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Tau deficiency leads to the upregulation of BAF-57, a protein involved in neuron-specific gene repression

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ABSTRACT

Although tau is mainly located in the cell cytoplasm, mostly bound to tubulin, it may also be found in the nucleus of neurons. Hence, we tested whether tau might play a role in regulating the expression of certain genes by comparing gene expression in mice containing or lacking the tau protein. Our results identified a significant difference in the expression of the *smarce1* gene, which codes for the BAF-57 protein, a protein involved in the repression of neuron specific genes. These data suggest a role for tau in neuron maturation.
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1. Introduction

Tau is a microtubule associated protein that promotes tubulin assembly in vitro [1], as well as stabilizing assembled microtubules in cultured cells [2] by suppressing microtubule dynamics [3]. Tau deficiency delays neurite extension in cultured neurons [4,5], although the tau deficient mice produced by gene-targeting are viable and they do not show important cytoskeletal abnormalities [5,6]. The mild phenotype of these mice could be due to functional redundancy between tau and other microtubule-associated proteins [7]. Nevertheless, delayed axonogenesis was observed in tau deficient neurons [6], a feature that has yet to be fully explained.

Tau can also bind to nucleic acids [8–11] and it has been observed in the nucleus of neuronal cells [12–14]. We confirmed that tau is indeed present in the nucleus of cells and hence, we tested the possible influence of the tau protein on gene expression showing that it can modify the expression of certain genes. We observed differences in the level of *smarce1* [15], which codes for BAF-57 [16], a protein involved in the repression of neuronal specific genes [17] and whose expression is stimulated in tau deficient mice.

2. Materials and methods

2.1. Animals

Tau$^{-/-}$ mice were generated as described previously [6], crossing heterozygous (*Tau*$^{+/-}$) mice to obtain homozygous tau knockout mice (*Tau*$^{-/-}$) and control littermates (*Tau*$^{+/+}$). 3 month-old male mice were used in the experiments carried out here. The animals were bred at the Centro de Biología Molecular "Severo Ochoa" (Madrid, Spain) and they were maintained in accordance with the institutional guidelines. Four to five mice were housed per cage in a temperature controlled environment on a 12/12 h light-dark cycle, with food and water available ad libitum.

2.2. Primary hippocampal cultures

Hippocampal cell cultures were prepared from 17-day-old mouse fetuses according to modified versions of established procedures [18]. Briefly, the hippocampus was dissected out from each pup and dissociated individually with the papain dissociation system (Worthington Biochemical Corp.; Lakewood, NJ). The cells recovered were plated on poly lysine-coated (1 mg/ml) coverslips

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at low density in 86% DMEM containing 10% horse serum, 0.5 mM Glutamine, 1% penicillin/streptomycin. After 4 h, the medium was replaced by the maintenance medium: Neurobasal medium (Invitrogen) supplemented with 2% B-27, 1% N2, 0.5 mM Glutamine, 1 mM pyruvate, 1% penicillin/streptomycin.

2.3. Immunofluorescence

After 24 and 48 h in culture, the hippocampal cells were fixed for 20 min with warm 4% paraformaldehyde in PBS containing 4% sucrose. After several washes with PBS, the cells were permeabilized in 0.1% Triton X-100 for 15 min and they were then treated with 1 M glycine for 30 min to eliminate any autofluorescence. After a brief wash with PBS, the cells were blocked with 1% BSA/PBS for 1 h and then they were exposed to the primary antibodies diluted in 1% BSA/PBS for 1 h: an antibody against Smi-31 (1:1000; Sternberger) as an axonal marker and anti-β-III-tubulin (1:2000; Covance) as a neuronal marker. Subsequently, the cultures were washed extensively and then incubated for 45 min with the appropriate secondary antibody conjugated with either fluorescein isothiocyanate or with TexasRed (Jackson Laboratories, West Grove, PA). After washing, the cells were mounted immediately with Fluorosave Reagent (Calbiochem) and the coverslips were analyzed on an Axioskop2 plus microscope (Zeiss) using an Image J Software to determine the axon length.

