Acupuncture and Assisted Reproductive Technology: The Effect on Clinical Pregnancy Rates

Hannah K. Holland
holland.hannah.k@gmail.com

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ACUPUNCTURE AND ASSISTED REPRODUCTIVE TECHNOLOGY: THE EFFECT ON CLINICAL PREGNANCY RATES

A Thesis Presented to
The Faculty of the School of Medicine
Yale University

In Candidacy for the degree of
Master of Medical Science

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Hannah K. Holland, PA-SII
Class of 2015
Yale Physician Associate Program

J. Ryan Martin, MD
Assistant Professor
Obstetrics, Gynecology, and Reproductive Services
# TABLE OF CONTENTS

Title Page.................................................................................................................i

Table of Contents....................................................................................................ii

Abstract..................................................................................................................v

Chapter 1: Introduction..............................................................................................1

  Background.............................................................................................................1

  Statement of the Problem.......................................................................................5

  Goals and Objectives.............................................................................................6

  Hypothesis.............................................................................................................6

  Definitions.............................................................................................................7

  References............................................................................................................8

Chapter 2: Review of the Literature...........................................................................11

  Introduction..........................................................................................................11

  Review of Empirical Studies..................................................................................12

    Mechanisms of Acupuncture..............................................................................12

    Positive Association of Acupuncture and Clinical Pregnancy Rates............15

    Negative Association of Acupuncture and Clinical Pregnancy Rates........17

    Equivocal Association of Acupuncture and Clinical Pregnancy Rates..17

  Review of Studies Regarding Possible Confounding Variables..................20

  Review of Relevant Methodology.......................................................................23

    Study Design and Study Setting.......................................................................23

    Sampling and Recruitment for Study Population........................................24

    Selection Criteria...............................................................................................25
Randomization and Blinding Techniques ......................................... 26
Interventions and Methods of Administration .................................. 27
Primary and Secondary Outcome Measures ...................................... 29
Sample Size and Statistical Significance .......................................... 30
Strengths and Limitations of Previous Studies .................................. 31
Conclusion ......................................................................................... 32
References ......................................................................................... 33
Chapter 3: Study Methods ................................................................. 38
Study Design ...................................................................................... 38
Study Population and Sampling ......................................................... 38
Subject Protection and Confidentiality .............................................. 38
Recruitment ...................................................................................... 40
Study Variables and Measures ......................................................... 40
  True Acupuncture Intervention ......................................................... 40
  Sham Acupuncture Intervention ...................................................... 41
  Standard Medical Care .................................................................. 42
  Primary and Secondary Outcome Measures .................................... 42
  Baseline Variables ........................................................................ 42
Methodology Considerations ............................................................ 43
  Assignment of Intervention ............................................................ 43
  Blinding of Intervention ................................................................. 43
  Blinding of Outcome ..................................................................... 43
  Adherence ...................................................................................... 44
ABSTRACT

Acupuncture has become an increasingly common concurrent therapy for women undergoing assisted reproductive technology. The effect of acupuncture on clinical pregnancy rates, however, remains unclear due to inconclusive study results and a lack of proper comparisons to all possible controls. In this three arm, single-blinded, randomized controlled trial, we propose to compare clinical pregnancy rates of women under age 35 undergoing in vitro fertilization with or without intracytoplasmic sperm injection in combination with either true acupuncture, sham acupuncture, or standard medical care. We hypothesize there will be a statistically significant difference in clinical pregnancy rates six to eight weeks following embryo transfer of women undergoing concurrent true acupuncture therapy. The results of this study will help to end the acupuncture debate and allow practitioners to accurately advise their patients about what may improve their chances of conception as they undergo the stressful and expensive treatment that is in vitro fertilization.
CHAPTER 1: INTRODUCTION

Background:

One in seven to ten couples in industrialized nations suffer from infertility as they attempt to build their family.\(^1\) Causes of infertility may include ovulatory disorders, tubal disease, peritoneal adhesions, endometriosis, uterine abnormalities, abnormalities of sperm, and advancing female or male age.\(^2,3\) Even with this range of possibilities, five to twelve percent of infertility cases remain unexplained after a thorough evaluation.\(^2,4\) Infertile couples or individuals often call upon assisted reproductive technologies, one subset of which is in vitro fertilization with or without the assistance of intracytoplasmic sperm injection, as they search for medical assistance to biologically parent a child.

In 2013, approximately 175,000 cycles of in vitro fertilization were completed in the United States.\(^4\) This process of in vitro fertilization, first successfully completed in 1978, involves harvesting oocytes, mixing of oocyte and spermatozoon (insemination), and waiting to allow the oocyte and spermatozoon to combine (fertilization).\(^5\) The oocyte may also be fertilized via intracytoplasmic sperm injection, during which a single sperm is injected directly into the oocyte.\(^6\) Once the oocyte and spermatozoon combine, an embryo forms and begins to grow.\(^7\) If an embryo is deemed of good quality, it is then transferred into the woman’s uterus.\(^7,8\) Regardless of the cause of infertility, in vitro fertilization with or without intracytoplasmic sperm injection treatment leads to the highest pregnancy rates per cycle when compared to other treatment options.\(^8\) The most recently available United States birth report, published in 2012, recorded the highest number of
babies born via in vitro fertilization to date, accounting for approximately 1.5% of all live births that year.4,9

In vitro fertilization carries with it many potential risks, including those in the physical, mental, and emotional realms. Some of the potential physical risks include bleeding, infection, anatomic structure damage, and ovarian hyperstimulation syndrome, a complication that may result in a build up of fluid in the abdomen and chest.7 Additionally, many women report increased amounts of stress and anxiety associated with this procedure, as being faced with barriers to constructing a biological family can be psychologically damaging.10-13 The financial burden of in vitro fertilization is another challenge that many women must confront. Although there is great state variability, it is estimated that approximately 75% of insurance companies do not cover the cost of in vitro fertilization.14 This results in a financial responsibility that the patient must assume, one that frequently exceeds $10,000 per in vitro fertilization cycle.8,14

In addition to the numerous risks and challenges associated with in vitro fertilization, successful pregnancy is not guaranteed. Current statistics indicate that for women under the age of 35, 46% of in vitro cycles result in a clinical pregnancy.4 As the ages of women increase, the rates of clinical pregnancies decrease due to reductions in oocyte quantity and quality.15 Women ages 35 to 37 achieve a clinical pregnancy in 38% of in vitro cycles; women ages 38 to 40 achieve a clinical pregnancy in 29% of in vitro cycles; women ages 41 to 42 achieve a clinical pregnancy in 19% of in vitro cycles; women over the age of 42 achieve a clinical pregnancy in 8.9% of in vitro cycles.4 The success rates of in vitro fertilization,
expressly in context of the numerous associated risks and challenges, highlight the need for practitioners and patients to do everything possible to increase a woman’s pregnancy success after undergoing this method of assisted reproductive technology.

One contemporaneous therapy that has been proposed to increase clinical pregnancy rates in women undergoing in vitro fertilization is acupuncture. Acupuncture is an integral part of traditional Chinese medicine, and its employment can be dated back at least 4,000 years. Its use has gained increased popularity in the Western world, including facets in dental medicine, pain management, and chemotherapy. Acupuncture involves inserting fine, sharp needles into the skin along pathways, or meridians, in an effort to alter the flow of energy, or Qi, throughout the body. Traditional Chinese medicine upholds that Qi must flow smoothly with strength and quality for health to be maintained.\(^{16}\) Along the meridians through which Qi flows are specific regions, known as acupuncture points or acupoints, that have unique electrical properties and connective tissue characteristics.\(^{17,18}\)

Specific acupoints are used to treat certain medical conditions; points related to the traditional Chinese medicine concept of the *Kidney* are linked to reproductive success.\(^{19-21}\) The *Kidney* in this context does not refer to the organ and associated functions from a biomedical perspective; rather, the *Kidney* underlies the fundamental constitution of a person and represents reproductive function in females. In the framework of traditional Chinese medicine, infertility represents a premature decline of *Kidney*-related functions that result from either constitutional,
lifestyle, or dietary factors. Additionally, stimulation of the Taiying meridians (spleen) and Yangming meridians (stomach and colon) will result in improved blood flow and increased energy in the uterus, thus potentially enhancing reproductive function.

In a standard acupuncture session, between four to ten acupoints are needled for ten to thirty minutes. The needles stimulate the body via either manual twirling or small amounts of electric current, which is known as electro-acupuncture. Acupuncturists attempt to produce the sensation of DeQi, which is a sense of heaviness, soreness, or numbness at the point of needling; this is regarded as a sign of correctly arousing the acupuncture point. Negative side effects of acupuncture are possible, most commonly minor bleeding and needling pain, though a prospective study has determined such adverse effects are rare; when practiced correctly, acupuncture is deemed one of the safer forms of medical intervention.

There are several theories that may support acupuncture and its potential utilization to increase pregnancy rates in in vitro fertilization. Acupuncture has been shown to alter plasma beta-endorphin levels, which in turn alter the release of gonadotropin-releasing hormone, pituitary gonadotropin secretion, and luteinizing hormone, thus impacting the hypothalamic-pituitary-ovarian axis. In animal studies, the effects of acupuncture on the hypothalamic-pituitary-adrenal and the hypothalamic-pituitary-gonadal axes have been shown to affect the release of corticotrophin-releasing factor and subsequently decrease stress responses; these decreasing stress responses have subsequently been linked to reproductive function.
and success.\textsuperscript{27,28} In addition, acupuncture has been shown to reduce uterine artery resistance, thus increasing blood flow, which may increase the likelihood of embryo implantation and potential successful pregnancy.\textsuperscript{29}

**Statement of the Problem:**

Conceiving a child with the assistance of in vitro fertilization is becoming increasingly common.\textsuperscript{9} It is hypothesized that the current societal trend of marrying and beginning a family later in life, potentially due to an increasing focus on education and career, coupled with advancements in birth control preventing accidental pregnancies, has spurred this increase in the utilization of in vitro fertilization.\textsuperscript{8,9,30} It is well accepted that as parental age increases, fertility decreases; this fertility-age association has been demonstrated in both males and females.\textsuperscript{3,31} With increasing utilization of in vitro fertilization and the potential physical, psychological, and financial stresses that coincide its employment, optimizing the chance of a clinical pregnancy is a shared goal between the patient and the practitioner.

A suggested strategy aimed at increasing clinical pregnancy rates in women undergoing in vitro fertilization is concurrent employment of acupuncture near the time of embryo transfer; however, data on the intersection of acupuncture and reproductive care is controversial and in need of further research. High-quality randomized controlled trials that study the effect of acupuncture on clinical pregnancy rates are lacking. Current data are conflicting, as these studies attest to various relationships between acupuncture and clinical pregnancy rates. Existing
research conclusions range from acupuncture having a positive to an equivocal to a negative effect on clinical pregnancy rates. In addition, previous studies lack randomization to all possible comparison groups—sham acupuncture and standard medical care—in a single, well-designed randomized controlled trial.

**Goals and Objectives:**

We propose to conduct a multicenter, three arm, single-blinded, randomized controlled trial comparing clinical pregnancy rates of women under age 35 undergoing in vitro fertilization with or without intracytoplasmic sperm injection in combination with either true acupuncture, sham acupuncture, or standard medical care. The goal of this study is to determine the efficacy of acupuncture on clinical pregnancy rates among women under age 35 undergoing in vitro fertilization with or without intracytoplasmic sperm injection. The objective of this study is to compare the different percentages of clinical pregnancy among the women in the true acupuncture group, the sham acupuncture group, and the standard medical care group.

