Published in final edited form as:

Neurosci Lett. 2011 January 7; 487(2): 177–181. doi:10.1016/j.neulet.2010.10.017.

Measurements of neuron soma size and density in rat dorsal striatum, nucleus accumbens core and nucleus accumbens shell: differences between striatal region and brain hemisphere, but not sex

John Meitzen, Kelsey R. Pflepsen, Christopher M. Stern, Robert L. Meisel, and Paul G. Mermelstein

Department of Neuroscience, University of Minnesota, Minneapolis, MN

Abstract

Both hemispheric bias and sex differences exist in striatal-mediated behaviors and pathologies. The extent to which these dimorphisms can be attributed to an underlying neuroanatomical difference is unclear. We therefore quantified neuron soma size and density in the dorsal striatum (CPu) as well as the core (AcbC) and shell (AcbS) subregions of the nucleus accumbens to determine whether these anatomical measurements differ by region, hemisphere, or sex in adult Sprague-Dawley rats. Neuron soma size was larger in the CPu than the AcbC or AcbS. Neuron density was greatest in the AcbS, intermediate in the AcbC, and least dense in the CPu. CPu neuron density was greater in the left in comparison to the right hemisphere. No attribute was sexually dimorphic. These results provide the first evidence that hemispheric bias in the striatum and striatal-mediated behaviors can be attributed to a lateralization in neuronal density within the CPu. In contrast, sexual dimorphisms appear mediated by factors other than gross anatomical differences.

Keywords

Striatum, sexual dimorphism; rat; nucleus accumbens, lateralization; morphometry

Lateralizations and sex differences in neural circuitry and behavior occur across animal taxa and brain region. These brain dimorphisms can take many forms, including but not limited to differences in neuroanatomy, neurochemistry, and electrophysiology [1,2]. In some model systems, these differences are dramatic and have a fairly straightforward connection to behavior [3,4]. In other areas, these differences may be more subtle, although no less important. One of these regions is the mammalian striatum, including the dorsal striatum (CPu) and the core and shell subregions of the nucleus accumbens (AcbC and AcbS, respectively). Though clear lateralizations [5–12] and sex differences [13–15] exist in the striatum and striatal-related behaviors and pathologies, whether this is reflected in striatal neuron morphology remains largely unknown.

Corresponding Author: John Meitzen, Ph.D., Dept. of Neuroscience, University of Minnesota, 6-145 Jackson Hall, 321 Church St SE, Minneapolis, MN, 55455, meitz010@umn.edu, Office Phone: 612-625-0784, Fax: 612-626-5009.

Publisher's Disclaimer: This is a PDF file of an unedited manuscript that has been accepted for publication. As a service to our customers we are providing this early version of the manuscript. The manuscript will undergo copyediting, typesetting, and review of the resulting proof before it is published in its final citable form. Please note that during the production process errors may be discovered which could affect the content, and all legal disclaimers that apply to the journal pertain.

Given that differences in neuronal soma size and density have previously been found between hemispheres and sexes in other brain regions, we decided to quantify these neuronal attributes in the CPu, AcbC and AcbS of adult gonadectomized male and female rats. Gonadectomized rats were used to avoid any potential confounds induced by steroid sex hormones, which exert activational effects on striatal neurons [14,16] [17] beyond baseline sex differences [14,15]. We found that neuron soma size was larger in the CPu than the AcbC or AcbS. Neuron density was greatest in the AcbS, intermediate in the AcbC, and least dense in the CPu. While no attribute was sexually dimorphic, one lateralization was detected: neuron density in the CPu was greater in the left versus right hemisphere. These results indicate that the different striatal regions can be distinguished based upon neuron soma size and density, provide an anatomic basis for previous findings regarding lateralization in the striatum and striatal-mediated behavior, and that sex differences in striatal-mediated behaviors are driven by factors other than neuron soma size and density.

