

François Giuliano<sup>1,2</sup>, Laurent Alexandre<sup>1</sup>, Stéphane Droupy<sup>2</sup>, Jacques Bernabé<sup>1</sup>.

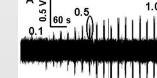
\* giuliano@cyber-sante.org

## ABSTRACT

**Introduction and Objective:** Lack of adequate and sensitive animal models to investigate the female sexual response has led to scarce knowledge of the physiology of female sexual reflexes. Here we describe an original model to study the effect of mechanical stimulation of the external genitalia on the afferent nerve activity in the L6 dorsal root, a major pathway for clitoral afferences.

**Methods:** Nine ovariectomized female rats were anesthetized (urethane, 1.2 g/kg) and placed on a stereotaxic frame. Platinum bipolar electrodes were placed on the L6 and S3 dorsal roots to record the electrical activity of spikes frequency. After laminectomy, the electrodes were placed on the clitoris and after clitoral stimulation. Mechanical stimulation of the clitoris was achieved by a stream generated by connecting a metal tube of 3 mm of diameter to a cylinder of compressed air and directed to the vulva from a distance of 2 mm. Pressure of stimulation, lasting 10 s each, was increased by 0.05 bar steps.

**Results:** Graded stimulation of the external genitalia (vulva) caused a graded increase in the afferent nerve activity detected in the L6 dorsal root. Fig. A corresponds to a typical neuron, with figures indicating stimulation at 0.1, 0.5 and 1.0 bar. The nerve activity during the stimulation at 0.5 and 1.0 bar is detailed by a quantification of spike frequency in raw recordings is given in B. As a matter of comparison, the stream of air was also applied on the anus.



**Conclusions:** This study demonstrates that afferent nerve activity recorded in L6 dorsal root is activated in response to mechanical stimulation of the external genitalia in female anesthetized rat. This may provide a unique model to study the sensory feedback from the female external genitalia to the central nervous system and will hopefully enhance our understanding of the physiology and pathophysiology of female sexual function.

## BACKGROUND

- Female genital sexual response is a complex vasculo-tissular event under neuro-endocrine control.
- Female sexual arousal responses are mainly the product of spinal reflex mechanisms, and are under descending excitatory and inhibitory control from supraspinal sites.
- Increasing genital (i.e. vaginal, labial and clitoral) blood engorgement for the treatment of female sexual dysfunctions did not appear as successful as increasing penile blood engorgement for the treatment of male sexual dysfunctions (Modelska K. and Cummings S. Female sexual dysfunction in postmenopausal women: systematic review of placebo-controlled trials. Am J Obstet Gynecol, 2003, 188, 286-293).
- In men, ability to get and maintain good erection : sufficient for feeling as being performant
- In women,
  - no such physiological performance
  - feeling of sexual arousal results more from cognitive processing of stimulus meaning and content, than from peripheral vasocongestive feedback .

Laan E et al Psychophysiology 1995;32:444-51

- discrepancies between subjective feeling of sexual arousal and the objective measurement of vaginal blood flow .

Tutten A et al Psychosom Med 1996, 58:234-41

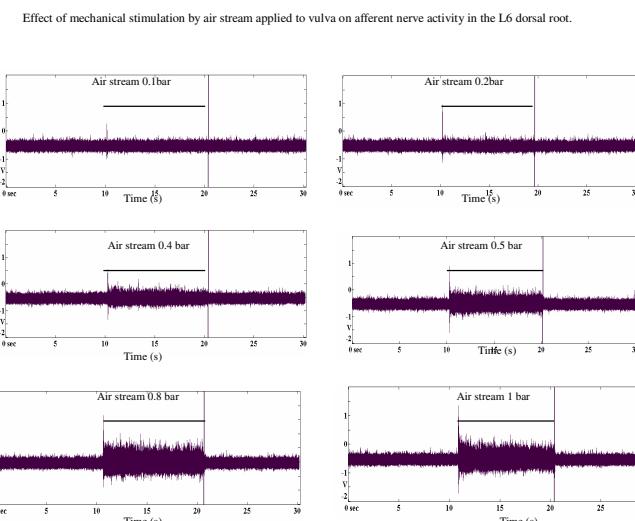
## OBJECTIVES

- >To design an experimental model to investigate the afferent innervation of the vulva which is involved in the regulation of female genital sexual response and thus link the female genital tract to the central nervous system.
- To determine the effect of mechanical stimulation of the external genitalia on afferent nerve acitivity in the L6 (equivalent of S2-S3 in humans) dorsal root in female rat.
- To compare the effect of external genitalia and anal stimulation on afferent nerve activity in the L6 dorsal root.

