
| **ICSI** | Intracytoplasmic sperm injection |
| **NRM** | Nucleus Raphe Magnus |
| **ITT** | Intention-to-treat |
| **IVF** | In vitro fertilization |
| **PAG** | Periaqueductal grey |
| **PCB** | Paracervical block |
| **POB** | Pre-ovarian block |
| **RCT** | Randomized controlled trial |
| **STAI** | State Trait Anxiety Inventory test |
| **VAS** | Visual Analouge Scale |

**Pain** Definition according to the International Association for the Study of Pain: "an unpleasant sensory and/or emotional experience associated with actual or potential tissue damage, or described in terms of such damage."

# INTRODUCTION

***Comments and experiences from some women in recovery on the day of oocyte retrieval:***

**"Everything went OK and the oocyte retrieval was not as painful as I expected."**
**"I am happy and relieved."**
**"I was panic-stricken."**
**"The local anesthesia hurt and the retrieval from the second ovary was painful otherwise it was OK."**
**"Thanks for all the consideration."**
**"The pain was tolerable but psychologically it was exhausting."**
**"I felt tense during the retrieval."**
**"The most effective pain relief was the feeling of support from the midwife; she devoted all her time to my well-being.**"
**"Some pain and discomfort but probably to a normal extent."**

Oocyte retrieval is the most painful part of an in vitro fertilization (IVF) treatment; while most women tolerate the pain well some women experience intolerable pain and great discomfort during the procedure. Pain perceived in conjunction with oocyte retrieval is usually described in terms of intensive menstrual pain. The pain is caused by the passage of the aspiration needle through the vaginal wall and the ovary capsule, and by mechanical stimulation of the ovary. The process may be more painful if the ovaries have adhesions or if they are stuck for example in the pouch of Douglas, behind or on top of the uterus. Pain during oocyte retrieval is more intermittent than continuous (Zelcer *et al*., 1992). When evaluating if women are satisfied with their pain relief during oocyte retrieval, studies show that women rate the degree of satisfaction as high although the pain levels are also high (Kwan *et al*., 2005). Infertility and IVF treatment itself are major stressors (Baram *et al*., 1988, Anderheim *et al*., 2005, Holter *et al*., 2006). Particularly stressful events in an IVF cycle include waiting for the results of fertilization, waiting for pregnancy particularly after, one or more unsuccessful treatment cycles or a terminated pregnancy. In one study (Hammarberg *et al*., 2001), 52% of women rated oocyte retrieval as extremely stressful or very stressful. Facilitating the oocyte retrieval procedure and minimizing pain and discomfort are important goals in IVF.

## Infertility

The World Health Organization (WHO, 2002) has identified infertility as "failure to become pregnant after one year of unprotected intercourse" and has recognised infertility as a reproductive

health disease. For many people, infertility is more than a disease. It may also be a social and public health issue and an individual problem. The prevalence of involuntarily childlessness, primary and secondary, varies from area to area, but in industrialised countries it is considered to affect 10-15% of couples of reproductive age.

Infertility is related to male or female factors or mixed factors, and in some cases infertility is unexplained. Common causes of infertility in females are ovulatory disorders, tubal factors, cervical factors, hormonal disturbances or endometriosis. Reasons for male infertility include abnormal sperm production involving a reduced number of sperm and/or decreased motility. In male disorders, azoospermia is presented in up to 10% of male infertility and caused by abnormal spermatogenesis, disorders of secretory function or genital tract obstruction (De Croo *et al*., 2000).

Reproduction has been a central issue for human beings since time in memorial. Today, couples in industrialised countries tend to postpone child birth and reduce family size. In Sweden, for first-time mothers the median age has increased by 4 years over the last 30 years (The National Board of Health and Welfare, 2005). Several studies have shown that couples postpone the birth of the first child until they feel prepared socially and financially to assume the responsibilities of parenting (Rasch and Knudsen, 2001, Lampic *et al*., 2006). In a Swedish study (Lampic *et al*., 2006) 47% of women intended to have children after 35 years of age and they were not aware of the age-related decline in female fecundity in the late 30s (Dunson *et al*., 2002). The decision to delay childbirth might increase the demand for medical advice and services for fertility treatment.

## Assisted reproductive technology

Assisted reproductive technology (ART) encompasses a wide range of techniques used for infertility treatment, which can be defined as treatments that include in vitro handling of human gametes for the purpose of establishing a pregnancy. The term ART includes techniques such as IVF and intra-cytoplasmic sperm injection (ICSI), embryo freezing, pre-implantation genetic diagnosis (PGD), gamete donation, and surrogate motherhood. The International Committee for Monitoring ART - World Health Organization (ICMART-WHO) - has not classified intrauterine insemination (IUI) as ART. However, in Europe, the European IVF Monitoring Consortium (EIM) has included IUI under ART because it involves fertility treatment and predisposes for risks such as multiple pregnancies (Andersen *et al*., 2007). Additionally, the European Union Tissue and Cell Directive have determines that all clinics performing ART must undergo accreditation (Directive, 2004).

The field of ART has been rapidly expanding since the first IVF child was born in England in 1978 (Steptoe and Edwards, 1978). In Scandinavia, the first IVF child was born in 1982 in Göteborg. Today, IVF is superior for infertility treatment, of both female and male origin. In 1992, Palermo *et al.* (Palermo *et al*., 1992) reported that the first child, using

ICSI, was born in Brussels, Belgium and soon after, in 1993, the first ICSI child was born in Scandinavia, in Göteborg. Worldwide more than 3.3 million children are estimated to have been born after IVF (Adamson *et al*., 2006).

The development of ART and other new technologies raises a number of ethical and social issues as well as medical and financial questions. Human gametes and embryos used for fertilization outside the body are available for testing, manipulation and research. In many countries, however, there is an absence of appropriate guidelines and regulations. Concerns about the safety and efficacy of ART have led a number of countries to collect data at national level reporting on outcome of IVF treatment. In Europe, the EIM reports the European results for ART collected mainly from existing national registers (Andersen *et al*., 2007). One problematic issue pointed out by EIM is that the quality of data varies between countries because of differences in data collection systems, coverage, definitions and validation. Totally, in Europe, during 2004, 785 clinics from 29 countries reported 367, 066 cycles performed using different types of ART (Andersen *et al*., 2007). In comparison, about 128, 000 cycles were reported from the USA in 2004 (Wright *et al*., 2007).

## IVF in Sweden

In Sweden, the legalisation concerning ART dates from 2002 (SOFS 2002:13). The National Board of Health and Welfare have registered data from the Swedish IVF clinics since 1982. This register includes variables such as infertility reason, age of women, number of cycles performed, number of oocyte retrievals performed and fertilization method used (IVF/ICSI). Further, number of embryos transferred, for single embryo transfers presented as number of elective and non-elective single embryo transfers, pregnancy rate, number of fetal sacks per pregnancy and live birth rate, presentation of number of singletons and multiple birth rates. Accumulated data has been collected per clinic on an annual basis. Stable pregnancy and delivery rates are noted, despite a decrease in the number of embryos transferred, resulting in a dramatic decrease in the multiple pregnancy rate (Karlström and Bergh, 2007). In 2007, about 9000 IVF treatments and 4000 frozen-thawed ET cycles were performed in Sweden (personal communication, P.O Karlström, 2008), and 3% of all deliveries were children born after IVF. The live birth rate is approximately 25% for fresh embryos, when using frozen-thawed embryos the corresponding rate is approximately 18%. Since 2007 a new National Quality Registry is established in Sweden. This registry is like other quality registries in Sweden, individually based and includes identified variables for all women and men undergoing IVF. This registry will be an important tool for follows of quality in IVF.

In 2008, there are 16 IVF clinics in Sweden, half of them are public funded and half of them are private clinics. IVF treatment is available for couples who live in stable relationships and have no children in the current relationship (the

last only for public clinics). The majority of regions in Sweden finance 2-3 IVF cycles but some regions have restricted treatment, allowing only one cycle. In early IVF the success rates were poor and several embryos were transferred to achieve a pregnancy. As IVF results gradually improved, multiple birth rates increased and became a problem. Today, among couples treated with ART worldwide, the multiple birth rates are approximately 25%.

The most important factor influencing the rate of multiple births is the number of embryos transferred. In Sweden, IVF clinics voluntarily shifted from three to two embryos in 1993, which eliminated most triplet pregnancies, although the twin rate still remained high. Studies on identifying women suitable for elective single embryo transfer (eSET) have been performed (Coetsier and Dhont, 1998, Strandell *et al*., 2000). Salumets *et al.* (Salumets *et al*., 2006) found the age of the woman and the embryo quality to be the most strongly predictive factor for multiple births and live birth rates. The first report concerning eSET came from Finland (Vilska *et al*., 1999). This observational study showed, for selected patients, similar pregnancy rates with one as with two embryos. A large randomized multi-centre study was initiated in Scandinavia in 2003 (Thurin *et al*., 2004). The aim was to show equivalence concerning live birth rates when comparing one fresh single embryo + one frozen-thawed embryo (1+1) with one fresh double embryo transfer (DET), (2+0). The study showed that cumulative live birth rates after one fresh and one frozen SET were not substantially lower than after one fresh DET. In 2003, parallel with the Scandinavian SET study, new guidelines recommendations from the Swedish National Board of Health and Welfare stated that eSET should be the method of choice.

In a review article (Bergh, 2005), including four randomized controlled trials (RCT) and seven observational studies, eSET was evaluated. Results from RCTs showed significantly lower birth rates after eSET than with DET, while the retrospective observational studies demonstrated similar live birth rates between eSET and DET. Another review article (Pandian *et al*., 2005) reported that eSET reduced the multiple birth rates, but also the live birth rate. If a frozen-thawed embryo was subsequently replaced comparable live birth rates as with DET were achieved. Lundin and Bergh (Lundin and Bergh, 2007) showed that it is possible to maintain similar live birth results and decrease multiple birth rates using eSET in a majority of women, but a higher number of frozen-thawed embryos were needed. A national wide report from Sweden (Karlström and Bergh, 2007) showed a dramatic decrease in multiple birth rates from 35% to about 5% when fewer embryos were replaced. Delivery rate were maintained at around 26%.

### Follow up of IVF children

Numerous publications have reported less favourable outcomes for IVF children compared to children born after spontaneous conception (Bergh *et al*., 1999, Helmerhorst *et al*., 2004, Jackson *et al*., 2004, Wennerholm and Bergh, 2004). An IVF registry with all children born after IVF was established in Sweden in the early 90ies. This registry has been cross linked with other population based registries (Medical Birth Registry, Cancer Registry, Malformation registry, Hospital discharge registry and Cause of death registry). The results from these crosslinkings have found that the main complications associated with IVF are multiple pregnancies and multiple births, with an increased risk for prematurity, low or very low birth weight and perinatal death (Bergh *et al*., 1999). An increased risk of prematurity and small-for-gestational age has however also been noted for IVF singletons (Bergh *et al*., 1999, Koudstaal *et al*., 2000).

In a Swedish population-based study, IVF children were found to be at increased risk of neurological complications (Strömberg *et al*., 2002). One explanation is the high frequency of twins born, but the IVF procedure per se or other factors not adjusted for cannot be excluded.

A slight increased risk for congenital malformations among IVF/ICSI children has been shown, in controlled studies and meta-analysis, as compared with spontaneously conceived children (Hansen *et al*., 2005, McDonald *et al*., 2005, Rimm *et al*., 2004). Whether the increased risk is due to the ovarian culture technique or the ovarian stimulation, or whether infertility *per se* is a risk factor, is still not clear. Studies published to date suggest the latter explanation; when risk figures have been adjusted for parental characteristics, the differences have no longer been significant. No difference in malformations between IVF and ICSI was found in a large Swedish register study (Källén *et al*., 2005).

## Pain

Conceptions and causes of pain have many explanations. Pain has thought to be caused by the intrusion of objects or spirits in the body, by magic influences from the dead or evil spirits or as a consequence of sin.

The International Association for the Study of Pain (IASP) (Merskey and Bogduk, 1994) defines pain as:

**”An unpleasant sensory and/or emotional experience associated with actual or potential tissue damage or described in terms of such damage.”**

According to the IASP definition, there is no distinct link between pain and injury.