2.4. Western blotting

Hippocampal and kidney extracts from 3 month-old mice were prepared in ice-cold extraction buffer consisting of: 20 mM HEPES pH 7.4, 100 mM NaCl, 10 mM NaF, 1% Triton X-100, 1 mM sodium orthovanadate, 10 mM EDTA, and protease inhibitors (10 μg/ml aprotinin, 10 μg/ml leupeptin and 10 μg/ml pepstatin). The tissue samples were homogenized at 4 °C and the protein content was determined by the Bradford method (Biorad). Subsequently, protein extracts and the subcellular fractions samples were resolved on 10% SDS polyacrylamide gels and transferred to a nitrocellulose membrane (Schleicher & Schuell). The membranes were probed with the following primary antibodies: anti-BAF57 (1:1000; Sigma); 7.51 (1:1000; from Dr. Claude Wischik, UK) that reacts with phosphorylated as well as non-phosphorylated forms of tau within the tubulin binding region; Tau-1 (1:1000; Chemicon) that detects dephosphorylated tau; Tau-5 (1:1000; Calbiochem) that recognizes the central region of tau in both the phosphorylated and the non-phposphorylated forms; AT100 (1:500; Innogenetics) to detect phosphorylated tau; anti α-tubulin (1:1000; Sigma) as a cytoplasmic marker; and anti-laminB1 (1:200; Santa Cruz) as a nuclear marker. The membranes were incubated with the antibodies at 4 °C overnight in 5% non-fat dried milk, and the immunoreactive bands were detected with the corresponding secondary antibodies (1/5000; GIBCO) and visualized by ECL (Amersham Biosciences). The quantification of BAF57 was performed by densitometric scanning and the values were normalized to those obtained with an anti-β-actin antibody (1:5000; Sigma) to correct for deviations in protein loading.

2.5. Subcellular fractionation

To prepare nuclear and cytosolic extracts, tissues were disrupted with 15 strokes of an ice Dounce-homogenizer in M-SHE buffer (210 mM manitol, 70 mM sucrose, 10 mM HEPES [pH 7.4], 1 mM EDTA, 1 mM EGTA) supplemented with protease inhibitors (1 mM phenylmethylsulfonyl fluoride, 10 μg/ml aprotinin, 10 μg/ml leupeptin, 10 μg/ml pepstatin), phosphatase inhibitors (1 mM sodium orthovanadate, 30 mM glycerophosphate, 5 mM pyrophosphate, 1 μM okadaic acid) and 1 mM dithiothreitol. The homogenate was placed on ice for 30 min and then centrifuged at 1200×*g* for 10 min at 4 °C. The resulting pellet was homogenized and centrifuged again as above, and the supernatant was added to that obtained in the first step to constitute the cytosolic fraction. The membrane fraction (pellet) was homogenized in M-SHE buffer and centrifuged at 1000×*g* for 10 min at 4 °C. Both the supernatant and the pellet's upper layer were discarded and the remaining pellet, which contained the nuclei, was resuspended in TSE buffer (10 mM Tris [pH 7.5], 300 mM sucrose, 1 mM EDTA) supplemented with 0.1% NP-40. This suspension was centrifuged at 8500×*g* for 10 min at 4 °C, and the resulting pellet was resuspended in TSE buffer supplemented with 1% NP-40 and centrifuged at 1500×*g* for 7 min at 4 °C. This last step was repeated to clean the nuclear fraction (the opaque layer at the bottom of the tube) of any other membranes (yellow layer). Finally, lysis buffer (50 mM Tris–HCl [pH 6.8], 100 mM β-mercaptoethanol, 2% SDS, 0.1% bromophenol blue, 10% glycerol) supplemented with protease and phosphatase inhibitors, was added to cytosolic and nuclear fractions, and the samples were boiled for 10 min before they were analyzed by SDS–PAGE.

2.6. Gene expression analysis

For transcriptional analysis, hippocampal tissue from $Tau^{-/-}$ mice and control littermates ($n = 3$ per group) was analyzed on an Affymetrix MOE 430A 2.0 GeneChip®, which analyzes 14,000 annotated mouse genes. The preparation of RNA, hybridization, staining, and scanning of the GeneChip® MOE 430A 2.0 was carried out by Progenika Biopharma (Derio, Spain).

