Could acupuncture at the so-called forbidden points be harmful to the health of pregnant Wistar rats?

André Vilella Guerreiro da Silva,1,2 Mary Uchiyama Nakamura,1 João Bosco Guerreiro da Silva,2 José Antonio Cordeiro,3 Glória Elisa Mendes,4 Luiz Lima5

ABSTRACT

Aim This study is the second report from a single experiment planned to determine if acupuncture at LI4 and SP6, or at the sacral points, could produce any harm in the pregnancy outcome of Wistar rats, as has been hypothesised in traditional knowledge. Here, we observe if these points can be harmful to the health of pregnant Wistar rats.

Methods A total of 48 pregnant Wistar rats were randomly divided into 4 groups: total control, where rats were left in cages without manipulation; anesthetised control, where rats were manipulated and anesthetised but did not receive electroacupuncture; and peripheral point and sacral point groups, where rats were anesthetised and received 6 sessions of electroacupuncture at 4 acupuncture points: LI4 and SP6, and BL27 and BL28, respectively. In the 19th day of pregnancy the rats were killed and examined. The primary endpoints were levels of biochemical parameters. No differences were also seen regarding the histological changes of liver and kidney.

Results There were no differences between the levels of biochemical parameters. No differences were also seen regarding the histopathological analysis.

Conclusions We found no evidence that acupuncture at the LI4, SP6 and sacral points could be harmful to the Wistar rat dams.

INTRODUCTION

Acupuncture has been offered as a medical practice in many Western countries and has even been adopted as a form of institutional healthcare in many of them.1 It is used in several diseases as a complementary or even a first choice treatment. Obstetrics is one such area where acupuncture seems to fit in very well.2 Being a non-pharmacological treatment, it avoids problems that many drugs may cause in this sensitive period of a woman’s life. Good results with different conditions (such as nausea and vomiting,3 low back pain,4 depression,5 insomnia6 and dyspeptic problems7) have encouraged its use by researchers and practitioners. Notwithstanding these results, concerns remain about using acupuncture in pregnancy because it has been hypothesised that some points are forbidden in pregnant women due to potential abortive effects. Based on ancient sources,8–10 many points are thought to be dangerous, but LI4 (Hoku), SP6 (Sanyin Jiao) and points in the sacral area are the most cited,11–14 but there is no scientific evidence for this issue. Acupuncture and mainly electroacupuncture have been carried out in anesthetised animals, following models for location acupuncture points in different mammals,15–17 when experiments in humans are not possible. Rats are the most commonly used animals for this purpose.18–20 This is a further report of an experiment conducted to investigate whether acupuncture at the so-called ‘forbidden points’ is be harmful to pregnancy.21 In this report we investigate the possible harmful effect to maternal health.

MATERIALS AND METHODS

This experiment, conducted according to the National Institute of Health guidelines for the use and care of animals, was approved by the Ethics in Animal Research Committee of Faculdade de Medicina de São José do Rio Preto (Rio Preto Medical College, São Paulo State, Brazil), process 001-0032224/2008. We randomly divided 48 female Wistar rats obtained from the animal care facility of

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Received 17 September 2012
Revised 19 December 2012
Accepted 14 January 2013
Published Online First
5 February 2013

our institution into 4 groups: (A) total control (rats were left in the cages without manipulation; n=12); (B) anesthetised control (rats were manipulated and anesthetised but did not receive acupuncture; n=12); (C) peripheral points (rats were anesthetised and received acupuncture at LI4 and SP6; n=12) and (D) sacral points (rats were anesthetised and received acupuncture in the lumbar sacral region at BL27 and BL28; n=12). The anesthetised groups received xylazine 10 mg/kg and ketamine 50 mg/kg, which provided satisfactory anaesthesia without adverse effects on gestation or the fetuses.22 The peripheral points were used according to a proposed transpositional acupuncture point system in a rat model,23 and BL27 and BL28 were chosen because they are located in the L6 and S1 spinal cord segments, respectively.

The rats were bred by caging female rats with proven fertile male rats. Day 0 of pregnancy was indicated by the presence of a sperm-positive vaginal smear, and copulation-confirmed rats were divided at random into the four groups. Animals were kept in group cages under steady conditions, 12:12 h light/dark cycle and a room temperature of 25±3°C. The animals received rat chow and water ad libitum.

