Review

Implications of the thyroid hormone on neuronal development with special emphasis on the calmodulin-kinase IV pathway☆

Joachim Krebs

NMR-based Structural Biology, MPI for Biophysical Chemistry, Göttingen, Germany

ARTICLE INFO

ABSTRACT

Thyroid hormones influence brain development through regulation of gene expression. This is especially true for Ca^{2+}-dependent regulation since a major pathway is controlled by the Ca^{2+}/calmodulin-dependent protein kinase IV (CaMKIV) which in turn is induced by the thyroid hormone T_3. In addition, CaMKIV is involved in regulation of alternative splicing of a number of protein isoforms, among them PMCA1a, the neuronal specific isoform of the plasma membrane calcium pump. On the other hand, hypothyroidism or CaMKIV deficiency can have a severe influence on brain development. This article is part of a Special Issue entitled: ECS Meeting edited by Claus Heizmann, Joachim Krebs and Jacques Haiech.

© 2016 Elsevier B.V. All rights reserved.

1. Introduction

Growth, differentiation and maturation of the mammalian brain has been shown to be under the control of different growth factors and hormones, among them the thyroid hormone 3,3′,5-triiodo-L-thyronine or T_3 [1], the active form of the thyroid hormone. T_3 regulates the transcription of specific T_3-responsive genes through the T_3-receptor which recognizes regulatory elements of the expressed gene [2] typical for the steroid/thyroid hormone superfamily (for a review see [3]). The action of thyroid hormone during brain development can be manifold influencing different aspects of neurogenesis, but also glial development and differentiation (for a recent review see [4]). During the development of the fetal brain the availability of T_3 is essential in the sense that thyroid hormone deficiency, even for a short period of time, may lead to irreversible brain damage [5,6]. Since fetal thyroid hormone derives from the maternal thyroid gland well before the fetal thyroid gland is operating, maternal hypothyroidism can cause severe defects during fetal brain development [7–9]. This is especially critical for the hippocampal region since expression of the thyroid receptor and availability of T_3 are crucial for the development of the hippocampus [10]. So it has been shown that rat embryos hypothyroid during embryonic days between E12 and E15 displayed defective migration pattern of hippocampal neurons [11]. Another area of brain development under T_3 control concerns the cerebellum which in rodents is largely postnatal [12]. Therefore, perinatal hypothyroidism dramatically affects cerebellar

development, especially axodendritic connections between Purkinje cells and granular neurons (for reviews see [13,14]).

One of the major Ca^{2+} signaling pathways in brain development is under the control of the calmodulin-dependent protein kinase IV (CaMKIV) (for review see [15,16]). In 1996 [17] we provided evidence for the first time that the expression of CaMKIV during an early stage of brain development is specifically induced by T_3 in a concentration and time dependent manner using a fetal rat telencephalon primary cell culture system which can grow and differentiate under chemically defined conditions [18]. This finding was later confirmed by others [19,20] including the identification of a 5′-flanking region of the CaMKIV gene responsive to T_3 and binding the thyroid hormone receptor [20]. In this review I will discuss the importance of the CaMKIV-dependent Ca^{2+}-signaling pathways for brain function and possible consequences of hypothyroidism for the malfunction of the enzyme.

2. General properties of CaMKIV

CaMKIV belongs to the family of multifunctional calmodulin-dependent protein Ser/Thr kinases including also CaMKI, CaMKII and CaMKK [21]. CaMKIV, of which two isoforms exist (α and β), was first discovered in the granular cells of the cerebellum and therefore was named CaM-kinase Gr [22], but was later renamed as CaM-Kinase IV (CaMKIV) [23,24]. The enzyme is highly expressed in specific tissues, especially in brain (here next to the granular cells of the cerebellum [22], also in other neuronal subpopulations, especially in neuron nuclei of the hippocampus [25]). To a lesser extent the enzyme is also detected in the thymus (here particularly in T-lymphocytes, not in B-lymphocytes or monocytes [26]), in testis, ovary, and in bone marrow-derived cells

[21]. It is predominantly located in the nucleus [16], but can also be found in the cytoplasm. CaMKIV is conserved among higher organisms including zebrafish [27] and *C. elegans* [28].

