

1 **Supplementary Information for:**

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3 **Oestradiol synthesized by female neurons generates sex**  
4 **differences in neuritogenesis**

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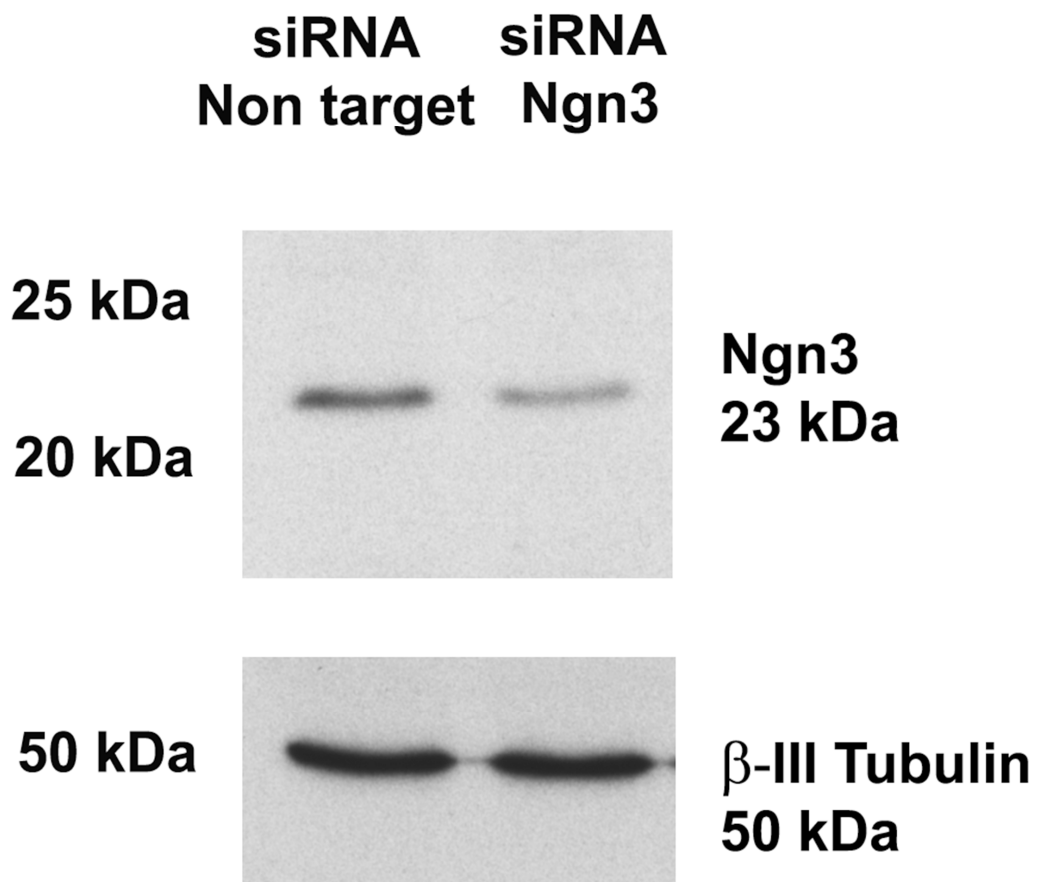
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1 **Supplementary Fig. S1**



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5 **Supplementary Fig. S1. Downregulation of Ngn3 expression by the action of**

6 **a specific Ngn3 siRNA.** The efficacy of Ngn3 silencing was verified by Western

7 blot. The band corresponding to the molecular weight of Ngn3 (23 kDa), decreases