2.7. RNA isolation and quantitative real-time PCR

RNA was isolated from the hippocampus of $Tau^{-/-}$ mice and control littermates ($n = 6$ animals per genotype) using the Absolutely RNA MiniPrep Kit (Stratagene). This RNA was used as a template for the first strand cDNA synthesis (Roche), which was then analyzed by quantitative real-time PCR with an applied biosystems ABI PRISM 7900HT real-time detection system with the SYBR green PCR master mix (Applied Biosystems), interpreting the results with the SDS 2.1 software. The primers used to amplify each gene were: 5′-CACTTCTCCCATCTTTCGCT-3′ and 5′-GGGACTTAATGGCAGCT GAG-3′ for *mapt*; 5′-TCCTTTTCCTCAAGTGATTTGG-3′ and 5′-AACA

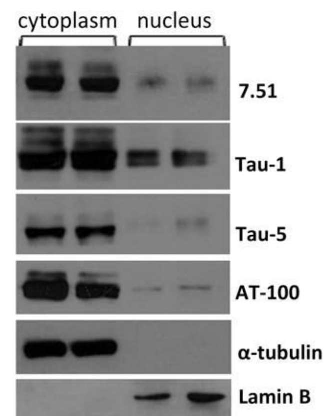


Fig. 1. Tau protein is not only present in the cytoplasm but also in the nucleus of cells. Western blot of the cytoplasmic and nuclear fractions from the hippocampus of two wild-type mice. The 7.51 antibody detected tau, both in the cytoplasm and in the nucleus, whereas tubulin was only present in the cytoplasmic fraction and laminB only in the nucleus. The interaction with other tau antibodies (Tau-1, Tau-5 and AT100) was also done, with similar result.

CATCTTTGTCTTATTTCTTTTCA-3′ for *calbindin*; 5′-ATGCCCTTCT CAGTCCTCTG-3′ and 5′-GGAAAGTCACAGGTGGAGGA-3′ for *lsm12′homolog*; 5′-TGGCTTTCCAAAGACACGAT-3′ and 5′-TCCATTA TAAGAACATTCCATTCCT-3′ for *smarce1*; 5′-TGGGAGTTCTGGT CAGGTTC-3′ and 5′-CCTAATCCACTGGGGACTGA-3′ for *map1lc3a*. Mean *Ct* values of gene expression were normalized to the mean expression of β-actin in each sample.

3. Results

3.1. Tau is located in the nucleus of neuronal cells

Tau protein was first discovered as a microtubule-associated protein [1] and as a result, it has been considered as a cytoplasmic protein like tubulin, the main component of microtubules. However,

Table 1
Overexpressed and downregulated genes detected in the Affymetrix Mouse GeneChip. These five genes were selected from 265 genes whose expression was significantly different in the hippocampus of $tau^{-/-}$ mice when compared to the wt mice (three animals per genotype).

Probeset ID	Gene title	Gene symbol	*P*-value	Fold change
Down regulated				
1424719	Microt. prot. tau	*mapt*	0.0000038	−5.42043
1448738	Calbidin 28 k	*calb1*	0.00196123	−1.26513
Overexpressed				
1427998	LSM12 homolog	*lsm12*	0.00126384	2.34947
1422676	SW1/SNF relat., matrix cell, actin (BAF57)	*smarce1*	0.0170026	1.16013
1451290	Microtubule assoc. prot. 1 light ch.3	*map1lc3a*	0.763173	1.14813