3. CaMKIV activity and its function in Ca^{2+}-dependent gene expression and alternative splicing

Like other protein kinases also CaMKIV is inhibited in the resting state by an autoinhibitory segment. The regulation of its activity is tightly controlled and a rather complex process [15,16,21]. The autoinhibition of the enzyme is released by binding of calmodulin in a Ca^{2+}-dependent manner leading to a basal activity of the enzyme. Due to binding of calmodulin CaMKIV undergoes a conformational change exposing the activation loop which contains a threonine (T^{200} in human, T^{196} in mouse [16]) to be phosphorylated by calmodulin-dependent protein kinase-kinase (CaMKK) [29]. This results in a 10–20 fold activation of CaMKIV [30]. Dephosphorylation of CaMKIV and thereby inactivation of the enzyme is mediated by protein phosphatase 2A (PP2A) which builds a tight complex with CaMKIV [31,32].

One of the main functions of CaMKIV is regulating Ca^{2+}-dependent gene expression. Therefore some of the major substrates are transcription factors such as CREB (cAMP/Ca^{2+} response element binding protein) [33–35], SRF (serum response factor) [36] and others. CREB is phosphorylated at Ser133 by the activated CaMKIV [33] enabling CREB to interact with the transcription activator CBP, the CREB binding protein [37]. In this way the recruitment of CBP to the promoter region of CREB target genes regulates CRE-mediated transcription [25,38].

In recent years it became evident that CaMKIV is also involved in the regulation of another important Ca^{2+} signaling pathway, i.e. alternative splicing [39]. This could became especially important for regulating the expression of the PMCA1a spliced isoform as outlined in detail below.

4. Alternative splicing of PMCA, the calcium pump of the plasma membrane

The plasma membrane calcium pump (PMCA) is one of the key player to control Ca^{2+} homeostasis of eucaryotic cells [40,41]. It is responsible for the fine tuning of the Ca^{2+} level in the cell, especially in selected sub-plasma membrane domains [42]. PMCA activity is regulated in many ways [41], but its major activator is the Ca^{2+}-binding protein calmodulin which interacts directly with PMCA through a defined calmodulin-binding domain [43], usually located at the C-terminus of the enzyme. After determination of the primary structure of PMCA from human [44] and rat [45] tissues it became clear that the enzyme is an essential component of all mammalian plasma membranes deriving from four different genes [46]. Additional isoforms of the protein are produced by alternative splicing of the primary transcripts [46,47]. For all PMCA genes two splice cites have been characterized (sites A and C [46]) located close to regulatory regions of the pump (Fig. 1). About 30 different spliced isoforms have been identified [48]. In the context of this review splice site C of PMCA is of special interest. This site is located in the middle of the calmodulin binding domain [49] giving rise to different spliced isoforms for the 4 different gene products. As will be clear later, splicing of PMCA1 at site "C" will display some special features which will be of interest for the development of the embryonal mammalian brain. As can be seen from Fig. 1 the splicing products 1a,b,c,d,e can be obtained. PMCA1b is the housekeeping form which can be found in all mammalian tissues [48,49]. The importance of PMCA1 is underlined by the fact that PMCA1 knock out mice are embryonically lethal [50] in contrast to the other ubiquitously expressed isoform, PMCA4, which exhibits no embryolethality [50]. In addition, in fetal brain development PMCA1 can be detected as early as day 9 or 10 dpc in mice or rats, respectively [48,49]. As can be noted from Fig. 1 the 1b isoform excludes an exon which has been identified as exon
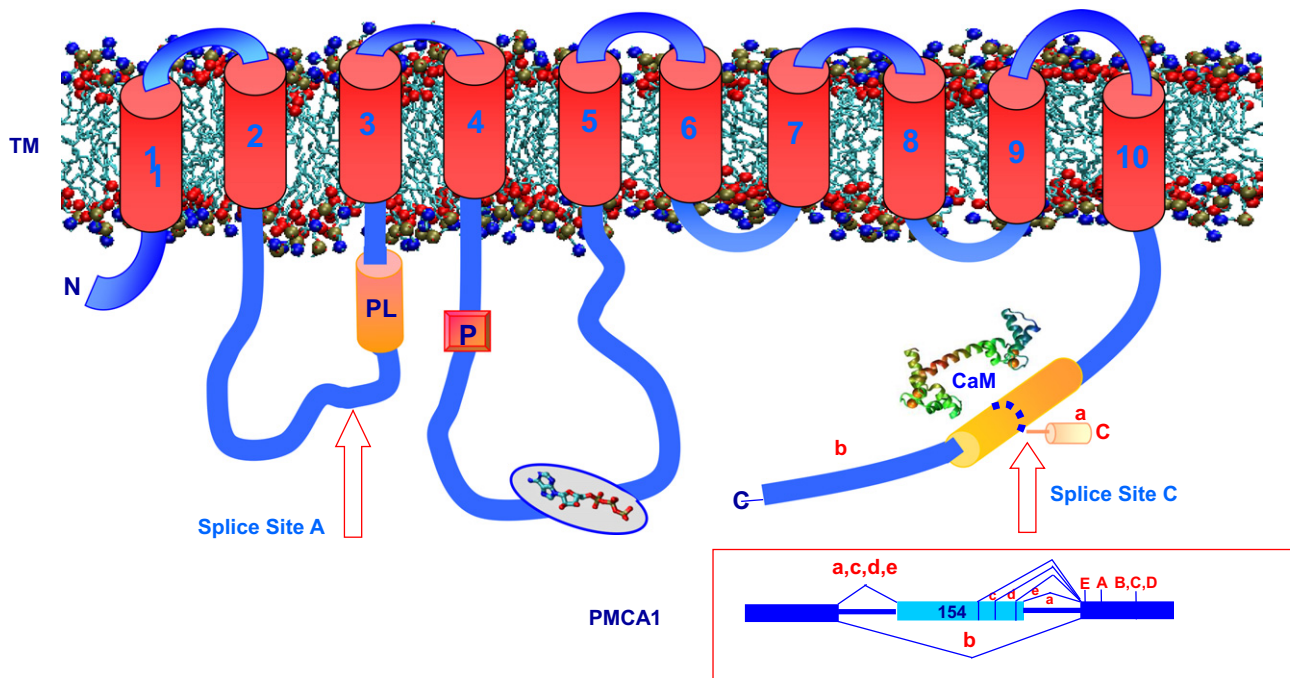

