



# Method to Measure Free Corticosterone in Mouse Serum

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## 1. Introduction

- Cortisol/Corticosterone (CORT)
  - Steroid “stress” hormones
  - Dysregulation -> Brain disorders
    - ex. anxiety = high CORT
- Corticosteroid-binding globulin (CBG)
  - Transport protein
  - Binds ~90% CORT at baseline
  - Only free CORT enters cells = active
  - Brain disorders and free CORT?
    - ex. anxiety -> changes in free CORT?
- Research often only assesses total CORT
  - Lack method to measure free CORT
- Aim:
  - To develop an accurate and reliable method of separating free from bound CORT in mouse serum using ultrafiltration

## 2. Methods

### Ultrafiltration Filters

- Separate free from bound molecules

### Spike

- An added known amount of free CORT

### Liquid chromatography-tandem mass spectrometry (LC-MS/MS)

- Detects CORT in samples using particle weight and charge

### CORT Enzyme Linked Immunosorbent Assay (ELISA)

- Detects CORT in samples using antibodies

### Experiment 1

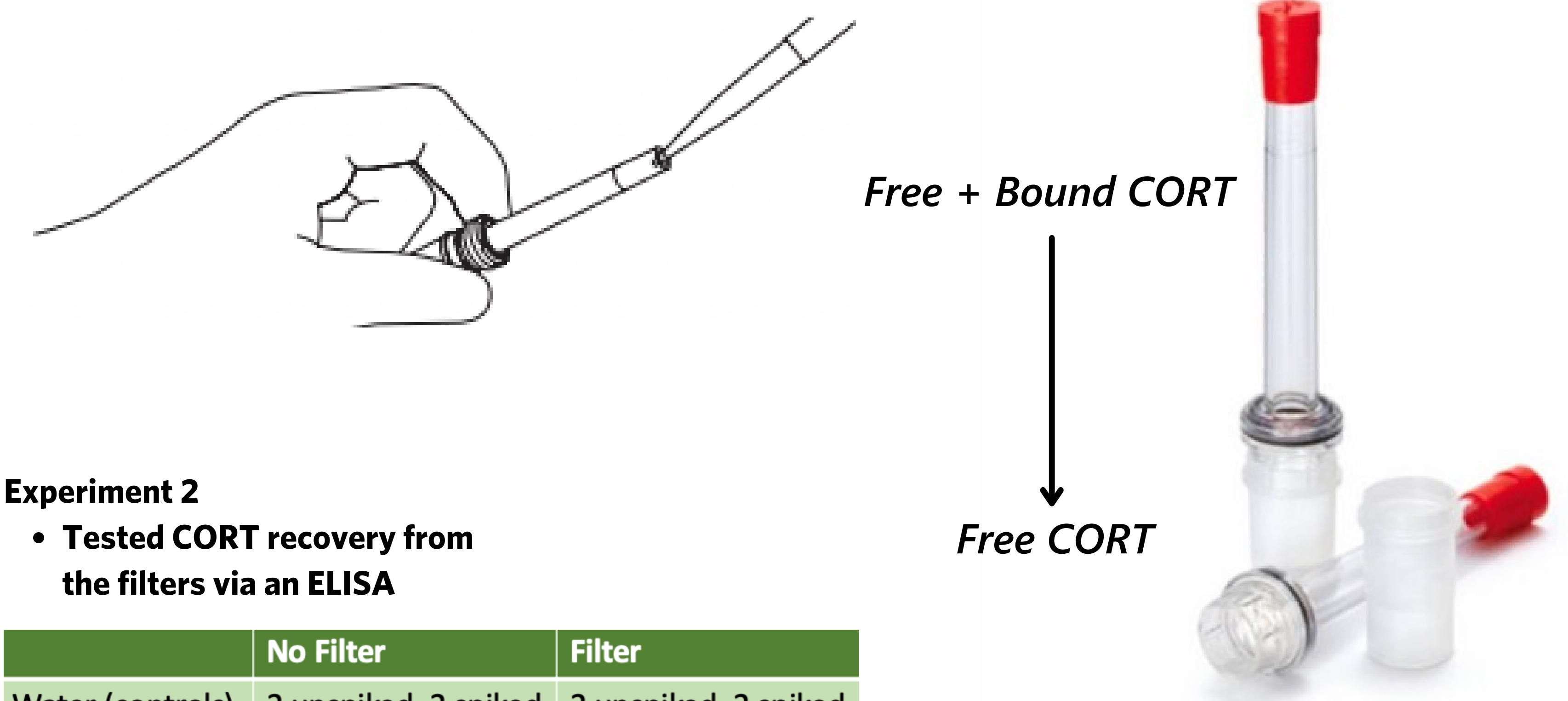
- Tested CORT recovery from the filters via LC-MS/MS

