## Sexual dimorphism of brain region volumes varies by season in ground squirrels. Andrew N. Iwaniuk\*, Robin J. Keeley, Danielle K. Burger, Deborah M. Saucier

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## Introduction

The brains of small mammals vary with both season and sex. Seasonally, many small mammals undergo significant changes in overall brain size such that it is smaller in fall/winter than in spring. Sex differences in the size of individual brain regions also occur. Generally, males have larger hippocampal volumes than females, although this does vary among rodent species. Despite these marked differences in brain size according to season and hippocampal (and some other brain region) volumes between the sexes, there are few studies that have tested for seasonal and sex differences in the brain. In fact, there are only two such studies on hippocampal volumes and no studies that we are aware of have tested for seasonal and sex differences in the volumes of non-hippocampal brain regions.. Here, we test for sexual dimorphism across several brain regions in a wild rodent, Richardson's ground squirrel (*Urocitellus richardsonii*).

## Richardson's Ground Squirrel

A polygyamous, social ground squirrel species that lives in matrilineal kinship groups. 32 wild squirrels were live trapped at their burrows during breeding (Feb-Apr) and non-breeding (Jul-Sep) seasons with 8 individuals of each sex for each season. All squirrels were deeply anaesthetised with sodium pentobarbital and transcardially perfused with 0.9% saline followed by 4% buffered paraformaldehyde. All brains were gelatin embedded, serially sectioned at 40 microns on a freezing stage microtome and every fourth series mounted onto gelatinized slides and stained with thionin.



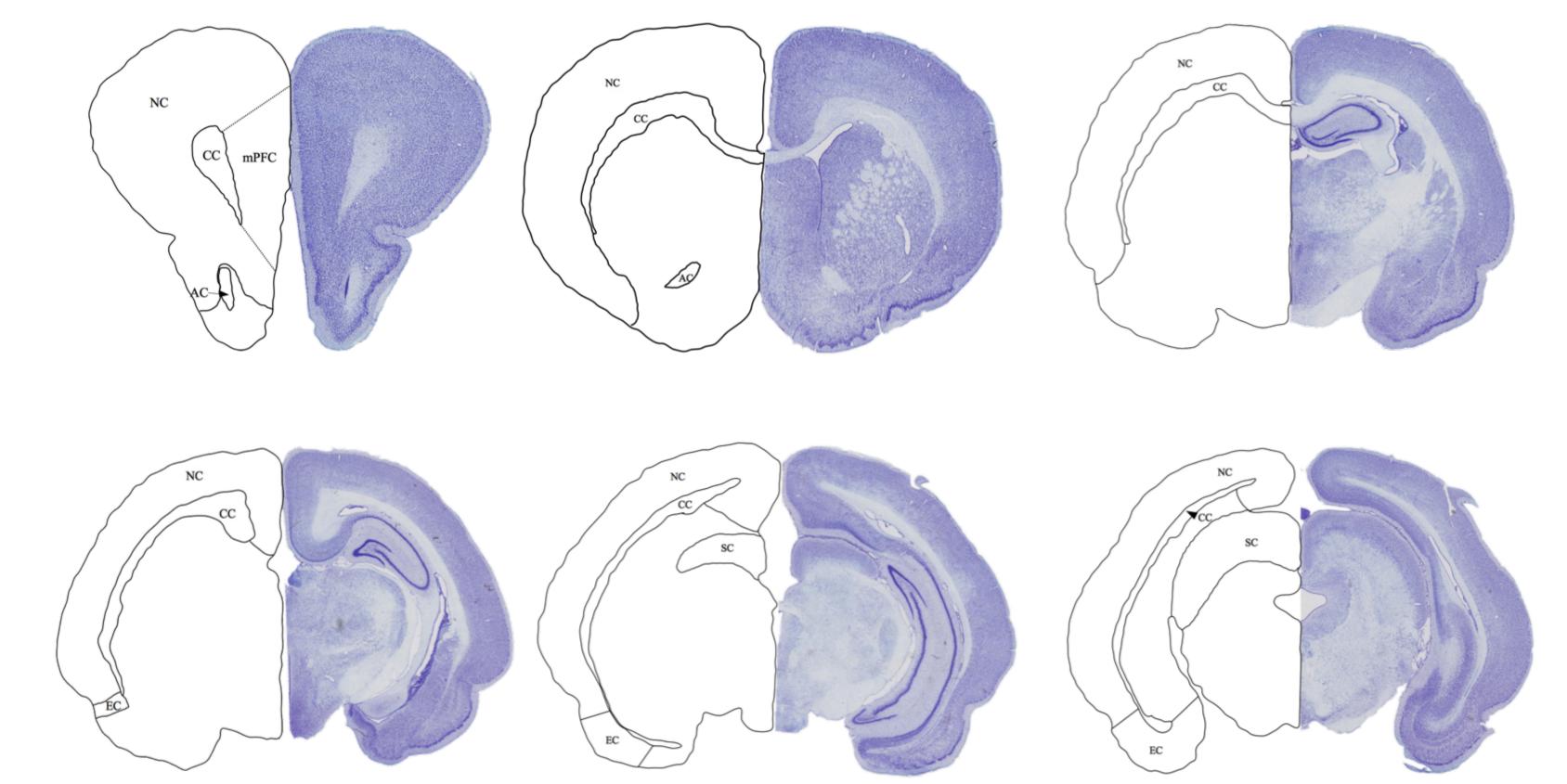
**Fig. 1** Adult Richardson's ground squirrel.

