Effects of unilateral or bilateral superior ovarian nerve section in prepubertal rats on the ovulatory response to gonadotrophin administration

L Morales, R Chávez, M E Ayala and R Domínguez

Biology of Reproduction Research Unit, Laboratory of Physiology of Reproduction, Facultad de Estudios Superiores Zaragoza, UNAM AP 9-020, CP 15000, México, D.F., México

(Requests for offprints should be addressed to L Morales)

Abstract

The modulating effects of ovarian innervation reaching the ovary through the suspensory ovarian nerve on the reactivity of the ovaries to gonadotrophins were analysed. Juvenile rats (32 days old), with or without unilateral or bilateral section of the superior ovarian nerve, were injected with 8 iu of pregnant mare serum gonadotrophin (PMSG), 10 iu of human chorionic gonadotrophin (hCG) or with 8 iu of PMSG followed 56 h later with 10 iu of hCG. Treatments were given immediately after surgery or 4 days later, and the rats were killed on the day of first vaginal oestrus. In rats with unilateral section, treatment with PMSG did not induce full ovulatory response by the denervated ovary whether the treatment was applied immediately or 4 days after surgery (0/11 rats treated immediately ovulated vs 5/5 (sham) and 11/12 (control, $P<0.05$ Fisher's exact probability test), and 4/19 did when treatment was done 4 days after surgery vs 8/10 (sham) and 11/12 (control, $P<0.05$). The rats with bilateral section receiving the same hormonal treatment, PMSG administration, ovulated. The number of ova shed by the left ovary was similar to those of the control, while the right ovary released fewer ova. Stimulation with hCG immediately after unilateral section did not induce ovulation in normal or denervated ovary. When the treatment was applied 4 days after surgery, ovulation was observed only in the innervated ovary. In the rats with bilateral section, hCG injection induced ovulation in both ovaries. In those rats with unilateral section of the superior ovarian nerve, the treatment with PMSG+hCG given immediately after surgery resulted in a compensatory ovulation by the innervated ovary (the number of ova shed/ovulating

animal was significantly higher than those released by control or sham-operated animals: left section, number of ova shed by the right ovary 7.6 ± 0.3 vs 5.5 ± 0.8 and 4.9 ± 1 respectively, $P<0.05$; right section, number of ova shed by the left ovary 10.2 ± 0.6 vs 4.4 ± 1.1 and 7.0 ± 0.9 , $P<0.05$), while the denervated one showed a lower ovulation rate as well as a smaller number of ova shed than those by the control animals. When the hormonal replacement was given 4 days after surgery, such compensatory ovulation was observed in the left ovary of those rats with a section of the right nerve (14.3 ± 2.6 vs 4.4 ± 1.1 and 6.5 ± 1.1 , $P<0.05$). When the PMSG+hCG treatment was applied to animals with bilateral section of the superior ovarian nerve, the ovulation rate by the right ovary was significantly lower than in control and sham-operated treated animals (2/10 vs 11/11 and 6/7, $P<0.05$). Because the ovaries receive innervation through the superior ovarian nerve, the ovarian plexus and the vagus nerve, the results obtained in unilateral denervated animals suggest that the innervation of the ovary via the superior ovarian nerve regulates in a stimulatory way the effects of gonadotrophin resulting in ovulation. The ovulation induced by hormonal treatment of rats with bilateral section of the superior ovarian nerve suggests that the effects of bilateral section on ovulation are not the addition of the effects of left and right denervation, implying the existence of a modulatory effect in gonadotrophin action on ovulation via other neural pathways.

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Introduction

The evidence obtained from pharmacological or surgical denervation suggests that ovarian innervation participates in the regulation of ovarian functions. The main neural information arriving at the ovaries is through the superior

ovarian nerve, ovarian plexus and vagus nerve (Burden 1978, Lawrence & Burden 1980).

Peripheral sympathetic denervation, induced by chronic guanethidine injection into prepubertal rats, results in a marked accumulation of small antral follicles and a relative paucity of large follicles (Lara *et al.* 1990), as well as an

increase in the number of ova shed by ovulating animals (Flores *et al.* 1990). The sympathetic denervation produced by bilateral section of the superior ovarian nerve results in an acute decrease in serum levels of progesterone (Erskine *et al.* 1988) or oestradiol (Aguado & Ojeda 1984). This evidence supports the idea that hormonal control of ovarian functions (hormonal secretion and ovulation) is modulated by ovarian innervation.

