

## Experimental Research

# Effect of instant moxibustion on the levels of prostaglandin and arginine vasopressin in the uterine tissues of dismenorrhea rats with cold-damp congealing and stagnation type<sup>\*</sup>

## 即时灸对寒湿凝滞型痛经大鼠子宫组织前列腺素及加压素含量的影响<sup>\*</sup>

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### ABSTRACT

**Objective** To observe the effect of instant moxibustion on the levels of prostaglandin E<sub>2</sub> (PGE<sub>2</sub>), prostaglandin F<sub>2α</sub> (PGE<sub>2α</sub>) and arginine vasopressin (AVP) in the uterine tissues of dismenorrhea rats with cold-damp congealing and stagnation type and to explore its possible mechanism. **Methods** Female Wistar rats were randomly divided into blank group, model group, herble medicine group, pre-moxibustion group and instant moxibustion group, with 9 rats in each group. Cold-damp congealing and stagnation type primary dismenorrhea models were established by adopting (0±1)°C ice water-extraction method combined with estradiol benzoate injection method. After modeling on the 8th day, in herble medicine group, *Tongjingbao* granules was given to the rats by intragastric administration. In pre-moxibustion group, mild moxibustion was carried out at “Shénquè” (神阙 CV 8) and “Guānyuán” (关元 CV 4) of the rats for 10 min at each acupoint. In instant moxibustion group, moxibustion as that in pre-moxibustion group was conducted for once after injection with oxytocin on the 11th day. ELISA was adopted to detect the levels of PGE<sub>2</sub> and PGE<sub>2α</sub> in the uterine tissues of rats, and radioimmunoassay was used for detection of AVP level in the uterine tissues of rats. **Results** Compared with the model group, the latent period of rats in herbal medicine group, pre-moxibustion group and instant moxibustion group obviously prolonged, the number of times of torsion reduced, and the total score of torsion decreased ( $P<0.01$ ); compared with herbal medicine group, the latent period of rats in instant moxibustion group obviously prolonged, and the total score of torsion decreased ( $P<0.05$  or  $P<0.01$ ); compared with pre-moxibustion group, the number of times of torsion of rats in instant moxibustion group reduced, and the total score of torsion decreased ( $P<0.01$ ). Compared with blank group, the levels of PGE<sub>2α</sub> and AVP and the ratio of PGE<sub>2α</sub> and PGE<sub>2</sub> in the uterine tissues of rats in model group significantly increased ( $P<0.01$ ), and the PGE<sub>2</sub> level significantly reduced ( $P<0.01$ ); compared with model group, the PGE<sub>2α</sub> level and the ratio of PGE<sub>2α</sub> and PGE<sub>2</sub> in the uterine tissues of rats in herble medicine group, pre-moxibustion group and instant moxibustion group obviously reduced ( $P<0.05$  or  $P<0.01$ ), the PGE<sub>2</sub> level obviously increased ( $P<0.01$ ), and the AVP level in the uterine tissues of rats in pre-moxibustion group and

instant moxibustion group obviously reduced ( $P<0.05$  or  $P<0.01$ ); compared with herbal medicine group, the levels of  $\text{PGE}_{2\alpha}$  and AVP and the ratio of  $\text{PGE}_{2\alpha}$  and  $\text{PGE}_2$  in the uterine tissues of rats in instant moxibustion group significantly reduced ( $P<0.05$  or  $P<0.01$ ); compared with pre-moxibustion group, the  $\text{PGE}_{2\alpha}$  level and the ratio of  $\text{PGE}_{2\alpha}$  and  $\text{PGE}_2$  in the uterine tissues of rats in instant moxibustion group obviously reduced ( $P<0.05$ ), and the  $\text{PGE}_2$  level obviously increased ( $P<0.01$ ). **Conclusion** Both pre-moxibustion and instant moxibustion can obviously inhibit spasmodic uterine smooth muscle contraction of rats with dismenorrhea, regulate imbalanced levels of  $\text{PGE}_{2\alpha}$  and  $\text{PGE}_2$ , reduce the AVP level, so as to improve the uterine hypoxia-ischemia, and play a role in alleviating pain. The efficacy of instant moxibustion was superior to that of pre-moxibustion.