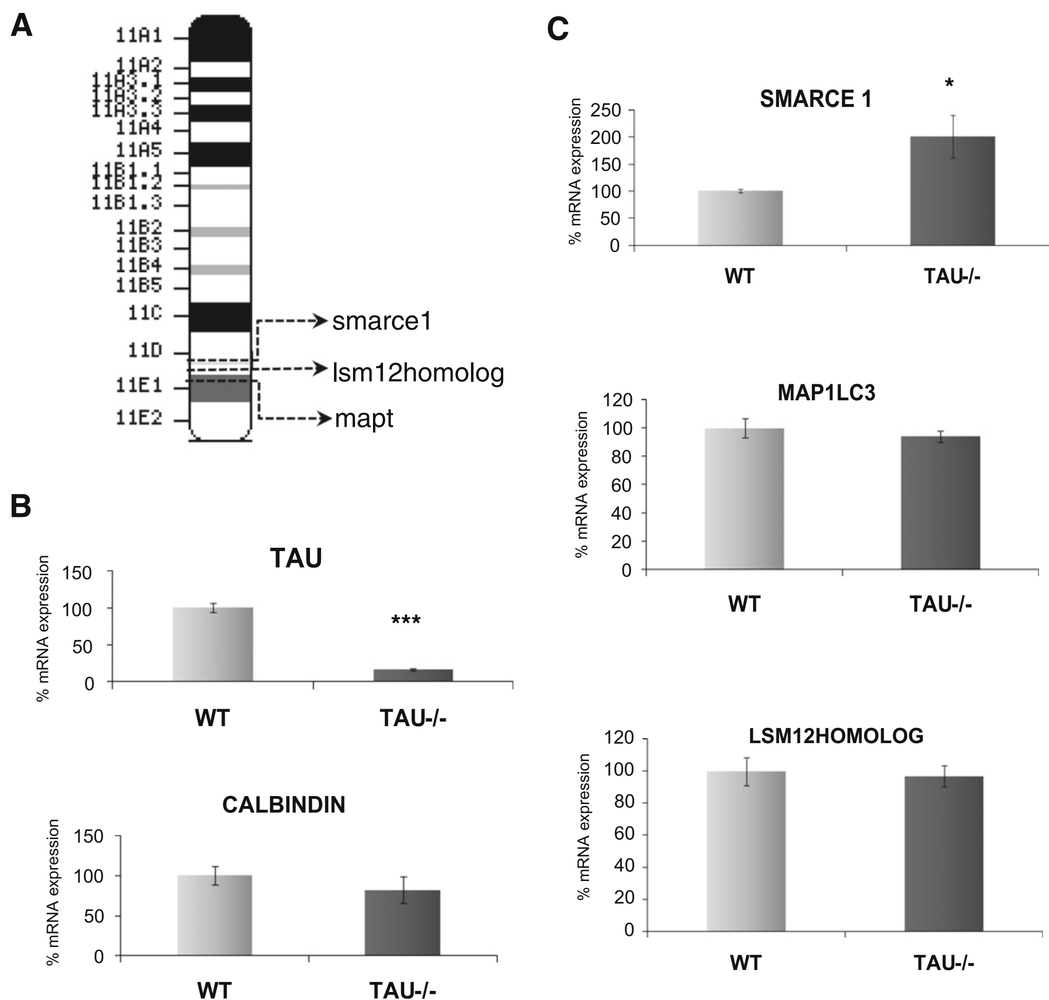


Fig. 2. Microarray validation by quantitative real-time PCR. (A) Mouse chromosome 11 ideogram showing the locations of *mapt*, *smarce1* and *lsm12 homologue*. Although these three genes are located in the same region, they are not contiguous. (B) Quantitative real-time PCR for downregulated genes. As expected, tau gene expression decreased significantly in tau deficient mice when compared to *wt* mice (fivefold; $^{***}p < 0.001$, Student's *t*-test). Calbindin mRNA levels were slightly lower in $Tau^{-/-}$ mice but the difference was not statistically significant. (C) Quantitative real-time PCR for upregulated genes. We confirmed *smarce1* gene was overexpressed (twofold; $^{*}p < 0.05$, Student's *t*-test) in $Tau^{-/-}$ mice when compared to *wt* mice. The expression of *map1lc3a* and the *lsm12 homologue* did not change significantly between $Tau^{-/-}$ and *wt* mice. Results are expressed as percentage of control. Data shown are the mean ± S.E. of six animals per genotype.

tau does not appear to be exclusively found in the cytoplasm and there are different reports indicating that a small proportion of tau is also present in the nucleus of cells [12–14]. We performed subcellular fractionation to confirm the presence of tau in the cell nucleus, as well as in the cytoplasm [19], using α-tubulin as a cytoplasmic marker and laminB as a marker of the nuclear fraction. Tau protein was clearly present in both cellular compartments, although there was less in the nuclear fraction. By contrast, tubulin was only present in the cytoplasmic fraction and laminB was only found in the nuclear fraction (Fig. 1).

Little is known about the possible function of tau in the nucleus [9], although it is known to be able to interact with chromatin and nucleic acids [10–14,20]. This nuclear localization may be compatible with a possible influence of the tau protein on gene expression. Thus, we studied whether tau might regulate the expression of specific genes.