We have previously shown that the effects of unilateral or bilateral ovarian denervation on ovulation are different. While bilateral section of the superior ovarian nerve performed in prepubertal and adult rats did not modify the number of ovulating rats (ovulation rate), nor the number of ova shed, unilateral section resulted in a diminution of the ovulation rate and the number of ova shed by the denervated ovary (Chávez *et al.* 1991, Morales *et al.* 1993). These results suggest that ovarian innervation through the superior ovarian nerve modulates the responsiveness of the ovarian compartments to gonadotrophins, in a stimulatory and asymmetrical manner (Chávez & Domínguez 1994, Domínguez *et al.* 1998).

Our objective was to analyse whether ovarian innervation reaching the ovary through the superior ovarian nerve modulates the reactivity of the ovary to gonadotrophins in a stimulatory way, and to compare the effects on ovulation of gonadotrophin administration to prepubertal rats with and without unilateral or bilateral section of the superior ovarian nerve. For that purpose, the animals were stimulated with pregnant mare serum gonadotrophin (PMSG) to induce follicular growth and oestrogen secretion or with human chorionic gonadotrophin (hCG) to induce oestrogen secretion without an initial acceleration of the follicular development. Both paradigms need endogenous luteinising hormone (LH) release to induce ovulation (D'Albora *et al.* 1974, Greenwald & Roy 1994). To bypass the neuroendocrine endogenous mechanisms regulating LH secretion, PMSG was injected, followed 56 h later by hCG.

Materials and Methods

Animal surgery and hormone treatment

Thirty-two-day old CIIZ-V strain female rats from our breeding stock were kept under conditions of controlled lighting (lights on from 0500 to 1900 h). The animals were kept in group cages (six rats per cage), with free access to food and tap water. All surgery was under ether anaesthesia. Groups of untouched-, sham-operated- and denervated-gonadotrophin treated rats were processed at the same time. Gonadotrophins were diluted in saline, and 0.5 ml was injected subcutaneously. Rats were inspected daily for vaginal opening, and once it occurred vaginal smears were obtained daily. On the morning of the day of the first vaginal oestrus, all animals were killed by bleeding under ether anaesthesia.

The animals were allotted at random to one of three groups.

Sham-operated group Forty-five rats were anaesthetised, laparotomised and the wound sealed. Three groups of animals were treated immediately after surgery with 8 iu of PMSG (Sigma Chemical Co., St Louis, MO, USA) (five rats); or 10 iu of hCG (Sigma) (five rats); or 8 iu of PMSG followed 56 h later by 10 iu of hCG (eight rats). Three other groups of sham-operated animals were left untouched for 3 days, and on day 4 they were treated with gonadotrophins in the same described sequence (ten treated with PMSG, ten with hCG and seven with PMSG+hCG).

Denervated animals One hundred and fifty-seven rats were used. A midline incision was made to provide access to the suspensory ligament (left, right or both) containing the superior ovarian nerve. It was then isolated from the surrounding tissue, and the nerve was sectioned as previously described (Chávez *et al.* 1991, Morales *et al.* 1993). When a bilateral section was performed, the first nerve sectioned was always the left one. All the animals were injected with PMSG, hCG or PMSG+hCG, immediately or on day 4 after surgery.

Control untouched group Thirty-six-day-old rats were injected with PMSG (24 animals), hCG (14 rats) or PMSG+hCG (22 animals).

Autopsy procedure

The animals were killed by an ether overdose. At autopsy, the right and left oviducts were dissected and the number of ova present was counted with the aid of a dissecting microscope. Ovaries and uterus were removed and weighed using a precision balance. The ovaries from non-ovulating rats were fixed in Bouin's fluid, embedded in paraffin, serial sectioned, stained with haematoxylin-eosin and inspected for the presence of fresh corpora lutea.

Statistical analysis

Data on the weight of the ovaries and uterus were analysed by multivariate analysis of variance (MANOVA) (for each time when the animals were treated, immediately after surgery and 4 days later, the independent variables were group and hormonal treatments), followed by Tukey's test. Ovulation rate (number of ovulating animals/number of treated one) (Chávez & Domínguez 1994) was analysed by Fisher's exact probability test or the Chi square test; number of ova shed/ovulating animal by the Kruskal-Wallis test followed by a Mann-Whitney U test. A *P* value of less than 0.05 was considered significant.