### Pain perception

Pain is considered a subjective experience with a number of dimensions. It is influenced by physiological, psychological, psychosocial and cultural factors. A painful experience is a complex entity with sensory, involved in pain perception act in a serial and a parallel way, discriminating and locating the original

stimulus, and integrating the affective feelings (Almeida *et al*., 2004). The great variability and complexity makes it complicated to compare pain in different individuals. The ability of a person to cope with pain is influenced by his or her personality. Expectations and desires are important psychological mediators of pain, underlying common human emotions, such as sadness, anxiety and relief (Price and Barrell, 2000). Individuals with depression and sleep disturbances have lower pain thresholds than individuals who do not suffer from these problems (Chiu *et al*., 2005). Memories of previous experiences are considered to affect an individual's dealing with pain (Hampton, 2005). Price *et al.* (Price *et al*., 1999) found that individuals remembered pain intensity as much greater than it actually was.

The knowledge of neurobiological and psychological mechanisms of placebo effects has increased during recent decades. Wager *et al.* (Wager *et al*., 2004) found that placebo analgesia is related to altered neural activity in pain processing areas in the brain. Decreased neural activity was also correlated with reduction in pain ratings. In a recently published study, Price *et al.* (Price *et al*., 2008) summarised that placebo response is associated with true neurobiological response. Currently, differences among women's and men's responses to pain and opiates are being studied (Dahan *et al*., 2008, Hurley and Adams, 2008). The underlying mechanisms why women and men respond different are not clear, although both biological (i.e. age, gonadal hormones and menstrual cycle) and psychosocial factors (such as sex and sex roles) are considered to be involved. Women have been reported to have greater sensitivity to pain intensity then men (Levine *et al*., 2006). Smith *et al.* (Smith *et al*., 2006) found lower pain thresholds in women during the low estradiol phase of the menstrual cycle.

Since the mid-1960s, the perception of pain has been separated into three dimensions (Table 1), (Fernandez and Turk, 1992, Melzack and Wall, 1965, Price *et al*., 1987):

**Table 1.** Dimensions of pain perception

| ***Dimensions of pain*** | ***Description and localisation*** |
|---|---|
| **Sensory-discriminative** | Intensity, duration and localisation of pain, projects to sensory cortex. |
| **Affective-emotional** | Emotional and behaviour response to pain, projects to sensory cortex. |
| **Cognitive-evaluating** | Previous experiences, thoughts and ideas, projects to pre-frontal cortex. |

**Table 2.** Classification of pain

| | |
|---|---|
| **Nociceptive pain** | Transient pain in response to a noxious stimulus into an intact nervous system. |
| **Inflammatory pain** | Spontaneous pain and hypersensitivity to pain in response to tissue damage and inflammation. |
| **Neuropathic pain** | Spontaneous pain and hypersensitivity to pain in association with damage to or a lesion of the nervous system. |
| **Functional pain** | Hypersensitivity to pain resulting from abnormal central processing of normal input. |

## Pain classification

The neurobiological mechanisms and the distinct types of pain are classified into different subgroups based on etiological factors (Table 2), (Woolf, 2004).

The innervations to uterus and vagina come from somatic segments; Thoracic spine12-Lumbar spine 2, Sacral spine 2-4 (Bonica, 1999). Pain perceived at oocyte retrieval is acute nociceptive pain caused by the passage of the aspiration needle through the vaginal wall and the ovary capsule, and by mechanical stimulation of the ovary.

The nociceptive system is a specialised high-threshold sensory system that mediates noxious stimuli and constitutes the sensory experience in acute pain. The system extends from the periphery through the spinal cord, brain stem and thalamus to the cerebral sensory cortex, where the sensation is perceived. The nociceptive system can be described as an alarm system that announces the presence of potential damage and promotes healing of the injured tissue (Woolf, 2004). Damage tissue increases sensitivity to pain, which prevents contact with or movement of the injured part until repair is complete. Multiple mechanisms producing pain have been identified (Julius and Basbaum, 2001, Woolf and Salter, 2000). The main mechanism is perception of noxious stimuli. Nociception is initiated in the peripheral terminals of the nociceptors, specialised neurons that respond to intense stimuli that may cause tissue damage. The nociceptors respond to mechanical pressure and to thermal and chemical stimuli, as when the aspiration needle penetrates the vaginal wall. Nociceptors activate myelinated (Aδ-fibres) and unmyelinated (C-fibres) axons into the dorsal root ganglion, and make synaptic contact with neurons in the dorsal horn which projects pain via multiple ascending pathways (tractus spinothalamicus, tractus spinomesencephalicus and tractus spinoreticularis) to the thalamus, and from the thalamus to the sensory cortex. The

pain sensation mediated from Aδ-fibres is sharp and distinct and may produce withdrawal or flexion reflexes. The C-fibres give rise to the secondary, aching or burning pain sensation, and are often referred to a large area even when the stimulus itself is localised. The somatosensory input and divergence to several areas in the brain processes and represents distinct dimensions of pain, i.e. pain sensation and pain unpleasantness. The somatosensory cortex processes the sensory-discriminative dimensions of pain such as pain intensity, location and duration. The affective and cognitive components of pain are projected to the limbic cortical areas and to the frontal cortex, and process avoidance and the unpleasantness associated with pain. The nucleus amygdale represents the autonomic responses of threatening stimuli that participate in parts of the affective dimension of pain. This also affects the initial feelings associated with acute pain (Price and Verne, 2002).

## Pain control

Pain is controlled by several endogenous inhibitory mechanisms that govern pain transmission to the different areas in the brain. These mechanisms are only briefly presented in this thesis. Main mechanisms for pain control are via 1) peripheral modulation, 2) segmental modulation in the spinal cord, 3) activation of descending inhibiting neuronal pathways, 4) psychological mechanisms, and 5) diffuse noxious inhibit control.

**1) Periphery level**

In the periphery, administrating e.g local analgesic will block local nociceptors and transmission to the spinal cord. Acupunture exerts effect from antidrome nerve impulses that in turn induce release of neueropeptides such as Substance P, vasoactive intestinal polypeptide, and calcitonin gene-related peptide, resulting in vasodilatation and increased nutrition blood flow in the periphery (Dawidson *et al*., 1997, Lundeberg, 1996, Sato *et al*., 2000).

**2) The gate control theory**

Melzack and Wall's (Melzack and Wall, 1965) concept of the gate control theory include pain modulation in the dorsal horn via gates that inhibit transmission of pain to the cortex. An open gate means that pain reaches the brain, while a closed gate inhibits pain in the dorsal horn. The gate control theory involves activation of sensory fibres (Aβ) by touch, pressure and vibration. When entering the dorsal horn interneuron's are activated and release γ-amino-butyric acid (GABA), which in turn inhibit transmission in Aδ-fibres and C-fibres both pre and postsynaptic, and pain is reduced.

**3) The descending pain inhibitory system**

Activation of Aδ-fibres and possibly C-fibres stimulate the descending system which is outgoing from neurons in the midbrain and in the brainstem nuclei; periaqueductal grey (PAG) to produce β-endorphin and nucleus raphe magnus

(NRM) to produce serotonin (5-HT) and noradrenalin (NA) which in turn project to the dorsal horn. In the dorsal horn interneuron's are activated that release encephalin and dynorfin which in turn inhibit transmission of pain.

**4) Psychological mechanisms**

Psychological mechanisms activate the endogenous inhibitory system in different ways. Stress increases the activity in the hypothalamus-pituitary-adrenal (HPA) axis. The limbic system, which represents the cognitive and affective dimensions of pain, modulates hypothalamus and are involved in the pain control (Price, 2000) (Fig. 1, 2).

**5) Diffuse noxious inhibit control**

Diffuse noxious inhibit control is a phenomenon that occur in relation to strong sensory inflow via activations of the nociceptive afferents and supraspinal pain inhibitory centres, which in turn project to the dorsal horn at every segmental level. Pain modulation is unspecific and is not related to the site of stimulation (Tousignant-Laflamme *et al*., 2008, Le Bars *et al*., 1979).