**Hypothesis:**

We hypothesize there will be a statistically significant difference in clinical pregnancy rates six to eight weeks following embryo transfer of women under age 35 undergoing concurrent true acupuncture therapy compared to the sham acupuncture group and the standard medical care group.
Definitions:

**Assisted reproductive technology:** procedures for the treatment of infertility in which both oocytes and sperm are handled outside the body; these include in vitro fertilization with transcervical embryo transfer, intracytoplasmic sperm injection, gamete and zygote intrafallopian transfer, frozen-embryo transfer, and donor-embryo transfer.\(^{32}\)

**Clinical pregnancy:** evidence of at least one gestational sac with fetal heart motion confirmed via transvaginal ultrasound six to eight weeks post embryo transfer.\(^{33}\)

**Infertility:** failure to conceive after twelve months of regular, unprotected sexual intercourse, or the occurrence of more than two consecutive natural miscarriages or stillbirths.\(^{34,35}\)

**Sham acupuncture:** needling with a sham needle (Streitberger placebo needle) where true skin penetration does not occur because the tip of the needle is blunted and retracts upon itself or needling with traditional acupuncture needles in an area not recommended by traditional Chinese medicine practitioners for fertility treatment.\(^{33,36}\)

**Standard medical care:** no true acupuncture or sham acupuncture treatment – following embryo transfer, the patient is placed on bed rest for 25 minutes (approximately the time period of one true acupuncture or sham acupuncture intervention minus needle insertion and removal time).\(^{37}\)

**True acupuncture:** traditional acupuncture practice where needles are inserted in the classic meridian points – needling can be achieved with a stimulation source of either fine needles, the hand, moxibustion with warming needles, or electrical
stimulation (acupuncture sources without needling – such as laser, acupressure, tap-pricking, point injection, or cupping on pricked superficial blood vessels – are not considered true acupuncture).33

References:

European Journal of Obstetrics & Gynecology and Reproductive Biology. 
14. Bitler MP, Schmidt L. Utilization of infertility treatments: the effects of 
15. Piette C, De Mouzon J, Bachelot A, Spira A. In-vitro fertilization: influence of 
17. Langevin HM, Yandow JA. Relationship of acupuncture points and meridians to 
connective tissue planes. The Anatomical Record. 2002;269(6):257-265.
impedance along connective tissue planes associated with acupuncture 
Sciences; 2011.
20. Lyttleton J. Treatment of infertility with Chinese medicine. Elsevier Health 
Sciences; 2013.
fertilization and acupuncture: clinical efficacy and mechanistic basis. 
22. Zollman C, Vickers A. ABC of complementary medicine: Users and 
practitioners of complementary medicine. BMJ: British Medical Journal. 
23. White A, Hayhoe S, Hart A, Ernst E. Survey of adverse events following 
acupuncture (SAFA): a prospective study of 32,000 consultations. 
24. Chen D SX, Cai MX. Clinical observation on treatment of functional 
anovulation by acupuncture prick. Zhongguo Zhong Xi Yi Jie He Za Zhi 
Janson PO. Effects of electro-acupuncture on anovulation in women with 
26. Chen B-Y. Acupuncture normalizes dysfunction of hypothalamic-pituitary-
ovarian axis. Acupuncture and Electro Therapeutics Research-the 
27. Stener-Victorin E, Lundeberg T, Waldenström U, Bileviciute-Ljungar I, Janson 
P. Effects of electro-acupuncture on corticotropin-releasing factor in rats with 
experimentally-induced polycystic ovaries. Neuropeptides. 


CHAPTER 2: REVIEW OF THE LITERATURE

Introduction:

Between December 2014 and April 2015, a comprehensive review of the medical literature applicable to this proposed randomized controlled trial was conducted. Utilizing the databases of PubMed and Ovid MEDLINE, the following keywords were searched in a variety of combinations: acupuncture, acupuncture therapy, anxiety, anxiety symptoms, assisted reproduction, assisted reproductive techniques, assisted reproductive technology, clinical pregnancy, clinical pregnancy rates, depression, depressive symptoms, fertility, finance, in vitro fertilization, infertility, intracytoplasmic sperm injection, pregnancy, pregnancy rates, psychology, psychological stress, and subfertility. All applicable articles written in the English language between January 1965 and April 2015 were examined for significance pertaining to the current topic and analyzed. Through the resulting literature review, the proposed mechanisms for the relationship between acupuncture and pregnancy rates and the existing conflicting data regarding acupuncture, in vitro fertilization with or without intracytoplasmic sperm injection, and pregnancy rates were analyzed. This literature review illustrates the need for a multicenter, randomized controlled trial to resolve the ambiguity surrounding the utilization of acupuncture therapy during in vitro fertilization.
Review of Empirical Studies:

Mechanisms of Acupuncture:

There are several theories that may support acupuncture and its potential utilization to influence pregnancy rates during in vitro fertilization. One notion is that acupuncture alters plasma beta-endorphin levels; after administration of electroacupuncture, plasma beta-endorphin levels increased from 65.59±24.15 pg/ml to 80.09±22.16 pg/ml (p>0.05) in women who did not experience ovulation and from 65.5±24.15 pg/ml to 38.86±10.11 pg/ml (p<0.05) in women who did experience ovulation. Plasma beta-endorphin levels, in turn, alter the release of gonadotropin-releasing hormone, pituitary gonadotropin secretion, and luteinizing hormone, thus impacting the hypothalamic-pituitary-ovarian axis. While simultaneously increasing the levels of hypothalamic/pituitary beta-endorphin and peripheral luteinizing hormone, electroacupuncture has also been shown to decrease the levels of hypothalamic gonadotropin-releasing hormone and pituitary luteinizing hormone in animal studies. These results suggest that the effects of acupuncture on the regulation of the hypothalamic-pituitary-ovarian axis may be exerted via the promotion of the hypothalamic-pituitary-adrenal axis through increasing the synthesis and secretion of adrenal steroid hormones. These androgens may then be transformed into estrogen in other tissues and therefore reset the negative feedback of estrogen on the hypothalamic-pituitary-ovarian axis. Furthermore, electroacupuncture may accelerate the release of brain and pituitary beta-endorphin, which subsequently inhibits the over-regular secretion of
gonadotropin-releasing hormone and luteinizing hormone that has erroneously normalized.¹

In additional animal models, the effects of acupuncture on the hypothalamic-pituitary-adrenal and the hypothalamic-pituitary-gonadal axes have been shown to affect the release of corticotrophin-releasing factor, a physical and emotional stress-related peptide linked to pathological changes in reproductive functions.⁴⁻⁷ One study demonstrated that in rats with experimentally-induced polycystic ovaries who received electroacupuncture treatments, the ovarian concentration of corticotrophin-releasing factor was significantly lower in both the experimentally-induced polycystic ovarian control group [p<0.01; confidence interval (CI)=0.03, 0.13] and the healthy control group (p<0.05; CI=-0.11, -0.008).⁴ Decreasing stress and its subsequent negative physiologic effects has been linked to improved reproductive function and pregnancy success in human subjects.⁸

Stress and anxiety, although harmful to obstetric outcomes, are very common in infertility patients, especially those undergoing in vitro fertilization.⁹ Potential reasons for this include the invasive nature of therapy, the awareness that in vitro fertilization is often the last hope to biologically parent a child, and the financial burden of treatment.¹⁰,¹¹ Moreover, anxiety and depression are related to discontinuation of assisted reproductive technologies after the first in vitro fertilization cycle and are related to lower pregnancy rates.¹²⁻¹⁵ Acute and chronic stress negatively affects pregnancy, live birth rates, and birth weights of children conceived via assisted reproductive technology, thus reducing stress and anxiety during the in vitro fertilization process is beneficial to patients and their potential
for reproductive success.\textsuperscript{15-18} Acupuncture is a proposed mechanism for reducing said stress and anxiety. True acupuncture treatment was associated with less perceived stress both before (p=0.01) and after (p=0.02) embryo transfer in an observational, prospective, cohort study.\textsuperscript{8} Additionally, in a randomized controlled trial conducted to study the effects of acupuncture on mitigating anxiety, true auricular acupuncture at the relaxation point demonstrated a significantly reduced anxiety level when compared to the sham auricular acupuncture control at both thirty minutes (p=0.007) and 24 hours (p=0.035) post-treatment.\textsuperscript{19}

Beyond its normalizing effects on the endocrinological and psychological aspects of reproduction, acupuncture has been shown to reduce uterine artery resistance, which increases blood flow to the uterus via decreased tonic activity in the vasoconstrictor fibers to the uterus and a general inhibition of the sympathetic system.\textsuperscript{20,21} The acupuncture group of a randomized controlled trial experienced a significant reduction in pulsatility index of both uterine arteries after electroacupuncture treatment when compared to the non-acupuncture control. In the acupuncture group, the right uterine artery pulsatility index decreased from 2.4±0.7 to 2.2±0.6 (p=0.01), and the left uterine artery pulsatility index decreased from 2.3±0.6 to 2.0±0.6 (p=0.002); in the control group, the right uterine artery pulsatility index decreased from 2.4±0.6 to 2.3±0.5 (p=0.44), and the left uterine artery pulsatility index decreased from 2.5±0.8 to 2.3±0.4 (p=0.27).\textsuperscript{22} Another randomized controlled trial demonstrated acupuncture’s reduction of nerve growth factor, a neurotrophin in both the sympathetic and the sensory nervous systems, in the ovaries, therefore indicating a reduction in peripheral sympathetic nerve
hyperactivity and contractility of the uterus.\textsuperscript{21} This acupuncture-induced increase in blood flow into a more relaxed uterus may create a more favorable endometrium, thus increasing the likelihood of embryo implantation and potential successful pregnancy.\textsuperscript{20-22}

Positive Association of Acupuncture and Clinical Pregnancy Rates:

In numerous studies, acupuncture has been associated with an increase in clinical pregnancy rates. In 2002, Paulus et al. conducted a randomized controlled trial of 160 patients with good quality embryos undergoing in vitro fertilization with or without intracytoplasmic sperm injection at a single clinic in Germany. Patients were randomized into either a true acupuncture group (n=80) or a standard of care group (n=80). Acupuncture was performed 25 minutes before and after embryo transfer while the standard of care group received no additional interventions. The same clinician, who was blinded to the patient’s acupuncture status, conducted all oocyte retrievals and embryo transfers; the same acupuncturist directed all acupuncture sessions. The true acupuncture group attained a pregnancy rate of 43\% in comparison to the standard of care group’s pregnancy rate of 26\% [p=0.03; odds ratio (OR)=2.08; CI=1.07, 4.04].\textsuperscript{23}

Dieterle et al. randomized 225 patients undergoing in vitro fertilization with or without intracytoplasmic sperm injection into a true acupuncture group (n=116) and a sham acupuncture group (n=109). Unlike similar studies, treatment included placing Chinese herbs in patients’ ears, and the sham acupuncture control group received true acupuncture at sites believed not to affect fertility. The true acupuncture group achieved a clinical pregnancy rate of 34\%; the sham
acupuncture group achieved a clinical pregnancy rate of 16% (p<0.01; OR=2.74; CI=1.44, 5.22).24

In a randomized controlled trial by Westergaard et al., 273 Danish women were randomized into a true acupuncture on the day of embryo transfer only group (n=95), a true acupuncture on the day of embryo transfer and again two days later group (n=91), or a standard of care group (n=87). Acupuncture was performed immediately before and after embryo transfer for the first two groups, with each session lasting 25 minutes; then, one 25 minute session was performed two days later in the second grouping only. Specially trained nurses performed all acupuncture treatments. Clinical pregnancy rates were significantly higher in the acupuncture group receiving treatment only on the day of embryo transfer as compared to the standard of care group. Clinical pregnancy rates of the acupuncture on the day of embryo transfer only group and the standard of care group were 39% and 24%, respectively (p=0.038). The clinical pregnancy rate in the acupuncture group receiving treatment on the day of embryo transfer in addition to two days post embryo transfer (36%) was higher than in the standard of care control, but the difference did not reach statistical significance (p>0.05). When the two acupuncture treatment groups were combined for statistical analysis, the total clinical pregnancy rate was 38% and achieved statistical significance over the 24% clinical pregnancy rate of the standard of care group (p<0.05; OR=1.90; CI=1.07, 3.37).25
Negative Association of Acupuncture and Clinical Pregnancy Rates:

Although several studies have demonstrated a positive association between acupuncture and clinical pregnancy rates, there has been a single report of a negative association between true acupuncture and clinical pregnancy rates. Craig et al. demonstrated this association in a multicenter, randomized controlled trial investigating clinical pregnancy rates and true acupuncture before and after embryo transfer. The participants were randomized into either an acupuncture group that performed one acupuncture treatment 25 minutes before and after embryo transfer (n=57) or a standard of care group (n=56). The intervention was provided by one of two licensed, board-certified acupuncturists utilizing a unique acupuncture protocol at an off-site acupuncture clinic located within five miles of the fertility centers. The true acupuncture group achieved a clinical pregnancy rate of 44%; comparatively, the standard of care group achieved a clinical pregnancy rate of 65% (p=0.045; OR=0.34; CI=0.15, 0.79). Unlike other investigations, this study included egg donation recipients, and acupuncture treatment was performed at an off-site location.

