The effects of the aromatase inhibitor, Letrozole, on lithium chloride (LiCl)-induced conditioned disgust behaviour (anticipatory nausea) in male rats

Vangel Matic, Indra R. Bishnoi[1,2], Klaus-Peter Ossenkopp[1,2], Martin Kavaliers[1,2]

[1]Graduate Program in Neuroscience, Western University, London, Ontario, Canada [2]Department of Psychology, Western University, London, Ontario, Canada

INTRODUCTION

- **Anticipatory Nausea** (AN) is a form of classical conditioning in which the effects of a nausea-inducing substance become associated with a specific environmental context and is suggested to have a higher incidence in females than males[1,2,3].
- AN can be represented by the occurrence of **conditioned gaping behavior** in rats (Fig. 1), displayed upon re-exposure to a context previously associated with a nausea-inducing toxin such as lithium chloride (LiCl)[4,5,6].
- **Estrogen** is suggested to be involved in mediating the female-biased sex difference in AN through its enhancement of hippocampal-dependent spatial memory[7,8,9].
- **Aromatase** is an enzyme involved in the synthesis of estrogens from androgens in estrogen-producing tissues in the body[10].
- **Purpose:** Investigate the role of letrozole, an aromatase inhibitor, in the learning of conditioned disgust
- **Hypothesis:** For rats conditioned with LiCl in a novel context, daily administration of letrozole for 10 days prior to LiCl-free context re-exposure will significantly reduce the frequency of conditioned gaping behaviour compared to controls.

METHODS

Animals
- 20 adult male Long-Evans rats

Conditioning Drugs
- Lithium chloride (LiCl) (128 mg/kg, 20 ml/kg)
- 0.9% NaCl (20 ml/kg)

Treatment
- Letrozole (Let) (1 mg/kg, 10 ml/kg)
- Vehicle (Veh) 0.9% NaCl 5% Ethanol, 5% Tween-80 (10 ml/kg)

Day 1	Day 2 CND 1	Day 3	Day 4 CND 2	Day 5	Day 6 CND 3	Day 7	Day 8 CND 4	Day 9	Day 10 EXT
Let or Veh	Let or Veh	Let or Veh	Let or Veh	Let or Veh	Let or Veh	Let or Veh	Let or Veh	Let or Veh	Let or Veh

Figure 3. Drug Administration Timeline

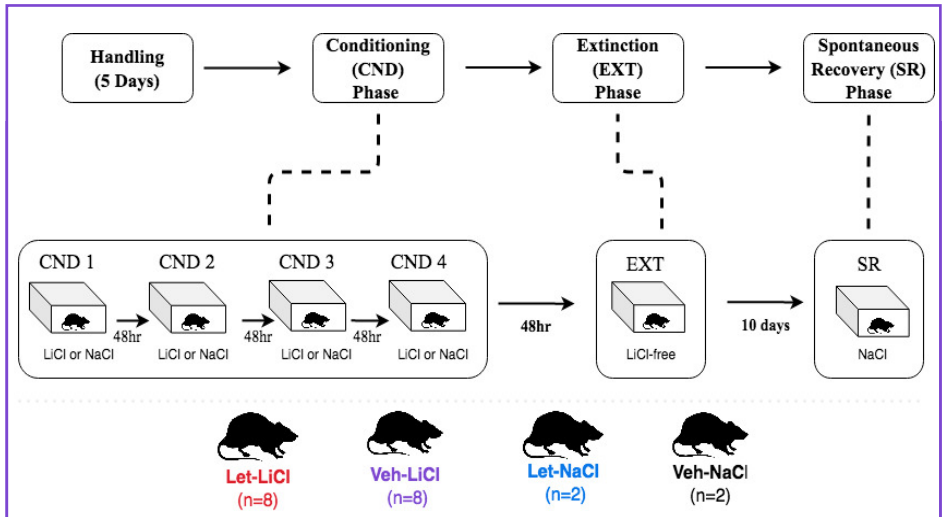

Figure 4. Experimental Procedure.

Figure 1. Gaping

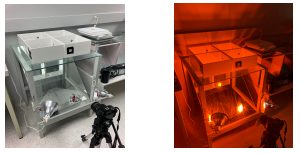

Figure 2. Conditioning Apparatus

DISCUSSION

- **Contrary to the hypothesis**, it was found that **letrozole administration did not significantly reduce the frequency of conditioned gaping behaviour** compared to controls.
- Letrozole administration in LiCl-conditioned rats showed a **non-significant enhancement of AN learning** (p = 0.06) compared to vehicle, suggesting that aromatase inhibition and decrease in estrogen and/or increase in testosterone may improve the learning of AN.
- **Limitations** of the present study include: i) the small sample size of NaCl control groups (n = 2), ii) the use of only one dose of letrozole (1 mg/kg), and iii) no measurement of testosterone or estrogen levels.
- Given the small sample size, the enhanced AN learning is marginally significant and **requires further exploration.**