Table 1 Ovulation rate (number of ovulating/number of treated) and mean $\pm$ S.E.M. number of ova shed in animals with unilateral or bilateral section of the superior ovarian nerve performed on 32-day-old rats treated with 8 iu PMSG immediately or 4 days later. Animals were autopsied at the first day of vaginal oestrus

Group	Immediately after surgery				Four days later			
	Ovulation rate		Number of ova shed		Ovulation rate		Number of ova shed	
	Left	Right	Left	Right	Left	Right	Left	Right
Control	11/12	11/12	6.6 $\pm$ 0.9	6.5 $\pm$ 1.2	11/12	11/12	6.6 $\pm$ 0.9	6.5 $\pm$ 1.2
Sham	4/5	5/5	5.0 $\pm$ 1.3	5.8 $\pm$ 0.4	8/10	8/10	4.7 $\pm$ 0.8	5.9 $\pm$ 0.9
Left section	0/5	1/5†*	0	7	2/9†*	9/9	3, 5	5.7 $\pm$ 0.7
Right section	4/6	0/6	3.8 $\pm$ 0.5†	0	9/10	2/10†*	6.1 $\pm$ 0.8	1, 2
Bilateral section	11/12	6/12†*	5.8 $\pm$ 0.6	2.8 $\pm$ 0.9†*	7/10	5/10†	7.5 $\pm$ 2.2	3.4 $\pm$ 1.3

† $P < 0.05$ vs control group (ovulation rate by Chi square test; number of ova shed by Kruskal–Wallis test).

* $P < 0.05$ vs sham group (ovulation rate by Chi square test; number of ova shed by Kruskal–Wallis test).

Table 2 Mean $\pm$ S.E.M. of the weight of ovaries (mg/100 g body weight) in animals with unilateral or bilateral section of the superior ovarian nerve performed on 32-day-old rats, treated with 8 iu PMSG, 10 iu hCG or PMSG plus hCG immediately or 4 days later. Animals were autopsied on the first day of vaginal oestrus

Group	Immediately after surgery			Four days later		
	PMSG	hCG	PMSG+hCG	PMSG	hCG	PMSG+hCG
Control	60.8 $\pm$ 4.0	37.5 $\pm$ 3.2	47.9 $\pm$ 2.1	60.8 $\pm$ 4.0	37.5 $\pm$ 3.2	47.9 $\pm$ 2.1
Sham	53.2 $\pm$ 7.3	65.6 $\pm$ 4.8†	65.3 $\pm$ 3.4†	41.7 $\pm$ 3.6†	47.9 $\pm$ 1.5†	71.4 $\pm$ 2.6†
Left section	42.3 $\pm$ 3.2†	61.7 $\pm$ 3.9†	73.1 $\pm$ 2.1†	56.6 $\pm$ 5.7	50.2 $\pm$ 2.4†	57.9 $\pm$ 5.3
Right section	41.9 $\pm$ 5.7†	55.7 $\pm$ 4.3†	60.2 $\pm$ 2.4†	50.8 $\pm$ 6.7	42.8 $\pm$ 1.3	66.6 $\pm$ 9.4
Bilateral section	67.8 $\pm$ 2.1	59.8 $\pm$ 4.0†	72.4 $\pm$ 4.0†	55.6 $\pm$ 2.8*	55.1 $\pm$ 2.9†*	60.6 $\pm$ 6.2

† $P < 0.05$ vs control group (MANOVA).

* $P < 0.05$ vs sham group (MANOVA).

Results

Vaginal opening occurred 72 h after the beginning of the hormone stimulation in all rats (control+PMSG 24/24, control+hCG 14/14, control+PMSG+hCG 22/22; sham+PMSG 15/15, sham+hCG 15/15, sham+PMSG+hCG 15/15; denervated+PMSG 52/52, denervated+hCG 48/48, denervated+PMSG+hCG 57/57), and all the animals presented vaginal oestrus.

Effects of PMSG injection

Immediately after surgery Eleven out of 12 untouched control animals and five out of five sham-operated rats treated with PMSG, ovulated. Differences in the number of ova shed by ovulating animals were not observed (13.0 ± 1.96 in untouched control animals, and 11.0 ± 0.63 in sham-operated (NS, Mann–Whitney U test).