Equivocal Association of Acupuncture and Clinical Pregnancy Rates:

Though trials have demonstrated both a positive association and a negative association between acupuncture surrounding embryo transfer and subsequent clinical pregnancy rates, many studies have demonstrated a lack of statistically significant effect. In a four-center, randomized controlled trial of fertility patients in Denmark by Andersen et al., 635 patients less than or equal to 37 years of age that were scheduled for embryo transfer were randomized into either a true
acupuncture group (n=314) or sham acupuncture group utilizing a Streitberger placebo needle (n=321). True acupuncture was performed by nurses who were either professional acupuncturists or by nurses who had received instruction and training by the acupuncturists prior to the trial, and participants were needled for 25 minutes before and after embryo transfer at points consistent with traditional Chinese medicine. Both the patients and the clinicians performing embryo transfer were blinded. The true acupuncture group achieved a clinical pregnancy rate of 32%, while the sham acupuncture group achieved a clinical pregnancy rate of 35% (p>0.05; OR=0.88; CI=0.64, 1.23).\textsuperscript{28} Similarly to the methodology of Andersen et al., Moy et al. randomized 161 patients less than or equal to 37 years of age into a true acupuncture group (n=87) or a sham acupuncture group (n=74). The clinical pregnancy rates of the true acupuncture group and sham acupuncture group were 45% and 53%, respectively (p=0.35; OR=0.74; CI=0.40, 1.39).\textsuperscript{29}

Paulus et al. furthered their investigation in 2003 by comparing a true acupuncture group (n=100) to a sham acupuncture group (n=100) while following the same protocol as their 2002 study.\textsuperscript{23,30} Subjects were needled 25 minutes pre and post embryo transfer by acupuncturists utilizing either a true needle or placebo needle at the same acupoints. Unlike their 2002 study, no statistically significant effect was observed. The true acupuncture group achieved a clinical pregnancy rate of 43% while the sham acupuncture group achieved a clinical pregnancy rate of 37% (p>0.05; OR=1.28; CI=0.73, 2.26).\textsuperscript{30}

Benson et al. conducted a randomized controlled trial at a single clinic in the United States during which fertility patients were randomized into a true
acupuncture group (n=53), a relaxation group (n=50), or a no treatment group (n=50). True needle acupuncture was performed for 25 minutes before and after embryo transfer, the relaxation group rested for 25 minutes before and after embryo transfer, and the no treatment group received the standard of care. Clinical pregnancy rates in the true acupuncture arm, relaxation arm, and no treatment arm were 55%, 42%, and 44%, respectively (p=0.24; OR=1.54; CI=0.17, 3.35). While the results did not have adequate power to achieve statistical significance, likely due to the small sample size, the clinical significance of an over 10% increase of clinical pregnancy rates in the true acupuncture group when compared to the relaxation group and no treatment group should be considered.

In an attempt to replicate the Paulus et al. (2002) study in the United States, Domar et al. randomized 146 patients to true acupuncture (n=78) or standard of care (n=68). Acupuncture was performed utilizing the Paulus protocol by a single acupuncturist 25 minutes before and after embryo transfer. For unclear reasons, the results were not replicable to the Paulus et al. (2002) study, as the true acupuncture group achieved a clinical pregnancy rate of 31% while the standard of care group achieved a clinical pregnancy rate of 34% (p=0.69; OR=0.87; CI=0.43, 1.74). This randomized controlled trial also investigated anxiety and optimism of patients by having subjects complete the Spielberger State Trait Anxiety Inventory (STAI) (Appendix V), the most widely used and reliable measure of both state and trait anxiety, and the Life Orientation Test-revised, a tool developed to assess individual differences in optimism and pessimism. When compared to the control subjects, the acupuncture patients reported significantly less anxiety after
embryo transfer, enjoyed their sessions more, felt more relaxed, and reported feeling significantly more optimistic about their chance of conception.³²

Similar to the Domar et al. trial, So et al. also investigated clinical pregnancy rate and anxiety levels; however, this trial utilized a true acupuncture group (n=185) and a sham acupuncture group (n=185) who received their true or placebo treatments 25 minutes before and after embryo transfer by a single, certified Chinese acupuncturist. While no significant difference in clinical pregnancy rates were detected between the groups, the true acupuncture group had a clinical pregnancy rate of 39% and the sham acupuncture group had a clinical pregnancy rate of 49% (p=0.059; OR=0.66; CI=0.44, 1.01). Additionally, both groups had a significant reduction of anxiety after treatment (p<0.001) when compared with anxiety levels before the acupuncture treatments.³⁵ Although the clinical pregnancy rates of the two groups were not statistically significant, the clinical significance of a 10% increase in clinical pregnancy rates in the sham acupuncture group compared to the true acupuncture group cannot be ignored, lending the possibility that sham acupuncture may not be completely inert.

**Review of Studies Regarding Possible Confounding Variables:**

Several confounding variables with the ability to influence outcomes for women undergoing in vitro fertilization with or without intracytoplasmic sperm injection have been identified, and thus need to be controlled for while investigating the relationship between acupuncture and clinical pregnancy rates. Age is a well-established deterrent to successful biological childbearing due to both female and
male factors. Increase in female age correspond to decreases in oocyte quantity and oocyte quality; increases in male age correspond to decreases in semen volume, sperm motility, and sperm morphology. Numerous investigations have demonstrated that the negative effects of female age can be largely overcome with the utilization of donated eggs; however, among subjects who received donated eggs, the pregnancy rates remained significantly lower in older women which suggests an age-related effect on endometrial receptivity. Regarding male age-related declines in fertility, these often can be largely overcome through the employment of intracytoplasmic sperm injection during the in vitro fertilization process.

Once age has been controlled for, the duration of infertility, the number of previous treatment cycles, the number of previous pregnancies, and the patient's body mass index (BMI) have also been shown to affect in vitro fertilization outcomes. In a retrospective analysis of over 35,000 in vitro fertilization cycles, Templeton et al. investigated numerous potential confounders thought to influence the success of in vitro fertilization, which were operationalized in this study as live birth rates. Increasing duration of infertility between one and twelve years demonstrated a significant decrease in age-adjusted live birth rates (p<0.001). Similarly, increases in in vitro fertilization attempts were associated with a decline of live birth rates (p<0.001) with the first attempt attaining the highest level of success. Women who achieved any previous clinical pregnancy had a significantly higher live birth rate than women with no previous clinical pregnancies (p=0.014),
yet a previous successful clinical pregnancy via in vitro fertilization was an even stronger predictor of future success.40

Beyond a woman’s obstetric history, BMI has also been identified as a potential confounder of successful pregnancy.44,45 Nichols et al. demonstrated that either extremes of weight, underweight (BMI<20 kg/m^2) or obese (BMI≥28 kg/m^2), were associated with a decrease in clinical pregnancy rates; however, differences among the weight groups existed with respect to primary diagnosis. Compared to normal-weight (20<BMI ≤27.9 kg/m^2) women, obese women were the least diagnosed with endometriosis (12% versus 29%; p<0.001) and most diagnosed with ovarian dysfunction (32% versus 15%; p=0.013). Underweight women were more likely to have a uterine problem (8.9% versus 0.3%; p=0.001) or unexplained infertility (6.7% versus 0.9%; p=0.003) compared to normal-weight women. Male factor infertility was least associated with underweight women compared to the other two groups (6.7% versus 17% and 10%, respectively; p=0.021). Obese women experienced a lower clinical pregnancy rate of 35% when compared to the normal-weight women clinical pregnancy rate of 52% (p=0.003); similarly, underweight women also experienced a lower clinical pregnancy rate of 36% when compared to the 52% clinical pregnancy rate of normal-weight women (p=0.049).45

In regards to the variances seen in infertility diagnoses, no statistically significant differences of in vitro fertilization success have been demonstrated. Two studies have demonstrated no difference by cause of infertility in both clinical pregnancy rates and live birth rates.40,46 Templeton et al. reported no significant differences in live birth rates per treatment cycle when comparing the infertility
diagnoses of tubal disease, endometriosis, cervical/uterine conditions, and unexplained infertility ($p=0.48$).

Similarly, Mahadevan et al. reported no significant differences in clinical pregnancy rates when comparing the infertility diagnoses of tubal blockage, endometriosis, male factor, and unexplained infertility ($p>0.05$).

Although these investigations do not reveal a statistically significant difference of in vitro fertilization success rates by infertility diagnoses, one could argue that the above-mentioned Nichols et al. study investigating BMI and clinical pregnancy rates was confounded by the statistical differences of primary infertility diagnoses among the weight groups.

It is thus best practice to ensure adequate randomization of both infertility diagnoses and BMI, along with age, the duration of infertility, the number of previous treatment cycles, and the number of previous pregnancies, in all study arms when investigating this topic of in vitro fertilization success.

**Review of Relevant Methodology:**

>This portion of the literature review includes methodology that is relevant to the proposed study. A more thorough explanation of the proposed study methods will be presented in Chapter 3.

**Study Design and Study Setting:**

The proposed study will be a multicenter, three arm, single-blinded randomized controlled trial examining the efficacy of true acupuncture compared to sham acupuncture and standard medical care on clinical pregnancy rates in women under age 35 undergoing in vitro fertilization with or without intracytoplasmic
sperm injection. Centers will include Boston IVF in Waltham, Massachusetts; Weill Medical College of Cornell University Center for Reproductive Medicine and Infertility in New York, New York; and Yale Fertility Center in New Haven, Connecticut. These sites were chosen due to their similar clinical pregnancy rates in women under age 35 undergoing in vitro fertilization and their standings as academic institutions. The previously described studies were all randomized controlled trials, the gold standard of study designs, yet some employed a multicenter design while others employed a single-center design. The following studies that demonstrated a positive association were all single-center designs: Paulus et al. (2002) and Dieterle et al. completed their studies at two university fertility centers in Germany; Westergaard et al. completed their study at a private fertility center in Denmark. Craig et al., which demonstrated a negative effect of true acupuncture compared to standard medical care, utilized a multicenter design comprising three fertility clinics in the United States. Of the studies that demonstrated an equivocal association, Andersen et al. was the sole multicenter design, consisting of four Danish fertility centers. The remaining studies with equivocal associations consisted of single-center designs in China, Germany, and the United States. The single-center designs allowed for more consistent implementation of study methods; however, the multicenter designs are more generalizable to a larger population of women undergoing in vitro fertilization.

**Sampling and Recruitment for Study Population:**

The patients in the proposed study will be recruited on a non-random, consecutive basis from Boston IVF, Weill Medical College of Cornell University
Center for Reproductive Medicine and Infertility, and Yale Fertility Center. Recruitment will occur over a 20-month period to obtain an adequate number of eligible participants and to allow sufficient time for data collection completion by 24 months. This recruitment method is similar to all previously described randomized controlled trials investigating acupuncture and clinical pregnancy rates.

**Selection Criteria:**

Inclusion criteria for the proposed study will consist of women less than 35 years of age who are undergoing in vitro fertilization with or without intracytoplasmic sperm injection Boston IVF, Weill Medical College of Cornell University Center for Reproductive Medicine and Infertility, and Yale Fertility Center. Exclusion criteria will include age greater than or equal to 35, use of donor eggs, use of frozen and thawed embryos, previous participation in this study, and a lack of written, informed consent. In comparison to the proposed study, the Paulus et al. studies included embryos only deemed of good quality, the Craig et al. study included use of donor eggs, and several studies utilized varying age restrictions on participants. In order to ensure comparable expected pregnancy rates for sample size calculations, probable embryo quality, and consistent endometrial receptivity based on age-related factors, we propose to investigate only women under age 35 who are undergoing in vitro fertilization with or without intracytoplasmic sperm injection.
Randomization and Blinding Techniques:

Randomization in the proposed study will be accomplished via a computer generated randomization list to a 1:1:1 allocation into the three treatment arms. The allocation will then be concealed in a sequentially-numbered, opaque, sealed envelope. This method is most similar to the Paulus et al. (2002), the Paulus et al. (2003), the Moy et al., and the So et al. studies. Of the investigations described, however, all were at low risk of selection bias due to their utilization of some method of computer-generated randomization except for the Benson et al., the Dieterle et al., and the Westergaard et al. studies. These three randomized controlled trials are at an unclear risk of selection bias secondary to not reporting their randomization techniques in detail; however, all of these studies reported no significant differences between the study groups for the noted demographic information.