Animals

The Institutional Animal Care and Use Committee at the University of Minnesota approved all procedures used in this study. Four adult male and four adult female Sprague-Dawley rats were purchased from Harlan Laboratories. Rats were gonadectomized on day 60 of life at Harlan Laboratories. The absence of gonads was verified post-mortem. Food and water were available *ad libitum*, and animals were maintained on a 14-h light, 10-h dark cycle in a climate controlled colony.

Brain Histology

On day 75 of life, animals were deeply anesthetized using pentobarbital (200 mg/kg, *i.p.*) and perfused transcardially with saline until liver clearance, and then with 300 ml of 4% paraformaldehyde over 12 minutes. The anticoagulant heparin (0.5 ml of 1000 UPS units/ml) was injected into the left ventricle prior to perfusion. Brains were post-fixed overnight in 4% paraformaldehyde at 4° C, cryoprotected in 30% sucrose solution in 0.1 M phosphate buffered-saline (PBS), and then sectioned coronally (50 µm) on a freezing microtome. The right hemisphere of each brain was nicked to distinguish hemispheres. Every other section was mounted onto slides, stained with cresyl violet, and imaged using a Leica DM4000B light microscope coupled to a Leica DFC500 digital camera. The cresyl violet stain was used to provide continuity with previous studies, because the stain effectively distinguishes between neurons and glia (discussed further below), and because other laboratories have found no difference in the morphology of D1 and D2 expressing medium spiny neurons [18]. Given the distribution of soma sizes measured by this study and the preponderance of medium spiny neurons in the striatum [19,20], the vast majority of the neurons measured in this study were medium spiny neurons.

Morphometry

Morphometric methods were adapted from previous studies [21–24], and followed the principles of unbiased stereology [25]. Neuronal density and soma size were measured in both hemispheres and along the rostral-caudal axis of the CPu, AcbC and AcbS. Neuron density was measured using protocols adapted from previous studies [24], including those that analyzed striatal tissue [22,23]. Briefly, following previous analyses [22], a large counting box (199 x 149 µm) was placed within the CPu, AcbC or AcbS in a systemic and random manner. A large box was used to minimize sampling variance by ensuring equal sampling of both patch and matrix. Patch and matrix medium spiny neurons have similar morphology and intrinsic membrane properties [26,27]. At least two of these large fields were counted in each hemisphere, and at least 200 neurons were counted per hemisphere per region. Neuronal nucleoli were used as the unit of count to obtain estimates of neuron

density that were unbiased by soma size or shape. Nucleoli that were bisected by either the left boundary or the upper boundary of the counting frame were counted, whereas those that were bisected by the right boundary or the lower boundary were not counted. Along the z-axis, nucleoli were counted if they were bisected by the upper boundary but not the lower. To account for sectioning artifacts at the tissue edge (e.g., lost nucleolus caps), nucleoli in the top and bottom 3 μm of the section were not included in any count. The size of neuronal nucleoli did not differ between groups, so it is unlikely that splitting errors introduced asymmetric bias to our data [28,29]. Density was calculated by dividing neuron count by the volume of the tissue sampled and averaging across sampling boxes. The reliability of our neuron density measurements was assessed by having an experimenter blindly re-measure neuron density in multiple fields across subjects. Neurons were distinguished from glia by the clear presence of a nucleolus, a well-defined nuclear envelope, nongranular cytoplasm, and/or an obvious axon hillock, as in previous studies [21,24,30,31]. Measurements were made blind to treatment group.

Methods for measuring neuron size were adapted from previous studies [21,23,24]. Neuron size was measured using the cross-sectional area of the soma. A minimum of 100 neurons per region were measured, and this sample size was sufficient to encompass the entire range of variability in striatum soma area. Neurons were distinguished from glia as described above, and all measurements were made blind to treatment group.