3.2. Changes in gene expression in tau deficient mice

We set out to look for possible differences in gene expression between mice expressing or lacking the tau protein by using a mouse GeneChip platform (Affymetrix) as a high throughput technique to analyze gene expression. When RNA pools from *Tau*$^{-/-}$ and *Tau*$^{+/+}$ mice were compared (Table 1) there was a dramatic decrease in tau gene expression in *Tau*$^{-/-}$ mice, as expected (which served as a positive control for the analysis). In addition, slight changes in the expression of other genes were found, including: *smarce1* (coding for the BAF57 protein), *calbindin*, *the lsm12 homologue*, or *map1lc3a*. Curiously, some of these genes are located on chromosome 11 like the *tau* gene, although they are not contiguous (Fig. 2A). Real-time PCR analysis was performed on these transcripts to validate the data obtained from the microarrays (Fig. 2B and C). No significant differences were found in the expression of each of these different genes, except for the *tau* gene and *smarcel*. When the accumulation of the corresponding proteins was assessed in western blots, clear differences were only found in the *tau* protein, as well as slight differences in *smarce1* expression. When the presence of the BAF57 protein (the product of *smarce1* gene) was further investigated, there was a difference in the expression of the isoform with lower electrophoretic mobility (Fig. 3A). This difference was only found in the brain but not in the kidney of *Tau*$^{-/-}$ mice (Fig. 3B) and indeed, the three main BAF57 isoforms found in adult mouse brain were evident (Fig. 3C). These isoforms contain some exons similar to those found for human BAF57 isoforms whereas in other tissues, like the kidney, only the slower migrating isoform was found in mice [21]. However, when we tested whether there were changes in this isoform in the kidney of *Tau*$^{-/-}$ mice, no differences were found in the only isoform present (the longest one, Fig. 3B).

All three mouse BAF57 isoforms were expressed in the brain of wild type and tau deficient mice (Fig. 3A) and there was an increase in the total amount of the three BAF57 isoforms in *Tau*$^{-/-}$ when compared to wild type mice. In addition, a decrease in the proportion of the two isoforms of higher electrophoretic mobility was observed, isoforms mainly present in neuronal tissue, when compared to the slower migrating ubiquitously expressed BAF57 isoform.

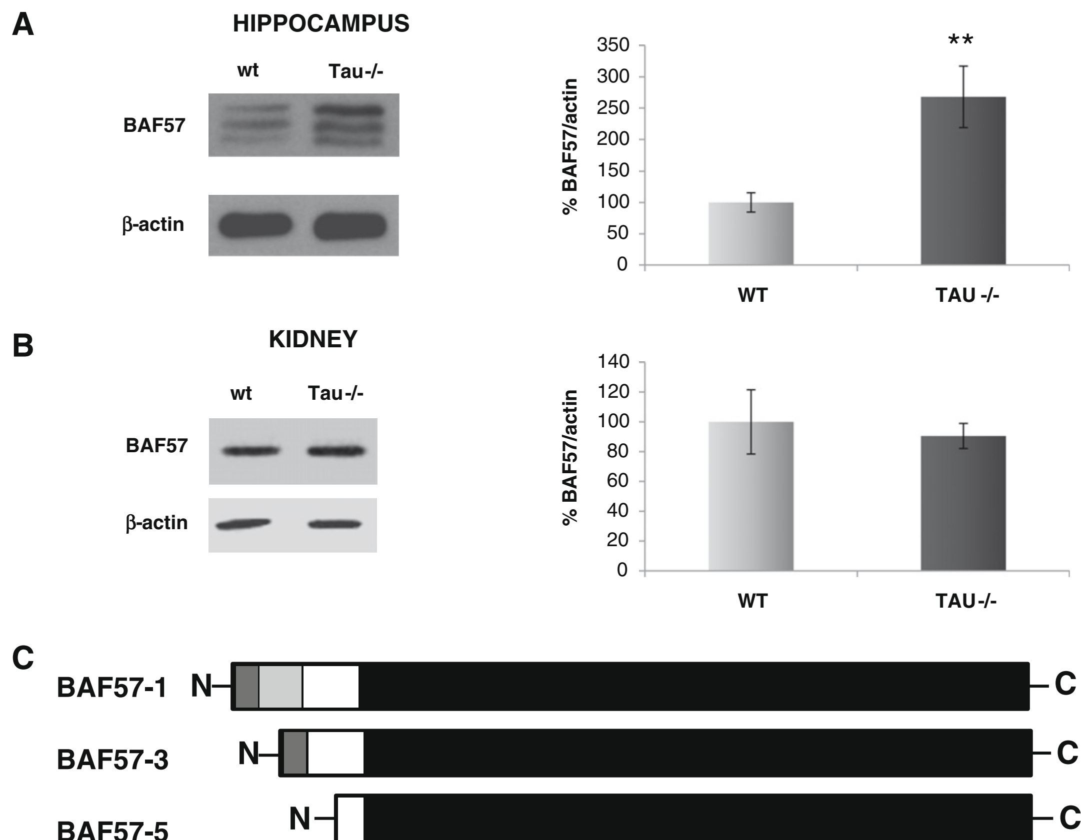
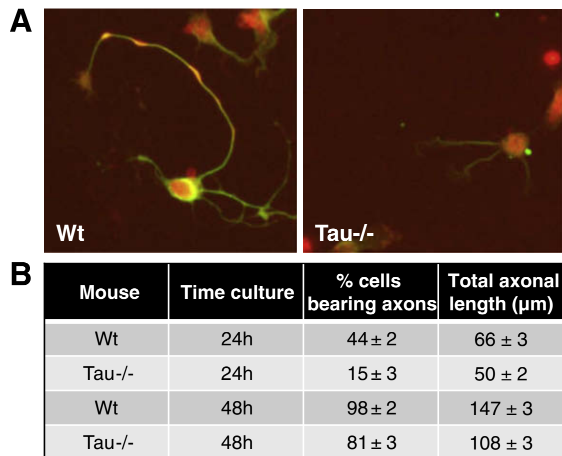