Electroacupuncture (EL 608, NKL, Brazil) with single-use, sterile, prepacked acupuncture needles with guide tubes, 25 mm×0.20 mm (Dongbang Acupuncture Inc., Boryeong City, South Korea) was used because it is believed to result in stronger stimulation than manual acupuncture. We used a frequency of 5 Hz, medium intensity, for 25 min. The entire treatment consisted of 6 sessions: 2 sessions in the first 7 days (implantation period), 2 sessions from the 8th to the 14th day (embryonic period), and the last 2 sessions between the 15th and 18th days (fetal developmental period).

The primary endpoints were the maternal biochemical changes represented by the levels of glucose, aspartate aminotransferase (AST), alanine aminotransferase (ALT) and creatinine, and the histological aspects of the liver and kidney. In the 19th day of pregnancy the dams were weighed, had a laparotomy performed after anaesthesia, had the product of conception or any other adverse effects. Food and water intake was comparable and showed no significant differences among the animals (data not shown).

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The primary endpoints were the maternal biochemical changes represented by the levels of glucose, aspartate aminotransferase (AST), alanine aminotransferase (ALT) and creatinine, and the histological aspects of the liver and kidney. In the 19th day of pregnancy the dams were weighed, had a laparotomy performed after anaesthesia, had the product of conception (as covered in a separate report), were canalised for blood collection and had their livers and right kidneys removed and fixed in 40 g/litre formic acid and processed for paraffin embedding. After that, rats were killed using sodium thiopental 100 mg/kg.

Tissue sections were obtained and stained with haematoxylin and eosin (H&E) for light microscopy and observed at 400× magnification. Using randomly selected high-power fields, the histological sections were analysed by a single pathologist, blinded to the groups. The liver was analysed with respect to the portal spaces (size, usual components—central vein, bile duct and arterioles), veins and centrilobular hepatocytic and sinusoidal trabecular architecture. The kidney was analysed with respect to the glomeruli (cellularity, amount of mesangial matrix, capillary loops), cortex and medulla in tubules, interstitium and vessels (arteries and arterioles). A total of 10 randomly selected areas of liver and kidney were examined and were scored as follows: 0, normal; 1, changes involving <25% of the tissue; 2, alterations involving >25% and <50% of the tissue; 3, similar changes involving >50% but <75% of the tissue; and 4, changes involving >75% of the tissue.

The Kruskal–Wallis test was used to compare the groups. If any difference was seen, it was calculated as between-group comparisons with Bonferroni correction.25 The adopted significance level was α=0.05.

### RESULTS

Results are summarised in table 1. All pregnant rats were healthy during the gestation period. None of them showed vaginal bleeding or expulsion of the products of conception or any other adverse effects. Food and water intake was comparable and showed no significant differences among the animals (data not shown).

There were no differences between the levels of glucose, ALT, AST and creatinine between groups.

There were no differences in all parameters observed on histopathological analysis of the liver and kidney, as shown in figures 1–4. They received scores of 0, which means no changes at all were seen. Minimal differences were a few vascular and/or ischaemic changes, in a greater or lesser extent attributed to delays in collection of material (eg, acute retraction of the tuft capillary, tubular or hepatocytic

### Table 1 Summarised data

<table>
<thead>
<tr>
<th>Factor</th>
<th>Group A: total control (n=12)</th>
<th>Group B: anesthetised control (n=12)</th>
<th>Group C: peripheral points (n=12)</th>
<th>Group D: sacral points (n=12)</th>
<th>p Value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Initial weight</td>
<td>212.4 (24.6)</td>
<td>218.6 (28.8)</td>
<td>210.1 (19.3)</td>
<td>216.5 (25.3)</td>
<td>0.42</td>
</tr>
<tr>
<td>Glucose</td>
<td>151.1 (30.0)</td>
<td>139.6 (20.4)</td>
<td>146.6 (26.0)</td>
<td>133.2 (21.6)</td>
<td>0.31</td>
</tr>
<tr>
<td>AST</td>
<td>58.5 (23.5)</td>
<td>56.6 (23.5)</td>
<td>54.5 (18.5)</td>
<td>61.6 (23.9)</td>
<td>0.23</td>
</tr>
<tr>
<td>ALT</td>
<td>43.6 (25.8)</td>
<td>50.22 (28.6)</td>
<td>45.0 (24.2)</td>
<td>37.4 (16.0)</td>
<td>0.13</td>
</tr>
<tr>
<td>Creatinine</td>
<td>0.3 (0.02)</td>
<td>1.9 (0.02)</td>
<td>0.56 (0.03)</td>
<td>0.46 (0.02)</td>
<td>0.46</td>
</tr>
</tbody>
</table>

Data shown are median (IQR). ALT, alanine aminotransferase; AST, aspartate aminotransferase.
degeneration, mild interstitial oedema), equally distributed between the groups.