Regarding randomization of potential confounding variables, the proposed study will account for age, duration of infertility, number of previous treatment cycles, number of previous pregnancies, BMI, and infertility diagnoses. Of the studies with a positive association, the Paulus et al. (2002) study did not report the duration of infertility, the number of previous treatment cycles, the number of previous pregnancies, or the BMI of the true acupuncture and the standard of care groups; however, there were no significant differences of infertility diagnoses between groups. The Dieterle et al. and Westergaard et al. studies reported on all potential confounders except the number of previous pregnancies and noted no significant differences between groups. For the only investigation that
demonstrated a negative association, the Craig et al. trial, the potential confounding variables of duration of infertility, number of previous treatment cycles, number of previous pregnancies, BMI, and infertility diagnoses were not noted. Of the studies with an equivocal association, Andersen et al. and So et al. did not account for number of previous pregnancies, while Moy et al. did not account for duration of infertility. Since both the Paulus et al. (2003) and the Benson et al. trials were published only as abstracts, specific demographic data were not available for their subjects. Finally, the Domar et al. trial did not note demographic data for the duration of infertility, the number of previous pregnancies, the BMI, or the infertility diagnoses.

Due to the nature of the interventions and their delivery, only single-blinding is possible in the proposed study. The providers completing the embryo transfer, ultrasound assessment, and any general care of the patient will not be aware of the patient’s treatment allocation. The patients undergoing true acupuncture and sham acupuncture will also be blinded to their treatment provision; however, those patients receiving standard medical care will be cognizant to their treatment arm. All subjects will be instructed not to discuss their study group with any clinical staff directly involved in their care. In previous studies investigating true versus sham acupuncture, double-blinding was achieved, and in those investigating true acupuncture versus standard of medical care, only single-blinding was possible.

Interventions and Methods of Administration:

The proposed study will have three treatment arms: true acupuncture, sham
acupuncture, and standard medical care. True acupuncture will occur with sterile disposable stainless steel needles at acupoints designated by traditional Chinese medicine principles to govern reproduction and will adhere to the second edition of the Standard Acupuncture Nomenclature.\textsuperscript{23,35,50-53} The depth of needle insertion into the skin will depend on the location of the acupoints, ranging from 10 to 20 millimeters. DeQi will be elicited during the initial insertion; after 10 minutes, DeQi will be maintained by rotating, lifting, and thrusting the needle handles. Needles will remain in place for a total of 25 minutes before being removed.\textsuperscript{35} Sham acupuncture will follow the same methodology as true acupuncture; however, instead of traditional needles, sterile Streitberger placebo needles will be utilized. This placebo needle has the same appearance as the true needle; however, the placebo needle is not fixed into the copper handle and the tip of the needle is blunt, thus when it is pushed into the skin, the needle will slide into the handle and the whole needle will appear shortened. To blind subjects receiving the sham acupuncture, this placebo will give patients a pricking penetration sensation.\textsuperscript{35,54} The same acupoints will be needled in both the true and sham acupuncture groups. All acupuncture treatments will utilize the same methodology and be performed on each site by a certified, Chinese acupuncturist with a degree in Chinese Medicine, at least three years prior clinical experience in fertility acupuncture, training in Human Subjects Protection, and training in the proposed study's procedures. Further controlling the true acupuncture and sham acupuncture interventions, no significant differences in side effects have been demonstrated between groups in previous investigations.\textsuperscript{35}
Regarding previous studies’ administration of true acupuncture and/or sham acupuncture, each study employed different criteria for the acupuncturists: the Paulus et al. investigations utilized “well-trained examiners”; Dieterle et al. and Domar et al. utilized a single acupuncturist without details of qualifications; Westergaard et al. and Andersen et al. utilized “specially trained nurses” or nurses who were professional acupuncturists; Moy et al. utilized numerous licensed acupuncturists; So et al. utilized a single certified Chinese acupuncturist with a degree in Chinese Medicine and three years experience; Craig et al. utilized two licensed, certified acupuncturists with greater than three years experience; and Benson et al.’s acupuncture administration techniques were not detailed due to publication as only an abstract.23-32,35

The acupoints utilized in the previous randomized controlled trials varied slightly among investigations. All focused on fertility acupoints as designated by traditional Chinese medicine. The most commonly utilized protocol, which the proposed study will follow, consisted of the following acupoints: Cx6, GV20, Liv3, LI4, LU7, SP6, SP8, SP10, ST29, and ST36.23,28,30,32,53,55 Additional points incorporated in the remaining randomized controlled trials include: Cv6, DU20, LR3, PC6, Ren3, RN6, K3, K13, and K111.24,25,27,29,35,55 For the specific acupoints of each empirical trial, please see Appendix III; for an explanation of acupoints, please see Appendix IV.

Primary and Secondary Outcome Measures:

The primary outcome measure in the proposed study is clinical pregnancy, which will be determined via transvaginal ultrasound six to eight weeks post
embryo transfer. All study arms will be evaluated for this outcome, similar to previous randomized controlled trials investigating this topic. The secondary outcome measures in the proposed study are anxiety symptoms and depressive symptoms. Analogous to the Domar et al. trial, the STAI (Appendix V), a widely used and reliable 40-item evaluation, will be used to measure both state and trait anxiety before and after embryo transfer and assigned treatment intervention or standard medical care on the day of embryo transfer. Similar to other studies investigating the psychological components of assisted reproductive technologies, depressive symptoms will be evaluated with the well-validated 20-item Zung Self-Rating Depression Scale (Appendix VI), before and after embryo transfer and assigned treatment intervention or standard medical care on the day of embryo transfer.

Sample Size and Statistical Significance:

Given our study design, we will be using two-sided hypothesis testing with an alpha of 5%, a beta of 20%, and a power of 80%. After the primary outcome measure was determined for the proposed study, the literature was reviewed to establish the effect size of true acupuncture on clinical pregnancy rates. Of the studies that found a statistically significant difference in clinical pregnancy rates, the differences ranged from 15% to 21%. Based on this, we aim to detect a 15% difference between true acupuncture and standard medical care. Via Power and Precision Version 4 software, this leads to an unadjusted sample size of 173 subjects per arm, consequently equaling 519 subjects in total. Based on previous research, the highly motivated nature of these patients, and only a six to eight week follow-up
To ensure adequate sample size, we will anticipate a conservative 10% dropout rate, projecting a sample size of 191 subjects per arm and 573 subjects in total. Further details pertaining to the sample size calculation and statistical significance are provided in Chapter 3 and Appendix VII.

Strengths and Limitations of Previous Studies:

Beyond the methodological strengths and limitations previously described, some general strengths and limitations of the previous randomized controlled trials ought to be noted. Of the studies demonstrating a positive association of acupuncture and clinical pregnancy rates, the Paulus et al. (2002) study was the first randomized controlled trial to demonstrate a statistically significant effect and thus spurred future research on the topic and laid the foundation for fertility acupoint determination in Western medical trials. The trial and its positive results, however, also prompted patients and clinicians to begin using acupuncture as a concurrent therapy to assisted reproduction, perhaps prematurely as the data currently remains inconclusive. A limitation of this study, along with the Paulus et al. (2003) trial, is the inclusion of only good quality embryos in the analysis, thus making them less generalizable to a larger portion of women undergoing in vitro fertilization with or without intracytoplasmic sperm injection. In the studies that established a positive association, Paulus et al. (2002), Dieterle et al., and Westergaard et al., all revealed clinical pregnancy rates in the standard medical care or sham acupuncture groups that were lower than one might expect based on national averages (26%, 16%, and 24%, respectively); however, this is difficult to extrapolate secondary to
the age range of patients in each treatment arm (ages 21 to 45) and the wide variations in clinical pregnancy rates expected based on distinct age groups.\textsuperscript{23-25,57}

In the Craig et al. trial, the only investigation to reveal a negative association of acupuncture and clinical pregnancy rates, clinical pregnancy rates of the standard of care group were 65\%, which are higher than one might expect based on national averages.\textsuperscript{26,27,57} Also, two unique characteristics of this study, off-site acupuncture and donor egg employment, make the comparison of this randomized controlled trial with others more challenging.\textsuperscript{26,27} One may speculate that the burden of driving to an unfamiliar location on the day on embryo transfer may have caused deleterious stress and hence outweighed any potential benefit of the actual acupuncture therapy.\textsuperscript{15-18,27} Additionally, the utilization of donor eggs renders this study less generalizable to all women undergoing in vitro fertilization with or without intracytoplasmic sperm injection and less analogous to the other randomized controlled trials investigating acupuncture and clinical pregnancy rates.\textsuperscript{27,40,41}

**Conclusion:**

The benefit of a standardized acupuncture protocol in women undergoing in vitro fertilization with or without intracytoplasmic sperm injection on the day of embryo transfer remains unclear. Existing research conclusions range from acupuncture having a positive to an equivocal to a negative effect on clinical pregnancy rates. Those studies that demonstrated positive effects have provided justification for concurrent true acupuncture therapy at fertility clinics and
acupuncture centers across the country, yet the efficacy of this treatment remains unknown. In addition, previous studies attained clinical pregnancy rates that vary considerably from both other trials and the expected national averages. Reasons for these variations remain unclear. Regarding methodology, designs of sham acupuncture and true acupuncture protocols differ among trials, thus rendering comparisons among studies problematic. Study designs also lack randomization to all possible comparison groups—sham acupuncture and standard medical care—in a single, well-designed randomized controlled trial.

A comprehensive review of the literature demonstrates the need for additional research regarding acupuncture and clinical pregnancy rates. The results of a well-designed, randomized controlled trial will help to more accurately advise patients and providers about appropriate concurrent therapy options while patients undergo the stressful and expensive treatment of in vitro fertilization. We therefore propose to conduct a multicenter, three arm, single-blinded, randomized controlled trial comparing clinical pregnancy rates of women under age 35 undergoing in vitro fertilization with or without intracytoplasmic sperm injection in combination with either true acupuncture, sham acupuncture, or standard medical care.

References:


33. Spielberger CD. *State - Trait Anxiety Inventory*. Wiley Online Library; 2010.


47. Technology SFAR. SART: Clinic Summary Report: Boston IVF (Waltham, Massachusetts). 2013;


CHAPTER 3: STUDY METHODS

Study Design:

The study design is a multicenter, three-arm, single-blinded, randomized controlled trial.

Study Population and Sampling:

Patients will be sampled on a non-random, consecutive basis from Boston IVF in Waltham, Massachusetts; Weill Medical College of Cornell University Center for Reproductive Medicine and Infertility in New York, New York; and Yale Fertility Center in New Haven, Connecticut. The inclusion criteria for the study will consist of women less than 35 years of age who are undergoing in vitro fertilization with or without intracytoplasmic sperm injection. Exclusion criteria will include age greater than or equal to 35, use of donor eggs, use of frozen and thawed embryos, previous participation in this study, and a lack of written, informed consent.

Subject Protection and Confidentiality:

All study personnel will complete Health Insurance Portability and Accountability Act (HIPAA) privacy training for research personnel and Yale Human Subjects Protection Training or the equivalent at their corresponding institution. Personnel must submit proof of attainment of these trainings to the Yale Human Investigation Committee and their corresponding site's institution prior to subject recruitment. An application to involve human subjects in research will then be submitted to the Yale Human Investigation Committee and the Yale Institutional
Review Board (IRB) for approval. Once approved at Yale, the approval letter will then be sent to Boston IVF and Weill Medical College of Cornell University Center for Reproductive Medicine and Infertility. These sites will next seek approval from their corresponding academic institutions; if protocol is approved, their approval letters will be submitted to the Yale Human Investigation Committee and recorded. The study will begin only after all approvals are obtained. Involved research personnel will access all subject electronic medical records on university-approved, encrypted, and secure electronic devices. Any protected health information not in electronic form will be stored in a locked cabinet within the locked office of the investigators on site grounds, to which only direct research staff will have access.