Fig. 3. The BAF57 protein is overexpressed in the hippocampus but not in the kidney of tau deficient mice. (A) Hippocampal extracts from *Tau*$^{-/-}$ and *wt* mice were analyzed by western blotting ($n = 6$ animals per genotype). Immunodetection with an anti-BAF57 antibody revealed three bands that corresponded to the three main BAF57 isoforms expressed in the mouse nervous system (see C). Total BAF57 was significantly increased in *Tau*$^{-/-}$ mice when compared to the *wt* mice ($^{**}p < 0.01$, Student's *t* test). (B) The antibody against BAF57 only detected one protein isoform in kidney extracts and the levels of BAF57 were similar in *Tau*$^{-/-}$ and *wt* mice. (C) Schematic structure of the three main BAF57 isoforms expressed in the mouse nervous system. The longest BAF57-1 isoform is ubiquitous, whereas BAF57-2 and BAF57-3 are specific to neural tissues [21].



Mouse	Time culture	% cells bearing axons	Total axonal length (μm)
Wt	24h	44 ± 2	66 ± 3
Tau-/-	24h	15 ± 3	50 ± 2
Wt	48h	98 ± 2	147 ± 3
Tau-/-	48h	81 ± 3	108 ± 3

Fig. 4. Axonogenesis is delayed in tau deficient neurons. (A) Immunofluorescence of 1DIV primary hippocampal neurons from $Tau^{-/-}$ mice and control littermates labeled with the Smi-31 (green) and β-III-tubulin (red) antibodies. (B) The percentage of cells bearing axons and the axon length were measured in $Tau^{-/-}$ and control neurons at 24 and 48 h (n = 80 cells per group).

3.3. Delayed axonogenesis in tau deficient neurons

BAF57 (BRG1-associated factor 57) is a component of the mammalian SWI/SNF ATP-dependent chromatin remodelling complex [16,22], which is involved in transcriptional regulation. Moreover, BAF57 interacts with CoREST, a protein that acts as a co-repressor of REST (RE1 silencing transcription factor), a transcription factor which represses neuronal genes in non-neuronal cells [17]. We have just indicated that an increase in BAF57 could produce a decrease (or delay) in the appearance of a neuronal morphology in tau deficient cells. Indeed, a delay in axon extension was reported earlier in primary cultures of tau deficient neurons [6]. These results have been now confirmed in similar experiments (Fig. 4), where a decrease in the proportion of cells bearing axons at stage III [23] was observed in primary hippocampal cultures of $Tau^{-/-}$ mice, although more than 80% of neurons from either source had extended neurites after 48 h in culture.

4. Discussion

We have shown here that the tau protein may be found in the cytoplasm or in the nucleus of a cell and that it may be capable of regulating gene expression. Thus, we tested the changes in gene expression in the *wt* and $Tau^{-/-}$ mice brain. In the absence of tau, changes in the expression of some genes were observed. As expected a dramatic decrease in *mapt* expression was seen, but also we detected changes in *smarce1*, *calbindin*, the *homologue of the lsm12′yeast* gene and in the *map1′light chain 3a* were also observed.

