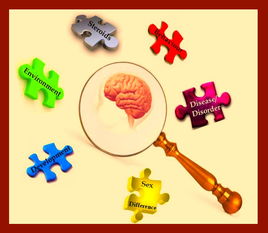

Tilting the axis: Devising a conceptual model for early life steroid deprivation and HPA axis programming in rodents

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Introduction

Objective

- To conceptualize a working model that will be employed to evaluate the effects early life alterations in steroid exposure in select brain areas on development of the hypothalamic-pituitary-adrenal (HPA) response to stress in a sex-dependent manner.
- **Brain areas of interest:** Hypothalamus-preoptic area (HPOA), amygdala (AMYG), hippocampus (HPC), and frontal cortex (FC).

Background

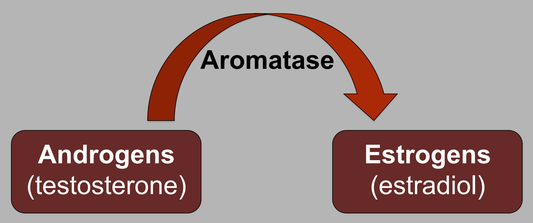
- In rodents, brain sensitivity to steroids is highest during a critical period of central nervous system (CNS) development, which extends from the 18th day of gestation to the 11th postnatal day (PND 11).[14]

Estradiol

- The initial mode of testosterone (T) action may involve its conversion to estradiol in discrete brain regions.[6]
- Estradiol receptors in the neonatal rat brain may mediate developmental effects of estrogen, as well as those of T.[14]

Aromatase activity (AA)

- In both the adult and perinatal rat brain, the aromatase enzyme is phylogenetically conserved in its ability to catalyze the conversion of androgens (testosterone) to estrogens.[6,11,13]



Best method to inhibit AA involves MS

- Maternal separation (MS) involves deprivation of feeding and warmth (physiological components) + maternal attention (psychological component).[4,5]
- It has been suggested that MS results in a compromised ability to cope with stressors.[8,9]
- MS during early postnatal development leads to an enduring dysregulation of HPA axis reactivity to stress.[2,3,10]

Impact of MS manipulation on AA

- Repeated separations may lead to permanent changes in brain development, as evidenced by behavioral, neurochemical and immunological abnormalities.[10]

Importance of estradiol and AA

- **Brain aromatase hypothesis:** The conversion of androgens to estrogens in or around target cells may account for specific neuroendocrine functions, such as brain differentiation.[6]
- More specifically, this process is important in the differentiation of neural structures in the HPOA and AMYG.[14] There is a greater distribution of AA in the hypothalamic and limbic nuclei.[11,12]
- Androgen-derived estrogens play a physiological role in the organizational actions of T on developing neurocircuitry and are necessary in adult male rats for the activation of sexual behavior.[1]

Methodology

I) Literature search

- **Online search was limited to the following MeSH terms:**
  - Aromatase
  - Brain/differentiation
  - Brain/estrogens; Brain/estradiol
  - Brain/androgens; Brain/testosterone
  - Rats/stress; Maternal separation
  - Rats/sex characteristics

Methodology

- **Several search engines and databases were consulted:**
  - PubMed
  - SCOPUS
  - ScienceDirect
  - Scirus

II) Fresh brain dissections

Image 1: Ventral view

Image 2: 1mm separates blades

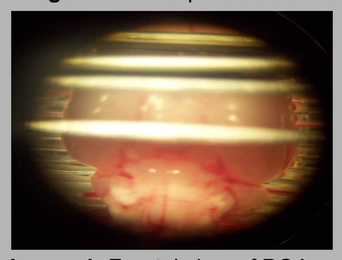


Image 3: Frontal view of AMYG

Image 4: Frontal view of POA

Results

Critical parameters of MS paradigm

1. **Frequency: MS on consecutive days (DMS)**
   Repeated MS based on a daily + predictable procedure.[10]
2. **Duration: 3 h**
   A minimum of 2 hours (h) is needed to induce an immediate effect on HPA responsiveness.[10]
3. **Age at separation: PND 1-5**
   Based on findings in early handling (EH) studies, the earlier the manipulation is carried out, the stronger the effects will be.[10]