Prior to participation in the study, all participants will be required to grant written, informed consent (**Appendix I**). The consent form will detail the following: research methodology and procedures, potential risks of study participation, potential benefits of study participation, means by which their records will be kept confidential, and the ability of any subject to withdraw from the trial for any reason at any time. A trained member of the research team will explain the form before it is signed and dated by the subject upon agreement to participate in the study. All subjects will have an opportunity to ask any questions and discuss any concerns prior to providing written consent. If for any reason a participant is unable to provide written, informed consent, she will be deemed ineligible for study participation.
**Recruitment:**

Recruitment will be directed towards all women less than 35 years of age who are undergoing in vitro fertilization with or without intracytoplasmic sperm injection at the above-mentioned centers. Trained research personnel will identify potential study participants on a consecutive basis at presentation for oocyte retrieval. Written, informed consent will be obtained prior to initial screening for eligibility criteria. Subjects who meet all inclusion criteria will then be randomized into the true acupuncture, the sham acupuncture, or the standard medical care study arm before their scheduled day of embryo transfer. Enrollment into the study, randomization, and initiation of the treatment interventions will occur on an ongoing basis to accommodate the constant influx of patients at each center.

**Study Variables and Measures:**

**True Acupuncture Intervention:**

On the day of embryo transfer, true acupuncture will occur for 25 minutes before and after embryo transfer with sterile disposable stainless steel needles at acupoints designated by traditional Chinese medicine principles to govern reproduction and will adhere to the second edition of the Standard Acupuncture Nomenclature. The acupoints are as follows: Cx6, GV20, Liv3, Li4, LU7, SP6, SP8, SP10, ST29, and ST36. The depth of needle insertion into the skin will depend on the location of the acupoints, ranging from 10 to 20 millimeters. DeQi will be elicited during the initial insertion; after 10 minutes, DeQi will be maintained by rotating, lifting, and thrusting the needle handles. Needles will remain in place for a
total of 25 minutes before being removed. All true acupuncture treatments will utilize the same methodology and be performed in a designated clinic room on each site by a certified, Chinese acupuncturist with a degree in Chinese Medicine, at least three years prior clinical experience in fertility acupuncture, training in Human Subjects Protection, and training in the proposed study’s procedures. To allow adequate time for needle insertion and removal, women will begin their pre-embryo transfer true acupuncture sessions 40 minutes prior to their scheduled embryo transfers.

**Sham Acupuncture Intervention:**

On the day of embryo transfer, sham acupuncture will occur for 25 minutes before and after embryo transfer with sterile Streitberger placebo needles at acupoints designated by traditional Chinese medicine principles to govern reproduction and will adhere to the second edition of the Standard Acupuncture Nomenclature. The acupoints are as follows: Cx6, GV20, Liv3, Li4, LU7, SP6, SP8, SP10, ST29, and ST36. To blind subjects receiving the sham acupuncture, this placebo will give patients a pricking penetration sensation. Needles will remain in place for a total of 25 minutes before being removed. All sham acupuncture treatments will utilize the same methodology and be performed in a designated clinic room on each site by a certified, Chinese acupuncturist with a degree in Chinese Medicine, at least three years prior clinical experience in fertility acupuncture, training in Human Subjects Protection, and training in the proposed study’s procedures. To allow adequate time for needle insertion and removal,
women will begin their pre-embryo transfer sham acupuncture sessions 40 minutes prior to their scheduled embryo transfers.

**Standard Medical Care:**

On the day of embryo transfer, no true acupuncture or sham acupuncture treatment will occur. In accordance with standard medical care, the patient will be placed on bed rest for 25 minutes following her embryo transfer. This corresponds to the time of the post embryo transfer true acupuncture and sham acupuncture sessions minus needle insertion and removal.

**Primary and Secondary Outcome Measures:**

The primary outcome measure is clinical pregnancy, which will be determined via transvaginal ultrasound six to eight weeks post embryo transfer. The secondary outcome measures are anxiety symptoms and depressive symptoms. The STAI (Appendix V) will be used to measure both state and trait anxiety before and after embryo transfer and the assigned treatment intervention or standard medical care on the day of embryo transfer. The Zung Self-Rating Depression Scale (Appendix VI) will be used to measure depressive symptoms before and after embryo transfer and assigned treatment intervention or standard medical care on the day of embryo transfer.

**Baseline Variables:**

This study will account for age, number of embryos transferred, presence or absence of intracytoplasmic sperm injection, duration of infertility, number of previous treatment cycles, number of previous pregnancies, BMI, and infertility diagnosis.
Methodology Considerations:

Assignment of Intervention:

Randomization will be accomplished via a computer generated randomization list to a 1:1:1 allocation into the three treatment arms. The allocation will then be concealed in a sequentially-numbered, opaque, sealed envelope by a member of the research staff who is removed from patient care. The envelopes will be ordered sequentially and then opened consecutively to assign subjects a study arm at enrollment.

Blinding of Intervention:

The providers completing the embryo transfer, ultrasound assessment, and any general care of the patient will not be aware of the patient’s treatment arm allocation. The patients undergoing true acupuncture and sham acupuncture will also be blinded to their treatment provision; however, those patients receiving standard medical care will be cognizant to their treatment arm. All subjects will be instructed not to discuss their study group with any clinical staff directly involved in their care.

Blinding of Outcome:

To ensure that the assessments of primary and secondary outcomes measures remain blinded, the research team member who prepared the randomization scheme and study envelopes will have no participation in data collection or analysis. Patient records will have identifying data removed and will be coded by a patient identification number. Investigators who have no direct patient contact and who have no knowledge of patient assignment will then indicate
if the subject attained a clinical pregnancy six to eight weeks following embryo transfer as assessed via a transvaginal ultrasound.

**Adherence:**

Adherence will be monitored by records of patients’ appointment attendance at the clinic and confirmation by acupuncturists’ documentation that true or sham acupuncture sessions were administered at the assigned time points.

**Monitoring of Adverse Events and Effects:**

An un-blinded, independent committee will review trial data for adverse events and effects at twelve-week intervals. If any statistically significant adverse effects are revealed among study groups, the trial will be discontinued immediately and subjects who have already participated will be informed of their treatment status.

**Data Collection:**

Pending IRB approval, recruitment and data collection will occur over a 20-month period to obtain an adequate number of eligible participants and to allow sufficient time for data collection completion by 24 months. Utilizing her assigned study identification number, research personnel will follow each subject until completion of a transvaginal ultrasound six to eight weeks following embryo transfer to determine the presence or absence of a clinical pregnancy. The secondary outcome measures will be gauged at the beginning of the embryo transfer appointment, thus before true acupuncture, sham acupuncture, or standard medical care administration. The secondary outcome measures will also be gauged
again immediately after the embryo transfer and true acupuncture, sham acupuncture, or standard medical care administration. After collection, data will be entered utilizing Qualtrics software and then stored on a secure Yale server until being exported for statistical analysis.

**Sample Size Calculation:**

We will be using two-sided hypothesis testing with an alpha of 5%, a beta of 20%, and a power of 80%. We aim to detect a 15% difference between true acupuncture and standard medical care. Via Power and Precision Version 4 software, this leads to an unadjusted sample size of 173 subjects per arm, consequently equaling 519 subjects in total. Based on previous research, the highly motivated nature of these patients, and only a six to eight week follow-up period per subject, we expect the loss to follow up rate to be very minor at less than 5%. To ensure adequate sample size, we will anticipate a conservative 10% dropout rate, projecting a sample size of 191 subjects per arm and 573 subjects in total. As previously stated, recruitment and data collection will occur over a 20-month period to obtain an adequate number of eligible participants. This will allow sufficient time for data collection completion by 24 months. Further details pertaining to the sample size calculation are provided in Appendix VII.

**Analysis:**

Data collected during this study will be analyzed using the Statistical Package for the Social Sciences Version 22. All data will be analyzed using an intention-to-
treat protocol. Baseline clinical and demographic data will be collected from all participants upon enrollment and compared among treatment arms utilizing standard parametric methods. Continuous variables among the three groups will be compared with Analysis of Variance (ANOVA) testing. If a statistically significant difference is found among the groups, pairwise Student $t$-tests will be performed between each of the pairs to elucidate which comparisons are significantly different. Categorical variables among the three groups will be compared using the Chi-square test; if a statistically significant difference is found among the groups, pairwise Chi-square tests will be performed between each of the pairs to elucidate which comparisons are significantly different. Additionally, if there are significant differences found in baseline characteristics among the groups after randomization, a regression analysis will be completed in addition to the above testing to control for confounding differences among groups measured at baseline.

The primary endpoint of the study, the proportion of clinical pregnancies in each treatment arm, will be evaluated using Chi-square testing. If a statistically significant difference is found among the three groups, pairwise Chi-square tests will be performed between each of the pairs to elucidate which comparisons are significantly different. The secondary outcomes among the three groups will be compared with ANOVA testing. If a statistically significant difference is found among the groups, pairwise Student $t$-tests will be performed between each of the pairs to elucidate which comparisons are significantly different. If the secondary outcomes are not normally distributed, the three groups will be compared with Kruskal-Wallis testing. If a statistically significant difference is found among the
groups, pairwise Mann-Whitney U testing will be performed between each of the pairs to elucidate which comparisons are significantly different.

**Timeline and Resources:**

Subjects will be recruited and enrolled on an ongoing basis for 20 months beginning in January 2016. The 20-month recruitment period will end in August 2017 to allow adequate time for the final subjects to be followed for six to eight weeks following their embryo transfers. Data collection will end in January 2018, and data analysis will occur in the final two months of the allotted 24-month time period and continue following completion of the trial.

The Principal Investigator of this study will be J. Ryan Martin, MD and the Co-Principal Investigator will be Hannah K. Holland, PA-SII. Each study site will have a designated primary investigator to oversee the trial at their corresponding site and to communicate with the Principal Investigators. Regarding personnel, one to two research assistants per site will be required to screen potential participants upon presentation for oocyte retrieval, obtain informed consent, gather baseline data, explain the secondary measures assessments, and perform other required tasks during the trial. A separate, non-blinded research assistant will be required to perform the randomization, prepare envelopes, and code all blinded data. One certified, Chinese acupuncturist with a degree in Chinese Medicine, at least three years prior clinical experience in fertility acupuncture, training in Human Subjects Protection, and training in the proposed study’s procedures will be employed per site to perform all true and sham acupuncture interventions. An oversight
committee consisting of a physician and a statistician will monitor safety of all sites at twelve-week intervals. Finally, a statistician will be consulted to assist with data analysis. Supplies for both types of acupuncture will include: approximately 3,900 sterile disposable stainless steel needles and approximately 3,900 sterile Streitberger placebo needles. The employment of research personnel and the acupuncturists, along with the supply materials, will need to be compensated by study funding.
CHAPTER 4: CONCLUSION

Advantages and Disadvantages:

Advantages of the proposed trial are its study design, study population, and large sample size. The single-blinded, randomized controlled trial design will minimize selection and information bias. Randomizing patients to all possible control groups—sham acupuncture and standard medical care—in a single, well-designed randomized controlled trial will provide a needed comparison of true acupuncture versus sham acupuncture versus standard medical care in the same study population. The multicenter nature of the investigation is also an advantage as it renders the study more generalizable and will add increased external validity to the results if a statistically significant effect is ascertained. The limited exclusion criteria of the study population will also increase study generalizability. While the main limitation of generalizability regarding the study population is age, as all women will be under 35 years of age, this will ensure comparable expected pregnancy rates, probable embryo quality, and consistent endometrial receptivity based on age-related factors among treatment arms.\textsuperscript{1-3} The large sample size will also increase generalizability of the study results and help to ensure adequate randomization.

The disadvantages of this trial must be noted. First, we recognize that the proposed study will be underpowered to detect a statistically significant effect size less than 15%; though, it is important to recognize that such an effect size may still hold great clinical significance to both providers and patients. Regarding the study design, subjects in the standard medical care arm will not lie down for a designated
amount of time before embryo transfer in accordance with current medical practice. Post embryo transfer, subjects in the standard medical care arm will not rest as long as the true acupuncture or sham acupuncture intervention groups once the time for needle insertion and removal is considered. We believe this is unlikely to affect results; however, one could argue this increased rest pre and post embryo transfer is a potential mechanism to influence clinical pregnancy rates. Also regarding the study design, the multicenter nature of the trial, despite being an advantage to generalizability and validity, does propose additional challenges for study methodologies. Extra care must be taken to ensure all interventions are performed consistently across all three sites. To ensure the acupuncture treatments are as similar as possible across all study sites, we will be using certified, Chinese acupuncturists with degrees in Chinese Medicine, at least three years prior clinical experience in fertility acupuncture, training in Human Subjects Protection, and training in the proposed study's procedures; however, individuals with this level of training may not be available in all areas of the country, thus limiting generalizability.

