

DEBATE—continued

Alternative treatments in reproductive medicine: much ado about nothing

Acupuncture—a method of treatment in reproductive medicine: lack of evidence of an effect does not equal evidence of the lack of an effect

Elisabet Stener-Victorin^{1,5}, Matts Wikland², Urban Waldenström³ and Thomas Lundeberg⁴

¹Department of Obstetrics and Gynaecology, Göteborg University, SE-413 45 Göteborg, ²Fertility Centre Scandinavia, SE-402 29 Göteborg, ³IVF centre Falun, SE-791 82 Falun and ⁴Department of Physiology and Pharmacology, Karolinska Institutet, Stockholm, SE-171 77 Stockholm, Sweden

To whom correspondence should be addressed. E-mail: elisabet.stener-victorin@medstud.gu.se

The increasing popularity of alternative treatments—methods of treatment that are not generally established in Western medicine—demands a serious debate about scientific documentation, efficacy and safety. It has been argued that there is no alternative medicine. There is only scientifically proven, evidence-based medicine supported by solid data, and we agree. Different methods of treatment, referred to as alternative treatments, are used by millions of patients every day which generates billions of dollars in health care expenditure each year. Therefore, it is important that physicians become more knowledgeable about different methods of treatment and increase their understanding of the possible benefits and limitations of each therapy. This article is intended to illustrate acupuncture in reproductive medicine today, as an example of a method of treatment that has become more established in Western medicine over the last decade. Discussed are the scientific documentation and aspects of acupuncture research, the physiological basis for the use of acupuncture, and evidence for the use of acupuncture in reproductive medicine. We are well aware that there are few well-designed papers on the effectiveness of different treatments in this field. However, we need to adhere to these principles, as we hope, do the readers of the present debate article.

Key words: acupuncture/evidence-based medicine/infertility safety/reproductive medicine

Introduction

Acupuncture is a method of treatment which dates back at least 3000 years. It is an integral part of Traditional Chinese Medicine (TCM), and over the last decade it has become established in Western medicine. Acupuncture may be exotic for some people, but to refer to it as a 'new method'—as did Renckens in the opening article of this Debate series—is not entirely correct (Renckens, 2002). Because of the lack of scientific documentation, however, there is scepticism over the effects claimed for acupuncture. The underlying mechanisms of acupuncture are often described in the language of TCM, and surprisingly few penetrating discussions have dealt with the physiological background of acupuncture.

Evidence-based medicine (EBM) and acupuncture

In debates on if and how alternative treatments, i.e. methods of treatment that are not generally established in Western medicine, have been scientifically tested, it must be stated that many treatments in conventional medicine have not been rigorously tested either, but the scientific community generally

acknowledges that this is a failing that needs to be remedied. It might also be argued that both conventional medicine and alternative medicine rely on anecdotes, some of which are published as case reports in peer-reviewed journals. We agree with the statement by Angell and Kassirer that there cannot be two kinds of medicine—conventional and alternative (Angell and Kassirer, 1998). There is only medicine that has been adequately tested and medicine that has not, medicine that works and medicine that may or may not work. Once a treatment has been tested rigorously, it no longer matters whether it was considered alternative at the outset. If it is found to be reasonably safe and effective, it should be accepted. But assertions, speculation, and testimonials are no substitutes for evidence. All methods of treatment referred to as alternative treatments should be subjected to scientific tests of efficacy no less rigorous than those required for conventional treatments.

Acupuncture is an example of a method of treatment that can be evidence-based if practitioners embrace standardization and conventional standard tools. The tools of evidence-based medicine (EBM) can help us to understand and explain the underlying mechanisms of the effects claimed for acupuncture.