Fig. 2. Field site for ground squirrel colonies in the foothills.



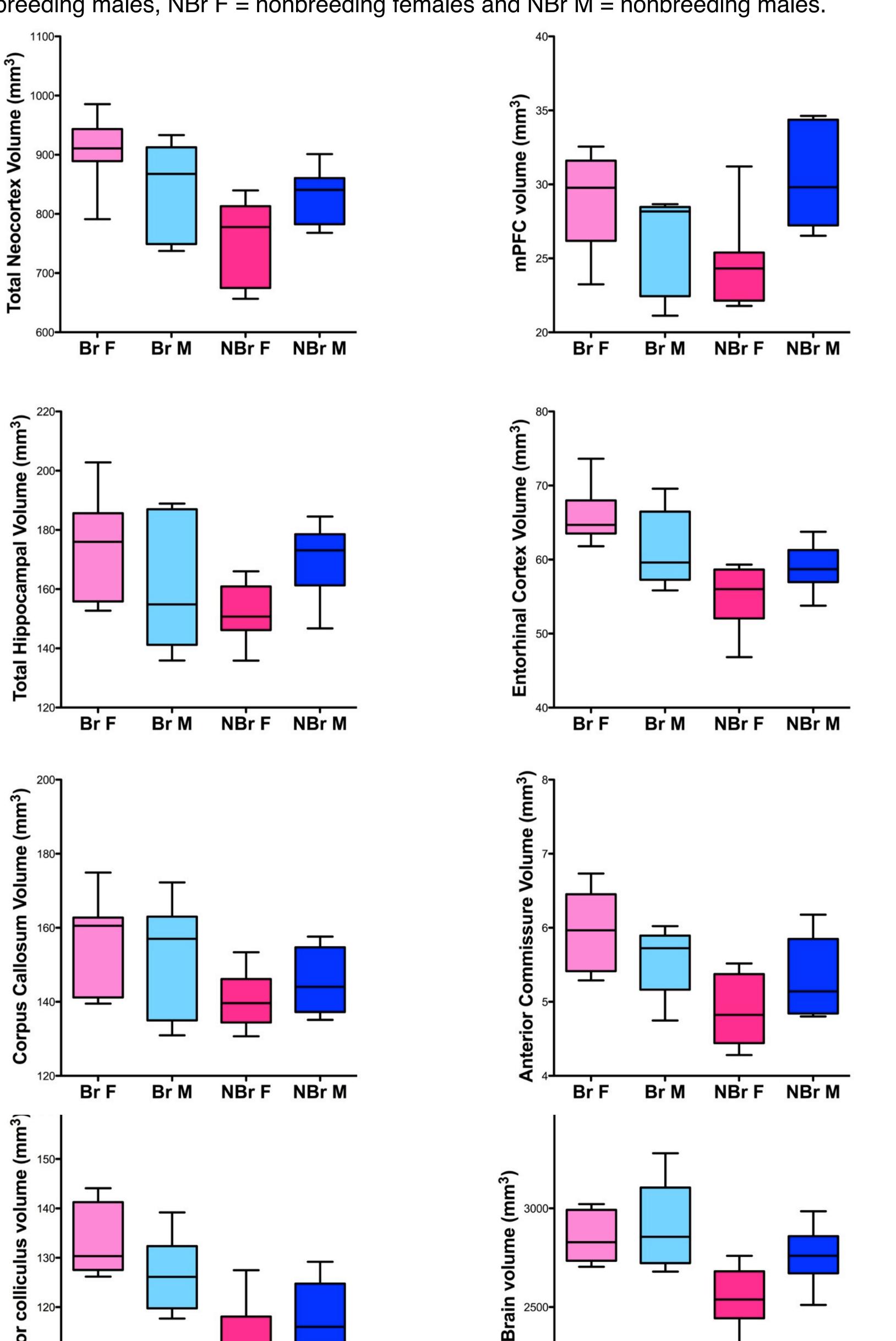
## Measurements

The volumes of 7 brain regions were measured with the Cavalieri method, as implemented in Stereoinvestigator. We predicted that total hippocampus and entorhinal cortex would exhibit sex and seasonal differences and corpus callosum to differ between the sexes, but we had no *a priori* predictions about the remaining regions.