Statistics

We used a one-way (two-tailed) ANOVA and Tukey's post-hoc tests to make comparisons between brain regions, and two-way repeated measures ANOVA and Tukey's post-hoc tests to make comparisons between sex and hemisphere within a brain region. Software used was Prism 4.03 (Graphpad, La Jolla, CA) and SigmaStat 3.00 (SPSS, Chicago, IL). Probability values less than 0.05 were considered *a priori* as significant. Data are presented as mean $\pm$ SEM.

Results

We first compared neuron soma size and density across striatal regions independently of sex or hemisphere using a one-way ANOVA with Tukey's post-hoc test. We found that neuron soma size was larger in the CPu compared to the AcbC or AcbS (Figure 1A; $p < 0.0001$ for both; $F_{2,21} = 36.70$), and did not differ between the AcbC or AcbS ($p > 0.05$ for both). Neuron density likewise varied across striatal regions. Neurons were denser in the AcbS compared to both the CPu and the AcbC (Figure 1B; $p < 0.001$ for both; $F_{2,21} = 59.84$), and in the AcbC compared to the CPu ($p < 0.001$). These data indicate that different striatal regions can be distinguished based upon neuron size and density.

After determining that neuron soma size and density differed between the CPu, AcbC and AcbS, we compared these measures between hemisphere and sex in each striatal region using two-way repeated measures ANOVAs (Table 1). We found no differences in neuron soma size between sex, hemisphere, or the interaction between sex and hemisphere in the CPu (Figure 2A), AcbC (Figure 2B) and AcbS (Figure 2C).

A difference was detected, however, in neuron density. Neuron density was lateralized in the CPu, with neurons being denser in the left hemisphere than the right (Figure 3A, $p < 0.05$; $F = 7.152$). No differences in neuron density were found between hemispheres in the AcbC (Figure 3B) or the AcbS (Figure 3C). We also found no differences in neuron density between sex or the interaction between hemisphere and sex in the CPu (Figure 3A), AcbC (Figure 3B; $p > 0.05$) and AcbS (Figure 3C).

Discussion

This study measured neuron soma size and density in three different regions of the striatum, the CPu, AcbC and AcbS, and then analyzed whether these neural attributes differed between sex and hemisphere. No differences in neuron size were detected between sex and hemisphere, while neuron density was mildly lateralized in the CPu but otherwise did not vary by sex or hemisphere. Here we first discuss this data with regards to differences between striatal regions, then with regards to sex, and then hemispheric lateralization.

Neuron soma size and density varied by striatal region, with neuron soma size increased in the CPu compared to the AcbC and AcbS, and neuron density being greatest in the AcbS, intermediate in the AcbC, and least in the CPu. These findings are similar to those of a previous morphometric study of neuron morphology in the nucleus accumbens, which found that the soma size of medium spiny neurons do not differ between the AcbC and the AcbS, but that neurons in the AcbC have greater dendritic lengths than those in the AcbS [32].

Dendritic length in AcbS and AcbC medium spiny neurons is not sexually dimorphic [15], though previous reports have identified sex differences in striatal neuron spine density [15], neurochemistry [13,14], gene expression [33], and responsiveness to steroid sex hormones [14,16]. All of these are relevant to the clear sex differences in striatal-mediated behaviors and pathologies [14]. In this study, neuron density and soma size were measured because these cellular attributes can dramatically differ between sexes [34,35];[36]. No differences in neuron density or soma size between sexes were detected, however, indicating that sex differences in striatal function and behavior are mediated through other mechanisms.