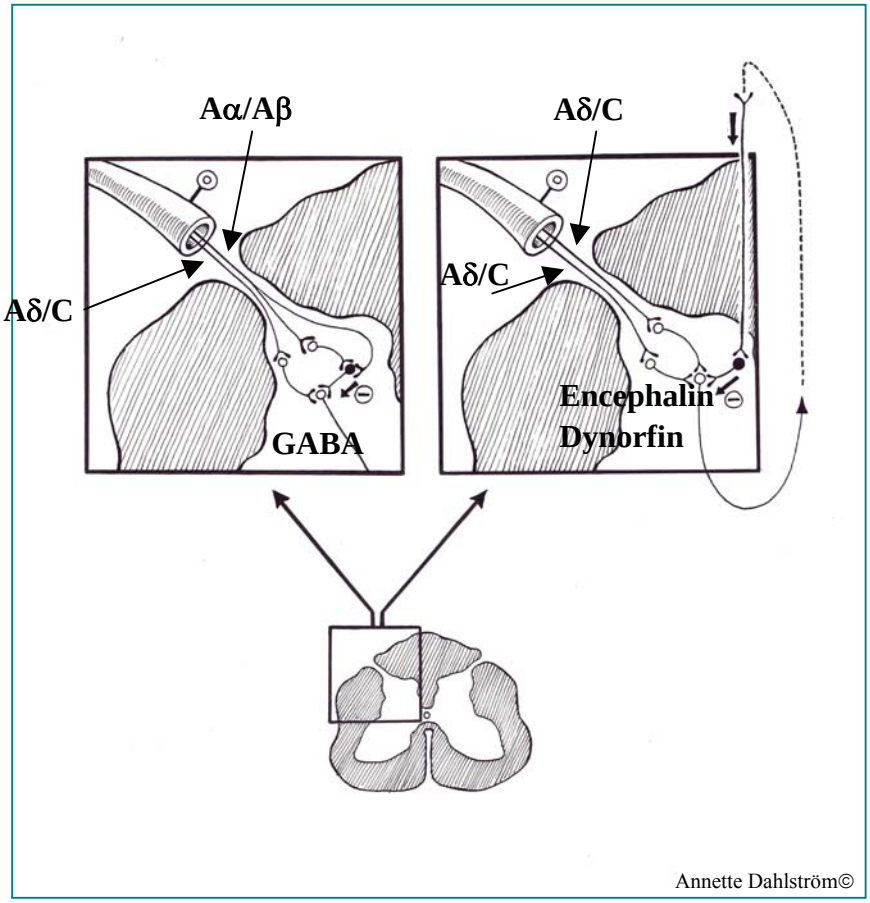


**Figure 1.** Pain modulation in the dorsal horn via gate control.

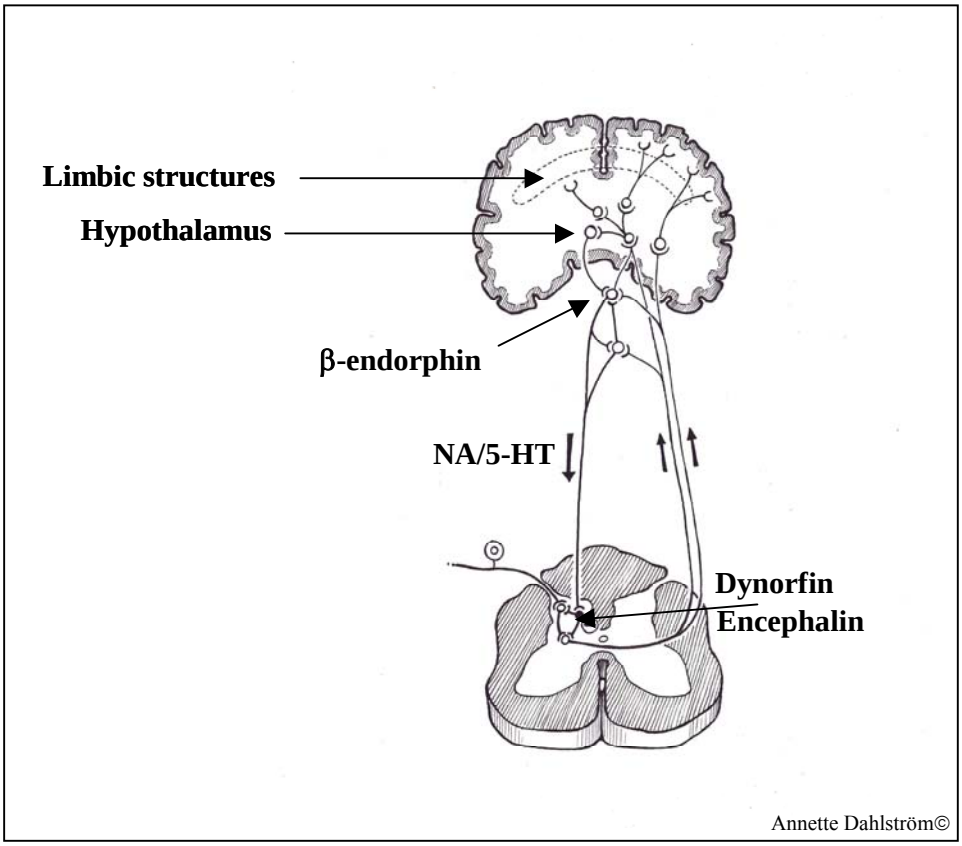


**Figure 2.** Pain modulation via descending pain inhibitoring systems.

## Pain reliving methods used in conjunction with oocyte retrieval

A satisfactory analgesic method for oocyte retrieval must provide adequate pain relief with rapid onset, rapid recovery and ease of administration and monitoring (Trout *et al*., 1998), and with no adverse toxics effect on the oocytes. The choice of analgesic has been influenced by concerns for potential adverse effects on fertilization and embryo development (Palot *et al.*, 1989, Wikland *et al*., 1990, Coetsier *et al*., 1992, Soussis *et al*., 1995, Christiaens *et al*., 1999). In animal studies, adverse effects on maturation and fertilization have been reported (Alsalili *et al*., 1997, Tatone *et al*., 1998, Janssenswillen *et al*., 1997).

Several analgesic methods are in use for oocyte retrieval today, including conscious sedation, local anesthesia, general anesthesia, epidural and spinal anesthesia and electro-acupuncture. To reduce anxiety, sedative premedication including benzodiazepams are commonly used. Today, there is insufficient evidence to determine the best method of pain relief for oocyte retrieval (Kwan *et al*., 2005, Stener-Victorin, 2005).

### General anesthesia

General anesthesia remains in use, but most clinics now use local anesthesia. General anesthesia requires highly specialised equipment, the presence of an anesthesiologist, a long recovery time in

relation to the short procedure and is expensive. Some studies have, also shown adverse effects on fertilization and embryo development (Christiaens *et al*., 1999, Gonen *et al*., 1995, Wilhelm *et al*., 2002, Bokhari and Pollard, 1998).

**Local anesthesia**

The most commonly used technique for local anesthesia when performing oocyte retrieval is paracervical block (PCB). PCB is used alone or with sedation. Lidocaine has been found in follicular fluid but is considered to not affect fertilization adversely (Wikland *et al*., 1990). The PCB is considered to produce effective analgesia during oocyte retrieval (Godoy *et al*., 1993, Corson *et al*., 1994, Ng *et al*., 1999). Different doses of lidocaine have been tested, in RCTs (Ng *et al*., 2000, Ng *et al*., 2003), to find the most effective dose for pain relief. No significant differences were found between the lowest dose of 50 mg and the highest dose of 200 mg. The lowest dose was recommended because of absence in improvement in effect on higher doses and potential dose-related risks. Another study, (Ng *et al*., 2001) compared the pain relieving effects of PCB + placebo with PCB + conscious sedation. Women who received PCB + placebo experienced 2.5 times higher levels of vaginal and abdominal pain than women who received PCB + conscious sedation. In an RCT (Tummon, 2004), non-invasive analgesia with lidocaine vaginal gel + conscious sedation was compared with PCB + conscious sedation as analgesic during oocyte retrieval. Women rated anxiety and pain levels repeatedly on VAS. The result showed that women using lidocaine vaginal gel rated pain significantly higher than women who used PCB, mean 73 ± 4.5 versus 47 ± 3.9 on VAS ($P = 0.001$).

**Conscious sedation**

Conscious sedation is commonly used for oocyte retrieval since it is effective, easy to use and there is usually no need for an assisting anesthesiologist and thus lower costs. Conscious sedation seems to be appropriate for most but not for all women. Pre-operative information is essential and should ensure that the woman fully understands the preparations and the oocyte retrieval procedure, and with the important aim of reducing pre-operative anxiety. Primarily, women must be willing and cooperative; it is not suitable for extremely anxious women. It is reported to be used in 84% of IVF clinics in the UK (Elkington *et al*., 2003), and in 95% of IVF centres in the USA (Ditkoff *et al*., 1997). In Sweden, conscious sedation is the dominant form of sedation and at most IVF clinics a physician or trained midwife/nurse from the fertility team administers the drugs.

Conscious sedation has been defined as "a technique in which the use of a drug or drugs produce a state of depression of the central nervous system enabling treatment to be carried out, but during which verbal contact with the patient is maintained through the period of sedation. The drugs and technique used to provide conscious sedation should carry a margin of safety wide enough to render loss of consciousness unlikely" (Skelly, 1996).

There is great variation in drugs and dosages used for conscious sedation in conjunction with oocyte retrieval. The most commonly used drugs for conscious sedation are opiates, since they produce analgesic effects and drowsiness rapidly. Drugs used include fentanyl, remifentanil and alfentanil. Benzodiazepines are used in conscious sedation owing to their sedative and amnesic effects. Nitrous oxide may also be used.

Conscious sedation has been compared with general anesthesia (Ben-Shlomo *et al*., 1999). The sedation group rated higher pain than the general anesthesia group. Despite significant differences in pain ratings, both groups reported satisfaction with modality of pain.

Physician-controlled versus patient-controlled analgesia/sedation has been evaluated in several studies (Zelcer *et al*., 1992, Bhattacharya *et al*., 1997, Thompson *et al*., 2000, Lok *et al*., 2002, Cook *et al*., 1993). In two studies, results showed no differences whereas two other studies gave an advantage to physician-controlled analgesia/sedation. Combined data of pain ratings also favoured the physician-controlled analgesia/sedation (Kwan *et al*., 2005). Women's satisfaction was comparable between the groups in these studies.

**Acupuncture**

Electro-acupuncture (EA) as analgesic in oocyte retrieval has showed similar pain relieving effects as conscious sedation using fast-acting opiates (Stener-Victorin *et al*., 1999, Stener-Victorin *et al*., 2003).

Acupuncture has also been used in addition to conscious sedation resulting in reduced pain intensity and analgesic consumption (Sator-Katzenschlager *et al*., 2006).The purposes of using acupuncture as an alternative/complement to opiates are fewer side-effects with shortened recovery. In some cases, the reason for using EA can be medical contraindications for opiates and the desires from women to have non-pharmacological analgesia. Pain relief produced by acupuncture is a result of activation of endogenous pain inhibiting mechanisms described above (Wang *et al*., 2008). The pain threshold becomes less sensitive to painful stimulus; both physiological and psychological mechanisms are involved. Acupuncture needles are placed at specific points in somatic segments related to the pain area, i.e. the ovaries and the uterus. Basic scientific research suggests that manual stimulation of acupuncture needles activates muscle afferents; depending on stimulation intensity, Aδ-fibres and C-fibers will transmit signals to the spinal cord and on to the central nervous system (Kagitani *et al*., 2005). In addition, distal points are used to prolong the pain inhibiting effects (Sandkuhler *et al*., 1997). Acupuncture needles can also be stimulated electrically, using electro-acupuncture. Low frequency (1–4 Hz) stimulation probably excites ergoreceptors during muscles contractions (Kaufman *et al*., 1984), and Aδ-fibres and C-fibres, and high frequency EA (80–120 Hz) excites Aβ-fibres and probably activates the previously described gate control theory. Endogenous opioids, which seem essential for activation of descending pain inhibitory pathways and inducing functional changes in organ systems, are

released during low and high frequency EA (Andersson and Lundeberg, 1995, Han, 2003).

The oxytocinergic system may also be involved in the modulation of stress, pain, and autonomic and immune functions. This system is activated by mild, non-painful, sensory stimulation such as 2 Hz EA, massage, vibration, and thermal stimulation (Uvnäs-Moberg *et al*., 1993). The placebo effect appears to be strongly involved in results of acupuncture treatment (Ernst, 2004, Lund and Lundeberg, 2006), related to expectations and interaction between care-giver and patients. Reports from China indicate that preparing patients mentally before surgery is essential when using acupuncture instead of anesthesia (Bonica, 1974).

In addition, acupuncture has been found to improve several gynecological and obstetric conditions such as pelvic girdle pain during pregnancy (Elden *et al*., 2005), nausea and vomiting associated with early pregnancy (Smith *et al*., 2002), pain during labour (Mårtensson *et al*., 2008, Ramnerö *et al*., 2002), dysmenorre (Witt *et al*., 2008), and vasomotor symptoms in postmenopausal women (Nedstrand *et al*., 2006, Wyon *et al*., 2004, Zaborowska *et al*., 2007). The role of acupuncture in fertility treatment is questioned. Studies have reported positive effects on pregnancy rates when using acupuncture as an adjuvant treatment to embryo transfer in conjunction with IVF (Dieterle *et al*., 2006, Paulus *et al*., 2002, Westergaard *et al*., 2006). One hypothesis for improved pregnancy rates after acupuncture is enhancement of uterine receptivity through increased blood flow, which in turn might improve endometrial receptivity (Stener-Victorin *et al*., 1996). In a systematic review and meta-analysis of Manheimer *et al.* (Manheimer *et al*., 2008,) which included seven RCTs and a total of 1366 women, the result showed improved pregnancy and live birth rates among women who received adjuvant acupuncture treatment to embryo transfer. Another systematic review and meta-analysis by El-Toukhy *et al.* (El-Toukhy *et al*., 2008), including thirteen trials and a total of 2500 women (five trials, n = 877, using acupuncture as pain relief method during oocyte retrieval and in 8 trials, n = 1623, when acupuncture was performed adjuvant to embryo transfer) did not show any significant improvement in pregnancy rate after adjuvant acupuncture.

# OBJECTIVES

The overall aim of this thesis was to evaluate and compare different methods for pain relief during oocyte retrieval and to study women's expectations and experiences of pain during oocyte retrieval in conjunction with in vitro fertilization.
The specific aims were:

- To compare the pain-relieving effects of EA related to oocyte retrieval, and to compare post-operative well-being.

- To test whether local analgesia with pre-ovarian block (POB) results in better pain relief than PCB during oocyte retrieval.

- To evaluate women's expectations and experiences of pain during oocyte retrieval, and to investigate whether preoperative information was sufficient, and which factors influenced women's subjective sense of security during oocyte retrieval.

- To investigate potential adverse effects of varied doses of an analgesic drug commonly used during oocyte retrieval – alfentanil – on fertilization rate and embryo development. Furthermore, potential adverse effects of benzo-diazepam and nitrous oxide were investigated.

# METHODOLOGICAL CONSIDERATIONS

### Settings

Papers I-IV were performed at the University of Gothenburg, Sweden. Paper II was performed at the University of Gothenburg and at the Stockholm University. The studies in this thesis were performed at Reproductive Medicine, Sahlgrenska University Hospital, Gothenburg, Fertility Centre, Carlanderska hospital (Paper II and III), Gothenburg and at Reproductive Medicine Centre, Karolinska University Hospital/Huddinge, Stockholm (Paper II).

### Ethics

The Regional Ethical Review Board in Gothenburg approved papers I-IV and the Regional Ethical Review Board in Stockholm also approved paper II. All the women gave their written informed consent (Papers I-III).