**Fig. 3.** Line drawings (left) and Nissl stained sections (right) of ground squirrel brains with the brain regions that we measured indicated as follows: AC = anterior commissure, CC = corpus callosum, EC = entorhinal cortex, mPFC = medial prefrontal cortex, NC = neocortex, SC = superior colliculus.

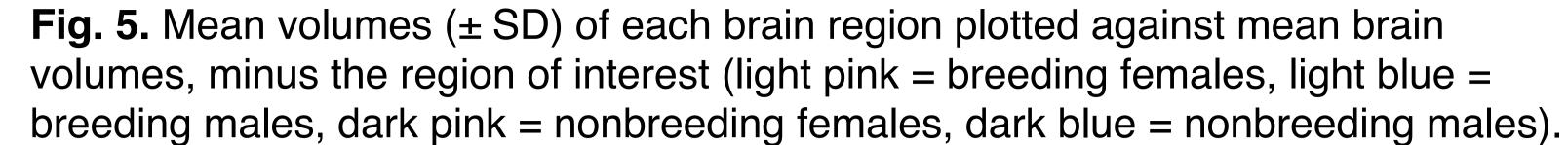
**Fig. 4.** Absolute volumes for the entire brain and all 7 brain regions shown as boxplots (min/max). Abbreviated group names are: Br F = breeding females, Br M = breeding males, NBr F = nonbreeding females and NBr M = nonbreeding males.