Regarding lateralization, we initially included an analysis of lateralization in this study because lateralized sex differences in dopamine levels have been reported in the striatum [37], and because other brain regions exhibit lateralized sexual dimorphisms in neuron size and regional volume [36]. In the rat striatum, however, no interactions between hemisphere and sex were detected (Table 1). This study did find that neuron density was lateralized in the CPu of both males and females, with neurons being denser in the left hemisphere than the right. This finding is consistent with earlier literature, which found that the majority of Sprague-Dawley rats show a right-sided bias in multiple behavioral tasks and assays [6–8], which is reflected by a number of lateralizations in the striatum that favor the left hemisphere. These include increased expression of dynorphin, GAD, and NR2A mRNA measured using hybridization histochemistry [10,11], and higher D2 dopamine receptor binding [38]. The increased number of neurons in the left hemisphere potentially accounts for these previous findings. Other measured lateralizations, however, cannot be explained by differences in striatal neuron density, including hemispheric differences in dopamine levels [5], adenylyl cyclase activity [39] and sensitivity to self-stimulation [40]. We thus speculate that projections into the striatum, particularly dopaminergic projections, might also exhibit lateralizations.

We conclude by noting that differences were detected in neuron soma size and density between striatal regions and brain hemispheres, but not between sexes. We found that neuron density was increased in the left hemisphere than the right, which is consistent with previous studies of striatal lateralization. We found no differences between males and females.

Research Highlights

- Neuron soma size was larger in the CPu than the AcbC or AcbS.

- Neuron density was greatest in the AcbS, intermediate in the AcbC, and least dense in the CPu.
- CPu neuron density was greater in the left than right hemisphere.
- Neuron soma size and density were not sexually dimorphic in the CPu, AcbC or AcbS.

Acknowledgments

We thank Valerie Hedges, Nancy Staffend, and Caroline Loftus for technical assistance, Katherine Ingram for help processing tissue, and Dr. Christopher K. Thompson for comments on a previous version of this manuscript. Grant Support: National Institutes of Health Grants NS41302 (P.G.M.), DA07234 (training grant supporting: J.M., C.M.S.), DA013680 (R.L.M.).

References

1. Tommasi L. Mechanisms and functions of brain and behavioural asymmetries. *Philos Trans R Soc Lond B Biol Sci.* 2009; 364:855–9. [PubMed: 19064348]
2. Cahill L. Why sex matters for neuroscience. *Nat Rev Neurosci.* 2006; 7:477–84. [PubMed: 16688123]
3. Wade J, Arnold AP. Sexual differentiation of the zebra finch song system. *Ann N Y Acad Sci.* 2004; 1016:540–59. [PubMed: 15313794]
4. Suthers RA. Peripheral control and lateralization of birdsong. *J Neurobiol.* 1997; 33:632–52. [PubMed: 9369464]
5. Zimmerberg B, Glick SD, Jerussi TP. Neurochemical correlate of a spatial preference in rats. *Science.* 1974; 185:623–5. [PubMed: 4858234]
6. Glick SD, Ross DA. Right-sided population bias and lateralization of activity in normal rats. *Brain Res.* 1981; 205:222–5. [PubMed: 7193500]
7. Castellano MA, Diaz-Palarea MD, Barroso J, Rodriguez M. Behavioral lateralization in rats and dopaminergic system: individual and population laterality. *Behav Neurosci.* 1989; 103:46–53. [PubMed: 2493792]
8. Castellano MA, Diaz-Palarea MD, Rodriguez M, Barroso J. Lateralization in male rats and dopaminergic system: evidence of right-side population bias. *Physiol Behav.* 1987; 40:607–12. [PubMed: 3671525]
9. Scholz VH, Flaherty AW, Kraft E, Keltner JR, Kwong KK, Chen YI, Rosen BR, Jenkins BG. Laterality, somatotopy and reproducibility of the basal ganglia and motor cortex during motor tasks. *Brain Res.* 2000; 879:204–15. [PubMed: 11011024]
10. Capper-Loup C, Kaelin-Lang A. Lateralization of dynorphin gene expression in the rat striatum. *Neurosci Lett.* 2008; 447:106–8. Epub 2008 Sep 30. [PubMed: 18838109]
11. Capper-Loup C, Rebell D, Kaelin-Lang A. Hemispheric lateralization of the corticostriatal glutamatergic system in the rat. *J Neural Transm.* 2009; 116:1053–7. Epub 2009 Jul 21. [PubMed: 19626270]
12. Kloppel S, Mangin JF, Vongersichten A, Frackowiak RS, Siebner HR. Nurture versus nature: long-term impact of forced right-handedness on structure of pericentral cortex and basal ganglia. *Journal of Neuroscience.* 2010; 30:3271–5. [PubMed: 20203186]
13. Nazarian A, Sun WL, Zhou L, Kemen LM, Jenab S, Quinones-Jenab V. Sex differences in basal and cocaine-induced alterations in PKA and CREB proteins in the nucleus accumbens. *Psychopharmacology (Berl).* 2009; 203:641–50. Epub 2008 Dec 4. [PubMed: 19052730]
14. Becker JB, Hu M. Sex differences in drug abuse. *Front Neuroendocrinol.* 2008; 29:36–47. Epub 2007 Aug 24. [PubMed: 17904621]
15. Forlano PM, Woolley CS. Quantitative analysis of pre- and postsynaptic sex differences in the nucleus accumbens. *J Comp Neurol.* 2010; 518:1330–48. [PubMed: 20151363]