Regarding the study population, the exclusion criteria of age greater or equal to 35 years does render this study only applicable to those women under age 35, further limiting generalizability. In addition, there is a disadvantage concerning our primary outcome. Live birth rate is the ultimate indicator of assisted reproductive technology success; however, due to time restrictions, this study seeks to examine clinical pregnancy rates, thus limiting the examination of definitive intervention efficacy. Lastly, this study is not designed to detect the biological mechanism(s) by
which true and/or sham acupuncture exert their effects on clinical pregnancy rates, a topic that is still a point of speculation within the scientific community.

**Clinical Significance:**

If the results of this proposed study support the previously stated hypothesis, there is potential for great clinical significance. Infertility is a common, and often a devastating, diagnosis that many choose to confront. One in seven to ten couples in industrialized nations suffer from infertility as they attempt to build their family and turn to assisted reproductive technology for assistance. In addition to the large financial burden associated with utilization of this technology, there also are numerous risks in the physical, mental, and emotional realms. Optimizing the chance of a clinical pregnancy is therefore a shared goal between the patient and the practitioner. Through the results of the proposed study, individuals and couples who face barriers to constructing their biological families may be presented with a contemporaneous intervention to improve their chances of a clinical pregnancy and potentially a live birth. These results will help to end the acupuncture and assisted reproductive technology debate, and most importantly, they will assist providers in more accurately advising their patients about appropriate concurrent therapy options while they undergo the stressful and expensive treatment that is in vitro fertilization.

**References:**


APPENDIX I: CONSENT FORM

HIC#: ---

CONSENT FOR PARTICIPATION IN THE ACUPUNCTURE AND ASSISTED REPRODUCTIVE TECHNOLOGY TRIAL

YALE UNIVERSITY SCHOOL OF MEDICINE – YALE FERTILITY CENTER

Study Title: Acupuncture and Assisted Reproductive Technology: The Effect on Clinical Pregnancy Rates

Principal Investigators: J. Ryan Martin, MD; Hannah K. Holland, PA-SII

Invitation to Participate and Description of Project

You are invited to participate in a research study designed to look at acupuncture and clinical pregnancy rates in women undergoing in vitro fertilization with or without intracytoplasmic sperm injection. You have been asked to participate because you are undergoing this procedure and are under 35 years of age. There will be approximately 573 participants in this study from Boston IVF in Waltham, Massachusetts; Weill Medical College of Cornell University Center for Reproductive Medicine and Infertility in New York, New York; and Yale Fertility Center in New Haven, Connecticut.

In order to decide whether or not you wish to be a part of this research study, you should know enough about its risks and benefits to make an informed decision. This consent form gives you detailed information about the research study, which a member of the research team will discuss with you. This discussion should go over all aspects of this research: its purpose, the procedures that will be performed, any risks of the procedures, possible benefits, and possible alternative treatments. Once you understand the study, you will be asked if you wish to participate; if so, you will be asked to sign this form.

Description of Procedures

If you agree to participate in this study, we will first confirm information about your demographics and obstetric/gynecologic history that you have already provided while being counseled for in vitro fertilization with or without intracytoplasmic sperm injection. Before the day of your embryo transfer appointment, you will be randomly assigned into one of three treatment arms: true acupuncture, sham (also known as placebo) acupuncture, or standard medical care. A specific person on the research staff will determine which group you are in by selecting an envelope that designates “true acupuncture”, “sham acupuncture”, or “standard medical care” inside it. No one directly involved with patient care (besides the certified acupuncturist if you are randomized to true acupuncture or sham acupuncture) will know your treatment status.

On the day of your embryo transfer appointment, every research subject will complete two questionnaires aimed to quantify differences in anxiety and depression. These questionnaires will ask you how often you experience certain emotions, thoughts, or actions. This is an example of a question you will be asked: “I feel hopeful about the future: a little of the time/some of the time/good part of the time/most of the time”.

After completing these two questionnaires, you will then undergo true acupuncture, sham
acupuncture, lor standard medical care depending on your treatment assignment. The true acupuncture patients will have true acupuncture performed on them by a certified Chinese acupuncture patient with a degree in Chinese Medicine, at least three years prior clinical experience in fertility acupuncture, training in/subject to protection, and training in the proposed study’s procedures for 25 minutes before and after embryo transfer. True acupuncture involves being needleless with sterile disposable stainless steel needles in a controlled environment. Needles are rotated, lifted, and thrust through the session. Needles will remain in place for a total of 25 minutes before being removed. The sham acupuncture patients will receive sham acupuncture performed on them by a licensed, certified Chinese acupuncture patient with a degree in Chinese Medicine, at least three years prior clinical experience in fertility acupuncture, training in/subject to protection, and training in the proposed study’s procedures for 25 minutes before and after embryo transfer. Sham acupuncture involves being needleless with sterile placebo needles, known as treiber or placebo needles. Needles are rotated, lifted, and thrust through the session. Needles will remain in place for a total of 25 minutes before being removed. The design of this study is such that the subject will be unaware if they are receiving true acupuncture or sham acupuncture.

The standard medical care patients will receive true acupuncture or sham acupuncture intervention before or after their embryo transfer. After embryo transfer, true acupuncture patients will have acupuncture designated as part of their treatment. Subjects will then complete the same two questionnaires aimed to quantify differences in anxiety and depression. Study subjects will then follow the normal protocol begun six to eight weeks after embryo transfer for a total of four to six determinations if they have achieved clinical pregnancy. These results will be recorded, as typical of all patients undergoing in vitro fertilization with or without intrauterine insemination, in the medical record. These results will also be recorded on study data collection forms for analysis. An identifiable patient information will be included in the study analysis for study results.

You will be told of any significant findings that are observed during the course of your participation in this study that may affect your willingness to continue to participate. A description of this clinical trial will be available on the website http://www.ClinicalTrials.gov, as required by U.S. Law. This website will not include information that can identify you. At most, the website will include a summary of the results. You can search this website at any time.

Risks and Inconveniences!

True acupuncture and sham acupuncture are considered relatively low risk therapies. The side effect profiles of both interventions are similar. A sense of heaviness, soreness, or numbness at the points of needling may be experienced, which is uncomfortable to some patients. The most common side effects of both true acupuncture and sham acupuncture include: itching, minor bleeding, needling pain, and throbbing. Other possible side effects include: dizziness, light headedness, fainting “or” passing out, headache, drowsiness, tiredness, nausea, and vomiting.

Page 2/6
Rarely, chest pain is a noted side effect. On the previous randomized controlled trial, discovered a statistically significant negative effect on clinical pregnancy rates with concurrent true acupuncture therapy. Upon your request, we will provide you a copy of this study, and we will be available to answer any questions or discuss any concerns that you may have. Additionally, though acupuncture therapy has been studied in research, participation in this study may still involve risks that are unknown at this time.

To mitigate risk, I, a certified Chinese acupuncture therapist with a degree in Chinese Medicine, at least three years' prior clinical experience in fertility acupuncture, training in Human Subjects Protection, and training in the proposed study's procedures, will perform true acupuncture and sham acupuncture therapy at the clinic location. Medical staff will be available on site at all times to answer any questions and address any side effects if you may be experiencing. If true acupuncture or sham acupuncture therapy is immediately stopped at any time upon your request. Also, lanun blinded, independent committee will review trial data for adverse effects and declare it twelve weeks. If any statistically significant adverse effects are revealed among study groups, the trial will be discontinued immediately, and subjects who have already participated will be informed of their treatment status.

Inconveniences in the proposed study include only an increased appointment length of ten minutes to the day of embryo transfer to complete the two questionnaires and undergo true acupuncture or sham acupuncture therapy if so assigned.

Benefits!

Benefits to participants in this study include those associated with achieving pregnancy and potentially live birth thereafter. Three previous randomized controlled trials have discovered a statistically significant positive effect on clinical pregnancy rates with concurrent true acupuncture therapy. Upon your request, we will provide you a copy of these studies, and we will be available to answer any questions or discuss any concerns that you may have. Acupuncture therapy has been associated with decreased stress and anxiety.

Additionally, there may be benefits to the assisted reproductive technology community as a whole. This study aims to better understand the effects of acupuncture on clinical pregnancy rates in women who are undergoing in vitro fertilization with or without intracytoplasmic sperm injection. We hope that the results of this study will aid in the general advancement of scientific knowledge related to this subject.

Economic Considerations!

There will be no direct compensation offered to participants in this study. All true acupuncture or sham acupuncture therapy will be provided at your free of cost. You will still be responsible for any copay required by your insurance company for standard treatment. If you have questions regarding your insurance coverage or policy, please call your insurance company directly. There will be no financial penalty for withdrawing from the study at any time.
Treatment Alternatives!

There are alternatives that should be considered. One alternative is to refuse participation in this study. You may then undergo true acupuncture therapy at your own discretion at a location not associated with this trial. You may also choose to not undergo any form of acupuncture therapy and proceed only with standard medical care. Please discuss any alternatives with your health care provider before you enroll or refuse participation in this study.

Confidentiality!

Any identifiable information that is obtained in connection with this study will remain confidential and will be disclosed only with your permission or as required by U.S. or State law. Examples of information that we are legally required to disclose include abuse of a child or elderly person, or certain reportable diseases.

Your electronic medical record will be contained on a university-owned or university-encrypted, secure, and approved devices. Health insurance portability and accountability act (HIPAA) standards will be met and maintained for all devices and will be accessed by personnel only if approved research personnel. Any information that is not relevant to this study will not be obtained from your medical record. Any protected health information not on electronic form will be stored in a locked cabinet within the locked office of the investigator(s) on site grounds. All records will be destroyed in accordance with HIPAA requirements. No record obtained during this study will be kept for 10 years prior to being destroyed. When the results of the research are published or discussed in conferences, no information will be included that could reveal your identity unless your specific consent for this activity is obtained.

Representatives from the Yale Human Research Protection Program, the Yale Human Investigation Review Committee (the committee that reviews, approves, and monitors research on human subjects) may inspect study records during internal auditing procedures. However, these individuals are required to keep all information confidential.

In Case of Injury!

If you are injured while on study, seek treatment and contact the study doctor as soon as you are able. Yale School of Medicine and Yale Fertility Center do not provide funds for the treatment of research-related injury. If you are injured as a result of your participation in this study, treatment will be provided. If you are injured, your insurance carrier will be expected to pay the costs of this treatment. No additional financial compensation for injury or lost wages is available.

You should give up any of your legal rights by signing this form.

Voluntary Participation and Withdrawal!

Participating in this study is voluntary. You are free to choose not to take part in this study. Refusing to participate will involve no penalty or loss of benefits to which you are otherwise entitled.
If you do become a subject, you are free to stop and withdraw from this study at any time during its course. If you withdraw from the study, you can call a member of the research team at any time and tell them that you longer want to take part. This will not affect your planned standard medical care. The researchers may withdraw you from participating in the research if necessary. This may occur if statistically significant adverse effects are discovered for the study as a whole, or if you ask an individual development of serious side effects from the possible interventions.

!! Withdrawing from the study will involve no penalty or loss of benefit(s) to which you are otherwise entitled.!! It will not harm your relationship with your own doctors or with Yale Fertility Center.!! We would still treat you with standard therapy or at your request, refer you to another clinic or doctor who can offer this treatment.!! When you withdraw from the study, in no new health information identifying you will be gathered later that date.!! Information that has already been gathered may still be used and given to others until the end of the research study, as necessary to ensure the integrity of the study land/or study oversight.!!!

Questions!
!!
We have used some technical terms in this form.!! Please feel free to ask about anything you do not understand and to consider this research and the consent form carefully as long as you feel is necessary.!! Before you make a decision.
!!
Authorization

I have read (or someone has read to me) this form, and I have decided to participate in the project described above. Its general purposes, the particulars of my involvement, and possible hazards and inconveniences have been explained to my satisfaction. My signature also indicates that I have received a copy of this consent form.

Name of Subject: _______________________________________________________________

Signature: _________________________________________________________________

Relationship: _____________________________________________________________

Date: _____________________________________________________________________

Signature of Principal Investigator __________________________ Date __________
or

Signature of Person Obtaining Consent __________________________ Date __________

If you have further questions about this project or if you have a research-related problem, you may contact the Principal Investigators: J. Ryan Martin, MD (203-785-4708) or Hannah K. Holland, PA-SII (814-460-8086).