## IVF treatment

### Ovarian stimulation

Two different techniques were used: IVF and ICSI. In standard IVF the aspirated oocytes are combined with the man's sperm in laboratory dishes. During ICSI a single sperm is injected into an oocyte. Women in the present studies underwent a stimulation protocol including down-regulation with a gonadotropin-releasing hormone agonist (Suprecur™ Hoechst, Frankfurt, Germany), beginning either in the follicular or the luteal phase. A minority of the women were treated according to an antagonist protocol (Cetrotide, MerckSerono, Geneva, Switzerland; Orgalutran, Organon, Oss, Netherlands. After down-regulation, stimulation was undertaken with recombinant follicle-stimulating hormone (Gonal-F™, MerckSerono, Geneva Switzerland; Puregon™, Organon, Oss, The Netherlands) or urinary-derived hormone (Menopur™, Ferring, Denmark). Monitoring was performed via vaginal ultrasound scans and serum estradiol measurements. When adequate stimulation was achieved, hCG (Ovitrelle, MerckSerono, Geneva Switzerland or Pregnyl, Organon, Oss, Netherlands) was administered, and oocyte retrieval was performed with a single lumen aspiration needle (Swemed Lab International AB, Billdal, Sweden). Oocyte retrieval was performed 38 hours after hCG administration using transvaginal sonographically guided puncture.

Fertilization was performed with conventional IVF or ICSI, using standard techniques. One or two embryos were transferred 2 or 3 days after oocyte retrieval using Wallace, Frydman TDT or Cook catheters. Luteal phase support was administered daily with progesterone. Pregnancy test was performed on day 14 (Paper II), or on day 19 (Paper I, III-IV) after embryo transfer. Clinical pregnancy was defined as an ultrasound verified pregnancy with fetal heartbeat, 5 weeks after embryo transfer.

## The oocyte retrieval procedure

Oocyte retrieval was initially carried out by laparoscopic surgery but the techniques soon changed to the less invasive transvaginal ultrasound-guided oocyte retrieval (Lenz *et al*., 1981, Wikland *et al*., 1987). Oocyte retrieval is usually performed without technical difficulties. The method is considered safe, since serious adverse events are rare (Ludwig *et al*., 2006). A single aspiration needle is preferable since it causes less pain, and fertilization and pregnancy rates do not differ compared with using a flash needle (Awonuga *et al*., 1996). Blood pressure, heart rate and oxygen saturation should be supervised throughout the procedure to monitor the woman's condition. When evaluating sedation and anesthetic practice for oocyte retrieval among clinics in United Kingdom, it was found that 4.8% of the clinics had no resuscitation trolley and 21.4% had no defibrillator (Yasmin *et al*., 2004).

Complications related to oocyte retrieval include vaginal or abdominal bleedings, pelvic infections and injury to pelvic structure, such as the ureter, the bowels or the appendix (Ludwig *et al*., 2006). Vaginal bleeding is the most common complication, reported in a range from 0.5% to 8.6% (Bergh and Lundkvist, 1992, Bennett *et al*., 1993, Ludwig *et al*., 2006). Most bleedings are treated with local pressure for a few minutes, although occasionally a tamponade or a suture is necessary. To prevent bleeding, repeated penetration of the vaginal wall and the ovaries should be avoided. Severe intra-abdominal bleedings have been reported in 0.08-0.2% (Dicker *et al*., 1993, Tureck *et al*., 1993, Govaerts *et al*., 1998). In a few cases bleeding has been associated with coagulation disorders (Battaglia *et al*., 2001, El-Shawarby *et al*., 2004). Initially, the transvaginal oocyte retrieval procedure was seen as a potential risk for pelvic infections. The incidence of pelvic infections and abscesses is however reported to be low, about 0.3-0.6% (Bergh and Lundkvist, 1992, Ashkenazi *et al*., 1994, Bennett *et al*., 1993, Dicker *et al*., 1993, Tureck *et al*., 1993). It has been suggested that a previous history of pelvic inflammatory disease increases the risk of pelvic infections after oocyte retrieval (Dicker *et al*., 1993). Benett *et al.* (Bennett *et al*., 1993) did not support this theory but advised against unnecessary needling of hydrosalpinges, cysts and endometriomas. Vaginal washing with normal saline solution is used but regular preventive antibiotic prior to ooccyte retrieval is regarded as unnecessary. Other rare side effects that have been reported are perforated appendix in conjunction to oocyte retrieval (Akman *et al*., 1995, Roest *et al*., 1996), an ureterovaginal fistula (von Eye Corleta *et al*., 2008) and ureteral damage (Fugita and Kavoussi, 2001, Ludwig *et al*., 2006, Miller *et al*., 2002).

## Subjects and design

General view over number of patients and study design in thesis

| | Paper I | Paper II | Paper III | Paper IV |
|---|---|---|---|---|
| No. of women | 160 | 183 | 124 | 663 |
| Study design | RCT, open | RCT, single-blinded | observational, prospective | observational, retrospective |
| Intervention | EA vs CA | POB vs PCB | | |

### *Paper I*

**Electro-acupuncture versus conventional analgesia**

This study was an open prospective RCT comparing EA with conventional analgesia (CA) regarding pain relieving effects and post-operative well-being in conjunction with oocyte retrieval. The study period was between March 2002 and October 2003. The women were informed about the study as they were about to start FSH stimulation. A total of 550 women were eligible for the study and 160 women were randomized, each woman contributed with one IVF-cycle to the study (Fig. 3). The inclusion criteria were a) willing to participate and b) knowledge of the Swedish language. Exclusion criteria were a) previous participation in the study, b) epilepsy, c) pacemaker, d) severe nickel allergy and e) hepatitis. Eighty women were allocated to EA and 80 to CA. Two women in the EA group were lost to follow up. The reasons were ovulation prior to oocyte retrieval (n = 1), resulting in withdrawal from EA prior to oocyte retrieval and missing assessments of VAS, and STAI (n = 1) due to administrative shortcomings.

### *Paper II*

**Pre-ovarian block versus paracervical block for oocyte retrieval**

This single-blind prospective multi-centre RCT examined whether local analgesia with POB improved pain relief as compared with local analgesia with PCB used in conjunction with oocyte retrieval. The study was performed between October 2004 and January 2005. In all, 554 women were assessed for eligibility and a total of 183 women were randomized, 96 women were allocated to POB and 87 to PCB (Fig. 4). The physician informed the women about the study when performing the last ultrasound, 2-5 days before oocyte retrieval. Women were eligible for randomization if they fulfilled the following inclusion criteria: a) signed the written consent, b) willing to participate and c) Swedish speaking. The exclusion criteria were: a) previous participation in the study, b) lidocaine allergy, c) only one ovary or abnormal position of ovaries and d) coasting >1 day (because of high risk of ovarian hyperstimulation syndrome), due to difficulties to separate pain due to oocyte retrieval and general pain. One woman in

the POB group had intolerable pain, received additional lidocaine, and did not complete VAS ratings but was included in the intention-to-treat analysis when possible.

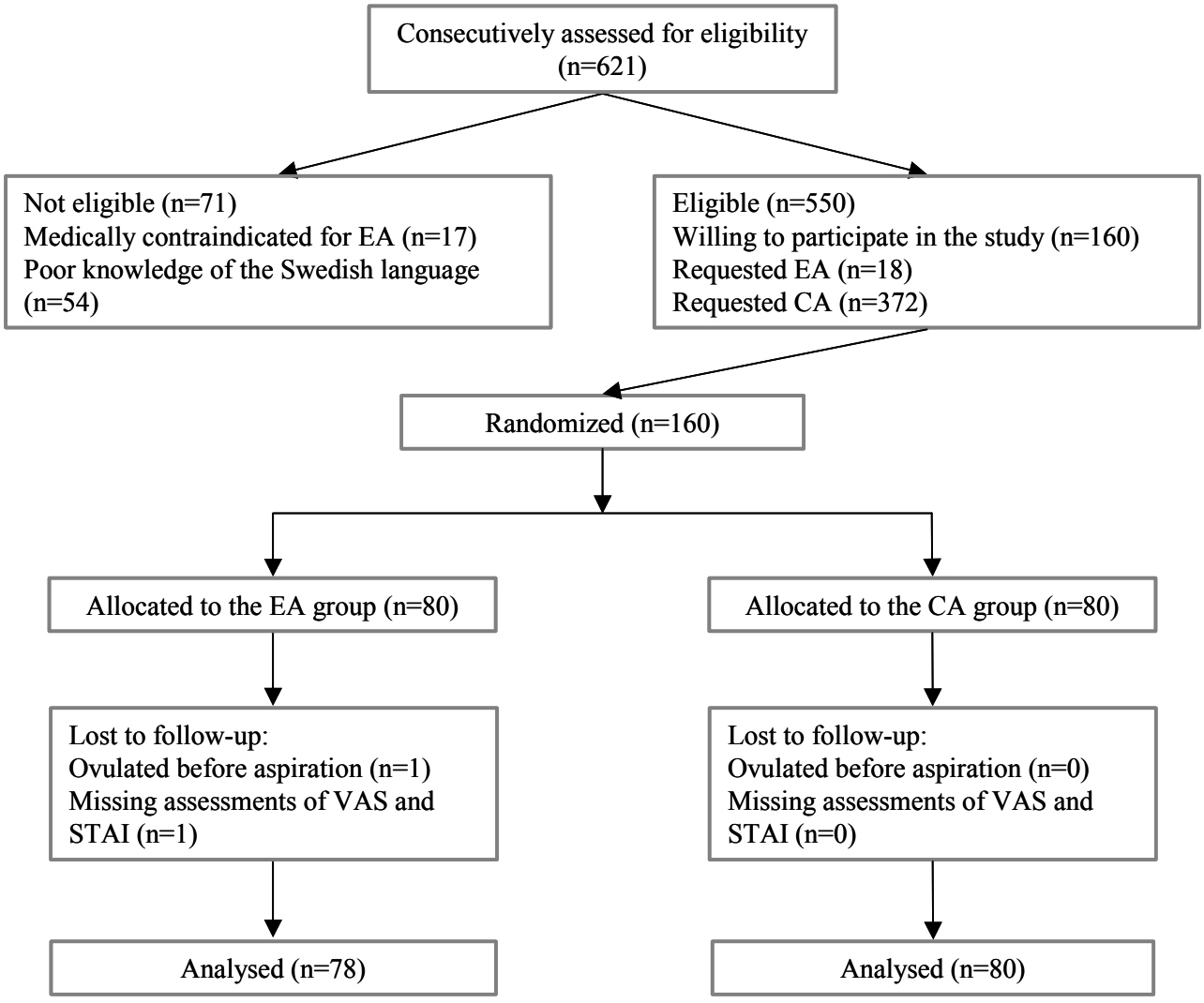


**Figure 3.** Flow chart of patients eligible for the study (paper I).

EA=electro-acupuncture, CA=conventional analgesia, STAI=state trait anxiety inventory test, VAS=visual analogue scale.