8 after the Ngn3 siRNA treatment respect to the non target siRNA in cultured

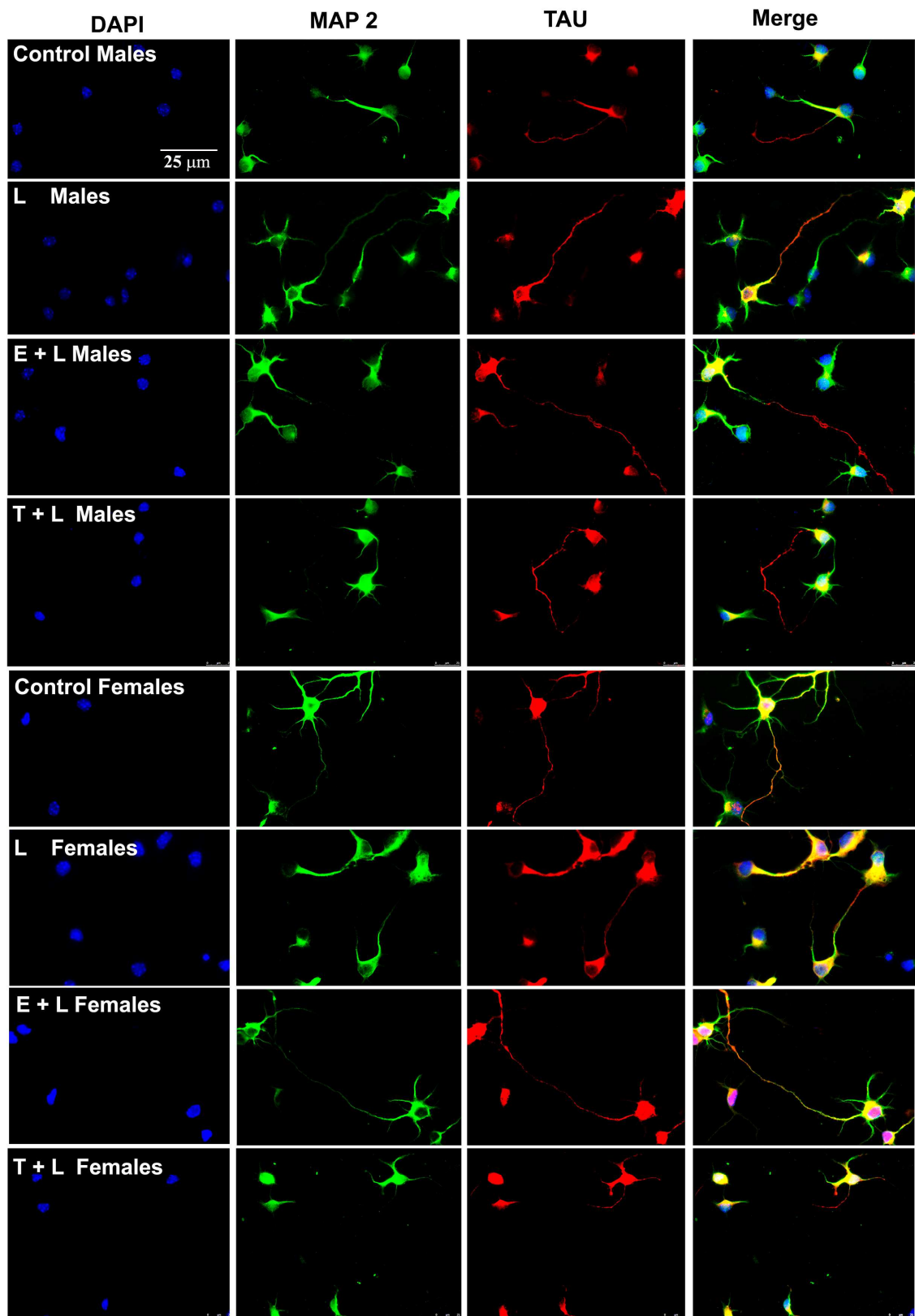
9 hippocampal neurons.