Fig. 1. Topology domains and splicing variants of the human PMCA1 isoform. The 10 transmembrane domains (TM) of the pump are numbered and indicated by red boxes. Splice sites "A" (first cytosolic loop) and "C" (C-terminal tail) are indicated by red arrows. Splice site "C" lies within the calmodulin-binding domain (yellow cylinder; defined by the structural model of CaM = calmodulin). The exon structure of splice site "C" affected by alternative splicing is shown for the PMCA1 gene. Constitutively spliced exons are indicated as dark blue boxes, alternatively inserted exons are shown in light blue; the resulting splice variants are labeled by their lower case symbols, the positions of the translation stop codons for each splice form are indicated by the corresponding capital letters. The sizes of the alternatively spliced exon is given as nucleotide numbers. PL = phospholipid binding domain; P = location of the aspartyl-phosphate formation. The figure was adopted from Fig. 1 of Ref. [46].

21 [47,50]. This results in an intact calmodulin-binding domain and a C-terminal sequence, highly conserved among the "b" isoforms of the different PMCA genes [46,48,51] since the reading frame remains intact. On the other hand, isoforms 1a,c,d,e include exon 21 containing 154 nucleotides in the entire exon. 1a and 1e include either all 154 nucleotides (1a) or 148 (1e), both isoforms resulting in a frame shift with a shorter C-terminal sequence due to an early stop codon, and a significantly altered calmodulin binding domain with a different calmodulin affinity. Due to internal cryptic donor splice sites within exon 21 alternative splicing can give rise to isoforms 1c (114 nt) and 1d (87 nt), both resulting in an intact reading frame [51].