If, after you have signed this form you have any questions about your privacy rights, please contact the Yale Privacy Officer at 203-432-5919. If you would like to talk with someone other than the researchers to discuss problems, concerns, and questions you may have concerning this research, or to discuss your rights as a research subject, you may contact the Yale Human Investigation Committee at 203-785-4688.

THIS FORM IS NOT VALID UNLESS THE FOLLOWING BOX HAS BEEN COMPLETED BY THE HIC OFFICE

THIS FORM IS VALID ONLY THROUGH: ________________________________

INITIALED: __________________________________

Page 6 of 6
APPENDIX II: DATA COLLECTION FORM

Study ID Number: ________________

Data Collection Form:

Subject Name: ________________________________

Date of Birth: ________________________________

Age: _________________________________________

Height: ________ Weight: ________ BMI: ________

____________________________________________________________________________________

Duration of Infertility: _________________________

Number of Previous Treatment Cycles: _______________

Number of Previous Pregnancies: ____________________

If Previously Pregnant, Conceived via ART: Yes No

Infertility Diagnosis: _______________________________

____________________________________________________________________________________

Date of Oocyte Retrieval: _________________________

Intracytoplasmic Sperm Injection: Yes No

____________________________________________________________________________________

Date of Embryo Transfer: _________________________

Number of Embryos Transferred: ___________________

STAI Score (pre-intervention): _____________________

Zung Self-Rating Depression Scale Score (pre-intervention): ____________

Intervention: True Acupuncture Sham Acupuncture Standard Medical Care

STAI Score (post-intervention): _____________________

Zung Self-Rating Depression Scale Score (post-intervention): ____________

____________________________________________________________________________________

1
Study ID Number: 

Date of Ultrasound: 

Clinical Pregnancy:  Yes  No

If Yes, Number of Gestational Sacs: 

### APPENDIX III: ACUPUNCTURE POINTS EMPLOYED IN EMPIRICAL STUDIES

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</table>

# APPENDIX IV: TREATMENT INTENDED FOR ACUPUNCTURE POINTS

<table>
<thead>
<tr>
<th>Acupuncture points</th>
<th>Intended treatment</th>
</tr>
</thead>
</table>
| **Cu6 (Neiguan)**  | Location: 2 cun above the transverse crease of the wrist, between the tendons of muscul and musculotendinous radialis.  
Indications: Cardiac pain, palpitation, stuffy chest, pain in the hypochondriac region, stomach ache, nausea, vomiting, hiccups, mental disorders, epilepsy, incontinence, irritability, malaria, contracture and pain in elbow and arm.  
Traditional action: Opens the chest, regulates Qi and blood, regulates and cleans the Triple Burner, calms the mind, regulates the terminal Yin, harmonizes the stomach. |
| **GV 20 (Baihui)** | Location: on the midline of the head, 7 cun directly above the posterior hairline, approximately on the midpoint of the line connecting the apexes of the two auricles.  
Indications: Headache, vertigo, tinnitus, nasal obstruction, aphasia by apoplexy, coma, mental disorders, prostate of the brain and the uterus.  
Traditional action: Clears the mind, lifts the spirits, tonifies yang, strengthens the ascending function of the spleen, eliminates interior wind, promotes respiration. |
| **Liv 2 (Xingjian)** | Location: on the dorsum of the foot between the 1st and 2nd toes, proximal to the margin of the web at the junction of the red and white skin.  
Point associations: Ying Spring point, Fire point.  
Actions and effects: Generally clears, LV Fire - extreme irritability, red face, eyes, tongue.  
Clears heat from the lower Jiao - burning urination.  
Useful for “true heat, false cold” - lack of Qi flow to the extremities (cold hands or feet). |
| **Liv 3 (Taiyin)**  | Location: on the dorsum of the foot in a depression distal to the junctions of the 1st and 2nd metatarsal bones.  
Point associations: Shu Stream point, Earth point, Yuan source point.  
Actions and effects: Generally, resolves stagnation and tonifies Yin - balancing for all LV pathologies.  
LV Qi Stagnation: LV Yang Rising - headaches, dizziness, cancer bone.  
Eye issues: blurred vision, red, swollen, painful eyes. |
### Table 2. Summary of the treatment intended for the respective acupuncture points (Continued)

<table>
<thead>
<tr>
<th>Point</th>
<th>Location</th>
<th>Precautions</th>
<th>Point associations</th>
<th>Acupuncture actions and effects</th>
</tr>
</thead>
</table>
| Menstrual issues from deficient blood, Yin, Qi, LV Qi stagnation - dysmenorrhea, amenorrhoea, PMS, breast tenderness. | Location: On the dorsum of the foot in a depression distal to the junctions of the 1st and 2nd metatarsal bones. | Nausea, vomiting, constipation, diarrhea with undigested food. | Point associations:  
Shu Stream point  
Earth point  
Yuan source point  | Calming point - anger, irritability, insomnia, anxiety. With LI 4, four gates treatment - powerfully effects the flow of Qi and blood in the body. |  
Actions and effects:  
Generally, resolves stagnation and tonifies Yin - balancing for all LV pathologies.  
LV Qi. Stagnation / LV Yang Rising - headaches, dizziness, canker sores.  
Ocular issues - blurred vision, red, swollen, painful eyes.  
Menstrual issues from deficient blood, Yin, Qi, or LV Qi stagnation - dysmenorrhea, amenorrhoea, PMS, breast tenderness.  
Genital issues - pain and swelling, hernia, impotence, seminal emission.  
Stagnation in the middle warmer - subcostal tension, chest or flank pain, swellings in the axillary region.  
Digestive issues from LV attacking ST/SP - nausea, vomiting, constipation, diarrhea with undigested food.  
Calming point - anger, irritability, insomnia, anxiety. With LI 4, four gates treatment - powerfully effects the flow of Qi and blood in the body. |  
Actions and effects:  
Releases the exterior for wind-cold or wind-heat syndromes.  
Strengthens the qi, improves immunity.  
Regulates the sweat glands, for excessive sweating tonify LI 4 then disperse KD 7 and vice versa.  
Any problem on the face - sense organs, mouth, teeth, jaw, toothache, allergies, rhinitis, hay fever, acne, eye problems, etc.  
Toothache use both LI 4 & ST 44 - LI for the lower jaw & ST for the upper jaw.  
Headache, especially frontal, sinus (yangming area).  
Chronic pain.  
Influence the circulation of Qi and blood - use the four gates, LI 4 & LV 3 to strongly move the Qi and blood in the body clearing stagnation and alleviating pain.  
Promote labor or for retained placenta. |
| LI 4 (Hegu)  | Location: in the middle of the 2nd metacarpal bone on the radial side. | No nausea, no needle in pregnancy. | Yuen source point  
Entry point  
Command point for face, nose, mouth and jaw | Releases the exterior for wind-cold or wind-heat syndromes.  
Strengthens the qi, improves immunity.  
Regulates the sweat glands, for excessive sweating tonify LI 4 then disperse KD 7 and vice versa.  
Any problem on the face - sense organs, mouth, teeth, jaw, toothache, allergies, rhinitis, hay fever, acne, eye problems, etc.  
Toothache use both LI 4 & ST 44 - LI for the lower jaw & ST for the upper jaw.  
Headache, especially frontal, sinus (yangming area).  
Chronic pain.  
Influence the circulation of Qi and blood - use the four gates, LI 4 & LV 3 to strongly move the Qi and blood in the body clearing stagnation and alleviating pain.  
Promote labor or for retained placenta. |
<table>
<thead>
<tr>
<th>Point</th>
<th>Location</th>
<th>Actions and effects</th>
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<tbody>
<tr>
<td>LI 10 (Shoushanli)</td>
<td>Location: 2 cm below LI 11 on the LI 5 to LI 11 line.</td>
<td>The following relationships exist between the ST and the LI and can be used to treat ST, LI and SI organ problems. ST: LI 8 &amp; SI 39. LI: LI 9 &amp; ST 37. ST: LI 10 &amp; ST 36. Shoulder, elbow and wrist pain issues, general aches in these areas. Less dispersive and more tonifying than other LI points. Epigastic and abdominal pain, ulcers, vomiting. Location: 2 cm below LI 11 on the LI 5 to LI 11 line.</td>
</tr>
<tr>
<td>SP 6 (Sanyinjiao)</td>
<td>Location: 3 cm directly above the tip of the medial malleolus on the posterior border of the tibia.</td>
<td>Precautions: no needle in pregnancy. Point associations: Intersection point of the SP, LV and KD (3 leg Yin meridians). Actions and effects: Tonify Yin and blood, all spleen disorders. Digestive disorders, sinking or prolapse. Gynecological issues, menopausal issues, difficult labor (expel fetus). Bleeding disorders, cool blood in hot skin diseases. Insomnia and other anxiety related emotions.</td>
</tr>
<tr>
<td>SP 8 (Diji)</td>
<td>Location: 3 cm below SP 9 on line connecting SP 9 and the tip of the medial malleolus.</td>
<td>Point associations: Xi Clavicle point. Actions and effects: Xi Clavicle point - acute and painful menstrual issues due to blood stagnation - clotting, fibroids, dysmenorrhea. Male infertility.</td>
</tr>
<tr>
<td>SP 10 (Xuehai)</td>
<td>Location: with knee flexed, 2 cm above the superior medial border of the patella on the bulge of the medial portion of quadriceps femoris (vastus medialis).</td>
<td>Actions and effects: Any gynecological issues originating from blood, heat, stasis and/or deficiency - irregular menstruation, cramping, PMS. Skin problems from damp-heat or hot blood.</td>
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<tr>
<td>ST 29 (Gulai)</td>
<td>Location: 2 cm lateral to the AML level with CV 3.</td>
<td>Actions and effects:</td>
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<tr>
<td><strong>Excess or Deficient disorders of the lower warmer - amenorrhoea, irregular menstruation, qi stagnation/masses</strong></td>
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<tr>
<td><strong>Running Right disorder.</strong></td>
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<tr>
<td><strong>ST 36 (Zusanli)</strong></td>
<td>Location: 3 cm below ST 35, one finger width lateral from the anterior border of the tibia.</td>
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<td></td>
<td>Point associations:</td>
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<td>He/Sea point</td>
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<td></td>
<td>Lower Lower He/Sea point of the ST</td>
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<td></td>
<td>Earth point</td>
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<tr>
<td></td>
<td>Sea of Water and Grain point</td>
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<tr>
<td></td>
<td>Command point of the abdomen</td>
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<td></td>
<td>Actions and effects:</td>
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<tr>
<td></td>
<td>Tonify deficient Qi or blood.</td>
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<td></td>
<td>Tonify Wei Qi.</td>
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<td>All issues involving the stomach or the spleen.</td>
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<td>Clear disorders along the course of the channel - breast problems, lower leg pain.</td>
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<td>Earth as the mother of Metal - will support lung function in cases of asthma, wheezing, dyspnoea.</td>
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<td></td>
<td>Psychological/Emotional disorders - PMS, depression, nervousness.</td>
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<tr>
<td><strong>PC 6 (Neiguan)</strong></td>
<td>Location: 2 cm above the wrist crease between the tendons of palmaris longus and flexor carpi radialis.</td>
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<td></td>
<td>Point associations:</td>
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<td></td>
<td>Luo Connecting point</td>
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<td></td>
<td>Yin Wei Maitre point coupled with SP 4</td>
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<td></td>
<td>Actions and effects:</td>
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<td></td>
<td>Similar to PC 3 but more for chronic heart symptoms from Qi stagnation.</td>
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<td></td>
<td>Opens and relaxes the chest, chest tightness, asthma, angina, palpitations.</td>
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<td>Insomnia, other spirit disorders of an excess or deficient nature, mania, nervousness, stress, poor memory.</td>
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<tr>
<td></td>
<td>Nausea, abdominal pain, motion sickness, vomiting, epigastric pain.</td>
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<tr>
<td></td>
<td>Carpal tunnel syndrome.</td>
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<tr>
<td><strong>Ren 3 (Zhongji)</strong></td>
<td>Location (zhongji): 1 cm superior to qugu. Regulates LR, warms KI, irregular menses.</td>
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<tr>
<td><strong>RN 4 (Guanyuan)</strong></td>
<td>Location: 1.5 cm lateral to the Du meridian, at the level of the lower border of the spinous process of the 5th lumbar vertebra.</td>
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<tr>
<td></td>
<td>Indications</td>
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<td>Low back pain, abdominal distension, diarrhoea, enuresis, olfactory, frequent urination.</td>
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<td>Traditional action</td>
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<td></td>
<td>Strengthens the lower back, removes obstructions from the channel.</td>
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<tr>
<td><strong>RN 6 (Qihai)</strong></td>
<td>Location: 1.5 cm lateral to the Du meridian, at the level of the lower border of the spinous process of the third lumbar vertebra.</td>
<td></td>
</tr>
<tr>
<td></td>
<td>Indications</td>
<td></td>
</tr>
<tr>
<td>Point</td>
<td>Description</td>
<td></td>
</tr>
<tr>
<td>-------</td>
<td>-------------</td>
<td></td>
</tr>
<tr>
<td>KI 3 (Taixi)</td>
<td>Location: in the depression between the medial malleolus and tendon calcaneus at the level with the tip of the medial malleolus. &lt;br&gt;Indications: Sore throat, toothache, deafness, tinnitus, dizziness, splitting of blood, asthma, thirst, irregular menstruation, insomnia, nocturnal emission, impotence, frequency of micturition, pain in the lower back. &lt;br&gt;Traditional action: Tonifies the kidneys, benefits essence, strengthens the lower back and knees, regulates the uterus.</td>
<td></td>
</tr>
<tr>
<td>KI 11 (Henggu)</td>
<td>Location: 1.5 cm posterior to Wuchi (UB 5), 1.5 cm lateral to the Du meridian. &lt;br&gt;Indications: Headache, blurring of vision, nasal obstruction. &lt;br&gt;Traditional action: Clears heat and eliminates vexation, brightens the eyes and opens the portals.</td>
<td></td>
</tr>
<tr>
<td>TE 5 (Wanguan)</td>
<td>Location: 1.5 cm lateral to the lower border of the spinous process of the eighth thoracic vertebra. &lt;br&gt;Indications: Diabetes, vomiting, abdominal pain, pain in the chest and hypochondriac region. &lt;br&gt;Traditional action: Relieves stagnation of blood.</td>
<td></td>
</tr>
</tbody>
</table>
## APPENDIX V: THE STATE TRAIT ANXIETY INVENTORY