Smarce1 encodes the BAF-57 protein, which interacts with coREST, a protein that forms a complex with the class I HDAC. The interaction of BAF-57 with coREST facilitates the activity of the transcriptional repressor REST, which blocks the expression of neuronal specific genes in non-neuronal cells [17]. Calbindin is a calcium binding protein that appears to buffer the excess of that cation [24]. Curiously, calbindin gene is one of the genes repressed by REST complex [25]. Thus, an increase in *smarcel* gene expression is compatible with a decrease in calbindin gene expression, as found in Table 1. The homologue of the Lsm-12 yeast gene appears to play a role in protein translation, although little is known about its possible function in neurons [26,27]. Finally, MAP1-light chain 3a is a cytoskeletal protein [28] and it has previously been suggested that the amount of MAP1A increases in tau deficient mice [5].

Here, we focused our analysis on the differential expression of *smarce1* since, unlike the other genes, the genechip data for this gene was validated by RT-PCR and in Western blots. Since the *smarce1* gene is located on mouse chromosome 11, like the *tau* gene, the differences in its expression may be due to an artifact produced when generating the $Tau^{-/-}$ mice [6]. However, the fact that these genes do not lie close together on chromosome 11 and that the differences for BAF57 (*smarcel* product) are specific to the brain and they are not found in the kidney, suggest that the changes in the level of *smarce1* gene expression in the brain could be produced specifically by the absence of tau.

In addition, the delay in axonogenesis observed in hippocampal neurons from $Tau^{-/-}$ mice suggests that the absence of tau protein facilitates a non-neuronal phenotype. This observation agrees well with previous findings indicating a delayed maturation of neuronal precursors in $Tau^{-/-}$ mice [29]. Tau is not essential for the non-neural phenotype, probably because REST expression might be important for the correct temporal execution of neuronal differentiation programs, although it is not required for neuronal commitment per se [30].

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References

[1] Weingarten, M.D., Lockwood, A.H., Hwo, S.Y. and Kirschner, M.W. (1975) A protein factor essential for microtubule assembly. Proc. Natl. Acad. Sci. USA 72, 1858–1862.
[2] Drubin, D.G. and Kirschner, M.W. (1986) Tau protein function in living cells. J. Cell Biol. 103, 2739–2746.
[3] Panda, D., Goode, B.L., Feinstein, S.C. and Wilson, L. (1995) Kinetic stabilization of microtubule dynamics at steady state by tau and microtubule-binding domains of tau. Biochemistry 34, 11117–11127.
[4] Caceres, A. and Kosik, K.S. (1990) Inhibition of neurite polarity by tau antisense oligonucleotides in primary cerebellar neurons. Nature 343, 461–463.
[5] Harada, A. et al. (1994) Altered microtubule organization in small-calibre axons of mice lacking tau protein. Nature 369, 488–491.
[6] Dawson, H.N., Ferreira, A., Eyster, M.V., Ghoshal, N., Binder, L.I. and Vitek, M.P. (2001) Inhibition of neuronal maturation in primary hippocampal neurons from tau deficient mice. J. Cell Sci. 114, 1179–1187.
[7] Takei, Y., Teng, J., Harada, A. and Hirokawa, N. (2000) Defects in axonal elongation and neuronal migration in mice with disrupted tau and map1b genes. J. Cell Biol. 150, 989–1000.
[8] Hua, Q., He, R.Q., Haque, N., Qu, M.H., del Carmen Alonso, A., Grundke-Iqbal, I. and Iqbal, K. (2003) Microtubule associated protein tau binds to double-stranded but not single-stranded DNA. Cell Mol. Life Sci. 60, 413–421.
[9] Sjoberg, M.K., Shestakova, E., Mansuroglu, Z., Maccioni, R.B. and Bonnefoy, E. (2006) Tau protein binds to pericentromeric DNA: a putative role for nuclear tau in nucleolar organization. J. Cell Sci. 119, 2025–2034.
[10] Villasante, A., Corces, V.G., Manso-Martinez, R. and Avila, J. (1981) Binding of microtubule protein to DNA and chromatin: possibility of simultaneous linkage of microtubule to nucleic and assembly of the microtubule structure. Nucleic Acids Res. 9, 895–908.
[11] Wei, Y., Qu, M.H., Wang, X.S., Chen, L., Wang, D.L., Liu, Y., Hua, Q. and He, R.Q. (2008) Binding to the minor groove of the double-strand, tau protein prevents DNA from damage by peroxidation. PLoS One 3, e2600.
[12] Greenwood, J.A. and Johnson, G.V. (1995) Localization and in situ phosphorylation state of nuclear tau. Exp. Cell Res. 220, 332–337.
[13] Loomis, P.A., Howard, T.H., Castleberry, R.P. and Binder, L.I. (1990) Identification of nuclear tau isoforms in human neuroblastoma cells. Proc. Natl. Acad. Sci. USA 87, 8422–8426.
[14] Thurston, V.C., Pena, P., Pestell, R. and Binder, L.I. (1997) Nucleolar localization of the microtubule-associated protein tau in neuroblastomas using sense and anti-sense transfection strategies. Cell Motil. Cytoskeleton 38, 100–110.
[15] Wattler, F., Wattler, S., Kelly, M., Skinner, H.B. and Nehls, M. (1999) Cloning, chromosomal location, and expression analysis of murine *smarce1*-related, a