5. Expression of PMCA1a in different brain regions and its possible regulation

As mentioned before PMCA1 can be detected in the brain at very early stages of its development [48,52]. Brandt and Neve [52] already noticed that in rat brain isoform PMCA1b is visible first, expression of isoform PMCA1a was later detectable at E16 [52]. These authors also noticed that during rat brain development into postnatal days the expression of the mRNA of PMCA1b declined whereas the expression of the mRNA of the PMCA1a isoform was steadily increasing suggesting that this isoform was important for the maturation of neurons and building of synaptic plasticity [52]. This view was later confirmed by several detailed studies [53–55]. As reported by Filoteo et al. [53] using antibodies specific for the different spliced isoforms PMCA1a was mainly detected in frontal cortex followed by the hippocampus and less identified in cerebellum. In contrast, PMCA1b was practically not detectable in those regions investigated. Kenyon et al. [54] pointed out that PMCA1 can be detected in all brain regions represented mainly by the spliced isoform 1a. As already reported by Brandt and Neve [52] PMCA1b is expressed at early stages of development of the rat brain but is progressively replaced by PMCA1a during the maturation process of the brain until it reaches a steady state level after birth. Even if Kenyon et al. [54] confirmed that PMCA1a is present at synapses they also reported that PMCA1a is expressed at higher levels in the soma or dendritic compartments of many neurons, but is absent in non-neuronal cells. It is interesting to note that in the cerebral cortex Kenyon et al. noticed an uneven distribution of PMCA1a indicating a possible concentration of the enzyme in specialized microdomains of the plasma membrane [54]. These authors also reported the presence of PMCA1a in all hippocampal regions indicating that the staining was much stronger in CA1 than in CA2 or 3. Using primary cultures derived from hippocampal neurons of 18 days old rat embryos Kip et al. [55] reported a similar shift in expression from PMCA1b to 1a during the development of hippocampal neurons. Interestingly, for PMCA2b which is abundantly expressed in hippocampal neurons such a shift to the spliced isoform 2a was not observed [55]. It is further of interest that next to a number of other areas of the brain PMCA1a was strongly expressed in the cerebellum, especially in the granular cells [55]. The most intriguing result reported by Kenyon et al. [54] was the finding that PMCA1b can only be detected early in the developing brain, but as soon as neurons mature and synapse plasticity increases isoform expression is switched from PMCA1b to 1a, even if PMCA1a is not directly involved in synapse-related processes. On the other hand, the neuron-specific replacement of PMCA1b by the 1a spliced isoform during brain development may be related to the specific differences in handling Ca^{2+} signaling. As already pointed out before, the two isoforms differ in their calmodulin-dependent regulation since PMCA1a due to alternative splicing results in a change in the reading frame within the middle of the calmodulin-binding domain leading to significant differences in the affinity for calmodulin between PMCA1b and 1a. This results in a low basal Ca^{2+} activity and high calmodulin sensitivity for PMCA1b in contrast to PMCA1a which has a much higher basal Ca^{2+} activity similar to the differences reported for PMCA4a and 4b [56]. Therefore it is of considerable interest to get information how this switch from PMCA1b to 1a is regulated

during brain development. Here I will discuss a possible mechanism how this change could occur.

6. Regulation of alternative splicing by CaMKIV

Black and his coworkers demonstrated that Ca^{2+}-dependent regulation of alternative splicing is under the control of CaMKIV [57–59]. In 2001 Xie and Black reported [57] that the splicing of the stress axis-regulated exon (STREX) of the calcium-activated potassium channel is repressed through the action of CaMKIV. They identified a CaMKIV-responsive RNA element (CaRRE1) mediating the alternative splicing of the pre-mRNA. Later it was shown [58,59] that the alternative splicing of the STREX exon occurs also in neurons controlled by the CaMKIV pathway [58]. Here the authors identified a CaMKIV-responsive consensus sequence (CACATNRTTAT) in a number of human genes (including PMCA1a, see below) responding to CaMKIV [58]. CaRRE1 is located at the 3'splice site which is a highly conserved region of introns [60]. The CA/AC rich motif is known to be critical for the binding of heterogenous nuclear ribonucleoproteins as trans-acting factors [61]. One of these hnRNPs, hnRNP L, has been identified as an essential component to suppress the STREX exon of the Ca^{2+}-dependent potassium channel in a CaMKIV dependent manner [62]. Important for this regulation is the phosphorylation of a conserved serine residue of hnRNP L by CaMKIV [63]. By examining the human genome Black and his co-workers [59] established $CACA(T/C)N_{1-4}A$ as a minimum consensus sequence matching the CARRE motif in a number of exons. These authors induced differentiation in P19 cells by depolarization obtaining cells with neuronal morphology. One of the spliced exons identified in these screening experiments was exon 21 of PMCA1 which was spliced in a CaMKIV dependent manner [59]. By analysing the pre-mRNA of PMCA1 a CARRE motif (CACATGTA) could be identified in exon 21 (see Ref. [47]). As shown by Strehler et al. [47] exon 21 of PMCA1 contains 2 internal splice donor sites with the CARRE consensus sequence located at the second site. If PMCA1a has to be expressed the total sequence of 154 nucleotides of exon 21 have to be included which means the 2 internal spliced donor sites have to be suppressed. This could be achieved by CaMKIV recognizing the CARRE consensus sequence at the second splice donor site, site one could be suppressed by the secondary structure of the pre-mRNA since it is known that those structures can influence the regulation of alternative splicing [64]. As already shown by the group of Black [59] expression of PMCA1a is increasing in a CaMKIV dependent way the more the P19 cells differentiate during depolarization. Similar observations have been made by Guerini et al. [65] for cerebellar granular cells which differentiate by depolarization in a Ca^{2+} dependent way inducing an increased expression of PMCA1a. By analysing the sequences of the other PMCA genes it was interesting to note that the CaMKIV-responsive CARRE motif could only be identified in PMCA1 indicating that such a CaMKIV-dependent regulation of alternative splicing is unique for PMCA1.