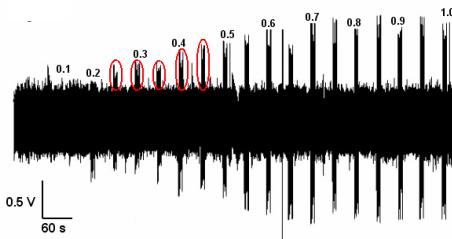
## METHODS

- Female Wistar rats (250-300g) were ovariectomized 7-10 days prior the experiment
- Rats were anesthetized with urethane (1.2g/kg) and placed on a stereotaxic frame and hold with two spinal contention forks placed on the L4 and S3 vertebrae.
- After laminectomy, platinum bipolar electrodes were placed on the L6 dorsal root.
- Mechanical stimulation of the external genitalia was performed with air stream.
- The air stream was generated by connecting a metal tube to a cylinder of compressed air and the stream was directed toward the vulva from a distance of 2 cm.
- The electrical activity on the L6 dorsal root was recorded (high pass 10KHz, low pass 300Hz, gain 10000)
- Electrical activity induced by air stream stimulation was discriminated with an amplitude window discriminator.
- Pressure stimulation was incremented by 0.05 bar step. Each stimulation lasted for 10 s.
- The frequency of spikes was determined for each pressure stimulation of vulva.
- To determine the effect of anal mechanical stimulation, the same procedure was performed with the air stream applied on the anus.

## RESULTS

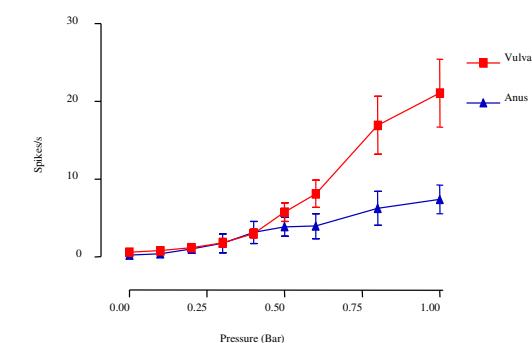


Recording of the electrical activity in the L6 dorsal root in a urethane-anesthetized female rat upon stimulation of the external genitalia of increasing intensity.



Pressure of stimulation was incremented by 0.05 bar step. Number corresponds to the air stream pressure (in bar) measured at the exit of the air cylinder for the corresponding response. Each stimulation lasts for 10 s. Only spikes of amplitude greater to the noise were quantified (e.g. red loops).

Effect of mechanical stimulation by air stream applied to vulva and anus on afferent nerve activity in the L6 dorsal root.



Neurograms obtained in 9 ovariectomized female rats were quantified and averaged. This quantification revealed a correlation between the strength of the stimulation (airstream) applied on the external genitalia (vulva) and the frequency of the spikes detected in the L6 dorsal root. As a matter of comparison, the air stream was also applied on the anus.

## CONCLUSION

- These experiments represent a preliminary attempt to investigate the afferent limb of the spinal reflex involved in genital sexual response in females.
- Afferent nerve activity recorded in L6 dorsal root is activated by mechanical stimulation of the external genitalia in female anesthetized rats.
- Further characterization of L6 dorsal root single afferent fibers innervating the vulva is mandatory. This will imply to study the responses of these afferent fibers to mechanical and thermal stimulation of the vulva
- Whether the vulvar sensory stimulation elicits a spinal reflex resulting in clitoral, labial and vaginal engorgement requires further investigation.