In those rats with section of the left superior ovarian nerve, a decrease in the ovulation rate occurred in both ovaries. None of the non-ovulating animals presented fresh corpora lutea in the ovaries. In the rats with section of the right superior ovarian nerve, a diminution in ovulation rate

by the denervated ovary was observed, while the number of ova shed decreased in both ovaries. The ovulation rate and number of ova shed by the left ovary were not affected by bilateral section of the superior ovarian nerve, while both parameters decreased in the right ovary (Table 1).

The weight of the ovaries in rats with unilateral section of the superior ovarian nerve was significantly lower than in untouched animals (Table 2). Such differences only occurred in the weight of the left ovary (left section: 19.4 ± 2.6 mg/100 g body weight; right section: 17.9 ± 1.8 vs 35.3 ± 4.8 , $P < 0.05$ MANOVA followed by Tukey's test).

The weight of the uterus in the sham-operated group was significantly lower than in untouched control animals (136.7 ± 7.3 mg/100 g body weight vs 167.5 ± 4.1 $P < 0.05$). Significant differences in the uterine weight between sham-operated and rats with unilateral or bilateral section of the superior ovarian nerve were not observed.

Four days after surgery Sham-operated animals ovulated in a similar way to controls. In rats with unilateral section of the superior ovarian nerve, the denervated ovary did not respond to PMSG treatment, while the innervated

Table 3 Ovulation rate (number of ovulating/number of treated) and mean $\pm$ S.E.M. number of ova shed in animals with unilateral or bilateral section of the superior ovarian nerve performed on 32-day-old rats treated with 10 iu hCG immediately or 4 days later. Animals were autopsied at the first day of vaginal oestrus

Group	Immediately after surgery				Four days later			
	Ovulation rate		Number of ova shed		Ovulation rate		Number of ova shed	
	Left	Right	Left	Right	Left	Right	Left	Right
Control	6/7	7/7	4.9 $\pm$ 1.1	5.4 $\pm$ 0.9	6/7	7/7	4.9 $\pm$ 1.1	5.4 $\pm$ 0.9
Sham	4/5	4/5	5.0 $\pm$ 1.0	6.0 $\pm$ 1.4	6/10	8/10	4.1 $\pm$ 1.2	5.3 $\pm$ 0.6
Left section	0/9	0/9	0	0	0/7	5/7	0	6.0 $\pm$ 0.5
Right section	0/8	0/8	0	0	6/8	2/8†*	6.0 $\pm$ 0.3	4, 4
Bilateral section	4/5	5/5	4.9 $\pm$ 0.9	5.9 $\pm$ 0.9	5/11	6/11	3.9 $\pm$ 1.3	3.2 $\pm$ 0.9

† $P < 0.05$ vs control group (Chi square test).

* $P < 0.05$ vs sham group (Chi square test).

gonad showed a similar response to that of sham-operated ones. In rats with bilateral section of the nerve, the number of ova shed by both ovaries was similar to that of sham-operated animals, while the ovulation rate by the right ovary was significantly lower (Table 1).

The weight of ovaries of rats with sham operation was lower than in untouched control animals injected with PMSG, but differences were not observed in rats with unilateral or bilateral section of the superior ovarian nerve (Table 2). Differences in the weight of the uterus were not observed.

Effects of hCG injection

Differences in ovulation rate and number of ova shed between untouched control and sham-operated rats treated with hCG immediately or 4 days after surgery were not observed.

Immediately after surgery The injection of hCG did not induce ovulation in those animals with unilateral section of the superior ovarian nerve, while all the animals with bilateral section did ovulate (Table 3). Fresh corpora lutea were not observed in the ovaries of non-ovulating animals.

The weight of the ovaries of sham-operated rats, and of rats with unilateral or bilateral section of the superior ovarian nerve and injected with hCG, was significantly higher than in untouched control rats (Table 2). Differences in the weight of the uterus were not observed.

Four days after surgery In all animals with section of the left superior ovarian nerve, the denervated ovary did not ovulate, while 71% of the innervated ovaries ovulated normally. When the animals had a section of the right nerve, hCG treatment induced ovulation in 75% of the left ovaries and in only 25% of the right ones. In those animals with bilateral section, hCG administration induced ovu-

lation in 55% of the animals (Table 3). Again, corpora lutea were not observed in the ovaries of non-ovulating animals.

The weight of the ovaries was significantly increased in sham-operated rats and rats with bilateral section, or section of the left nerve and injection of hCG (Table 2).