What could be the advantage for the brain development to switch from PMCA1b to PMCA1a as neuronal differentiation improves? This is most certainly connected with the significant difference in the C-terminal amino acid sequence between the 2 isoforms since due to the inclusion of exon 21 in PMCA1a the reading frame has changed leading to an early stop codon and a significantly reduced C-terminal sequence (see Ref. [48]). In addition, the calmodulin sensitivity for PMCA1a is much lower, but the basal Ca^{2+} activity is higher for PMCA1a. This might be an advantage for fast spiking neurons since PMCA1a could be more efficient in fast changing conditions to control Ca^{2+} homeostasis. This conclusion is supported by findings that if in fast spiking interneurons in which parvalbumin (PV) plays an essential role as Ca^{2+} buffer (for a recent review see [66]) PV is knocked down Ca^{2+} extrusion systems are increased, especially PMCA [67] indicating an important control function for PMCA1a in those neurons.

As indicated before the availability of the thyroid hormone T_3 is critical for the development of the brain. So it has been demonstrated by

Morte et al. [19] that gene expression in the developing brain of rats controlled by the CaMKIV signaling pathway is regulated by thyroid hormone. Since it is well known that the developing fetal brain is totally dependent on the maternal supply of the thyroid hormone [7,68] the findings reported by Zhang et al. [8] are of interest. These authors demonstrated that due to maternal hypothyroidism the expression of CaMKIV and its activating kinase CaMKK have been significantly reduced leading to the impairment of the CREB-dependent signaling pathways which are associated with synaptic plasticity, learning and memory [8]. Since the expression of PMCA1a is controlled by CaMKIV it can be assumed that a significant reduction of CaMKIV and its controlled pathways may lead to a severe reduction of PMCA1a which could impair Ca^{2+} homeostasis during neuronal development and synaptic formation. Similar conclusions could be made considering observations made with CaMKIV$^{-/-}$ mice which show severe hippocampal deficits influencing memory function [69] and cerebellar malfunction [70].

7. Consequences of CaMKIV$^{-/-}$ for neural development

CaMKIV as well as PMCA1a are highly expressed in the cerebellum and in hippocampus as two examples of well studied areas of the brain [22,25,53,54]. By investigating CaMKIV null mice Ribar et al. reported [71] locomotor defects indicating a malfunction of the cerebellum. The authors further observed a severe reduction of mature Purkinje neurons together with a significant decrease of p-CREB, phosphorylated at Ser-133 [71], a major target of CaMKIV to control Ca^{2+}-dependent gene transcription [25] and synaptic transmission. In a later study Means and his coworkers [72] reported a similar impairment for the development of cerebellar granule cells in mice deficient of either CaMKIV or CaMKK2, the activator kinase of CaMKIV. These results underline the importance of Ca^{2+} signaling via the CaMKK2-CaMKIV-pCREB pathway for the synergistic development and maturation of cerebellar granule and Purkinje cells to be able to form synaptic connections with each other.

Also in the hippocampus the CaMKIV regulated pathway plays a critical role to enable synaptic plasticity and cognitive functions such as learning and memory [73] since CaMKIV null mice [69] or transgenic mice containing a dominant-negative form of CaMKIV [74] displayed severe hippocampal defects resulting in an impairment of memory function. An interesting aspect was recently discussed by Tsien and his co-workers [75] who investigated CaMK pathways in parvalbumin (PV) containing GABAergic hippocampal neurons (PV(+)). Their activity is crucial for the morphological development and the synaptic plasticity of the hippocampus. Cohen et al. reported [75] that CaMKIV is responsible for the rate-limiting phosphorylation of CREB in PV(+) cells thereby regulating the dynamics of Ca^{2+} signaling in fast-firing PV(+) cells [75]. In this context it is interesting to note that grid cells in the medial entorhinal cortex which are responsible for spatial orientation target PV(+) interneurons to tune their firing activity [76].