The practice of EBM means integrating individual expertise with the best available external clinical evidence from systematic research, and this clinically relevant research often comes from basic science (Sackett, 1997). We read the debate article with regret since the author did not appear to have taken the pains to read the documentation about the acupuncture mechanisms or articles referred to. As an example, Renckens (Renckens, 2002) refers to a study in a category that uniformly reports favourable outcomes of electro-acupuncture (EA) (Chen, 1997) and argues that conclusions were not drawn from a randomized clinical trial (RCT) of normal quality. This is correct since this study was conducted on ovariectomized rats (Chen, 1997). The aim was to explain the mechanisms of EA in the regulation of the function of the hypothalamus–pituitary–gonadal (HPG) axis, which was related to effects of EA previously reported in women with polycystic ovarian syndrome (PCOS) by the same group (Chen and Yu, 1991; Chen, 1997).

We agree that the quality of clinical acupuncture studies is generally less than high and that properly designed and conducted RCTs in acupuncture are lacking. However, in recent years the effect of acupuncture on different conditions (pain and diseases) has been studied from a Western scientific perspective, and the results show that acupuncture has both a physiological and a psychological impact (Andersson and Lundeberg, 1995).

RCT, experimental trials, and acupuncture

RCTs are valued in medicine because they can test for causality, determine effect size, assess the risks and benefits of treatments, and minimise selection and measurement bias. Difficulties in designing appropriate experimental protocols for acupuncture trials have so far hampered clinical investigations. Many methods of treatment that are not generally established in Western medicine are difficult to blind or have no satisfactory placebos, but these methodological problems can be overcome. One example is the development of a ‘placebo needle’ for use in acupuncture research—the placebo looks exactly like an acupuncture needle, and it causes the same tap penetration sensation (Streitberger and Kleinhenz, 1998). For a placebo control for acupuncture to be effective, Streitberger stated that patients should not be able to distinguish the placebo treatment from tap penetration of a needle (Streitberger, 1988) and that patients should experience placebo penetration in the same therapeutic setting as in acupuncture (Streitberger and Kleinhenz, 1998). It has been shown that invasive acupuncture is more effective than the Streitberger placebo needle (Kleinhenz *et al.*, 1999). Even if there are objections to the use of a placebo needle, it may well be the best tool to date to use in designing controlled comparative studies on the efficacy of acupuncture needle penetration.

Attitudes to acupuncture and the psychological preparation before treatment are thought to influence the outcome. The power of the psycho-neuro-immunological effect of acupuncture could be very high in certain circumstances, and it is difficult to control these factors in the clinical situation. Kaptchuk discussed the importance of using a third no-treatment, no-placebo arm to differentiate between the

perception of a placebo effect and the ordinary natural history of a condition (Kaptchuk, 1998).

The use of experimental trials, in both animals and humans, enables us to evaluate the effects and mechanisms of acupuncture in a structured manner. The use of anaesthesia in animal models is another way to exclude many environmental variables. The ideal way to establish acupuncture procedures that are effective would be to conduct controlled comparative studies that consider every relevant variable of treatment. Such studies have been informally reviewed (Ceccherelli *et al.*, 2000). Moreover, if acupuncture can be established to be superior to placebo in some conditions, acupuncture might be considered to be ‘credible’. Placebo-controlled trials would then not be necessary for other similar conditions, and studies could then concentrate on direct comparisons with other treatment options. It seems sensible to aim at accumulating this type of trial in those clinical areas where trials already exist, and acupuncture appears to have a good effect size, such as in acute pain (Ernst and Pittler, 1998). Whatever the methodological design chosen, it is crucial that it is adequately described to make it possible to interpret the outcomes, and if necessary, to repeat the trial.

Is there a physiological basis for the use of acupuncture?