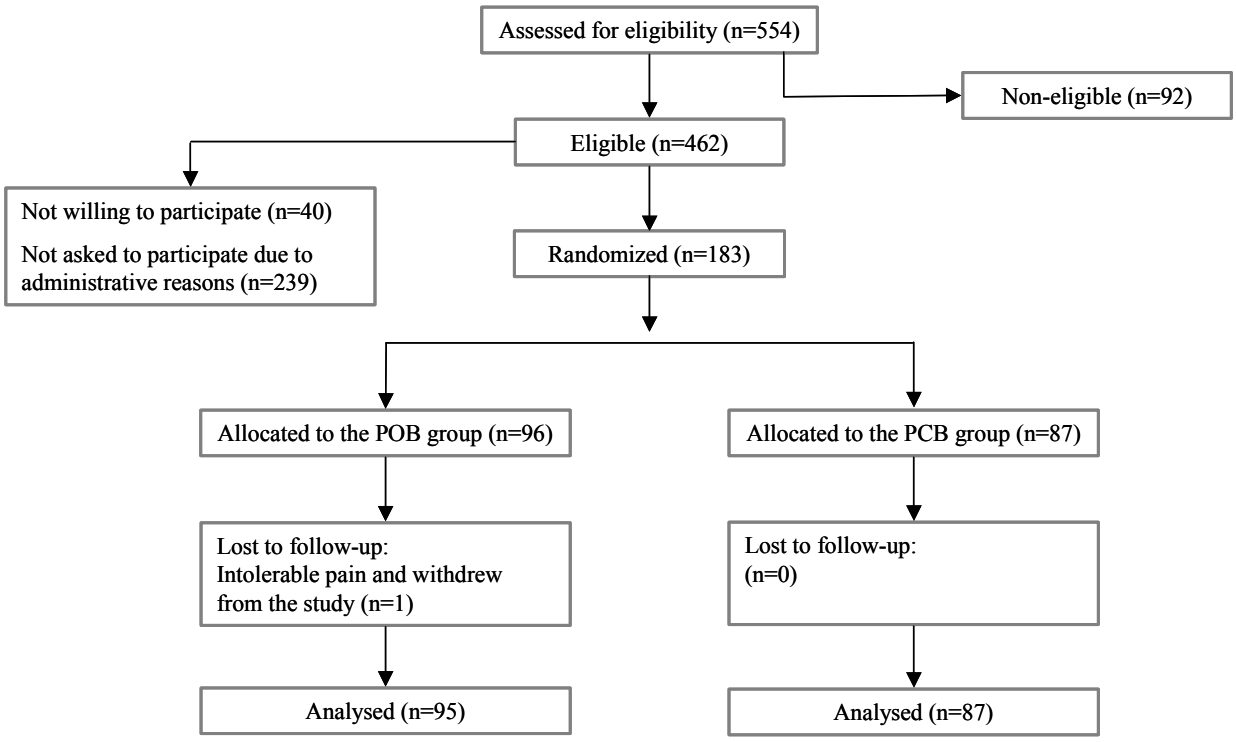


**Figure 4.** Flow chart of patients randomized for the study (paper II).

## *Paper III*

### Pain aspects in oocyte aspiration for IVF

This study was an observational study with the primary aim of evaluating women's expectations concerning pain before oocyte retrieval as compared with the pain experienced during surgery. The study period was between November 2004 and January 2005. Further aims were to investigate whether the pre-operative information was sufficient, and what influenced the women's sense of security during surgery. One hundred twenty-four women were included in the study, representing women from two clinics in Göteborg who took part in an RCT (Paper II). Information about the study was given by a midwife 2-5 days before oocyte retrieval. The inclusion criteria were: a) willing to participate and b) Swedish speaking. Exclusion criteria were: a) hypersensitivity to lidocaine and b) previous participation in the study.

## *Paper IV*

### Effect of alfentanil dosage during oocyte retrieval on fertilization and embryo quality

The objective of this observational retrospective study was to examine whether different doses of alfentanil adversely affected fertilization rates and embryo quality. For measurements, data

was collected from the clinic's database and women's hospital records, for completion of data (smoking habits, weight and length given BMI) approximately 30 women were interviewed over the phone. A total of 891 IVF-cycles (including oocyte retrievals) were performed, and 841 IVF cycles were included in the present study. Fifty cycles were excluded, forty-five owing to oocyte donation, freezing of all embryos due to cancer treatment or PGD, 1 owing to general anesthesia and 4 due to missing documentation concerning alfentanil doses. In total, 663 women contributed to these 841 IVF-cycles. The main analysis included only one cycle per woman, the woman's first oocyte retrieval during the study period. The women were divided in two groups according to dose of alfentanil, a low-dose group (≤0.5 mg alfentanil, n = 370) and a high-dose group (>0.5 mg alfentanil, n = 293).

# Measurements

### Visual Analogue Scale (VAS)

Women rated their expectations and experiences of pain using the VAS (Papers I-III), which consists of a 100 mm horizontal line on paper (0-100) (Altman, 1996). VAS was used since it is designed to provide information about internal subjective feelings such as preoperative anxiety (Lee and Kieckhefer, 1989, Kindler *et al*., 2000).The questions used in the study were tested on 10 women, non-participants, to find out whether the questions were correctly interpreted. VAS ratings were assessed in a standardised way, i.e. if a rating was between numbers it was assessed by always choosing the lower number.

Anxiety was measured, using VAS; prior to randomization in Papers I-II, since the studies were balanced for degree of anxiety. For anxiety, scale end-points ranged from 0 "not worried at all" to 100 "extremely worried". Women were defined as not anxious on levels of ≤22 mm or anxious at VAS levels of >22 mm. The level defined for anxiety was based on results from an earlier study (Stener-Victorin *et al*., 1999) in which the median level of stress prior to oocyte retrieval was 22 millimetres on VAS. We considered stress to correspond with anxiety.

### State Trait Anxiety Inventory test (STAI)

STAI is a validated self-reported questionnaire consisting of two 20-item scales that evaluates momentarily experienced pain (state anxiety, form Y-1) and general tendencies of an individual to restrain experienced anxiety (trait anxiety, form Y-2) (Spielberger, 1999). The anxiety scores range from 20 to 80; higher scores indicate a higher degree of anxiety. STAI was used for measurement of well-being pre-operatively, 60 minutes post-operatively and at recovery (Paper I). In thesis, post-operative well-being is defined as a feeling of comfort and security, which by nature is a subjective experience.

**The McGill Pain Questionnaire**

The McGill Pain Questionnaire is a standardised tested self-reported questionnaire that measures subjective pain experience and pain characteristics (Paper II) (Melzack, 1975), (http://community.ocr.org.uk/core/community/public/download_file?rid=474), describing the sensory, affective and evaluative dimensions of pain. In the study, a Swedish version of the short form of the McGill Pain Questionnaire was used, which has shown both validity and reliability (Burckhardt and Bjelle, 1994).The patient choose 15 words, 11 of which describe sensory and four which describe affective qualities of pain. For each word, the intensity of the pain is assigned (0, none; 1, mild; 2, moderate; 3, severe). The total number of words can thus range from 0-15, and total pain intensity can range from 0-45. Sensory pain intensity ranges from 0-33, and the affective pain intensity range from 0-12.

**Self-constructed questionnaire**

A self-constructed questionnaire with multiple-choice questions was used in paper III for evaluating women's satisfaction with the pre-operative information and describing what issues influenced their sense of security during oocyte retrieval. Questions were tested on 10 women (non-participants) and among five members of staff before the study started to assess whether interpretation of the questions was correct. This questionnaire was constructed to measure specific issues related to the clinical situation.

**Lidocaine concentrations**

In paper II, serum and follicular fluid lidocaine concentrations were measured in a random sample from one centre consisting of 15 women. Follicular fluid was collected from the first follicle and the last follicle from the ovary that was first punctured. A sample of blood/serum was collected from the woman's venous catheter at the same time as the follicular fluid was collected. The serum was analysed for lidocaine concentration. Lidocaine concentration was analysed using alkaline extraction to organic solvent and re-extraction to an acid water base, which was injected to a reverse phase HPLC system with ultraviolet detection. Detection limit was 0.02 µmol/l (0.0000046µg/ml).

## Statistics

**Table 3.** Statistical methods used in papers I-IV

| Methods | I | II | III | IV |
|---|---|---|---|---|
| *Descriptive statistics* | | | | |
| Mean (SD) | x | x | x | x |
| Median (min-max) | x | x | x | x |
| Number of patients (%) | x | x | x | x |
| *Analytical statistics* | | | | |
| Fishers's exact test | x | x | x | x |
| Chi-square test | | | x | x |
| Mann Whitney *U*-test | x | x | x | x |
| Wilcoxon signed rank test | | | x | x |
| Spearman's rank correlation test | x | | | |
| Multiple linear regression | x | x | x | |
| Logistic Regression | x | | | x |
| Intention to treat | x | x | | |
| Per protocol | x | | | |

An overview of the statistical methods used in this thesis is presented in Table 3. Analyses in paper I-IV were made using the Statistical Package for Social Science (SPSS) version 12.0 and SAS version 8.0 and 9.0.

All tests were two-sided, and $P < 0.05$ was considered statistically significant.

For descriptive statistics, means, standard deviations, medians, minimum and maximum, numbers of patients and percentages were used. Dichotomous variables were analysed using Fisher's exact test (paper I-IV), and Mantel-Haenszel Chi-square test was used for ordered categorical variables (paper III-IV). Univariate analysis was performed with Spearman's rank correlation test for association between continuous variables in paper I.

**Handling of comparisons between-group differences**

The non-parametric Mann Whitney *U*-test was used for comparisons of continuous variables (Papers I-IV). Ordinal data for VAS ratings concerning pain experience (Papers I-III) were analysed using the Mann Whitney *U*-test.

**Analysis of within-group differences**

The non-parametric Wilcoxon signed ranked test was used in paper III for paired comparisons of VAS-ratings regarding the women's expectations and experiences of pain relief before and after oocyte retrieval. In paper IV, Wilcoxon signed ranked test was used for analysis of a subgroup of women (n=65) who had repeated oocyte retrievals during the study period.

**Regression analyses**

In paper I, p-values were adjusted using logistic regression analysis because the number of oocytes retrieved differed significantly between groups and might have influenced experienced pain.

In paper I, a stepwise linear regression analysis was made also on the outcomes pain and tiredness as the dependent variables. Blom's rank method was used to achieve a normal distribution of the dependent variables (Blom, 1958).

In paper II, a stepwise linear forward regression analysis was used to examine which variables were related the dependent variable overall pain. To obtain a normal distribution of overall pain, the square root of overall pain on both sides was used for the regression analysis.

In paper III, a stepwise linear forward regression analysis was used to examine which variables were independently associated with mean pain.

To adjust for potential confounders for end-points in paper IV, such as age, BMI, smoking habits, previous pregnancies and deliveries logistic regression was used.

**Intention-to-treat analysis and per protocol analysis**

In papers I and II variables were analysed primarily according to intention-to-treat, which is a strict analysis by randomization group regardless of subsequent protocol violations, to avoid introduction of bias. In paper I, a per protocol analysis was performed for those women who completed to the protocol to which they were randomly assigned.

**Sample size calculation**

Sample size was calculated *a priori* for paper I and II.

***Paper I)*** Sample size was calculated for a difference in post-operative well-being and for equal levels of mean pain during oocyte retrieval between the EA and the CA groups. The assumptions for the primary end-points were: if STAI measurements of well-being 60 minutes post-operatively had an expected SD of 10.0 in each group, 68 patients would be needed in each group to show a significant difference between groups of 5.0 with a power of 80% and a significance level of 0.05. To demonstrate equal levels of pain directly related to oocyte retrieval and measured with VAS, the assumptions were as follows: given an SD of 18.0 in each group concerning pain assessed by VAS directly after oocyte retrieval, 80 patients in each group would be needed to show that the upper limit of the 95 % confidence interval for the differences in means between the groups would not exceed 11.0, with a probability of 0.80.

***Paper II)*** The primary outcome was vaginal and abdominal pain during the entire oocyte retrieval process measured with VAS directly after the procedure defined as "overall pain both sides" during oocyte retrieval. Assuming overall pain both sides in the PCB group to be 30 mm on VAS, with an SD of 20 mm, 90 patients in each group would be needed to show a decrease in overall pain both sides of 9 mm (30 %) with an 80% power and a significance level of 0.05.

***Paper IV)*** A retrospectively analysis based on group sizes of 370 and 293 patients allows for detecting an 11% difference in fertilization rate and GQE rate to assume a fertilization rate of 60% and a GQE rate of 60% in the low-dose alfentanil group (power 80%, $\alpha$ level 5%).

### Randomization process

***Paper I)*** The randomization was performed by the study midwife, generated from a computerised list, using sealed opaque envelopes, and taking into consideration that EA (if allocated) was administered 30-45 minutes before onset of oocyte retrieval. Randomization was performed in the proportion 1:1, and stratified for degree of anxiety, rated by VAS upon arrival at the clinic. Low degree of anxiety was defined as $\leq$ 22 millimetres on VAS and high degree if VAS was above 22 millimetres. Blinding of the woman or the physician was not possible. However, the study coordinator who evaluated the women's VAS ratings was blinded to allocation group.

***Paper II)*** Randomization was performed prior to oocyte retrieval in the proportion 1:1, using an Internet-based central randomization programme. A variation of optimal allocation according to Pocock's sequential randomization method (Pocock 1983) was used. The study was balanced for the following prognostic variables: age, number of previously completed IVF cycles, degree of anxiety, estimated number of follicles, BMI, pre-medication, and centre.

The randomization procedure used information from these variables for all previously included women, when a new woman was randomized to one of the groups.

A written report was generated from the computer with the woman's identification number in the study and randomized treatment. If the Internet or the computer was down, envelopes for emergency randomization were used. The women were blinded to which method was used until a pregnancy test was performed. The study coordinator who evaluated the VAS ratings was also blinded.