In summary, I outlined in this review that the CaMKIV controlled pathway is essential for Ca^{2+}-dependent gene expression. This is important for neuronal development contributing to synaptic plasticity and cognitive functions such as learning and memory [77]. In addition, CaMKIV is also a unique regulator of alternative splicing [78] which becomes important for the expression of the spliced isoform PMCA1a [39], the latter has the special function to control Ca^{2+} homeostasis in the developing brain [54]. Both regulatory roles of CaMKIV are probably dependent on the induction by the thyroid hormone T_3 [17] (see Fig. 2) since maternal hypothyroidism results in a significant reduction of CaMKIV and CaMKK in the fetal brain [8] leading to the impairment of its neurodevelopment. Therefore it would be important to analyse in more detail the role of PMCA1a during brain development, whether its expression is influenced by maternal hypothyroidism or by deficiency in CaMKIV or CaMKK2 expression, i.e. what consequence has an impairment of PMCA1a expression for brain development.
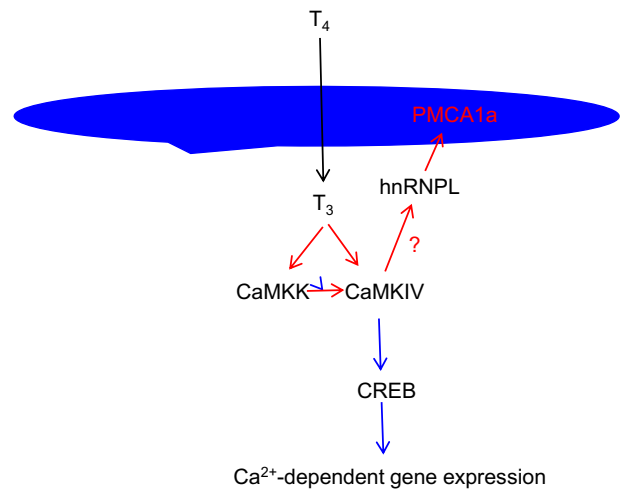

Fig. 2. Schematic view of different signal transduction pathways of CaMKIV as induced by the thyroid hormone T_3. The question mark indicating that hnRNPL could be the *trans*-acting factor under the control of CaMKIV to induce the splicing of PMCA1 resulting in PMCA1a is a possible assumption. This is based on the findings reported by Xie and co-workers who demonstrated that the STREX exon of the Ca^{2+}-dependent potassium channel is suppressed in a CaMKIV/hnRNPL dependent manner [62,63]. The following abbreviations have been used: CaMKIV = Ca^{2+}/calmodulin-dependent protein kinase IV; CaMKK = Ca^{2+}/calmodulin-dependent protein kinase kinase; CREB = cAMP/Ca^{2+}-dependent regulatory element binding protein; hnRNPL = heterogenous nuclear ribonucleoprotein L.

Conflict of interest

The author declares no conflicts of interest.

Transparency document

The Transparency document associated with this article can be found, in online version.

References

[1] R.S. Balazs, S. Kovacs, W.A. Cocks, A.L. Johnson, J.-T. Eayrs, Effect of thyroid hormone on the biochemical maturation of rat brain: postnatal brain formation, Brain Res. 25 (1971) 555–570.

[2] N. Viguerie, D. Langin, Effect of thyroid hormone on gene expression, Curr. Opin. Clin. Nutr. Metab. Care 6 (2003) 377–381.

[3] R.M. Evans, The steroid and thyroid hormone receptor superfamily, Science 240 (1988) 889–895.

[4] J. Bernal, in: L.J. De Groot, et al., (Eds.), Thyroid Hormones in Brain Development and Function, MDText.com.Inc., South Dartmouth (MA) 2015, pp. 2000–2015.

[5] K.W. Anderson, C.M. Schoonover, S.A. Jones, Control of thyroid hormone action in the developing rat brain, Thyroid 13 (2003) 1039–1056.

[6] G. Morreale de Escobar, M.J. Obregon, F. Escobar del Rey, Role of thyroid hormone during early brain development, Eur. J. Endocrinol. 151 (Suppl. 3) (2004) (U25–37).

