Histone deacetylase gene expression is regulated by sex chromosome complement in the amygdala of the developing mouse brain

Camila Sosa[1], Carla D. Cisternas[1, 2, 3], Romina Conti[2], María J. Cambiasso[1, 2]

[1]Instituto de Investigación Médica M y M Ferreyra, INIMEC-CONICET-UNC, Córdoba, Argentina.
[2]Cátedra de Biología Celular, Facultad de Odontología, Universidad Nacional de Córdoba, Argentina.
[3] Cátedra de Fisiología Animal, Facultad de Ciencias Exactas, Físicas y Naturales, Universidad Nacional de Córdoba, Argentina.

INTRODUCTION

- Although the role of gonadal hormones is undeniable, several studies have shown that sex chromosomes have gonadal-independent effects on sexually dimorphic gene expression in the mouse brain. [1,2,3]
- Both, hormonal and genetic factors contribute to regulate gene expression in developing amygdala neurons before the critical period. [4]
- Epigenetic mechanisms, such as histone modifications, have recently been proposed as mediators of sexual differentiation of the rodent brain. [5 6 7]
- Histone deacetylation implicates the action of histones deacetylases (Hdacs) which main function is to remove acetyl groups from histones, causing condensation of chromatin and usually leads to decrease gene expression.[8]
- It has been proposed that Hdac activity during the early postnatal period plays a crucial role in brain sexual differentiation via modifications of histone acetylation status.[9]

Aim

To analyze the contribution of the sex chromosome complement on the gene expression of the Hdacs in the mouse amygdala.

Hypothesis

The sex chromosomes determine a differential expression of Hdacs before the critical period of sexual differentiation in the amygdala of developing mouse brain.

MATERIALS AND METHODS

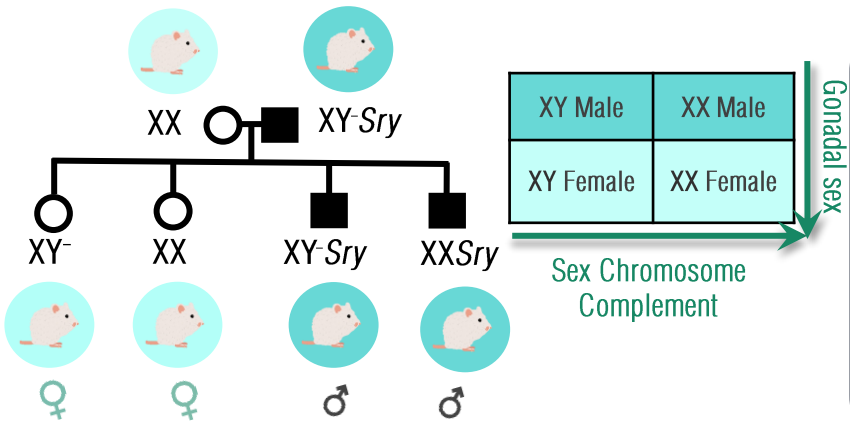

Figure 1. Four Core Genotypes mouse model

We used the "Four Core Genotypes" mouse model, which allows the evaluation of gonadal sex, sex chromosome complement, and their interaction (Figure 1). [10]

Amygdala tissue were obtained from E15 mice separately according to the sex and genotype. The brain was dissected out and the meninges were removed. The amygdala region was transversally sliced between the optic chiasma and the mammilary bodies.

RNA was extracted from amygdala of untreated E15 mice. cDNA was obtained by reverse transcription and Real-Time PCR was performed using SYBR® Green PCR Master Mix for *Hdac1, 2, 8, 4 and 6.* Relative mRNA expression level was calculated using *Gapdh* as housekeeping gene with the ΔΔCt method.

RESULTS

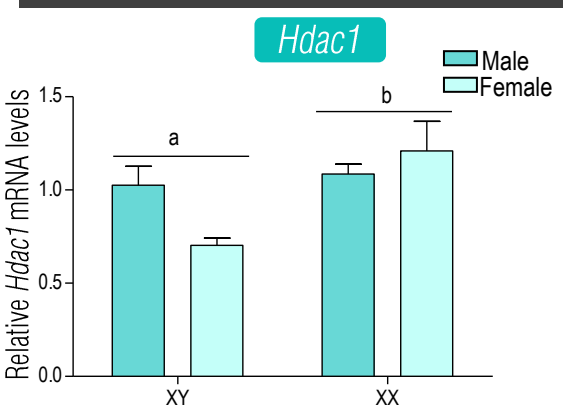

Figure 2. *Hdac1* mRNA levels in the amygdala of E15 mice. ANOVA:F (1,13)=7.52; p=0.016. Different letters indicate significant differences with $p < 0.05$.