## Interventions

### Paper I - Electro-acupuncture versus conventional analgesia

The women were randomized to either a) conventional analgesia or b) electro-acupuncture.

All women in both groups used a heat therapy pillow on the abdomen to reduce stress and pain pre-, per- and post-operatively. Prior to initiation of the oocyte retrieval, women in both groups received a local analgesic with PCB consisting of lidocaine 0.5%, 5 ml on each side, in total 50 mg (Xylocain™ 10 mg/ml, AstraZeneca Sverige AB). Both groups received additional analgesics, with opiates or nitrous oxide (AGA, Stenungsund, Sweden), if needed to achieve sufficient pain relief during oocyte retrieval.

### Conventional analgesia

Women were offered a sedative pre-medication of 0.5 mg oral flunitrazepam (Fluscand™ 0.5 mg; Pharmacemie BV, Harleem, the Netherlands) and rectal paracetamol, 1 g (Panodil® 1 g; GlaxoSmith Klane, Täby, Sweden).

In the operating theatre, 0.5 mg alfentanil (Rapifen™ 0.5 mg/ml; Janssen-Cilag AB, Sollentuna, Sweden) was administrated intravenously as a bolus dose before initiation of oocyte retrieval.

**Electro-acupuncture**

Electro-acupuncture was administered approximately 30-45 minutes before oocyte retrieval and terminated directly after the last follicle was punctured. No sedative pre-medication was offered. The electro-acupuncture was given according to a protocol fixed in advanced, which was the same for all women in the EA group. The acupuncture points were selected in the abdominal muscles and at points below the elbow and the knee bilaterally. At the top of the cranium, one point was chosen to increase relaxation based on traditional Chinese medicine. The exact positions, stimulations and innervations of the selected points are presented in Table 4, Fig. 7. The acupuncture needles used were Hegu Xeno: Hegu AB, Landsbro, Sweden: size 0.30 x 30 or 50 millimetres (mm). Needles were inserted intra-muscularly to a depth of 15-30 mm. The acupuncture needles in the abdominal muscles were stimulated electrically with high frequency (80 Hz) with the aim to activate the gate-control mechanism at the segment level corresponding to the ovaries and the uterus and the central descending pain inhibitory system that involves the β-endorphinergic system. The intensity was high, giving non-painful paraestehsia. The acupuncture point in the hands was electrically stimulated with low frequency (2 Hz) to induce non-painful local muscle contractions with the aim to stimulate the release of β-endorphin centrally and to activate the descending pain inhibitory systems previously described. The acupuncture points below knee and at the top of the cranium were manipulated by hand every ten minutes to evoke needle sensations and increase activity in the afferent nerves and central pain systems (Fig. 5). The electrical stimulator used was CEFAR, ACUS 4, Cefar, and Lund, Sweden, with square-wave pulses (0.18 ms duration) with alternating polarity for the high frequency (80 Hz) and for the low-frequency (2 Hz), burst pulses (a burst length of 0.1 s and a burst frequency of 80 Hz) (Fig. 6).

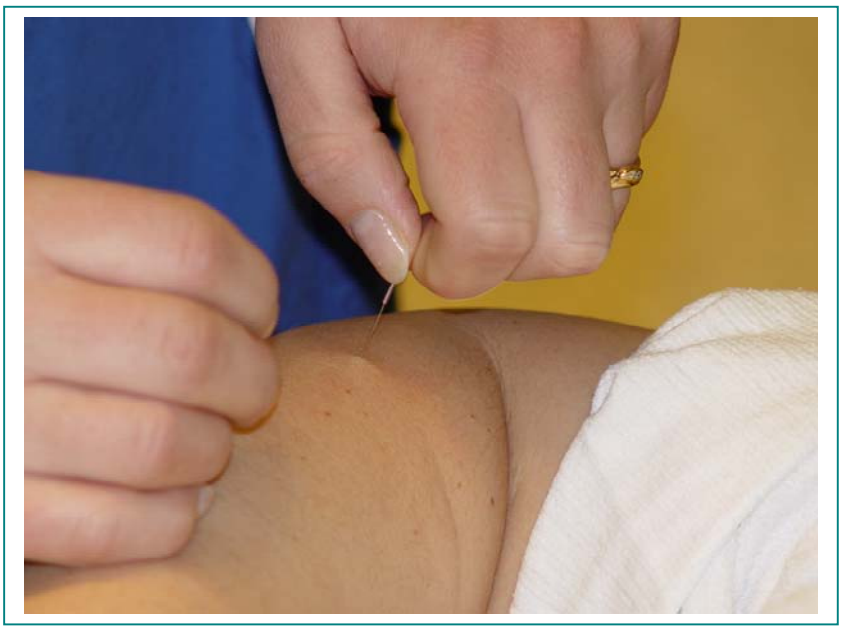

**Figure 5.** Manual stimulation of acupuncture needle.


**Figure 6.** The electrical acupuncture stimulator used in the study (paper I).

**Table 4.** The position and innervation of acupuncture points and type of stimulation used for pain relief combined with PCB for oocyte retrieval.

| Points | Stimulation | Segmental innervation | Muscle localisation |
|---|---|---|---|
| LI 4 bilateral | EA, 2 Hz | Nn.ulnaris, medianus (C8, Th 1) | Mm.interosseus dorsalis I, lumbricalis II, adductor pollicis |
| LI 10 bilateral | EA, 2 Hz | N. radialis (C6-7) | M. extensor carpi raialis longus |
| ST 29 bilateral | EA, 80 Hz | N. thoracius (Th 6-12) | M. rectus abdominis |
| KI 11 bilateral | EA, 80 Hz | N. thoracius (Th 6-12), sucostalis (Th 12) | Vagina m. rectus abdominis |
| GV 20 | manual | Nn. trigeminus (V), occipitalis minor (C2), occipitalis major (C2-3) | Aponeurosis epicrani |
| ST 36 bilateral | manual | N.peroneus profundus (L4-5) | M. tibialis anterior |

EA = electro-acupuncture; LI = large intestine; KI = kidney; GV = governor vessel; ST = stomach.

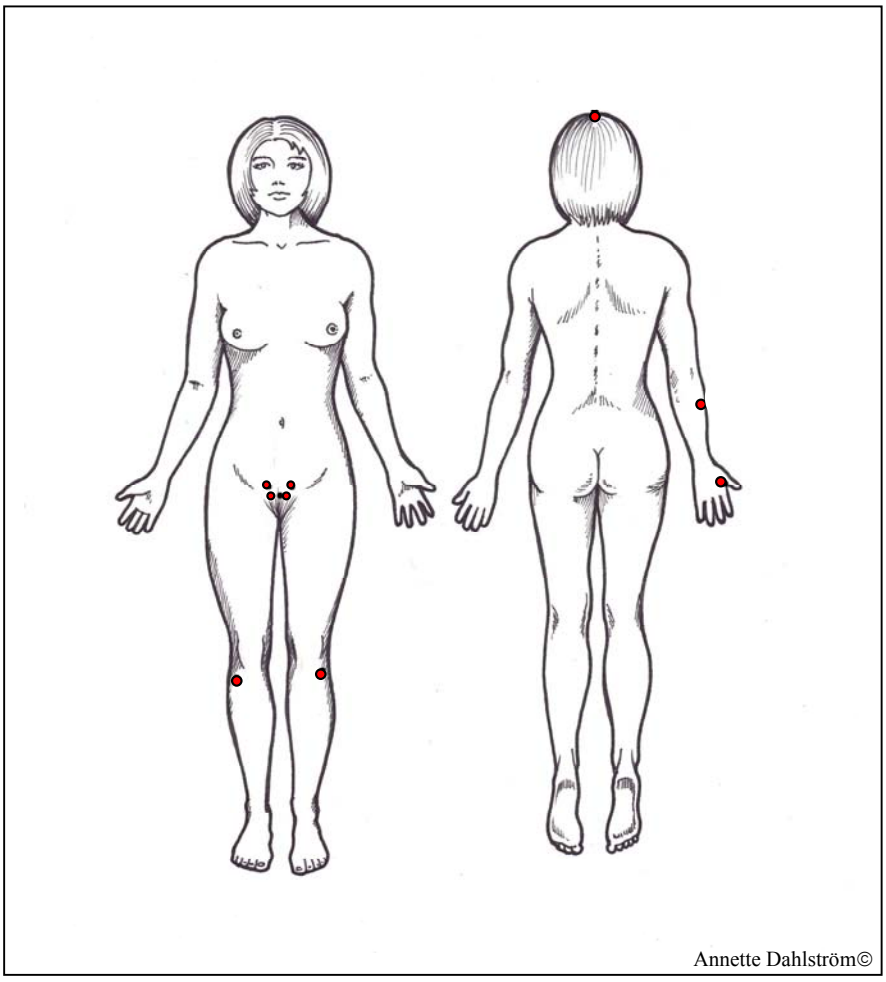


**Figure 7.** The locations of the selected acupuncture points.

**Paper II – Pre-ovarian block versus paracervical block**

Women were randomized, in a ratio of 1:1, to receive either PCB or POB as pain relief during oocyte retrieval. A few physicians, from each centre were selected to take part in the study and were trained to administer the POB appropriately.

**Paracervical block (PCB)**

The PCB was administered at the 2, 4, 6 and 9 o'clock positions, in a concentration of 1% lidocaine ((Xylaocain™ 10 mg/ml, Astra Zeneca Sverige AB), 2.5 ml for each injection, in total 100 mg. The needle used had a diameter of 0.9 mm and a length of 120 mm (Mediplast AB, Malmö, Sweden).

**Pre-ovarian block (POB)**

The POB consisting of 1% lidocaine, was administered on each side, 5 ml, and a total of 100 mg. The lidocaine was administered under ultrasound guidance in the vaginal wall and between the vaginal wall and the peritoneal surface near the ovary, where the needle used for oocyte retrieval was inserted (Fig. 8). A specific needle, POB™ (Swemed Lab Intl. AB) with a diameter of 1.2 mm and a length of 260 mm was used.

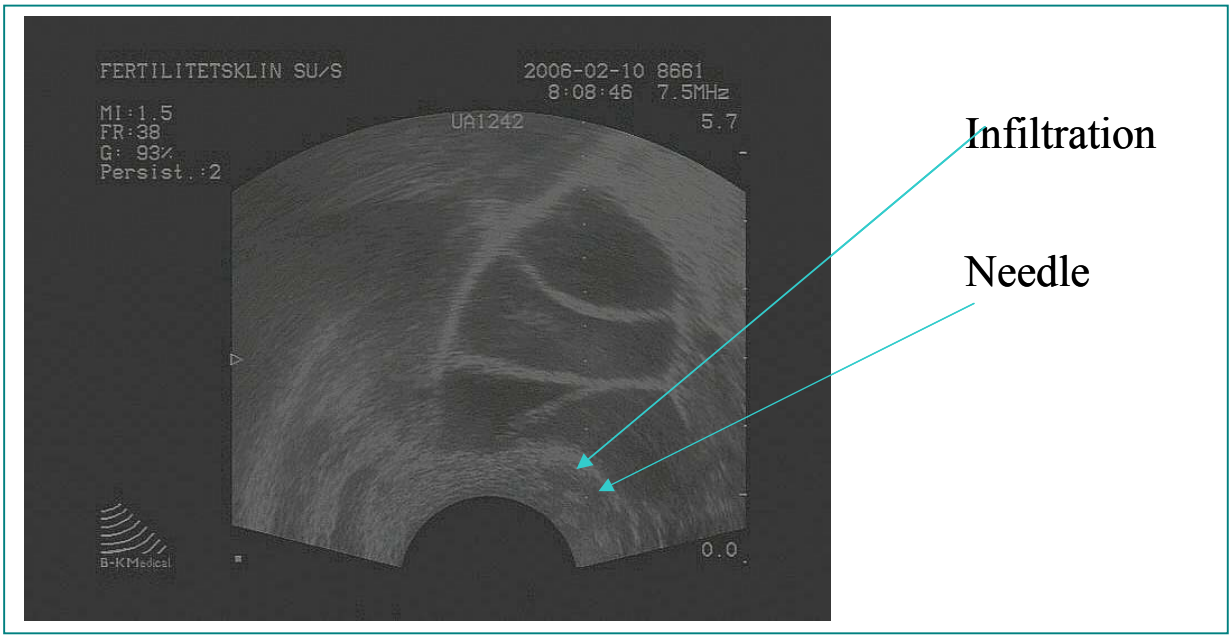


**Figure 8.** Administration of the POB.

# RESULTS AND COMMENTS

## Paper I

### Electro-acupuncture versus conventional analgesia

The primary aim was to evaluate the pain-relieving effects, and to evaluate post-operative well-being comparing EA with CA.