One question, among others, is vital before any judgements can be made about the validity of acupuncture as a treatment method: ‘Is there a physiological basis for the use of acupuncture?’ An understanding of the physiology of acupuncture is important in the refutation of the argument that the effect of acupuncture is implausible because it relies on concepts of changing the flow of energy in meridians. Needle insertion into the skin and deeper tissues results in a particular pattern of afferent activity in peripheral nerves. Basic scientific research suggests that manual stimulation of the acupuncture needle activates muscle afferents (mainly A-delta and possibly C-fibres). Another mode of stimulation is through the application of electrical stimulation, as in EA. Low-frequency EA (1–4 Hz) stimulation of the needles probably excites a group of receptors, found in muscles, denoted ergoreceptors (Kaufman *et al.*, 1984), which are physiologically activated during muscle contractions. Both EA and muscle exercise release endogenous opioids and oxytocin, which seem to be essential in the induction of functional changes in different organ systems (Andersson and Lundeberg, 1995). In this respect, particular interest has been dedicated to β -endorphin, an endogenous opioid with high affinity for the μ -receptor (Basbaum and Fields, 1984).

There is evidence that this hypothalamic β -endorphin system has a central role in mediating the pain-relieving effect of acupuncture (Wang *et al.*, 1990a,b) as well as the changes seen in autonomic functions after acupuncture. The latter changes are probably caused by an inhibition of the vasomotor centre (VMC) (Andersson and Lundeberg, 1995). This would result in a sustained decrease in general sympathetic tone with vasodilatation, increased skin temperature, and decreased blood pressure (Andersson and Lundeberg, 1995).

β -endorphin is also released into the blood from the hypothalamus via the anterior pituitary (Crine *et al.*, 1978).

This release is regulated by corticotrophin-releasing factor (CRF), which is produced and released from the paraventricular nucleus of the hypothalamus. CRF promotes the release of β -endorphin, adrenocorticotrophic hormone (ACTH), and melanocyte-stimulating hormone in equimolar amounts through stimulation of the synthesis of their precursor, pro-opiomelanocortin. These hormones exert their effects in different target organs via the bloodstream.

Stress increases the activity of the hypothalamic–pituitary–adrenal (HPA) axis and decreases reproductive functions. This suggests a close relationship between hormones of the HPA axis and those of the hypothalamic–pituitary–gonadal (HPG) axis. CRF, ACTH, β -endorphin, and adrenal corticosteroids play an important role in modulating the effect of stress on reproductive functions (Rivier and Rivest, 1991). Via this route, acupuncture may exert an effect on both the HPA axis and the HPG axis. Acupuncture has been shown to alter plasma β -endorphin levels and may thereby have an effect on the release of hypothalamic gonadotrophin-releasing hormone (GnRH) and pituitary gonadotrophin secretion (Chen and Yu 1991; Stener-Victorin *et al.*, 2000, 2002).

The two central β -endorphinergic systems described above operate independently, but both can be stimulated by afferent nerve activity (i.e. manual acupuncture, EA, and muscle exercise) (Andersson and Lundeberg, 1995).

Another system that may be involved in the modulation of stress, pain, autonomic and immune functions is the oxytocinergic system. 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). Interestingly, oxytocin effects β -endorphin release (Pettersson *et al.*, 1996). It is possible that oxytocin may account for some of the effects seen in rats that were given acupuncture including anxiolysis, increased pain withdrawal thresholds, and exploratory behaviour (Uvnäs-Moberg *et al.*, 1993).

In extensive studies by Sato and collaborators, it has been shown that both mild, ‘non-painful’, and strong, ‘painful’, acupuncture stimulation modulate spinal reflexes (Sato *et al.*, 1997). Strong, ‘painful’ stimulation results in increased sympathetic tone and pain. Mild, ‘non-painful’ stimulation has been shown to activate inhibitory systems in the spinal cord, resulting in segmental inhibition of sympathetic outflow (Sato *et al.*, 1997) and pain pathways, as predicted by the gate control theory (Melzack and Wall, 1965).

It has been suggested that mild, ‘non-painful’ stimulation of muscle afferents results in a decrease of glutamate and aspartate content in the dorsal horn, which may be mediated by a β -aminobutyric acid ergic mechanism (Lundeberg, 1996). Sandkuehler has demonstrated a long-term depression of synaptic transmission in the dorsal horn by low-frequency stimulation of A δ -fibres (Sandkuehler *et al.*, 1997). This depression of synaptic transmission was reduced or abolished by a blockage of N-methyl-D-aspartic acid receptors and may underlie segmental pain inhibition (Sandkuehler, 1996).