16. Mermelstein PG, Becker JB, Surmeier DJ. Estradiol reduces calcium currents in rat neostriatal neurons via a membrane receptor. *J Neurosci*. 1996; 16:595–604. [PubMed: 8551343]
17. Grove-Strawser D, Boulware MI, Mermelstein PG. Membrane estrogen receptors activate the metabotropic glutamate receptors mGluR5 and mGluR3 to bidirectionally regulate CREB phosphorylation in female rat striatal neurons. *Neuroscience* 2010. 2010 Epub ahead of print.
18. Surmeier DJ, Ding J, Day M, Wang Z, Shen W. D1 and D2 dopamine-receptor modulation of striatal glutamatergic signaling in striatal medium spiny neurons. *Trends Neurosci*. 2007; 30:228–35. Epub 2007 Apr 3. [PubMed: 17408758]
19. Tepper JM, Bolam JP. Functional diversity and specificity of neostriatal interneurons. *Curr Opin Neurobiol*. 2004; 14:685–92. [PubMed: 15582369]
20. Farries MA, Meitzen J, Perkel DJ. Electrophysiological properties of neurons in the basal ganglia of the domestic chick: conservation and divergence in the evolution of the avian basal ganglia. *J Neurophysiol*. 2005; 94:454–67. [PubMed: 15772239]
21. Meitzen J I, Moore T, Lent K, Brenowitz EA, Perkel DJ. Steroid hormones act transsynaptically within the forebrain to regulate neuronal phenotype and song stereotypy. *J Neurosci*. 2007; 27:12045–57. [PubMed: 17978046]
22. Rosen GD, Williams RW. Complex trait analysis of the mouse striatum: independent QTLs modulate volume and neuron number. *BMC Neurosci*. 2001; 2:5. Epub 2001 Apr 17. [PubMed: 11319941]
23. Thompson CK, Brenowitz EA. Seasonal change in neuron size and spacing but not neuronal recruitment in a basal ganglia nucleus in the avian song control system. *J Comp Neurol*. 2005; 481:276–83. [PubMed: 15593375]
24. Tramontin AD, Smith GT, Breuner CW, Brenowitz EA. Seasonal plasticity and sexual dimorphism in the avian song control system: stereological measurement of neuron density and number. *J Comp Neurol*. 1998; 396:186–92. [PubMed: 9634141]
25. Schmitz C, Hof PR. Design-based stereology in neuroscience. *Neuroscience*. 2005; 130:813–31. [PubMed: 15652981]
26. Kawaguchi Y, Wilson CJ, Emson PC. Intracellular recording of identified neostriatal patch and matrix spiny cells in a slice preparation preserving cortical inputs. *J Neurophysiol*. 1989; 62:1052–68. [PubMed: 2585039]
27. Kawaguchi Y, Wilson CJ, Emson PC. Projection subtypes of rat neostriatal matrix cells revealed by intracellular injection of biocytin. *J Neurosci*. 1990; 10:3421–38. [PubMed: 1698947]
28. Saper CB. Any way you cut it: a new journal policy for the use of unbiased counting methods. *J Comp Neurol*. 1996; 364:5. [PubMed: 8789271]
29. Guillery RW, Herrup K. Quantification without pontification: choosing a method for counting objects in sectioned tissues. *J Comp Neurol*. 1997; 386:2–7. [PubMed: 9303520]
30. Markham JA, Morris JR, Juraska JM. Neuron number decreases in the rat ventral, but not dorsal, medial prefrontal cortex between adolescence and adulthood. *Neuroscience*. 2007; 144:961–8. Epub 2006 Nov 28. [PubMed: 17137726]
31. Rubinow MJ, Juraska JM. Neuron and glia numbers in the basolateral nucleus of the amygdala from preweaning through old age in male and female rats: a stereological study. *J Comp Neurol*. 2009; 512:717–25. [PubMed: 19065620]
32. Meredith GE, Agolia R, Arts MP, Groenewegen HJ, Zahm DS. Morphological differences between projection neurons of the core and shell in the nucleus accumbens of the rat. *Neuroscience*. 1992; 50:149–62. [PubMed: 1383869]
33. Chen X, Grisham W, Arnold AP. X chromosome number causes sex differences in gene expression in adult mouse striatum. *Eur J Neurosci*. 2009; 29:768–76. [PubMed: 19250439]
34. Nottebohm F, Arnold AP. Sexual dimorphism in vocal control areas of the songbird brain. *Science*. 1976; 194:211–3. [PubMed: 959852]
35. Ball GF, MacDougall-Shackleton SA. Sex differences in songbirds 25 years later: what have we learned and where do we go? *Microsc Res Tech*. 2001; 54:327–34. [PubMed: 11668646]
36. Cooke BM. Steroid-dependent plasticity in the medial amygdala. *Neuroscience*. 2006; 138:997–1005. Epub 2005 Dec 5. [PubMed: 16330154]