A total of 160 women were randomized, 80 to the EA group and 80 to the CA group. The majority of women in both groups had their first IVF-treatment. The surgery time was significantly longer in the EA group, which was probably because the women in this group had significantly more follicles punctured (15.8 ± 9.4 versus 13.4 ± 8.6, $P = 0.042$) and more egg retrieved (12.5 ± 6.7 versus 10.5 ± 6.4, $P = 0.029$) than the women in the CA group. Adjusting for the difference in number of oocytes did not alter results of the VAS assessments. Number of embryo transfers and pregnancy rates were comparable between the groups.

An analysis of those women who declined participation in the study was performed. No differences were found in primary and secondary infertility, parity and number of IVF-cycles performed compared with women in the study. Infertility reason differed significantly between participants and non-participants.

### VAS ratings

Degree of anxiety was similar between the groups prior to oocyte retrieval. The women who had EA as pain relief during surgery rated pain significantly higher than the women who had CA (48.5 ± 26.8 versus 29.8 ± 23.4 ($P < 0.0001$, adjusted $P < 0.0001$). The CA group was, however, significantly more tired and more confused after oocyte retrieval. At recovery, women in the CA group reported that they were significantly more satisfied with the procedure than women in the EA group.

### Well-being

Well-being was similar between groups during the entire day of the oocyte retrieval.

### Variables independently associated with pain and tiredness

Linear regression analyses were performed for the dependent variables pain and tiredness. Analgesic group (EA or CA) and rated levels of state anxiety (momentarily experienced anxiety) preoperatively were found to be independently associated with pain during the oocyte retrieval. Lower age, number of oocytes retrieved and type of analgesia were independently correlated with tiredness.

**Conclusions:** EA cannot generally be recommended as a pain-relieving method during oocyte retrieval but might be an alternative for women who desire a non-pharmacological method. One advantage of EA is less post-operative pain and confusion than with CA.

**Comments:** The significant difference in pain ratings, to the disadvantage of EA was unexpected, since previous studies

have shown comparable pain levels between EA and CA (Stener-Victorin *et al*., 1999, Stener-Victorin *et al*., 2003). The difference between previous studies and the present study was the pre-medication given to the CA group. When sedatives are given, pain ratings may be affected. Humadain *et al.* (Humaidan and Stener-Victorin, 2004) reported significant differences in ratings of maximum pain levels between the EA and the CA groups, to the advantage of CA. This was interpreted as a consequence of the pre-medication used in the CA group, which may have interfered with pain perception. In the present study, no differences were found in pain ratings between the women in the CA group who used premedication (n = 55), and those who did not (n = 25). Women not using premedication in the control group still had significantly lower levels of pain in comparison with the EA group. One explanation for the negative results of EA in our study might be that we recruited a more unselected population due to the positive results of EA obtained in the previous studies. However, the questions regarding pain levels and the protocol used for acupuncture treatment in the various studies were heterogeneous which makes comparisons difficult. In a latter study, by Sator-Katzenschlager *et al*. (Sator-Katzenschlager *et al*., 2006) auricular low-frequency EA was given in combination with patient-controlled analgesia with remifentanil. They found that EA reduced pain intensity and analgesic consumption and increased subjective well-being during and after the procedure compared with patient-controlled analgesia alone. In our study, women walked to the operating theatre with needles inserted, which implied some discomfort and might have influenced their pain perception (Cohen *et al*., 1999, Harro *et al*., 1993, Widerström-Noga *et al*., 1998). One advantage of auricular EA is the absence of needles inserted into the body and the lack of body-wrapping wires. The basic idea of an RCT is to compare groups of patients who differ only with respect to their treatment. The results of analyses are used to make inferences about the population, thus representative sample is necessary. One important issue to assess is how many patients were eligible i.e. satisfied the inclusion/exclusion criteria, and how many of those eligble were included in the study, since this influences the generalizability of results. All eligble women in our IVF-programme were asked consecutively to participate in the study. Twentynine percent of eligble women were willing to participate, 3% declined randomization, due to requesting EA. Sixty-eight percent refused to participate due to requesting CA. Eight percent of the women assessed for eligibility were excluded since they had poor knowledge of the Swedish language. One limitation in our study was that we did not note whether women used or had used acupuncture treatment in another context, another limitation was that we did not note whether women had chronic pains or used pain killers on a regular basis.

## Paper II

### Pre-ovarian block versus paracervical block

This study aimed to test whether analgesia with POB resulted in greater pain relief than PCB during oocyte retrieval. A total of 183 women were randomized to local analgesia either with POB or PCB. There were no differences in the women's characteristics, and no differences were found in number of oocytes collected, use of opiates, surgery time, care time, fertilization rate and number of good quality embryos, or clinical pregnancy rate between the groups.

### Pain ratings

The primary end-point in this study was "overall pain both sides" measured directly after the oocyte retrieval procedure. Measurements of pain in conjunction with administration of lidocaine, using the POB and PCB, were comparable between groups. The POB group rated pain significantly higher when the needle was inserted through the vaginal wall on the first side than the PCB group. On the second side no difference was found. All other pain ratings during the entire oocyte retrieval including "overall pain both sides" showed no significant differences between groups. With exception for overall pain 20 minutes post-operative, all other pain ratings for overall pain and maximum pain post-operatively were comparable between the POB and the PCB group. Current pain post-operatively did not differ between groups. Measurements of pain intensity, total number of words chosen, present pain intensity, sensory and affective intensity, using the Short-Form McGill Pain Questionnaire showed no differences between groups.

### Regression analysis

BMI, number of follicles punctured and anxiety were found to be independently associated with overall pain during oocyte retrieval, adjusted $r^2 = 0.114$.

### Lidocaine concentrations

In a random sample of women (n = 15), from one centre, serum and follicular fluid lidocaine concentrations were analysed. The concentrations in serum and follicular fluid at the time the first follicle was punctured did not differ significantly between POB and PCB, but the results showed great variations, both in serum and in follicular fluid in both groups.

**Conclusions:** No differences were found in overall pain experienced during the entire oocyte retrieval procedure with POB compared with PCB.

**Comments:** The initial clinical impression by using POB was an excellent pain relief effect exceeding that of PCB. Findings in this RCT did not confirm this; both techniques produced satisfactory pain relief. This further supports the importance of performing appropriate scientific controlled studies before new treatment is implemented. The introduction of new treatments often begins in the clinic on a small number of patients to see what happens. These types of studies are usually uncontrolled and open. They often tend to give over-optimistic and hence biased results (Altman, 1996). It is

possible that the investigator's enthusiasm influences patient's well-being, especially when subjective symptoms, such as pain, are involved. The golden standard when comparing effects in a clinical trial is considered to be the design of an RCT, double-blinded (both patients and physicians). The main reason for using an RCT is to prevent bias, and to compare treatments between groups that do not differ in any systematic way.

## Paper III

### Pain aspects in oocyte aspiration for IVF

The primary aim of this study was to evaluate women's expectations concerning pain during oocyte retrieval, and to evaluate whether the pain experienced was in accordance with the women's expectations.

In total 124 women participated in this observational study, and the response rate to the questionnaires was 100%.

### Expectations and experiences of pain from oocyte retrieval

Measurements of the primary end-point, expectations of pain compared with experienced pain, showed that oocyte retrieval was significantly less painful than the women expected before the surgery. In addition, the women rated that they obtained adequate pain relief during the procedure. We also asked the women, in advance, to rate how much pain would be acceptable for them during surgery, and then we compared this with the experienced pain. Our findings showed that the pain experience during the oocyte retrieval procedure was considerably lower than women rated as acceptable, median 48.5 (5-94) versus 20.5 (1-98) on VAS ($P$ <0.0001). Concerning their ability influence pain management, the women's ratings were comparable before and after oocyte retrieval.

### Anxious women

The definitions used, in this thesis, of anxious and not anxious women are described under methodological considerations concerning VAS (see page 28). When comparing anxious women with not anxious women, several significant differences in pain ratings were found. Anxious women expected significantly more pain prior to oocyte retrieval, median 52.0 (19-96) compared with 34.0 (2-78) on VAS ($P$ <0.0001), and they also rated their experienced mean pain during oocyte retrieval significantly higher than not anxious women, median 30.1 (1-76) compared with 13.5 (1-98) on VAS ($P$ = 0.014). The anxious women in our study were significantly younger.

### Satisfaction

Overall the women rated their satisfaction with the pre-operative information as high. Women pointed out the staff's competence as being of most importance for their sense of security during oocyte retrieval.

**Conclusions:** The women rated oocyte retrieval as less painful than they had expected before the surgery. Women who are about to start IVF treatment should be given this information, since it might reduce their apprehension about the level of pain they could expect in conjunction with oocyte retrieval.

**Comments:** This study was conducted because many women often express strong worries concerning pain during oocyte retrieval. These findings, relating to both anxious and not anxious women, confirmed what many women express after oocyte retrieval: "this went better than expected" or from the staff point: "it is not that bad, you will manage". Most women are reassured by encouragement and support from their partner and the staff, but some women suffer from high anxiety. In turn, high-anxiety patients are known to have lower pain thresholds than low-anxiety patients (Caumo *et al*., 2001, Klages *et al*., 2004, Tang and Gibson, 2005). However, the problematic issue still remains of how to help those women who do not manage to cope with the pain. VAS has been shown to be a useful tool for identifying pre-operatively anxious patients (Kindler *et al*., 2000), but tends to provide a relatively high number of false-positive patients. Otherwise, when using self-reported scales, respondents tend to give less negative responses concerning issues that might stimulate negative thoughts and feelings, while in a conversation or interview the respondents tend to give a more negative picture of the situation (Forsberg-Wärleby, 2002). The quantitative analysis used answered the question of whether experienced pain was in accordance with expectations before surgery, presented as median and means. Using a qualitative design might have added other dimensions and a deeper understanding of women's fear in the specific situation. Waiting for surgery or on invasive procedures is considered stressful, and anxiety affects both physiological and psychological factors. Music has been used to promote relaxation and reduce anxiety. The theoretical basis of music as a tool to decrease anxiety is that it affects the autonomic nervous system possibly involving modulation of neurotransmittors (Cooke *et al*., 2005). The use of the sedative anxiolytic has been questioned, especially when used for short outpatient procedures, since it is associated with more drowsiness and a prolonged recovery. Ng *et al.* (Ng *et al*., 2002) in a double-blind RCT evaluated the effects of anxiloytic premedication on pre-operative anxiety and abdominal pain in conjunction with oocyte retrieval. Women were allocated, 30 minutes before surgery, to receive either premedication with pethidine and promethazine or placebo with normal saline. Degree of anxiety was measured at baseline and prior to onset of oocyte retrieval, performing comparisons within subjects. Pre-operative anxiety was significantly higher in the placebo group before surgery as compared with the premedication group, and abdominal pain during the procedure was significantly lower in the premedication group. However, women in both groups reported similar satisfaction scores regarding pain control, but the premedication group complained significantly more about drowsiness post-operatively than the women in the placebo group.

**Paper IV**

**Effects of alfentanil dosage during oocyte retrieval on fertilization and embryo quality**

In this retrospective observational study, the specific aim was to investigate potential adverse effects of varying doses of an analgesic drug commonly used during oocyte retrieval - alfentanil - on fertilization rate and embryo quality. A total of 663 women who underwent oocyte retrieval were subsequently included. Comparisons were performed between a high-dose group of alfentanil (>0.5 mg alfentanil) and a low-dose of alfentanil (≤0.5 mg alfentanil).

Women in the high-dose group were significantly younger than women in the low-dose group. No other between-group differences in patient characteristics were found.