This mechanism is strictly topographically related, and an optimal effect is obtained by stimulating the somatic segments related to the pain area and/or the innervation of the affected organ (Andersson and Lundeberg, 1995). It should be stressed

that all of these modulating systems are under central control (Andersson and Lundeberg, 1995; Sandkuehler 1996b; Sato *et al.*, 1997).

In the periphery, acupuncture exerts effects by antidromic nerve impulses. These antidromic impulses induce the release of substance P (SP), vasoactive intestinal polypeptide (VIP), and calcitonin gene-related peptide (CGRP), which are likely to result in vasodilatation and in increased nutrition from improved blood flow (Dawidson *et al.*, 1998; Lundeberg *et al.*, 1988; Sato *et al.*, 2000).

It has been demonstrated that intense painful stimulation results in the activation of supraspinal pain inhibitory centres, and this mechanism is denoted diffuse noxious inhibitory controls (DNIC) or counter-irritation (Le Bars *et al.*, 1979). The pain-modulating effect is related to the release of endogenous opioids (Willer *et al.*, 1990) and is not specific, nor is it related to the site of stimulation. DNIC is seen following the activation of nociceptive afferents, and the mechanism involves supraspinal pathways, with descending projections to the dorsal horn at every level. The effect of DNIC can be blocked by naloxone (Le Bars *et al.*, 1992). This system is probably involved in the mechanism of acupuncture analgesia, and the effect has been observed in both humans and animals (Le Bars *et al.*, 1979, 1992). It is unlikely that DNIC is reinforced only by strong placebo, as stated by Renckens (Renckens, 2002) who appears to have overlooked the physiological significance of the DNIC phenomena.

Psychological impact of acupuncture

It is important to keep in mind that neurophysiological and humoral events are related to psychological factors. Undoubtedly, sensory stimulation—and particularly acupuncture—has the potential to produce strong placebo effects. Acupuncture is probably one of the most effective non-pharmacological methods in terms of activating placebo effects. Indeed, acupuncture works by stimulating endogenous opioids and so, it appears, does the placebo effect (Amanzio and Benedetti, 1999). To achieve optimal acupuncture therapy, physiological and psychological factors must interact in synergy, utilising their respective endogenous mechanisms efficiently (Andersson and Lundeberg, 1995; Thomas and Lundeberg, 1996).

Reports from China have stressed the importance of psychologically preparing patients before surgery when acupuncture analgesia is to be used (Bonica, 1974). The purpose of this preparation was to increase the effectiveness of the procedure and possibly to enhance the magnitude of the analgesia. The importance of psychological factors before and during acupuncture treatment is further supported by the findings of increased concentrations of cholecystokinin (CCK) in both animals and humans during anxiety and panic attacks (Cohen *et al.*, 1999). Since CCK is an endogenous opioid antagonist, increased concentrations may reduce or completely negate the positive effects of acupuncture treatment. Recently it was reported that anxiety decreased pain thresholds (Rhudy and Meagher, 2000).

The therapeutic response depends on the complicated interaction between patient factors and expectations, therapist factors and expectations, and treatment factors, including the

specific and non-specific effects of treatment (Filshie and Cummings, 1999). In conclusion, treatment outcome depends on the patient's responsiveness to the whole of the therapeutic encounter (Thomas and Lundeborg, 1996).

Is there evidence for the use of acupuncture in reproductive medicine?

The use of acupuncture in reproductive medicine has not been well investigated to date. Many childless couples hoping for pregnancy try acupuncture but there is no scientific documentation that establishes whether acupuncture affects the pregnancy rate, although empirical evidence suggests that this may be the case. Below are just three applications chosen as examples for discussion.