[7] G. Morreale de Escobar, R. Pastor, M.J. Obregon, F. Escobar del Rey, Effect of maternal hypothyroidism on the weight and thyroid hormone content of rat embryonic tissues, Endocrinology 117 (1985) 1890–1901.

[8] Y. Zhang, Y. Fan, X. Yu, X. Wang, S. Bao, J. Li, C. Fan, Z. Shan, W. Teng, Maternal subclinical hypothyroidism impairs neurodevelopment in rat offspring by inhibiting the CREB signaling pathway, Mol. Neurobiol. 52 (2015) 432–441.

[9] H. Min, J. Dong, Y. Wang, Y. Wang, W. Teng, Q. Xi, J. Chen, Maternal Hypothyroxinemia-induced neurodevelopmental impairments in the progeny, Mol. Neurobiol. 53 (2016) 1613–1624.

[10] J. Gong, W. Liu, J. Dong, Y. Wang, H. Xu, W. Wei, J. Zhong, Q. Xi, J. Chen, Developmental deficiency and hypothyroidism impair neural development in rat hippocampus: involvement of doublecortin and NCAM-180, BMC Neurosci. 11 (2010) 50–59.

[11] E. Auso, R. Lavado-Autric, E. Cuevas, F.E. del Rey, G. Morreale de Escobar, P. Berbel, A moderate and transient deficiency of maternal thyroid function at the beginning of fetal neocortigogenesis alters neuronal migration, Endocrinology 145 (2004) 4037–4047.

[12] J. Altman, Morphological development of the rat cerebellum and some of its mechanisms, Exp. Brain Res. 16 (1982) 8–49.

[13] J. Legrand, Morphogenetic actions of thyroid hormones, Trends Neurosci. 2 (1979) 234–236.

[14] G.W. Anderson, Thyroid hormone and cerebellar development, Cerebellum 7 (2008) 60–74.

[15] J. Krebs, Calmodulin-dependent protein kinase IV: regulation of function and expression, Biochim. Biophys. Acta 1448 (1998) 183–189.

[16] H. Naz, A. Islam, F. Ahmad, M.I. Hassan, Calcium/calmodulin-dependent protein kinase IV: a multifunctional enzyme and potential therapeutic target, Prog. Biophys. Mol. Biol. 121 (2016) 54–65, http://dx.doi.org/10.1016/j.pbiomolbio.2015.12.016.

[17] J. Krebs, R.L. Means, P. Honegger, Induction of calmodulin kinase IV by the thyroid hormone during the development of rat brain, J. Biol. Chem. 271 (1996) 11055–11058.

[18] P. Honegger, D. Lenoir, P. Favrod, Growth and differentiation of aggregating fetal brain cells in a serum-free defined medium, Nature 282 (1979) 305–308.

[19] B. Morte, et al., Thyroid hormone regulation of gene expression in the developing rat fetal cerebral cortex: prominent role of the Ca2+/calmodulin-dependent protein kinase pathway, Endocrine 151 (2010) 810–820.

[20] Y.-Y. Liu, G.A. Brent, A complex deoxyribonucleic acid response element in the rat Ca2+/calmodulin-dependent protein kinase IV gene 5′-flanking region mediates thyroid hormone induction and chicken ovalbumin upstream promoter transcription factor 1 repression, Mol. Endocrinol. 16 (2002) 2439–2451.

[21] F.A. Chow, A.R. Means, The calcium/calmodulin-dependent protein kinase cascade, in: J. Krebs, M. Michalak (Eds.), Calcium, a Matter of Life or Death, Elsevier, Amsterdam 2007, pp. 345–364.

[22] C.A. Ohmstede, K.F. Jensen, N.E. Sahyoun, Ca2+/calmodulin-dependent protein kinase enriched in cerebellar granule cells. Identification of a novel neuronal calmodulin-dependent protein kinase, J. Biol. Chem. 264 (1989) 5866–5875.

[23] J.M. Sikela, M.L. Law, F.-T. Kao, J.A. Hartz, Q. Wei, W.H. Hahn, Chromosomal localization of the human gene for brain Ca2+/calmodulin-dependent protein kinase type IV, Genomics 4 (1989) 21–27.

[24] A.R. Means, F. Cruzalegui, B. LeMagueresse, D. Needleman, G. Slaughter, T. Ono, A novel Ca2+/calmodulin-dependent protein kinase and a male germ cell-specific calmodulin-binding protein are derived from the same gene, Mol. Cell. Biol. 11 (1991) 3960–3971.