new member of the high-mobility 365 group gene family. Genomics 60, 172–178.

[16] Wang, W., Chi, T., Xue, Y., Zhou, S., Kuo, A. and Crabtree, G.R. (1998) Architectural DNA binding by a high-mobility-group/kinesin-like subunit in mammalian SWI/SNF-related complexes. Proc. Natl. Acad. Sci. USA 95, 492–498.

[17] Battaglioli, E., Andres, M.E., Rose, D.W., Chenoweth, J.G., Rosenfeld, M.G., Anderson, M.E. and Mandel, G. (2002) REST repression of neuronal genes requires components of the hSWI.SNF complex. J. Biol. Chem. 277, 41038–41045.

[18] Banker, G.A. and Cowan, W.M. (1977) Rat hippocampal neurons in dispersed cell culture. Brain Res. 126, 397–342.

[19] Gómez de Barreda, E. et al. (2010) Tau-knockout mice show reduced GSK3-induced hippocampal degeneration and learning deficits. Neurobiol. Dis. 37, 622–629.

[20] Rossi, G. et al. (2008) A new function of microtubule-associated protein tau: involvement in chromosome stability. Cell Cycle 7, 1788–1794.

[21] Kazantseva, A., Sepp, M., Kazantseva, J., Sadam, H., Pruunsild, P., Timmusk, T., Neuman, T. and Palm, K. (2009) N-Terminally truncated Baf57 isoforms contribute to the diversity of Swi/Snf complexes in neurons. J. Neurochem. 109, 807–818.

[22] Wang, W. et al. (1996) Purification and biochemical heterogeneity of the mammalian SWI-SNF complex. EMBO J. 15, 5370–5382.

[23] Dotti, C.G., Banker, G.A. and Binder, L.I. (1987) The expression and distribution of the microtubule-associated proteins tau and microtubule-associated protein 2 in hippocampal neurons in the rat in situ and in cell culture. Neuroscience 23, 121–130.

[24] Hartmann, J. and Konnerth, A. (2005) Determinants of postsynaptic Ca^{2+} signaling in Purkinje neurons. Cell Calcium 37, 459–466.

[25] Ballas, N., Grunseich, C., Lu, D.D., Speh, J.C. and Mandel, G. (2005) REST and its corepressors mediate plasticity of neuronal gene chromatin throughout neurogenesis. Cell 121, 645–657.

[26] Albrecht, M. and Lengauer, T. (2004) Novel Sm-like proteins with long C-terminal tails and associated methyltransferases. FEBS Lett. 569, 18–26.

[27] Fleischer, T.C., Weaver, C.M., McAfee, K.J., Jennings, J.L. and Link, A.J. (2006) Systematic identification and functional screens of uncharacterized proteins associated with eukaryotic ribosomal complexes. Genes Dev. 20, 1294–1307.

[28] Mann, S.S. and Hammarback, J.A. (1994) Molecular characterization of light chain 3. A microtubule binding subunit of MAP1A and MAP1B. J. Biol. Chem. 269, 11492–11497.

[29] Fuster-Matanzo, A., Gómez de Barreda, E., Dawson, H.N., Vitek, M.P., Avila, J. and Hernandez, F. (2009) Function of tau protein in adult newborn neurons. FEBS Lett. 583, 3063–3068.

[30] Jorgensen, H.F. et al. (2009) REST selectively represses a subset of RE1-containing neuronal genes in mouse embryonic stem cells. Development 136, 715–721.