**Fertilization rates and embryo quality**

Between the low and high-dose groups, no differences in mean fertilization rate, 0.6 ± 0.3 versus 0.6 ± 0.2 ($P$ = 0.678, adjusted $P$ = 0.937, 95% CI for the difference - 0.041; 0.044) or mean GQE rate, 0.6 ± 0.3 versus 0.5 ± 0.3 ($P$ = 0.207, adjusted $P$ = 0.179, 95% CI for the difference -0.015; 0.078) respectively, were found. The high-dose group used nitrous oxide more frequently than the low-dose group. Fertilization rate and GQE rate were comparable between women who used and women who did not use nitrous oxide. Benzodiazepam was used in low and similar frequencies in both groups. In a subgroup of 65 women who underwent repeated oocyte retrieval with different doses of alfentanil, a paired comparison of low versus high doses of alfentanil on fertilizaton and GQE rates were made. No difference in fertilization rate and GQE rate was found in this analysis, either. Significantly more women in the low-dose group had no embryos transferred owing to failed cleavage/poor embryo quality 15.1% versus 7.8% ($P$ = 0.005 adjusted $P$ = 0.004). In the high-dose group, significantly more women had all their embryos cryopreserved owing to threatening ovarian hyperstimulation syndrome. Mean number of embryos transferred, pregnancy rate, and live birth rate were comparable between the low and the high-dose groups of alfentanil.

**Conclusions:** This study found no negative influence on fertilization, embryo development, or pregnancy rate that correlated with higher doses of alfentanil administered in conjunction with oocyte retrieval. Although few patients, no adverse effect of benzodiazepam and nitrous oxide were detected. Women can be offered the doses of alfentanil needed to obtain pain relief.

**Comments:** Although RCTs is considered a superior design for intervention studies to produce the most reliable results, observational studies may well be used to investigate potential adverse effects of drugs (Vandenbroucke, 2004). It could be ethically problematic to randomise women in need of pain relief to receive a predetermined dose; in the present study patients received the amount of drug they required. It is also easier to include a sufficient number of individuals in an

Observational study. One strength of this study is the paired comparison of patients who underwent two IVF cycles with different doses of alfentanil. A weakness of the study is the absence of a group of women who underwent oocyte retrieval without alfentanil; thus a negative effect of alfentanil *per se* cannot be ruled out. Several factors, such as the women's age and the reason for infertility, influence IVF success. In the present study, numbers of oocytes retrieved, numbers of cleaved embryos, and numbers of GQEs were significantly higher in women administered a high dose of alfentanil than in women administered a low dose. This corresponds to the fact that women in the high-dose group were significantly younger than women in the low-dose group. Younger age is usually associated with a stronger response to ovarian stimulation resulting in more follicles. It is reasonable to believe that with more follicles punctured during oocyte retrieval, and thereby more oocytes retrieved; higher doses of alfentanil are required for adequate analgesia. We chose fertilization rate and GQE rate as primary end-points, assuming that when laboratory conditions were similar, these rates would be independent of the number of oocytes retrieved (Bosch *et al.*, 2008), and we adjusted *P*-values for age. No adverse effects of the higher amount of alfentanil on cleavage or embryo quality were found. Significantly more women in the low-dose group had no embryo transfer owing to failed cleavage/poor embryo quality, 15.1% versus 7.8% ($P = 0.005$ adjusted $P = 0.004$). This is probably attributeable to the lower number of retrieved oocytes in this group.

# GENERAL DISCUSSION

***Measurements of pain***

The VAS was chosen for measurements since it has been shown to be sensitive to pain intensity (Fernandez and Turk, 1992), and is designed to provide information about subjective feelings such as anxiety (Kindler *et al*., 2000, Lee and Kieckhefer, 1989). Previous studies have measured pain in conjunction with oocyte retrieval using VAS (Stener-Victorin *et al*., 1999, Stener-Victorin *et al*., 2003, Humaidan and Stener-Victorin, 2004, Ng *et al*., 2001, Ng *et al*., 2003, Ng *et al*., 1999, Ng *et al*., 2000, Sator-Katzenschlager *et al*., 2006, Tummon, 2004). It is considered easy to use in the clinical situation, although its precision has been questioned. One criticism is the limitations of predetermined lower and upper limits (Svensson, 1998) and important information might be missed. The handling of VAS is also a matter of debate. We handled ratings of VAS as a numerical scale; another way would have been to dichotomize data into increase or decrease, for instance in paper III, comparing baseline measurements (expectations of pain) with assessed measurements of experienced pain. In this case, the magnitude of clinical relevance of the findings might be lost since any change, small or large would only be assessed as increased or decreased. In addition, comparison with other studies would have been more difficult since the most common way to present findings of VAS is using it as a continuous variable.

Pain was measured using self-assessment reports in all studies in this thesis. Pain is a subjective experience, and therefore the most reliable method of assessing pain is to ask the woman about her pain. Gohar *et al.* (Gohar *et al*., 1993) showed that physician observer assessments and women's self-assessments of pain in conjunction with oocyte retrieval did not correspond. The physicians rated pain intensity lower than women did. In paper II, participating physicians had the clinical impression that POB gave better pain relief than PCB. Women undergoing IVF may be highly motivated and reluctant to show their emotions.

Baseline measurements of pain are considered important since they make with-in comparisons of pain possible. If baseline measurements had been performed, in papers I-II, it might have been possible to explain some of the variability in pain. It is possible to believe that women in the EA group had some more abdominal discomfort preoperatively since they had significantly more follicles and egg retrieved than women in the CA group (paper I).

The great heterogeneity in studies has been pointed out as a problem when evaluating effects of pain relieving methods used for oocyte retrieval (Stener-Victorin, 2005, Kwan *et al*., 2005). Studies have concentrated on analgesic effects and women's satisfaction, while timing of pain measurements and tools used have not been in focus. Timing of measurements is important. Especially acute pain should be assessed as soon as

possible; otherwise measurements tend to be imprecise (Stener-Victorin, 2005, Hampton, 2005). The timing of pain measurements was in this thesis determined in advance, to be assessed at defined timepoints, pre-, per- and post-operatively in paper II, and pre- and post-operatively in papers I and III.

Although one tends to think of pain as a homogenous sensory entity, pain may vary in intensity, quality, duration and referral area, all of which can be classified in different categories for better understanding of patient's experience of pain (intensity – mild/moderate/severe; quality – sharp/ burning/dull; duration – transient/inter-mittent/persistent; referral area- superficial or deep/localised or diffuse). Furthermore it is essential when measuring and treating pain to distinguish between pain threshold and tolerance level. The pain threshold is the minimum amount of physical stimulation sufficient to evoke a pain sensation, and represents the sensory-discriminative dimension. The tolerance level is the point at which further stimulation is unacceptable, and encompasses the affective-emotional components of pain. In paper II, the McGill Pain Questionnaire was used as a complement to VAS, giving both the sensory-discriminative and the affective-emotional dimensions of pain (Melzack, 1975). This was also used by Tummon (Tummon, 2004) when comparing lidocaine vaginal gel with PCB.

## Measurements of well-being and satisfaction

In paper I, well-being was measured to determine whether the clinical impression from previously performed studies, a better post-operative well-being for women who had EA as pain relief during oocyte retrieval than women who had CA, was correct (Stener-Victorin *et al*., 1999, Stener-Victorin *et al*., 2003). We used the STAI since it has been shown to produce both validity and reliability, and have been used extendedly in research work and in studies including surgery patients (Badura-Brzoza *et al*., 2008, Hermes *et al*., 2007, Mancuso *et al*., 2008, Oztekin *et al*., 2008). The study did not find a significant difference in well-being between the groups. It is possible that a more specific measurement for assessing well-being corresponding to the effects of acupuncture would have been to measure physiological effects rather than psychological effects of autonomic responses, or perhaps measuring both psychological and physiological effects. Neuroendocrine functions could have been assessed by measuring cortisol in saliva.

Several studies have reported a high level of satisfaction among women, in conjunction with oocyte retrieval, in spite of experienced pain being high (Kwan *et al*., 2005). This might reflect the sense of the overall success of the procedure when oocytes are collected, overriding pain. In paper I, 50% of women stated that they would be willing to use EA again despite significantly higher levels of pain than using CA.

### *Comments on RCTs*

The great advantage of RCTs is that this design takes care of confounders both of known and unknown origin. In paper I, randomization was stratified for degree of

anxiety since anxiety was assumed to be and important predictor of pain. Later logistic regression analysis was used for adjustment of pain ratings since women in the EA group had significantly more egg retrieved than women in the CA group. We missed to stratify for the number of follicles noticed at the last ultrasound investigation, which might have balanced the groups concerning the number of oocytes retrieved. Acupuncture treatment often produces strong interaction between the caregiver and the patient, which is difficult to control for. To minimize the introduction of bias, the midwife who administered EA did not assist the woman in the operating theatre. In paper II, several variables were identified as potential predictors and the so called "minimization technique" was used to obtain an optimal allocation between groups (Pocock, 1983). Other factors that could have had an effect on the results were the qualifications of the midwives and physicians when administrating EA (paper I) or local anesthesia (paper II).

**Sample size calculation**

The power of a study is the possibility to detect an effect of a specified size if it exists in the population. Calculating in advance, the necessary sample size gives a high probability of finding a true effect of a given magnitude (Altman, 1996). One important factor that should be taken into consideration when calculating sample size is clinical relevance.

In paper I, sample sized was calculated for the primary aims; pain and well-being. Concerning pain, the SD was based on a previous study, which compared the pain relieving effects of EA and PCB compared with and alfentanil and PCB, by Stener-Victorin *et al.* (Stener-Victorin *et al*., 1999). No previous study had evaluated well-being in this context. We used stress as a comparable variable to well-being, and our SD was based on the studies of Gallinelli *et al.* 2001, concerning immunological changes and stress in IVF, and Verhaak *et al.* 2001 regarding stress and marital satisfaction among women during their first IVF treatment (Gallinelli *et al*., 2001, Verhaak *et al*., 2001).

In paper II, a 30% decrease in overall pain both sides was considered clinically relevant corresponding to better pain relief with POB than PCB. The SD was based on the findings of paper I in this thesis, for momentary pain and operation pain in the CA group.

**Implications for future research**

Studying pain objectively is a complex issue since pain is a subjective experience. As McCaffery (McCaffery, 1979) stated: "pain is whatever the patient says it is and existing whenever she says it does."

Guidelines and standardization for timing of pain measurements and instruments used are further steps in the direction of making studies more homogenous.

Quantitative studies dominate research in the context of pain studies in ART. In future research, it would be interesting to introduce and interfere quantitative studies with qualitative study design, which might contribute to a deeper understanding concerning women's experience of pain that may otherwise go undetected. Currently, no method of pain relief can be regarded as being superior to another

(Kwan *et al.*, 2005, Stener-Victorin, 2005). This may reflect the subjective nature of pain and that the optimal method of pain relief may also be individual.

# CONCLUSIONS

*The conclusions drawn from this thesis are as follows:*

**Paper I)** EA cannot generally be recommended as a pain-reliving method for oocyte retrieval but might be an alternative for women who wish to try a non-pharmacological method. An advantage of EA is less post-operative tiredness and confusion, as compared with CA.

**Paper II)** No differences were found in overall pain experienced during the entire oocyte retrieval procedure with POB as compared with PCB. No advantage was found with the new POB technique. Both POB and PCB combined with alfentanil are safe methods with rapid onset, rapid recovery and ease of administration and monitoring. No differences on fertilization and embryo development were found between the groups.

**Paper III)** Women rated oocyte retrieval as less painful than they had expected before surgery. Women who are about to start IVF treatment should be given this information, since it might reduce their apprehension about the level of pain they could expect in conjunction with the oocyte retrieval procedure. Satisfaction with pre-operative information was high. Staff's competence was considered as most important for sense of security.

**Paper IV)** This study found no negative influence on fertilization, embryo development, or pregnancy rate in conjunction with higher doses of alfentanil administered at oocyte retrieval. Although few patients, no adverse effects of benzodiazepam and nitrous oxide were detected. Women can be offered the dose of alfentanil that is needed to obtain adequate pain relief.

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