**SELF-EVALUATION QUESTIONNAIRE (STAI) Form Y-1**

Please provide the following information:

Name ___________________________ Date ___________ S _____

Age ___________ Gender (Circle) M F T _____

**DIRECTIONS:**

A number of statements which people have used to describe themselves are given below. Read each statement and then circle the appropriate number to the right of the statement to indicate how you feel right now, that is, *at this moment*. There are no right or wrong answers. Do not spend too much time on any one statement but give the answer which seems to describe your present feelings best.

1. I feel calm ................................................................. 1 2 3 4
2. I feel secure ............................................................... 1 2 3 4
3. I am tense ................................................................ 1 2 3 4
4. I feel strained ............................................................. 1 2 3 4
5. I feel at ease ............................................................... 1 2 3 4
6. I feel upset .................................................................. 1 2 3 4
7. I am presently worrying over possible misfortunes ......... 1 2 3 4
8. I feel satisfied ............................................................. 1 2 3 4
9. I feel frightened ......................................................... 1 2 3 4
10. I feel comfortable ...................................................... 1 2 3 4
11. I feel self-confident ................................................... 1 2 3 4
12. I feel nervous ............................................................ 1 2 3 4
13. I am jittery ................................................................. 1 2 3 4
14. I feel indecisive .......................................................... 1 2 3 4
15. I am relaxed .............................................................. 1 2 3 4
16. I feel content ............................................................ 1 2 3 4
17. I am worried ............................................................. 1 2 3 4
18. I feel confused .......................................................... 1 2 3 4
19. I feel steady ............................................................. 1 2 3 4
20. I feel pleasant .......................................................... 1 2 3 4
# SELF-EVALUATION QUESTIONNAIRE

**STAI Form Y-2**

Name_________________________ Date__________

**DIRECTIONS**

A number of statements which people have used to describe themselves are given below. Read each statement and then circle the appropriate number to the right of the statement to indicate how you generally feel. There are no right or wrong answers. Do not spend too much time on any one statement but give the answer which seems to describe how you generally feel.

<table>
<thead>
<tr>
<th>Statement</th>
<th>ALMOST NEVER</th>
<th>ALMOST ALWAYS</th>
</tr>
</thead>
<tbody>
<tr>
<td>21. I feel pleasant.</td>
<td>1</td>
<td>2</td>
</tr>
<tr>
<td>22. I feel nervous and restless.</td>
<td>1</td>
<td>2</td>
</tr>
<tr>
<td>23. I feel satisfied with myself.</td>
<td>1</td>
<td>2</td>
</tr>
<tr>
<td>24. I wish I could be as happy as others seem to be.</td>
<td>1</td>
<td>2</td>
</tr>
<tr>
<td>25. I feel like a failure.</td>
<td>1</td>
<td>2</td>
</tr>
<tr>
<td>26. I feel rested.</td>
<td>1</td>
<td>2</td>
</tr>
<tr>
<td>27. I am “calm, cool, and collected”.</td>
<td>1</td>
<td>2</td>
</tr>
<tr>
<td>28. I feel that difficulties are piling up so that I cannot overcome them</td>
<td>1</td>
<td>2</td>
</tr>
<tr>
<td>29. I worry too much over something that really doesn’t matter.</td>
<td>1</td>
<td>2</td>
</tr>
<tr>
<td>30. I am happy.</td>
<td>1</td>
<td>2</td>
</tr>
<tr>
<td>31. I have disturbing thoughts.</td>
<td>1</td>
<td>2</td>
</tr>
<tr>
<td>32. I lack self-confidence.</td>
<td>1</td>
<td>2</td>
</tr>
<tr>
<td>33. I feel secure.</td>
<td>1</td>
<td>2</td>
</tr>
<tr>
<td>34. I make decisions easily.</td>
<td>1</td>
<td>2</td>
</tr>
<tr>
<td>35. I feel inadequate.</td>
<td>1</td>
<td>2</td>
</tr>
<tr>
<td>36. I am content.</td>
<td>1</td>
<td>2</td>
</tr>
<tr>
<td>37. Some unimportant thought runs through my mind and bothers me.</td>
<td>1</td>
<td>2</td>
</tr>
<tr>
<td>38. I take disappointments so keenly that I can’t put them out of my mind</td>
<td>1</td>
<td>2</td>
</tr>
<tr>
<td>39. I am a steady person.</td>
<td>1</td>
<td>2</td>
</tr>
<tr>
<td>40. I get in a state of tension or turmoil as I think over my recent concerns and interests</td>
<td>1</td>
<td>2</td>
</tr>
</tbody>
</table>
# State-Trait Anxiety Inventory for Adults Scoring Key (Form Y-1, Y-2)

Developed by Charles D. Spielberger in collaboration with R.L. Gorsuch, R. Lushene, P.R. Vagg, and G.A. Jacobs

To use this stencil, fold this sheet in half and line up with the appropriate test side, either Form Y-1 or Form Y-2. Simply total the scoring weights shown on the stencil for each response category. For example, for question #1, if the respondent marked 3, then the weight would be 2. Refer to the manual for appropriate normative data.

<table>
<thead>
<tr>
<th>Form Y-1</th>
<th>Form Y-2</th>
</tr>
</thead>
<tbody>
<tr>
<td>1.</td>
<td>21.</td>
</tr>
<tr>
<td>2.</td>
<td>22.</td>
</tr>
<tr>
<td>3.</td>
<td>23.</td>
</tr>
<tr>
<td>4.</td>
<td>24.</td>
</tr>
<tr>
<td>5.</td>
<td>25.</td>
</tr>
<tr>
<td>7.</td>
<td>27.</td>
</tr>
<tr>
<td>8.</td>
<td>28.</td>
</tr>
<tr>
<td>9.</td>
<td>29.</td>
</tr>
<tr>
<td>10.</td>
<td>30.</td>
</tr>
<tr>
<td>11.</td>
<td>31.</td>
</tr>
<tr>
<td>12.</td>
<td>32.</td>
</tr>
<tr>
<td>13.</td>
<td>33.</td>
</tr>
<tr>
<td>14.</td>
<td>34.</td>
</tr>
<tr>
<td>15.</td>
<td>35.</td>
</tr>
<tr>
<td>16.</td>
<td>36.</td>
</tr>
<tr>
<td>17.</td>
<td>37.</td>
</tr>
<tr>
<td>18.</td>
<td>38.</td>
</tr>
<tr>
<td>19.</td>
<td>39.</td>
</tr>
<tr>
<td>20.</td>
<td>40.</td>
</tr>
</tbody>
</table>
APPENDIX VI: THE ZUNG SELF-RATING DEPRESSION SCALE

ZUNG SELF-RATING DEPRESSION SCALE

<table>
<thead>
<tr>
<th>Make check mark (✓) in appropriate column.</th>
<th>A little of the time</th>
<th>Some of the time</th>
<th>Good part of the time</th>
<th>Most of the time</th>
</tr>
</thead>
<tbody>
<tr>
<td>1. I feel down-hearted and blue</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>2. Morning is when I feel the best</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>3. I have crying spells or feel like it</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>4. I have trouble sleeping at night</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>5. I eat as much as I used to</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>6. I still enjoy sex</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>7. I notice that I am losing weight</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>8. I have trouble with constipation</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>9. My heart beats faster than usual</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>10. I get tired for no reason</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>11. My mind is as clear as it used to be</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>12. I find it easy to do the things I used to</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>13. I am restless and can't keep still</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>14. I feel hopeful about the future</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>15. I am more irritable than usual</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>16. I find it easy to make decisions</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>17. I feel that I am useful and needed</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>18. My life is pretty full</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>19. I feel that others would be better off if I were dead</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>20. I still enjoy the things I used to do</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

Adapted from Zung, A self-rating depression scale, Arch Gen Psychiatry, 1965; 12: 63-70.
KEY TO SCORING THE ZUNG SELF-RATING DEPRESSION SCALE

Consult this key for the value (1-4) that correlates with patients’ responses to each statement. Add up the numbers for a total score. Most people with depression score between 50 and 69. The highest possible score is 80.

<table>
<thead>
<tr>
<th>Make check mark (✓) in appropriate column.</th>
<th>A little of the time</th>
<th>Some of the time</th>
<th>Good part of the time</th>
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<tbody>
<tr>
<td>1. I feel down-hearted and blue</td>
<td>1</td>
<td>2</td>
<td>3</td>
<td>4</td>
</tr>
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<td>3</td>
<td>2</td>
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<td>3</td>
<td>4</td>
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<td>3</td>
<td>4</td>
</tr>
<tr>
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<td>4</td>
<td>3</td>
<td>2</td>
<td>1</td>
</tr>
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<td>1</td>
</tr>
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<td>2</td>
<td>3</td>
<td>4</td>
</tr>
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<td>2</td>
<td>3</td>
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<td>2</td>
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<td>3</td>
<td>2</td>
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<td>2</td>
<td>3</td>
<td>4</td>
</tr>
<tr>
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<td>3</td>
<td>2</td>
<td>1</td>
</tr>
</tbody>
</table>

Adapted from Zung.


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### APPENDIX VII: SAMPLE SIZE CALCULATION

<table>
<thead>
<tr>
<th>Group</th>
<th>Proportion Positive</th>
<th>N Per Group</th>
<th>Standard Error</th>
<th>95% Lower</th>
<th>95% Upper</th>
</tr>
</thead>
<tbody>
<tr>
<td>Population 1</td>
<td>0.46</td>
<td>173</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Population 2</td>
<td>0.61</td>
<td>173</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Rate Difference</td>
<td>-0.15</td>
<td>346</td>
<td>0.06</td>
<td>0.25</td>
<td>0.05</td>
</tr>
</tbody>
</table>

Alpha: 0.050, tails: 2

Power: 80%
BIBLIOGRAPHY

19. CDC. 2012 Assisted Reproductive Technology Fertility Clinic Success Rates Report. Atlanta, Georgia Centers for Disease Control and Prevention, American Society for Reproductive Medicine, Society for Assisted Reproductive Technology; 2012.


January 14, 2015.


65. Langevin HM, Yandow JA. Relationship of acupuncture points and meridians to connective tissue planes. The Anatomical Record. 2002;269(6):257-265.


115. Spielberger CD. *State-Trait Anxiety Inventory*. Wiley Online Library; 2010.


117. Stener-Victorin E, Humaidan P. Use of acupuncture in female infertility and a