Effects of PMSG+hCG injection

Differences were not observed in the ovulation rate and number of ova shed between untouched control and sham-operated rats treated with PMSG and hCG immediately or 4 days after surgery,

Immediately after surgery In those rats with section of the left superior ovarian nerve, the sequential administration of PMSG+hCG induced ovulation in 66% of the left ovaries, but the number of ova shed was significantly lower than in sham-operated animals. The right ovary in these animals released more ova than did sham-operated ones. When the section was performed in the right ovary, the ovulation rate and number of ova shed by the denervated ovary (right) were significantly lower than in sham-operated animals. When the animals were submitted to bilateral nerve section, the sequential administration of PMSG+hCG elicited an ovulatory response similar to that seen in the sham-operated group (Table 4).

The increase in the ovarian weight induced by PMSG+hCG administration was higher in sham-operated rats and rats with unilateral or bilateral section of the superior ovarian nerve than in untouched control animals (Table 2).

Four days after surgery In those animals with unilateral section of the superior ovarian nerve, the injection of PMSG+hCG did not induce ovulation by the denervated ovary. In those rats with section of the right nerve, the number of ova shed by the left ovary was significantly higher than in sham-operated ones. In rats with bilateral section of the nerve, the hormonal treatment induced

Table 4 Ovulation rate (number of ovulating/number of treated) and mean $\pm$ S.E.M. number of ova shed in animals with unilateral or bilateral section of the superior ovarian nerve performed on 32-day-old rats treated with 8 iu PMSG plus 10 iu hCG immediately or 4 days later. Animals were autopsied at the first day of vaginal oestrus

Group	Immediately after surgery				Four days later			
	Ovulation rate		Number of ova shed		Ovulation rate		Number of ova shed	
	Left	Right	Left	Right	Left	Right	Left	Right
Control	10/11	11/11	4.4 $\pm$ 1.1	5.5 $\pm$ 0.8	10/11	11/11	4.4 $\pm$ 1.1	5.5 $\pm$ 0.8
Sham	8/8	7/8	7.0 $\pm$ 0.9	4.9 $\pm$ 1.0	6/7	6/7	6.5 $\pm$ 1.1	6.2 $\pm$ 0.7
Left section	6/9	9/9	1.8 $\pm$ 0.5†*	7.6 $\pm$ 0.3*	3/8†	6/8	2.5 $\pm$ 1.2*	7.2 $\pm$ 0.7
Right section	12/12	3/12†*	10.2 $\pm$ 0.6†*	0.8 $\pm$ 0.4†*	8/11	0/11	14.3 $\pm$ 2.6†*	0
Bilateral section	5/7	4/7	5.5 $\pm$ 1.1	5.2 $\pm$ 1.7	7/10	2/10†*	5.8 $\pm$ 1.7	7, 6

† $P < 0.05$ vs control group (ovulation rate by Chi square test; number of ova shed by Kruskal–Wallis test).

* $P < 0.05$ vs sham group (ovulation rate by Chi square test; number of ova shed by Kruskal–Wallis test).

ovulation in 70% of the left ovaries, but only in 20% of the right ones (Table 4). As in PMSG- or hCG-treated animals, the ovaries of the non-ovulating animals did not present fresh corpora lutea.

When the ovulatory reactivity of the right and left ovaries in rats with bilateral section of the superior ovarian nerve to different treatments was compared, a significant difference was observed: in PMSG-treated animals (immediately after surgery and 4 days after), the left ovary ovulated in 18/22 rats treated vs the right ovary in 11/22 rats ($P < 0.05$, Chi square test); when the animals were injected with PMSG+hCG, the left ovary ovulated in 12/17 rats vs 6/17 for the right one ($P < 0.01$, Chi square test); such differences were not observed in hCG-treated animals (left ovary ovulated in 9/16 rats vs 11/16 animals by the right ovary, NS).

The increase induced by PMSG+hCG administration in the weight of the ovaries of rats with uni- or bilateral section of the nerve was similar to that induced in the untouched animals, and it was higher in sham-operated ones (Table 2).

Discussion

The results obtained in the present study suggest that the innervation reaching the ovary via the superior ovarian nerve regulates, in a stimulatory way, the effects of gonadotrophin, resulting in ovulation. The results obtained by Mayerhofer *et al.* (1997) suggested that in the ovaries of 2-day-old rats, norepinephrine and vasoactive intestinal polypeptide regulate the synthesis of follicle-stimulating hormone receptors in a stimulatory way, supporting such an interpretation.