Additional parameters

- **Litter size: Approximately 10 pups per dam (10 dams in total)**
- **Litter sex composition: Equal ratios of males to females**
  Male and female pups are exposed early in infancy to a different pattern of maternal behaviour.[10] In addition, males are more susceptible to early environmental disturbances.[10]
- **Cross-fostering: Perform on PND 1**
  Random redistribution of pups among dams eliminates possible effects of genetics, prenatal factors and maternal behaviour.[10]

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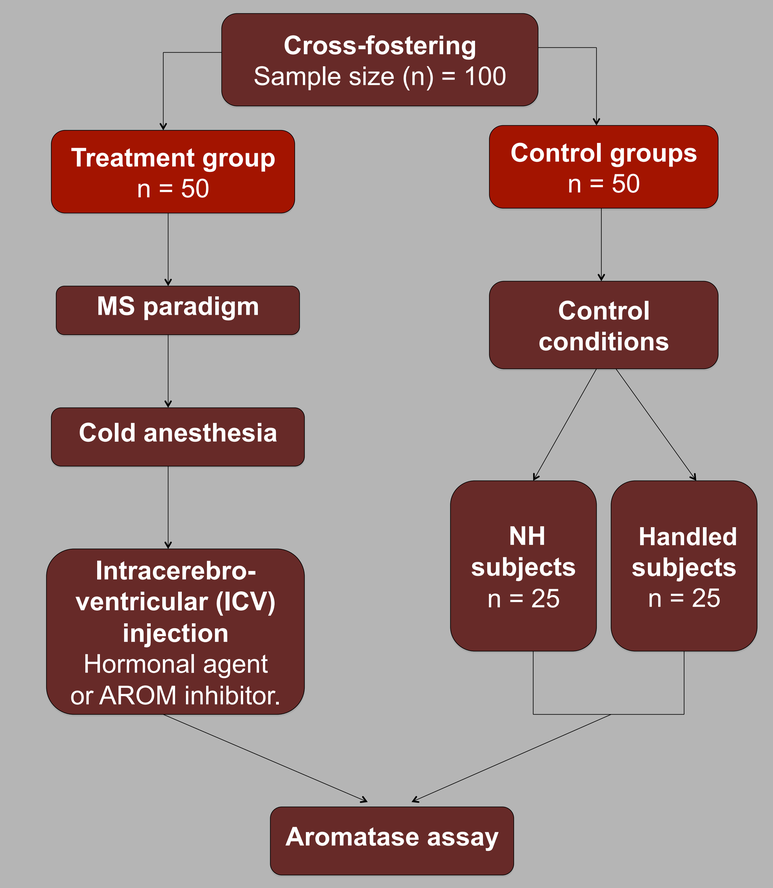
Results: Working model



Note:

- Behavioural tests will be administered using an independent sample of periadolescent rats undergoing a similar treatment paradigm.

Testing methods:

1. Open field paradigm
2. Elevated plus maze paradigm

Conclusion

Advanced search techniques

1. Craft a clear and concise description of the topic of interest.
2. Employ a wide range of relevant databases.
3. Employ a wide range of relevant search terms.
   - Appropriate use of synonyms.
4. Employ a wide range of appropriate search techniques.
   - Alternative search operations.
5. Record references in an accurate and consistent manner.
   - Keep a record of each search.
6. Identify gaps in the literature.

Study limitations

I) Statistical power of total sample size.
II) Variability of patterns in maternal response[10]:
- Frequency + duration of nursing
- Licking
- Nest-building
- Other activities: Eating, drinking, sniffing, grooming, and undirected locomotor activity.

III) Accessibility of apparatus necessary for conducting the aromatase assay.

Future directions

- Funding has recently been approved for conducting the aromatase assay.
- Following ethics approval, this conceptual model will be carried out in our laboratory setting.