**KEY WORDS:** Instant effect; cold-damp congealing and stagnation type; dismenorrhea; prostaglandin; arginine vasopressin

Primary dismenorrhea is also called functional painful menstruation, which is one of the common and frequent diseases in adolescent females. It is believed in modern medicine that primary dismenorrhea is not only related with neuroendocrine system, but also related to such factors as heredity, immunity, metabolism, and environment, etc<sup>[1]</sup>. It has been reported according to multiple literature that moxibustion intervention has satisfactory efficacy in treatment of primary dismenorrhea, which are highly favored by most patients<sup>[2-4]</sup>. It was found based on previous clinical studies that most patients with dismenorrhea manifested as cold-damp congealing and stagnation, and good efficacy has been obtained by adopting moxibustion at Shénquè (神阙 CV 8) and Guānyuán (关元 CV 4)<sup>[5-7]</sup>. The effect of instant moxibustion on the levels of  $\text{PGE}_2$ ,  $\text{PGE}_{2\alpha}$  and AVP in the uterine tissues of rats with primary painful menstruation was observed, in order to explore the efficacy difference and the possible mechanism of instant moxibustion on primary painful menstruation.

## MATERIALS AND METHODS

### Laboratory animals

Sixty clean, healthy, mature and unmated Wistar female rats, with the age of 8-10 weeks and weight of  $(200\pm 20)$  g, were provided by the animal experiment center of Hebei Medical University (certification No. 1509045). The rats were fed in the clean animal laboratory with the room temperature of  $(23\pm 2)$  °C, humidity of  $(45\pm 5)\%$ , and light and dark duration of 12 h, respectively, and they were permitted to drink and eat at liberty. The disposal of laboratory animals was reviewed and approved by the Ethics Committee of Hebei University of Chinese Medicine.

### Main reagents and instruments

Prostaglandin  $\text{E}_2$  radioimmunoassay kits (Beijing Puerweiyue Bio-Technology Co., Ltd.), rats prostaglandin  $\text{F}_{2\alpha}$  ( $\text{PGE}_{2\alpha}$ ) ELISA kits (Shanghai Westang Bio-Technology Co., Ltd.), arginine vasopressin radioimmunoassay kits (Beijing Huabulite Institute of Biotechnology), estradiol benzoate injection (Inner Mongolia Chifeng Boen Pharmaceutical Co., Ltd.), oxytocin injection (Inner Mongolia Chifeng Boen Pharmaceutical Co., Ltd.), Tongjingbao granules (Henan Wanxi Pharmaceutical Co., Ltd.), 7 mm (diameter) moxa sticks (Nanyang Hanyi Moxibustion Technology Development Co., Ltd.), TD10001 model electronic balance (Tianjin Balance Instrument Co., Ltd.), TDL-5-A model centrifuge (Shanghai Anting Scientific Instruments Factory), SHB-D model circulating water vacuum pump (Zhengzhou GreatWall), FJ-2021 model  $\gamma$ -radioimmunoassay counter (Xi'an Two-six-two Factory).

### Modeling and grouping

After adaptive feed for 7 d, the 60 Wistar female rats were screened for 4 d by adopting vaginal smear test<sup>[8-9]</sup>. The rats without diestrus or in the same period all the time were eliminated, and 45 rats were selected finally. The 45 rats were randomly divided into blank group, model group, herbal medicine group, pre-moxibustion group and instant moxibustion group with 9 rats in each group. Cold-damp congealing and stagnation type painful menstruation models were established by adopting  $(0\pm 1)$  °C ice water-extraction method combined with estradiol benzoate injection method<sup>[10-11]</sup> in all the groups except blank group. The posterior limbs and hypogastrium of rats were immersed into  $(0\pm 1)$  °C ice-water mixture (room temperature  $23\pm 2$  °C) for cold stimulus, once

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