RESULTS

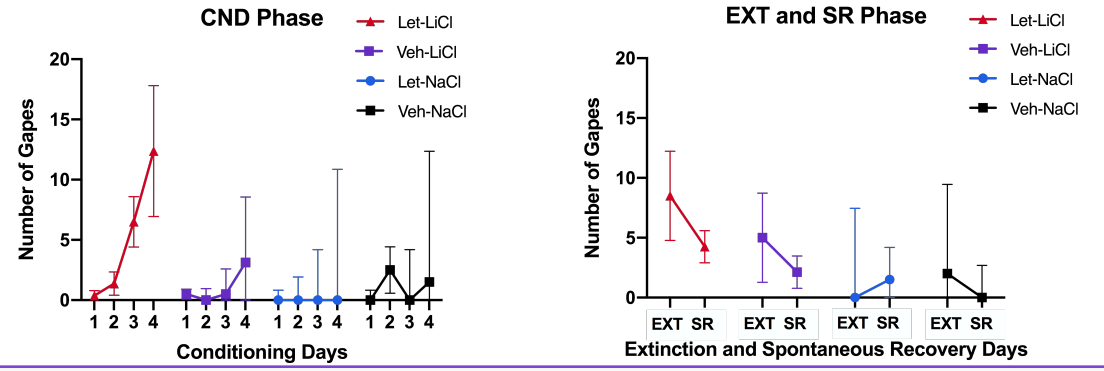

Figure 5. Mean (± S.E.M.) frequency of scored gaping behaviour of four groups [Let-LiCl (n=8), Veh-LiCl (n=8), Let-NaCl (n=2), Veh-NaCl (n=2)] through the conditioning (CND), extinction (EXT) and spontaneous recovery (SR) phases of the experiment. Let-LiCl group displayed a greater frequency of gaping behaviour on CND 3 (p = 0.06), CND 4 (p = 0.246), EXT (p = 0.323), and SR (p = 0.176), compared to other groups

References

[1]Matteson, S., Roscoe, J., Hickok, J., & Morrow, G. R. (2002). The role of behavioral conditioning in the development of nausea. American Journal of Obstetrics and Gynecology, 186(5, Supplement 2), S239–S243. https://doi.org/10.1067/mob.2002.122597
[2]Roscoe, J. A., Morrow, G. R., Aapro, M. S., Molassiotis, A., & Olver, I. (2011). Anticipatory Nausea and Vomiting. Supportive Care in Cancer, 19(10), 1533–1538. https://doi.org/10.1007/s00520-010-0980-0
[3]*Hilarius, D. L., Kloeg, P. H., van der Wall, E., van den Heuvel, J. J. G., Gundy, C. M., & Aaronson, N. K. (2012). Chemotherapy-induced nausea and vomiting in daily clinical practice: A community hospital-based study. Supportive Care in Cancer, 20(1), 107–117.*
[4]Limebeer, C. L., Hall, G., & Parker, L. A. (2006). Exposure to a lithium-paired context elicits gaping in rats: A model of anticipatory nausea. Physiology & Behavior, 88(4), 398–403. https://doi.org/10.1016/j.physbeh.2006.04.014
[5]Parker, L. A., Rana, S. A., & Limebeer, C. L. (2008). Conditioned nausea in rats: Assessment by conditioned disgust reactions, rather than conditioned taste avoidance. Canadian Journal of Experimental Psychology/Revue Canadienne de Psychologie Expérimentale, 62(3), 198–209. http://dx.doi.org.proxy1.lib.uwo.ca/10.1037/a0012531
[6]Limebeer, C. L., Krohn, J. P., Cross-Mellor, S., Litt, D. E., Ossenkopp, K.-P., & Parker, L. A. (2008). Exposure to a context previously associated with nausea elicits conditioned gaping in rats: A model of anticipatory nausea. Behavioural Brain Research, 187(1), 33–40. https://doi.org/10.1016/j.bbr.2007.08.024
[7]Cloutier, C. J., Zevy, D. L., Kavaliers, M., & Ossenkopp, K.-P. (2018). Conditioned disgust in rats (anticipatory nausea) to a context paired with the effects of the toxin LiCl: Influence of sex and the estrous cycle. Pharmacology Biochemistry and Behavior, 173, 51–57.
[8]Sandstrom, N. J., & Williams, C. L. (2004). Spatial memory retention is enhanced by acute and continuous estradiol replacement. Hormones and Behavior, 45(2), 128–135.
[9]Kranjac, D., McLinden, K. A., Deodati, L. E., Papini, M. R., Chumley, M. J., & Boehm, G. W. (2012). Peripheral bacterial endotoxin administration triggers both memory consolidation and reconsolidation deficits in mice. Brain, Behavior, and Immunity, 26(1), 109–121.
[10]Kokras, N., Pastromas, N., Papasava, D., de Bournonville, C., Cornil, C. A., & Dalla, C. (2018). Sex differences in behavioral and neurochemical effects of gonadectomy and aromatase inhibition in rats. Psychoneuroendocrinology, 87, 93–107.