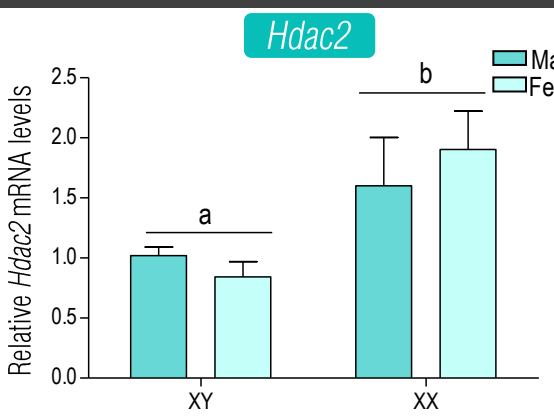

Figure 3. *Hdac2* mRNA levels in the amygdala of E15 mice. ANOVA:F(1,20)=12.93; p=0,0018. Different letters indicate significant differences with $p < 0.05$.

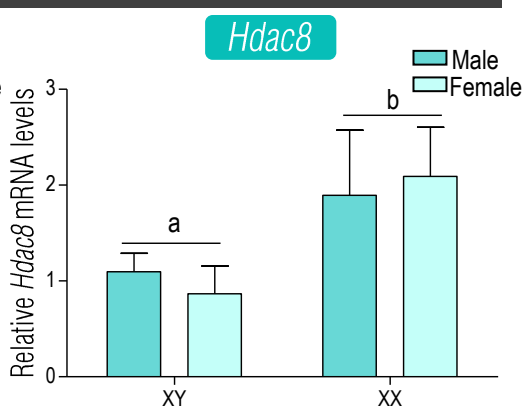

Figure 4. *Hdac8* mRNA levels in the amygdala of E15 mice. ANOVA:F (1,19)=4.77; p=0.04. Different letters indicate significant differences with $p < 0.05$.

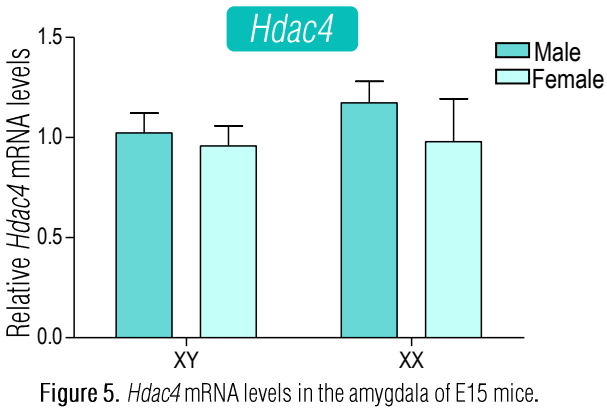

Figure 5. *Hdac4* mRNA levels in the amygdala of E15 mice.

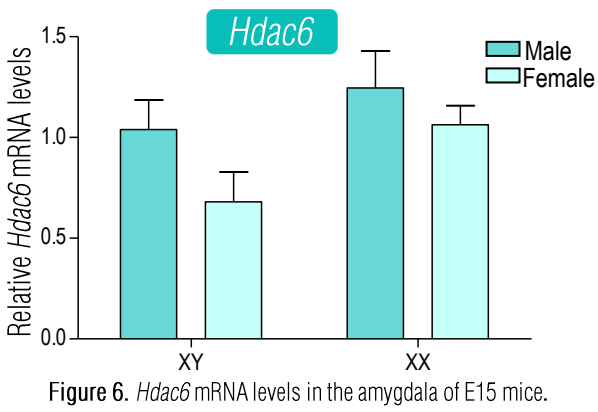

Figure 6. *Hdac6* mRNA levels in the amygdala of E15 mice.

CONCLUSIONS

- Class I Hdacs, *Hdac1, 2* and *8* expression levels were higher in amygdala derived from XX embryos compared to XY, irrespectively of gonadal type.
- No differences were observed in Class II Hdacs *Hdac4* and *6.*

In summary, these results suggest that sex chromosome complement might determine a higher "histone deacetylation" by Class I Hdac in specific areas of the XX brain.

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CONICET INSTITUTO FERREYRA UNC Universidad Nacional de Córdoba

I N I M E C

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