Circulatory effects of acupuncture—uterine artery blood flow impedance

The cardiovascular effects of acupuncture have been extensively studied and can be referred to a clinical area where trials already exist. Successful IVF and embryo transfer require optimal endometrial receptivity at the time of implantation. Blood flow impedance in the uterine arteries—measured by transvaginal ultrasonography expressed as a resistance index, the pulsatility index (PI), distal to the point of sampling—is considered valuable in assessing endometrial receptivity. In an uncontrolled study (as pointed out by Renckens) we showed that repeated EA treatments reduced a high PI value in the uterine arteries to normal levels (Stener-Victorin *et al.*, 1996). This is an interesting finding, but comparative RCTs with conventional treatments, such as low-dose aspirin, are needed.

EA as anaesthesia during oocyte aspiration

Renckens (Renckens, 2002) wonders whether the use of EA as anaesthesia during oocyte aspiration still persists in Gothenburg (Göteborg). Yes it does, as a method of pain relief during oocyte aspiration, and it works extremely well for patients who prefer trying this option to alfentanil. That EA induces adequate analgesia during a minor operation is not a new observation and can be referred to a clinical area where trials already exist (Ernst and Pittler, 1998). We therefore chose to compare EA with conventional anaesthetics (Stener-Victorin *et al.*, 1999), and we have recently finished a multi-centre follow-up study with 280 patients. These studies show that EA is as good an anaesthetic method as conventional anaesthetics during oocyte aspiration. In addition, women who received EA have less abdominal pain and nausea and were less stressed 2 h after aspiration. EA in combination with conventional anaesthetics has been shown to reduce the consumption of anaesthetics by 50% (Wang *et al.* 1994). Such a reduction is most likely preferable because the fast acting opiate, alfentanil, has been found in the follicular fluid shortly after i.v. injection (Soussis *et al.* 1995). In conclusion, EA is a valuable alternative or complement to conventional anaesthesia during oocyte aspiration.

Acupuncture effects on hormonal disturbances such as PCOS and anovulation

Most studies in this area use the approach of a trial of single intervention with variables analysed on an intention-to-treat

basis. Unfortunately, poor design and a lack of valid outcome measures and diagnostic criteria, which make it difficult to interpret the results, flaw most of them. The studies all report favourable outcomes such as ovulation induction; a regulatory effect on gonadotrophins, estrogens, and neuropeptides; and a tendency, for example, for miscarriage rates to be lower compared with hormonal treatment (Chen and Yu, 1991; Gerhard and Postneek, 1992; Xiaoming *et al.*, 1993).

Recently, we conducted a study on women with well defined and diagnosed PCOS and anovulation to elucidate the effect of repeated EA treatments on endocrinological and neuro-endocrinological parameters as well as on anovulation (Stener-Victorin *et al.*, 2000). This study showed that repeated EA treatments exert long-lasting effects on both endocrinological parameters as well as anovulation. These results are in accordance with previous results, but it is obvious that randomized, comparative studies are needed to verify the results and to exclude non-specific effects. However, these studies do not enlighten possible underlying mechanisms of EA. These are, for obvious reasons, difficult to study because tissue samples from the ovaries and central nervous system are unobtainable.

Experimentally induced polycystic ovaries (PCO) by a single i.m. injection of estradiol valerate (EV) in rats share many endocrinological and morphological characteristics of human PCO. Therefore, we used the steroid induced rat PCO model to investigate the effects and possible mechanisms of repeated EA treatments during light anaesthesia by analysing CRF (Stener-Victorin *et al.*, 2002) and nerve growth factor (NGF) (Stener-Victorin *et al.*, 2000) in the ovaries, the adrenal glands, and the central nervous system. The results indicate that EA modulates activity in the sympathetic nervous system and that there is a functional interaction between activity in the nervous and the endocrine systems.