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1 **Supplementary Fig. S2**

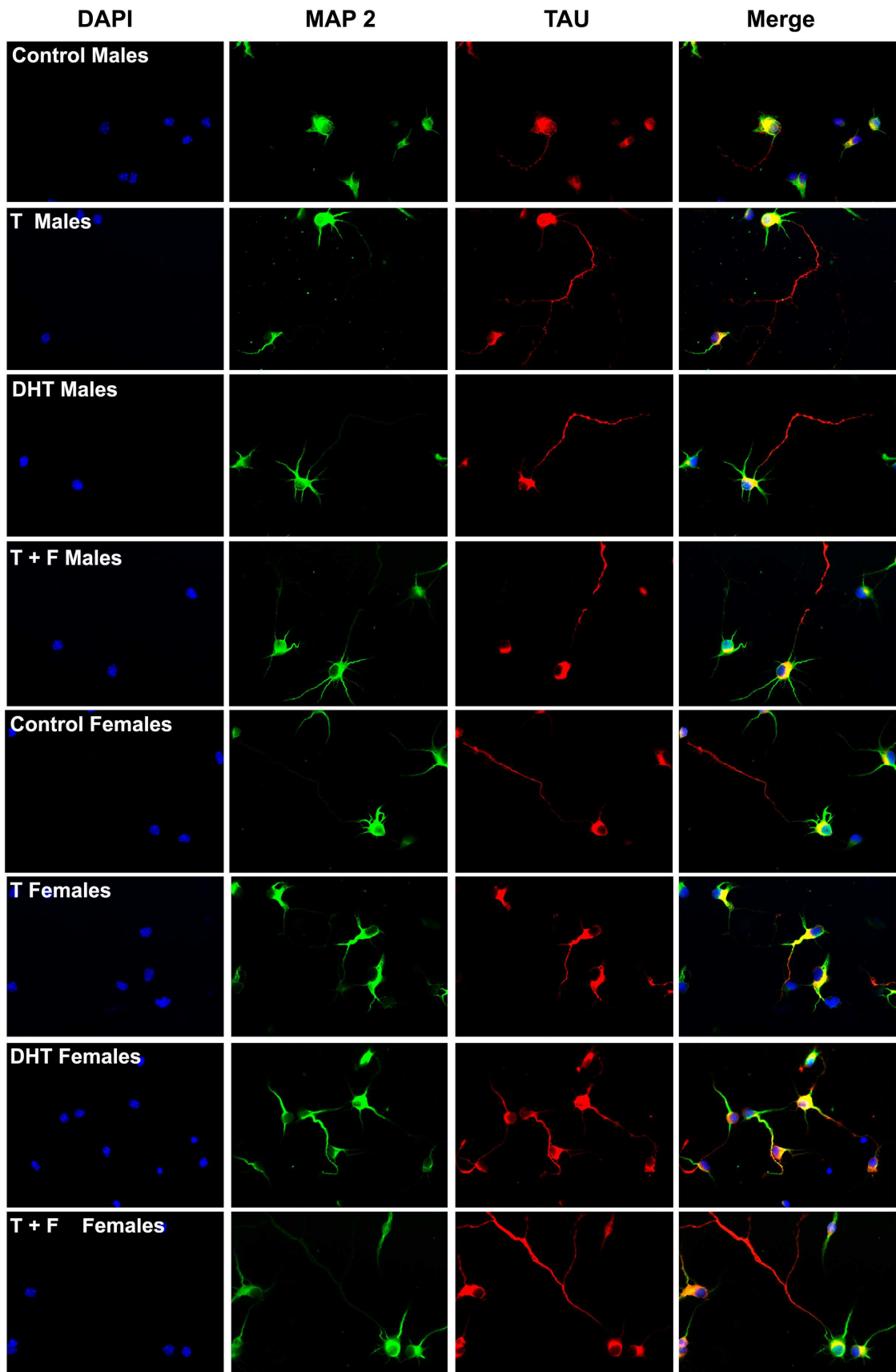


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1 **Supplementary Fig. S2. Representative examples of male and female**  
2 **neurons treated with letrozole (L).** Hippocampal neuronal cultured were treated  
3 at 1 DIV with L, L+E2 and L+T and immunostained for MAP 2 (green), TAU (red)  
4 and DAPI (blue) at 2 DIV.

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1 **Supplementary Fig. S3**



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1 **Supplementary Fig. S3. Representative examples of male and female**  
2 **neurons treated with testosterone (T), dihydrotestosterone (DHT) and**  
3 **finasteride (F).** Hippocampal neuronal cultured were treated at 1 DIV with T, DHT  
4 and T+F and immunostained for MAP 2 (green), TAU (red) and DAPI (blue) at 2  
5 DIV.

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1 **Supplementary Table 1. Levels of steroids in the culture medium of male and**  
 2 **female hippocampal neurons from E17 embryos**

<b>Steroid</b>	<b>Sex</b>	<b>Levels in culture medium (pg/ml)</b>
<b>Pregnenolone</b>	<b>Males</b>	<b>9.136 ± 1.764</b>
	<b>Females</b>	<b>12.02 ± 1.852</b>
<b>Progesterone</b>	<b>Males</b>	<b>2404 ± 216</b>
	<b>Females</b>	<b>2393 ± 200</b>
<b>Dihydroprogesterone</b>	<b>Males</b>	<b>2026 ± 98</b>
	<b>Females</b>	<b>2034 ± 85</b>
<b>Isopregnanolone</b>	<b>Males</b>	<b>61.92 ± 12.79</b>
	<b>Females</b>	<b>63.81 ± 8.319</b>
<b>Allopregnanolone</b>	<b>Males</b>	<b>39.65 ± 8.371</b>
	<b>Females</b>	<b>32.09 ± 1.854</b>
<b>Testosterone</b>	<b>Males</b>	<b>2.349 ± 0.420</b>
	<b>Females</b>	<b>2.720 ± 0.403</b>
<b>Dihydrotestosterone</b>	<b>Males</b>	<b>2.840 ± 1.226</b>
	<b>Females</b>	<b>1.280 ± 0.631</b>
<b>Oestradiol</b>	<b>Males</b>	<b>4.816 ± 0.594</b>
	<b>Females</b>	<b>4.253 ± 0.843</b>

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 4 Data are mean±SEM from 8 cultures.  
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