	No Filter	Filter
Water (controls)	3 unspiked, 3 spiked	3 unspiked, 3 spiked
Serum	5 unspiked, 5 spiked	5 unspiked, 5 spiked

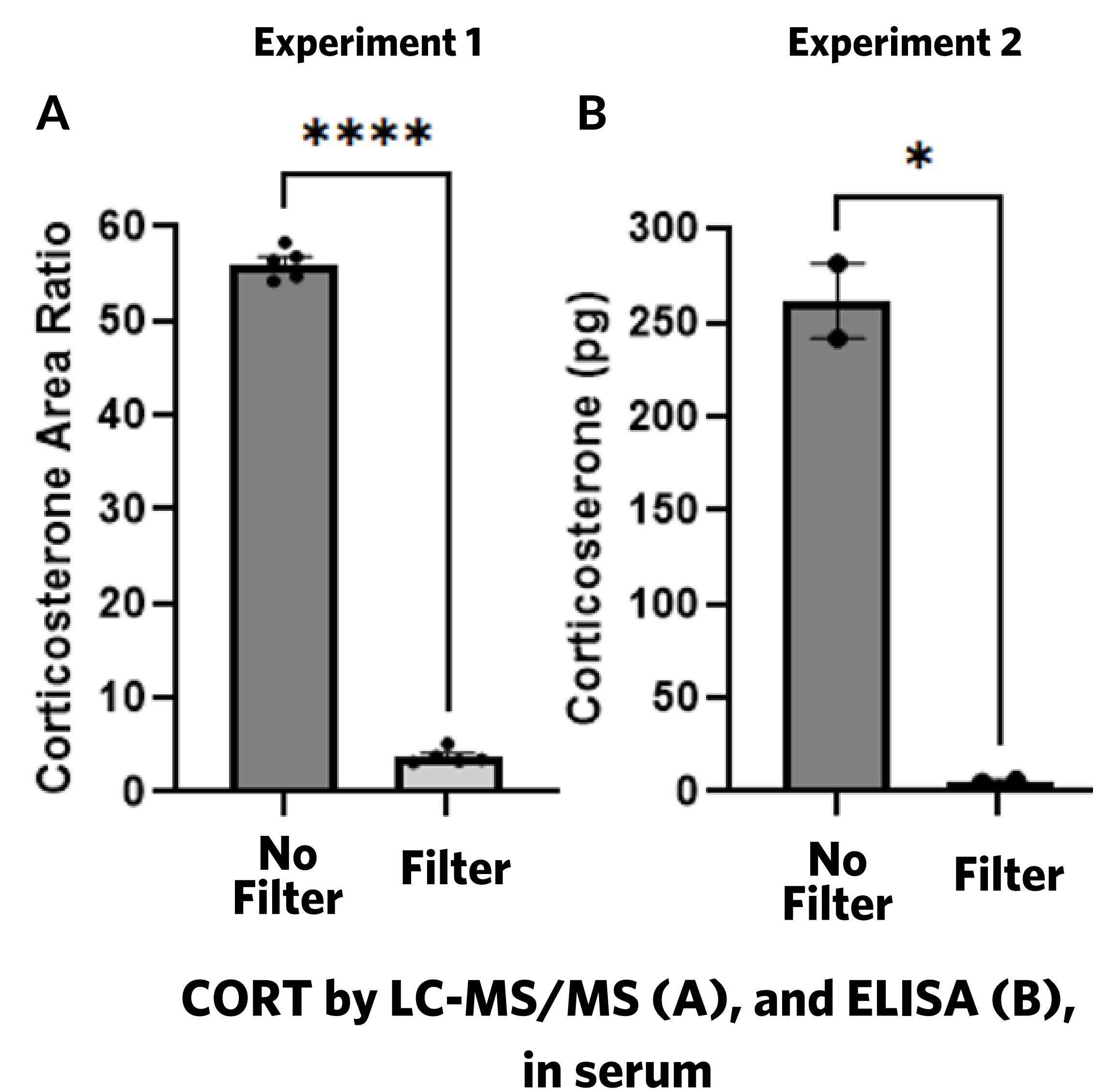
### Experiment 2

- Tested CORT recovery from the filters via an ELISA

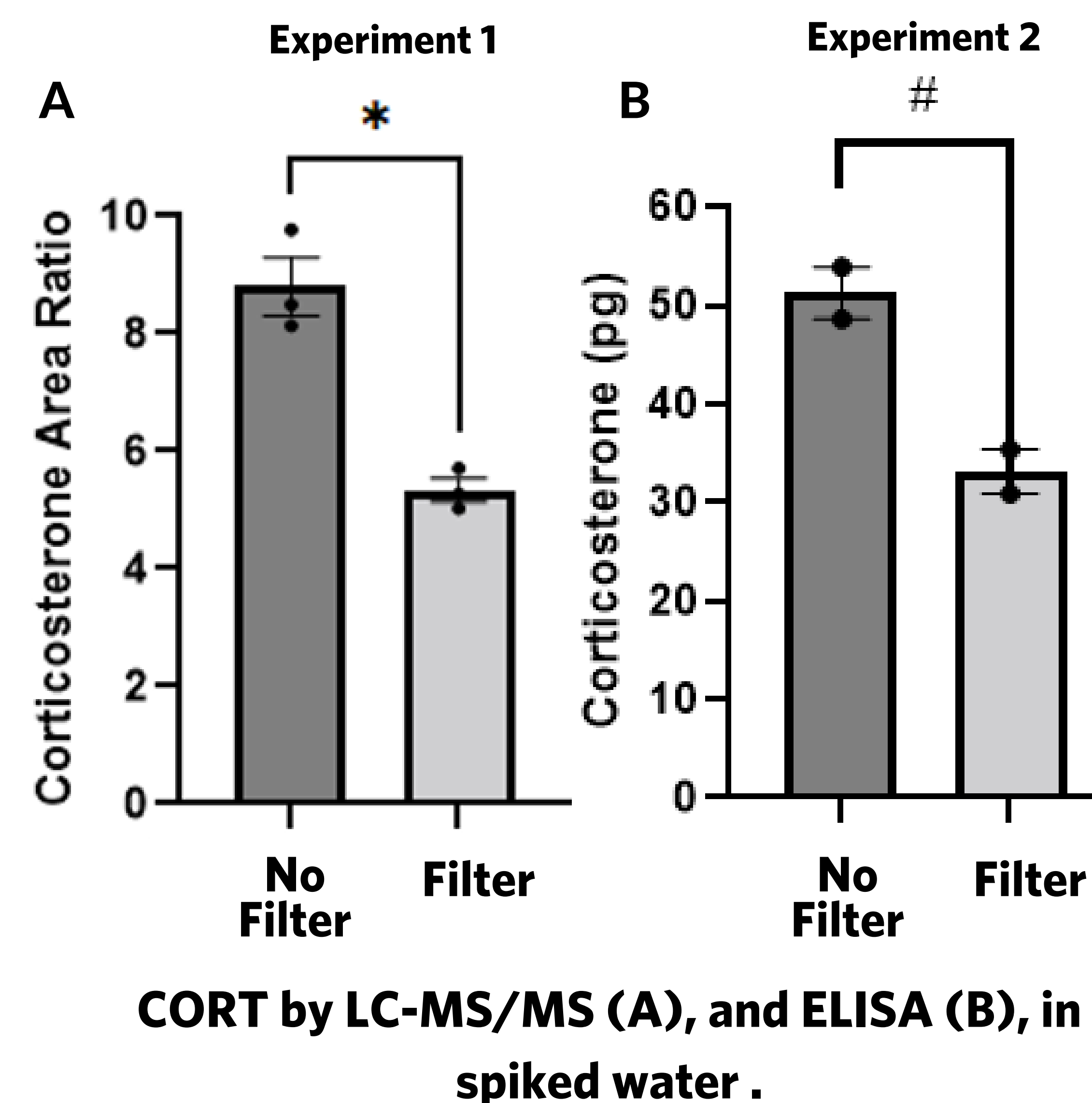
	No Filter	Filter
Water (controls)	2 unspiked, 2 spiked	2 unspiked, 2 spiked
Serum	2 unspiked, 2 spiked	2 unspiked, 2 spiked



## 3. Results



\*\*\*\* =  $p < 0.0001$ , \* =  $p < 0.05$



\* =  $p < 0.05$ , # =  $p < 0.1$

## 4. Discussion

- The CORT in filtered serum is significantly less (about 10%) than CORT of un-filtered serum
  - As expected (~90% CORT bound to CBG)
- Some CORT is being lost in spiked water samples going through the filters
  - CORT is being left behind in the filter with the current protocol

## 5. Conclusions

- Potential for ultrafiltration filters to separate free from bound hormones
  - Need to refine filtration method and quantifying CORT
- If we can validate a method:
  - Novel ultrafiltration method may be key in better revealing role of bound and free hormones on brain function

## 6. Acknowledgements

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- Lab site: <https://somalab.psych.ubc.ca/>
- UBC Honours BNS program
- My email: [annamaz@student.ubc.ca](mailto:annamaz@student.ubc.ca)