In conclusion, it appears that acupuncture may have a beneficial effect on women with PCOS and anovulation, supported by both clinical and experimental evidence. Therefore, acupuncture may be a suitable alternative or complement to pharmacological induction of ovulation in a woman with PCOS, with no negative side-effects. However, there is a need for more RCTs in well-defined diagnoses.

Conclusion

Despite lack of evidence for the effectiveness of alternative medicines it cannot be ignored that 40% of the people in the United States use some form of alternative treatment (Eisenberg *et al.*, 1998). Whether there is a true effect or not, it is a compelling reason for the scientific community to investigate the method. No other body is better qualified for the task. Furthermore, acupuncture is a very safe intervention in the hands of competent practitioners (Vincent, 2001). The dangers of many orthodox procedures are certainly greater, though no easy comparisons can be made. On the other hand, it must be pointed out that unless substantiated by research, the therapeutic use and acceptance of acupuncture cannot be extended in the future with confidence. In any case, the arguments against alternative treatments in the Renckens' article (Renckens, 2002) are not relevant and have low, if any, scientific level. It

is, of course, unethical to promise cure and recovery when the method used lacks evidence of an effect. On the other hand, it is also unethical to disallow a method that demonstrably works.

We do agree that there are few well-designed papers on the effectiveness of methods of treatment that are not generally established in Western medicine, but we *do not* agree that it can best be summarised as ‘much ado about nothing’. We need to stick to basics and to have open scientific minds. We hope that this also applies to the readers of the present debate article.