37. Robinson TE, Becker JB, Ramirez VD. Sex differences in amphetamine-elicited rotational behavior and the lateralization of striatal dopamine in rats. *Brain Res Bull.* 1980; 5:539–45. [PubMed: 7191765]
38. Schneider LH, Murphy RB, Coons EE. Lateralization of striatal dopamine (D2) receptors in normal rats. *Neurosci Lett.* 1982; 33:281–4. [PubMed: 7162690]
39. Jerussi TP, Glick SD, Johnson CL. Reciprocity of pre- and postsynaptic mechanisms involved in rotation as revealed by dopamine metabolism and adenylate cyclase stimulation. *Brain Res.* 1977; 129:385–8. [PubMed: 884513]
40. Glick SD, Weaver LM, Meibach RC. Lateralization of reward in rats: differences in reinforcing thresholds. *Science.* 1980; 207:1093–5. [PubMed: 7355277]

Figure 1.

Neuron soma size and density differed between the CPu, AcbC and AcbS. A) Neuron soma size was significantly increased in the CPu compared to the AcbC and AcbS ($p < 0.0001$ for both). B) Neurons were denser in the AcbS compared to both the CPu and the AcbC ($p < 0.001$ for both), and in the AcbC compared to the CPu ($p < 0.001$).

Figure 2.

Neuron soma size did not differ by sex or hemisphere. For all subpanels: Left: Neuron soma size compared by sex. Middle: Neuron soma size compared between hemispheres in males. Right: Neuron soma size compared between hemispheres in females. A) CPu. B) AcbC. C) AcbS. For all comparisons $p > 0.05$.

Figure 3. Neuron density was lateralized in the CPu but otherwise did not vary by sex or hemisphere. For all subpanels: Left: Neuron density compared by sex. Middle: Neuron density compared between hemispheres in males. Right: Neuron density compared between hemispheres in females. A) CPu. Neurons were significantly denser in the left hemisphere compared to the right in both males and females ($p=0.037$). B) AcbC. C) AcbS. Except for that noted above, for all comparisons $p>0.05$.