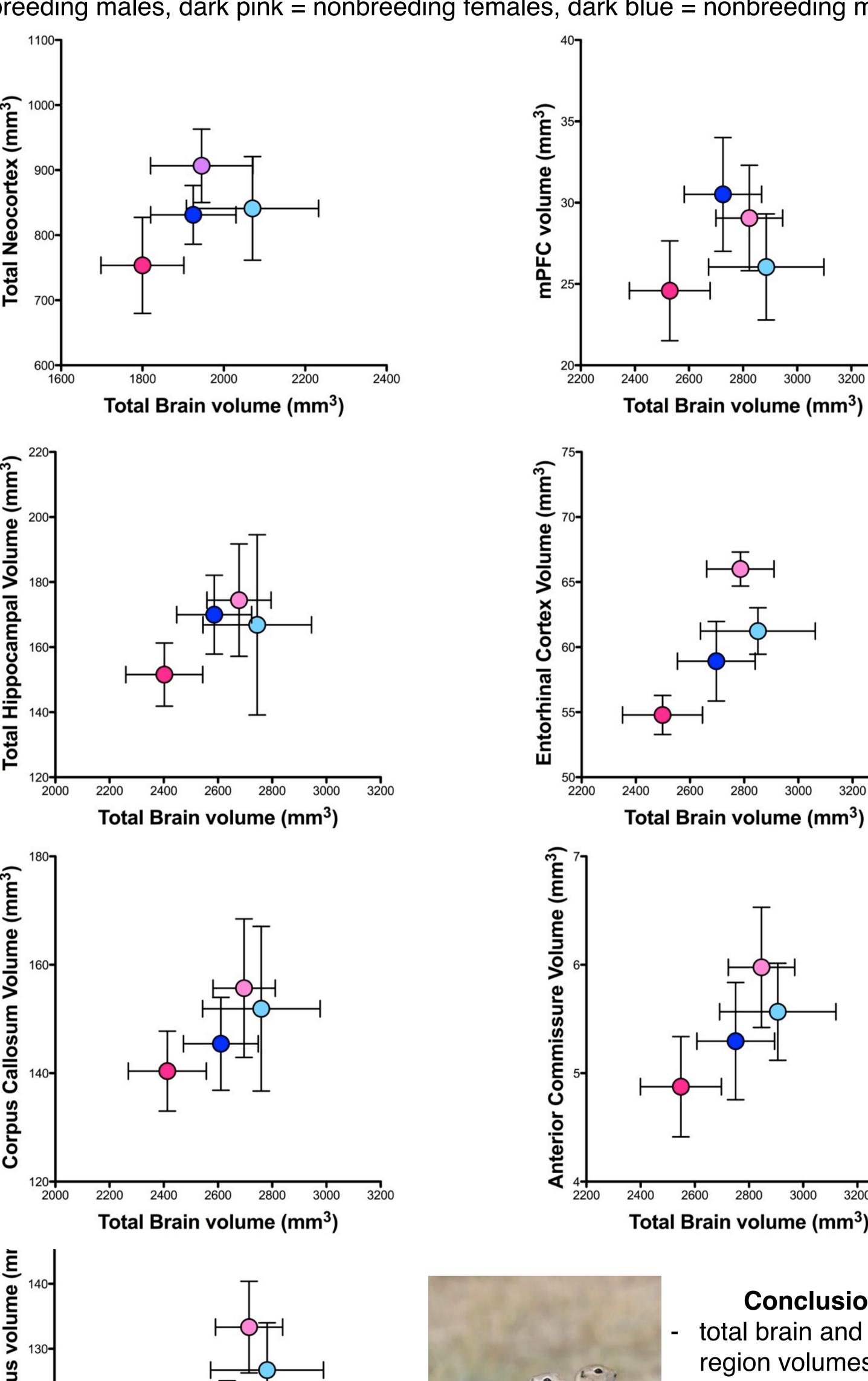


Overall, breeding females had significantly larger brain region volumes than nonbreeding females, except for the corpus callosum (only an overall effect of season). Brain volume was also significantly larger in breeding than non-breeding squirrels and in males than in females.

BrM NBrF NBrM

BrM NBrF NBrM





Conclusion
 total brain and brain region volumes vary by season, but only in females.
 this likely reflects sex

- this likely reflects sex differences in growth patterns.

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Total Brain volume (mm<sup>3</sup>)