References

- Amanzio, M. and Benedetti, F. (1999) Neuropharmacological dissection of placebo analgesia: expectation-activated opioid systems versus conditioning-activated specific subsystems. *J. Neurosci.*, **19**, 484–494.
- Andersson, S. and Lundeberg, T. (1995) Acupuncture—from empiricism to science: functional background to acupuncture effects in pain and disease. *Med. Hypotheses.*, **45**, 271–281.
- Angell, M. and Kassirer, J.P. (1998) Alternative medicine—the risks of untested and unregulated remedies. *N. Engl. J. Med.*, **339**, 839–841.
- Basbaum, A.I. and Fields, H.L. (1984) Endogenous pain control systems: Brain-stem spinal pathways and endorphin circuitry. *Ann. Rev. Neurosci.*, **7**, 309–338.
- Bonica, J.J. (1974) Anesthesiology in The People’s Republic of China. *Anesthesiol.*, **40**, 175–186.
- Ceccherelli, F., Gagliardi, G., Rossato, M. and Giron, G. (2000) Variables of stimulation and placebo in acupuncture reflexotherapy. *J. Altern. Complement. Med.*, **6**, 275–279.
- Chen, B.Y. (1997) Acupuncture normalizes dysfunction of hypothalamic-pituitary-ovarian axis. *Acupunct. Electrother. Res.*, **22**, 97–108.
- Chen, B.Y. and Yu, J. (1991) Relationship between blood radioimmunoreactive beta-endorphin and hand skin temperature during the electro-acupuncture induction of ovulation. *Acupunct. Electrother. Res.*, **16**, 1–5.
- Cohen, H., Kaplan, Z. and Kotler, M. (1999) CCK-antagonists in a rat exposed to acute stress: implication for anxiety associated with post-traumatic stress disorder. *Depress. Anxiety*, **10**, 8–17.
- Crine, P., Gianoulakis, C. and Seidah, N.G. (1978) Biosynthesis of beta-endorphin from beta-lipotropin and a larger molecular weight precursor in rat pars intermedia. *Proc. Natl Acad. Sci. USA*, **75**, 4719–4723.
- Dawidson, I., Angmar-Mansson, B., Blom, M., Theodorsson, E. and Lundeberg, T. (1998) The influence of sensory stimulation (acupuncture) on the release of neuropeptides in the saliva of healthy subjects. *Life Sci*, **63**, 659–674.
- Eisenberg, D.M., Davis, R.B., Ettner, S.L., Appel, S., Wilkey, S., Van Rompay, M. and Kessler, R.C. (1998) Trends in alternative medicine use in the United States, 1990–1997: results of a follow-up national survey. *JAMA*, **280**, 1569–1575.
- Ernst, E. and Pittler, M.H. (1998) The effectiveness of acupuncture in treating acute dental pain: a systematic review. *Br. Dent. J.*, **184**, 443–447.
- Filshie, J. and Cummings, M., (1999) Western medical acupuncture. In: E. Ernst and A. White (Eds.), *Acupuncture: a scientific appraisal. Butterworth-Heinemann, Oxford*, pp. 31–59.
- Gerhard, I. and Postneek, F. (1992) Auricular acupuncture in the treatment of female infertility. *Gynecol. Endocrinology*, **6**, 171–181.
- Kapchuk, T.J. (1998) Powerful placebo: the dark side of the randomised controlled trial. *Lancet*, **351**, 1722–1725.
- Kaufman, M.P., Waldrop, T.G., Rybycki, K.J., Ordway, G.A. and Mitchell, J.H. (1984) Effects of static and rhythmic twitch contractions on the discharge of group III and IV muscle afferents. *Cardiovasc. Rec.*, **18**, 663–668.
- Kleinhenz, J., Streitberger, K., Windeler, J., Gussbacher, A., Mavridis, G. and Martin, E. (1999) Randomised clinical trial comparing the effects of acupuncture and a newly designed placebo needle in rotator cuff tendinitis. *Pain*, **83**, 235–241.
- Le Bars, D., Dickenson, A.H. and Besson, J.M. (1979) Diffuse noxious inhibitory controls (DNIC). II. Lack of effects on non-convergent neurones, supraspinal involvement and theoretical implications. *Pain.*, **6**, 305–327.
- Le Bars, D., Willer, J.C. and De Broucker, T. (1992) Morphine blocks descending pain inhibitory controls in humans. *Pain.*, **48**, 13–20.
- Lundeberg, T., Kjartansson, J. and Samuelsson, U. (1988) Effect of electrical nerve stimulation on healing of ischaemic skin flaps. *Lancet*, **ii**, 712–714.
- Lundeberg, T. (1996) Electrical stimulation techniques [comment], *Lancet*, **348**, 1672–1673.
- Melzack, and Wall, P.D. (1965) Pain mechanism: A new theory. *Science*, **150**, 971–979.
- Petersson, M., Alster, P., Lundeberg, T. and Uvnäs Moberg, K. (1996) Oxytocin increases nociceptive thresholds in a long-term perspective in female and male rats. *Neurosci. Lett.*, **212**, 87–90.