Table 1**Measurements and Statistics**

Neuron soma size is presented in μm^2 . Neuron density is presented in neurons/ $\text{mm}^3 \times 10^3$. A 1-way ANOVA was used to compare values of neuron soma size and density across brain regions ("Global" row). In the Global section, different superscript letters denote significant differences across rows as determined by post-hoc tests. A 2-way repeated measured ANOVA was used to test the contributions of sex, hemisphere, and the interaction of sex and hemisphere to neuron size and density for each specific striatal region. Bold text indicates a significant finding. Abbreviations: M, male; F, female; L, left; R, right.

	CPu	AcbC	Acbs
Global			
Neuron Soma Size	81 ± 1^a	74 ± 1^b	72 ± 1^b
Statistics	$F_{2,23}=36.7$; $p<0.0001$	$F_{2,23}=36.7$; $p<0.0001$	$F_{2,23}=36.7$; $p<0.0001$
Neuron Density	110 ± 6^a	154 ± 8^b	220 ± 7^c
Statistics	$F_{2,23}=59.8$; $p<0.0001$	$F_{2,23}=59.8$; $p<0.0001$	$F_{2,23}=59.8$; $p<0.0001$
Sex			
Neuron Soma Size	M: 82 ± 1; F: 80 ± 2	M: 74 ± 1; F: 73 ± 1	M: 71 ± 1; F: 72 ± 1
Statistics	$F_{1,15}=0.554$; $p=0.485$	$F_{1,15}=0.0393$; $p=0.849$	$F_{1,15}=0.656$; $p=0.449$
Neuron Density	M: 119 ± 11; F: 102 ± 4	M: 147 ± 6; F: 160 ± 16	M: 210 ± 6; F: 231 ± 10
Statistics	$F_{1,15}=2.465$; $p=0.167$	$F_{1,15}=0.575$; $p=0.477$	$F_{1,15}=3.923$; $p=0.095$
Hemisphere			
Neuron Soma Size	L: 83 ± 2; R: 80 ± 1	L: 73 ± 1; R: 74 ± 1	L: 72 ± 1; R: 72 ± 1
Statistics	$F_{1,15}=0.947$; $p=0.368$	$F_{1,15}=4.410$; $p=0.080$	$F_{1,15}=0.052$; $p=0.827$
Neuron Density	L: 115 ± 7; R: 106 ± 6	L: 149 ± 8; R: 158 ± 10	L: 222 ± 10; R: 215 ± 8
Statistics	$F_{1,15}=7.152$; $p=0.037$	$F_{1,15}=0.958$; $p=0.366$	$F_{1,15}=0.771$; $p=0.414$
Sex x Hemisphere			
Neuron Soma Size	ML: 85 ± 2; MR: 79 ± 2; FL: 80 ± 2; FR: 80 ± 2	ML: 74 ± 1; MR: 74 ± 1; FL: 72 ± 1; FR: 74 ± 1	ML: 71 ± 2; MR: 71 ± 1; FL: 72 ± 1; FR: 72 ± 1
Statistics	$F_{1,15}=3.105$; $p=0.129$	$F_{1,15}=3.384$; $p=0.115$	$F_{1,15}=0.060$; $p=0.815$
Neuron Density	ML: 124 ± 13; MR: 114 ± 9; FL: 106 ± 5; FR: 98 ± 4	ML: 143 ± 13; MR: 151 ± 3; FL: 155 ± 12; FR: 165 ± 21	ML: 214 ± 12; MR: 196 ± 6; FL: 229 ± 16; FR: 233 ± 5
Statistics	$F_{1,15}=0.003$; $p=0.957$	$F_{1,15}=0.010$; $p=0.925$	$F_{1,15}=1.875$; $p=0.220$