**DISCUSSION**

Data from this study demonstrates that acupuncture at the so-called ‘forbidden points’—LI4, SP6 and the sacral points—resulted in no differences in the evaluated parameters when compared with total control and anesthetised control groups.

Glucose, ALT, AST and creatinine are some of the most important serum biochemical parameters to evaluate normal liver and kidney function and their possible injury by a chemical or a physical agent.26 We did not find any differences between the groups for any of these four parameters. We also could not find any differences on histopathological analysis of the livers and kidneys, except for some changes assumed to be related to delays in collect the material, randomly distributed between the groups.

The concept of ‘forbidden (acupuncture) points in pregnancy’ has been discussed recently elsewhere.27 Among acupuncture points, LI4, SP6 and points in the sacral and lower abdomen area are the ones most cited as ‘dangerous’, mostly by traditional sources.11–14 It has been shown however that there is no plausible mechanism for risk.28 Despite these traditional concerns,29 acupuncture has even been indicated as an option for threatenend miscarriages.30

In a recent paper we found that acupuncture at the LI4, SP6 and sacral points did not have any harmful effects on the pregnancy outcome in female Wistar rats.21 There were no differences in the size of the litters, number of implantations, resorptions, malformations and weight of rat pups.

Acupuncture at these points could produce uterine contractions, but only in post-date pregnant women,31 labour and fetal death cases.32 In some experiments it could protect from preterm labour, natural33 or provoked by oxytocin,34 showing a regulative effect on abnormal electrohysterograms of pregnant rats provoked by oxytocin and progesterone injection.35

Even with several indications that the acupuncture effect would not be deleterious in pregnancy, many acupuncturists are still worried about the idea of these points being harmful because of the strength of traditional knowledge. Could this concern be linked to
the health of the mother and not the pregnancy itself? That was the hypothesis we tested in this paper.

Rat pregnancy lasts 21 days. We performed 6 sessions covering the implantation period (first 7 days), the embryonic period (from the 8th to the 14th day) and the fetal development period (15th to 18th days), with 2 sessions each period.

Even considering the remote possibility that this procedure could provoke, via the so-called 'forbidden points in pregnancy', any harm to the pregnant rats, we did not find any difference in the evaluated parameters. To the best of our knowledge, this is the first study to observe the possible harmful effects of acupuncture in these polemic points in maternal development. However, our findings are in agreement with other studies that have shown acupuncture may regulate some parameters in the liver and kidney such as glucose, AST, ALT, and creatinine, and may even have a protective effect in the liver and kidney from lesions caused by carbon tetrachloride and lipopolysaccharides.

Acupuncture, stimulating the nervous system, by segmental and extrasegmental neuromodulation and other central nervous system effects, plays a homeostatic role. It can control heart rate, blood pressure, body temperature, granulocytes and lymphocytes, and mucociliary transport. It means that, irrespective of condition, this method of treatment seems to normalise the patterns of these parameters. Why could these not happen in a normal pregnancy? In our sample we did not find any harm in dams treated by acupuncture at LI4, SP6 and sacral points.

Our study has limitations. In respect to Ethics in Animal Research we had to use a limited number of animals. It is also not simple to extrapolate data from rat pregnancies to human pregnancies. Adverse effects are very complex in real life, and not finding them is not a guarantee that they do not exist. We believe, however, that our study can provide some input into the controversial issue of the safety of acupuncture in pregnancy.

CONCLUSIONS

In this study we did not see any ill effects provoked by acupuncture use of LI4, SP6, or sacral points on the health of pregnant Wistar rats.

Summary points

- Acupuncture is reported to have adverse effects in pregnancy.
- A small study found no adverse effects on rat foetuses.
- This paper reports also no adverse effects found on the pregnant rats.

Contributors AVG and GEM performed the laboratory work, JAC performed the statistical tests, LL performed the histopathological work, AVG, MUN and JBGdS planned the protocol. All authors wrote and revised the manuscript.

Competing interests None.

Provenance and peer review Not commissioned; externally peer reviewed.

REFERENCES

18 Higashimura Y, Shimou R, Maruyama H et al. Electro-acupuncture improves responsiveness to insulin via...


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*Acupunct Med* 2013 31: 202-206 originally published online February 5, 2013
doi: 10.1136/acupmed-2012-010246

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