- Renckens, C.N.M. (2002) Alternative treatments in reproductive medicine: much ado about nothing. *Hum. Reprod.*, **17**, 528–533.
- Rhudy, J.L. and Meagher, M.W. (2000) Fear and anxiety: divergent effects on human pain thresholds. *Pain*, **84**, 65–75.
- Rivier, C. and Rivest, S. (1991) Effects of stress on the activity of hypothalamic-pituitary-gonadal axis: peripheral and central mechanisms. *Biol. Reprod.*, **45**, 523–532.
- Sackett, D.L. (1997) Evidence-based medicine. *Semin. Perinatol.*, **21**, 3–5.
- Sandkuehler, J. (1996) The organization and function of endogenous antinociceptive systems. *Progr. Neurobiol.*, **50**, 49–81.
- Sandkuehler, J., Chen, J.G., Cheng, G. and Randic, M. (1997) Low frequency stimulation of afferent Aδ-fibers induces long-term depression of primary afferent synapses with substantia gelatinosa neurons in the rat. *J. Neurosci.*, **17**, 6483–6491.
- Sato, A., Sato, Y. and Schmidt, R.F. (1997) *The Impact of Somatosensory Input on Autonomic Functions*. Springer-Verlag, Heidelberg, 325 pp.
- Sato, A., Sato, Y., Shimura, M. and Uchida, S. (2000) Calcitonin gene-related peptide produces skeletal muscle vasodilation following antidromic stimulation of unmyelinated afferents in the dorsal root in rats. *Neurosci. Lett.*, **283**, 137–140.
- Soussis, I., Boyd, O., Paraschos, T., Duffy, S., Bower, S., Troughton, P., Lowe, J. and Grounds, R. (1995) Follicular fluid levels of midazolam, fentanyl, and alfentanil during transvaginal oocyte retrieval. *Fertil. Steril.*, **64**, 1003–1007.
- Stener-Victorin, E., Waldenström, U., Andersson, S.A. and Wikland, M. (1996) Reduction of blood flow impedance in the uterine arteries of infertile women with electro-acupuncture. *Hum. Reprod.*, **11**, 1314–1317.
- Stener-Victorin, E., Waldenström, U., Nilsson, L., Wikland, M. and Janson, P.O. (1999) A prospective randomized study of electro-acupuncture versus alfentanil as anaesthesia during oocyte aspiration in in-vitro fertilization. *Hum. Reprod.*, **14**, 2480–2484.
- Stener-Victorin, E., Lundeberg, T., Waldenström, U., Manni, L., Aloe, L., Gunnarsson, S. and Janson, P. (2000) Effects of electro-acupuncture on nerve growth factor in rats with experimentally induced polycystic ovaries. *Biol. Reprod.*, **63**, 1507–1513.
- Stener-Victorin, E., Waldenström, U., Tagnfors, U., Lundeberg, T., Lindstedt, G. and Janson, P.O. (2000) Effects of electro-acupuncture on anovulation in women with polycystic ovary syndrome. *Acta Obstet. Gynecol. Scand.*, **79**, 180–188.
- Stener-Victorin, E., Lundeberg, T., Waldenström, U., Bileviciute-Ljungar, I. and Janson, P. (2002) Effects of electro-acupuncture on corticotropin releasing-factor (CRF) in rats with experimentally induced polycystic ovaries. *Neuropep.*, **35**, 1–5.
- Streitberger, K. and Kleinhenz, J. (1998) Introducing a placebo needle into acupuncture research. *Lancet*, **352**, 364–5.
- Thomas, M. and Lundeberg, T. (1996) Does acupuncture work? *Pain Clinic Updates*, **4**, 1–4.
- Uvnäs-Moberg, K., Bruzelius, G., Alster, P. and Lundeberg, T. (1993) The antinociceptive effect of non-noxious sensory stimulation is mediated partly through oxytocinergic mechanisms. *Acta Physiol. Scand.*, **149**, 199–204.
- Vincent, C. (2001) The safety of acupuncture. *Br. Med. J.*, **323**, 467–468.
- Wang, B.G., Wang, E.Z., Chen, X.Z., Sun, F.L. and E.H., Y. (1994) Han’s Acupoint Nerve Stimulator (HANS) in combination with enflurane for anesthesia in cranial operations. *Chin. J. Anesthesiol.*, **14**, 427–429.
- Wang, Q., Mao, L. and Han, J. (1990a) The arcuate nucleus of hypothalamus mediates low but not high frequency electroacupuncture analgesia in rats. *Brain Res.*, **513**, 60–66.
- Wang, Q., Mao, L.-M. and Han, J.-S. (1990b) The role of periaqueductal gray in mediation of analgesia produced by different frequencies of electroacupuncture stimulation in rats. *Int. J. Neurosci.*, **53**, 167–172.
- Willer, J.C., Le Bars, D. and De Broucker, T. (1990) Diffuse noxious inhibitory controls in man: involvement of an opioidergic link. *Eur. J. Pharmacol.*, **182**, 347–355.
- Xiaoming, M.O., Ding, L.I., Yunxing, P.U., Guifang, X.I., Xiuzhen, L.E. and Zhimin, F.U. (1993) Clinical studies on the mechanism for acupuncture stimulation of ovulation. *J. Trad. Chin. Med.*, **13**, 115–119.