Previous reports showed that in the prepubertal rat, the bilateral superior ovarian nerve section provoked a marked decrease in ovarian norepinephrine content (Aguado & Ojeda 1984, Selstam *et al.* 1985, Chávez *et al.* 1994). This

decrease was accompanied by an almost 2-fold increase in β -adrenergic receptor number, while the affinity of receptors was similar in sham-operated rats (Aguado & Ojeda 1984). A luteinising hormone-releasing hormone content in denervated ovary was also observed by Marchetti *et al.* (1987). Such a norepinephrine content decrease did not affect spontaneous and induced ovulation, since in the rat with and without gonadotrophin stimulation, the bilateral section of the superior ovarian nerve altered neither the time of vaginal opening, nor that of the first ovulation. These results suggest that sympathetic information which travels along the suspensory ligament does not play an obligatory role in the ovulation process (Aguado & Ojeda 1984, Selstam *et al.* 1985). Another possibility is that 'the effects of bilateral section on ovulation are not the addition of the effects of left and right denervation, implying the existence of a modulatory effect in gonadotrophin action on ovulation via other neural pathways' (Morales *et al.* 1993).

Because, in the animals with bilateral section and administration of PMSG+hCG, the ovulatory response was normal compared with the sham group, we presume that the development of denervation supersensitivity by the increase in β -adrenergic receptor number (Aguado & Ojeda 1984) might compensate for the loss of neural input to the ovary. At present, we do not have a satisfactory explanation for the low ovulation rate by the right ovary observed in those rats injected with PMSG+hCG 4 days after bilateral denervation. The differences in the ovulatory response by the left and right ovary in rats with bilateral section of the superior ovarian nerve support our previous statement that the differences in ovulatory ability by the left and right ovary are related to ovarian innervation (Domínguez *et al.* 1989), because the ovaries are still receiving innervation through the ovarian plexus and the vagus nerve (Burden 1978). Another possibility is the existence of some kind of blood-borne information between the ovaries, as was proposed previously (D'Albora *et al.* 1992).

In rats with unilateral section, the sequential injection of PMSG+hCG did not induce normal ovulation by the denervated ovary, while the innervated ovary performed normally or showed superovulation. We can presume that the unilateral section of the superior ovarian nerve results in an ovarian supersensitivity; for which, therefore, the lack of ovulatory response by the denervated ovary cannot be explained in the same way. These results support the idea that there is neural communication between both ovaries (Gerendai *et al.* 1978, Domínguez *et al.* 1989, Morales *et al.* 1993), which would be via multiple pathways. When one of them is severed, the lack of information originating in this ovary induces an inhibitory signal arising from the innervated ovary reaching the partially denervated ovary through one of the other neural pathways. Such an interpretation agrees with the results of Klein *et al.* (1989) that the stimulation of the superior ovarian nerve results in an electrical signal in the ovarian plexus nerve.

When the animals were treated with PMSG or hCG to induce ovulation, it was necessary that the endogenous neuroendocrine mechanisms regulating the LH pre-ovulatory surge were working normally (Greenwald & Roy 1994). The lack of, or very low, ovulation by the denervated ovary observed in those animals with unilateral section of the superior ovarian nerve injected with PMSG, supports the interpretation that neural signals reaching the ovary through such a nerve play a role in the regulation of the reactivity of the ovary to gonadotrophins. Because, in the animals with section of the left ovarian nerve or bilateral section (in all animals the left nerve was sectioned first), PMSG did not induce full ovulation, we can presume that such neural signals, provoked in this experiment by cutting the nerve, initiated in the left ovary and sent to the right one, play an inhibitory role on the response of the right ovary to endogenous gonadotrophins, or inhibit the relay of a neural signal to the central nervous system arriving at the hypothalamus, as was proposed previously (Domínguez *et al.* 1998).

When ovulation was induced by hCG administration 4 days after surgery, the results were similar to those in the animals treated with PMSG. The lack of ovulation response to hCG administration in the denervated ovary of prepubertal rats with unilateral section of the superior ovarian nerve, is similar to the results obtained in adult rats with the same treatment (Chávez *et al.* 1991).

The differences observed between the response to the injection of PMSG or hCG immediately after surgery in animals with section of the right ovarian superior nerve could be explained by the results of Roth & Jones (1992) in the lizard, where one ovary is less sensitive to gonadotrophin administration. Another possibility is that such a variation is explained by the differences described in neuronal mechanisms involved in signal transduction for PMSG and hCG (Hsueh *et al.* 1994).

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