[25] H. Bito, K. Deisseroth, R.W. Tsien, CREB Phosphorylation and dephosphorylation: a Ca(2+)-and stimulus duration-dependent switch for hippocampal gene expression, Cell 87 (1996) 1203–1214.

[26] H. Hanissian, M. Frangakis, M.M. Bland, S. Jawahar, T.A. Chatila, Expression of a Ca2+/calmodulin-dependent protein kinase, CaM kinase-Gr, in human T lymphocytes. Regulation of kinase activity by T cell receptor signaling, J. Biol. Chem. 268 (1993) 20055–20063.

[27] T. Nimura, Y. Sugiyama, N. Sueyoshi, Y. Shigery, A. Ishida, I. Kameshita, A minimum size homologue of Ca2+/calmodulin-dependent protein kinase IV naturally occuring in zebrafish, J. Biochem. 147 (2010) 857–865.

[28] Y. Kimura, et al., A CaMK cascade activates CRE-mediated transcription in neurons of Caenorhabditis elegans, EMBO Rep. 3 (2002) 962–966.

[29] M.A. Selbert, K.A. Anderson, Q.H. Huang, E.G. Goldstein, A.R. Means, A.M. Edelman, Phosphorylation and activation of Ca(2+)-calmodulin-dependent protein kinase IV by Ca(2+)-calmodulin-dependent protein kinase 1a kinase. Phosphorylation of threonine 196 is essential for activation, J. Biol. Chem. 270 (1995) 17616–17621.

[30] H. Tokumitsu, G.A. Wayman, M. Muramatsu, T.R. Soderling, Calcium/calmodulin-dependent protein kinase kinase: identification of regulatory domains, Biochemistry 36 (1997) 12823–12827.

[31] R.S. Westphal, K.A. Anderson, A.R. Means, B.E. Wadzinski, A signaling complex of Ca2+−calmodulin-dependent protein kinase IV and protein phosphatase 2A, Science 280 (1998) 1258–1261.

[32] K.A. Anderson, P.K. Noeldner, K. Reece, B.E. Wadzinski, A.R. Means, Regulation and function of the calcium/calmodulin-dependent protein kinase IV/protein serine/threonine phosphatase 2A signaling complex, J. Biol. Chem. 279 (2004) 31708–31716.

[33] R.P. Matthews, C.R. Guthrie, L.M. Wailes, X. Zhao, A.R. Means, G.S. McKnight, Calcium/calmodulin-dependent protein kinase types II and IV differentially regulate CREB-dependent gene expression, Mol. Cell. Biol. 14 (1994) 6107–6116.

[34] H. Enslén, P. Sun, D. Brickey, S.H. Soderling, E. Klamo, T.R. Soderling, Characterization of Ca2+/calmodulin-dependent protein kinase IV. Role in transcriptional regulation, J. Biol. Chem. 269 (1994) 15520–15527.

[35] M. Sheng, M.A. Thompson, M.E. Greenberg, CREB: a Ca(2+)-regulated transcription factor phosphorylated by calmodulin-dependent kinases, Science 252 (1991) 1427–1430.

[36] C.K. Miranti, D.D. Ginty, G. Huang, T. Chatila, M.E. Greenberg, Calcium activates serum response factor-dependent transcription by a Ras and Elk-1 independent mechanism that involves a Ca2+/calmodulin-dependent kinase, Mol. Cell. Biol. 15 (1995) 3672–3684.

[37] J.C. Chrivia, R.P. Kwok, N. Lamb, M. Hagiwara, M.R. Montminy, R.H. Goodman, Phosphorylated CREB binds specifically to the nuclear protein CBP, Nature 365 (1993) 855–859.

[38] H. Fukushima, R. Maeda, R. Suzuki, A. Suzuki, et al., Upregulation of calcium/calmodulin-dependent protein kinase IV improves memory formation and rescues memory loss with aging, J. Neurosci. 28 (2008) 9910–9919.

[39] J. Krebs, The influence of calcium signaling on the regulation of alternative splicing, Biochim. Biophys. Acta 1793 (2009) 979–984.

[40] J. Krebs, L.B. Agellon, M. Michalak, Ca2+ homeostasis and endoplasmic reticulum (ER) stress: an integrated view of calcium signaling, Biochem. Biophys. Res. Commun. 460 (2015) 114–121.

[41] M. Brini, T. Cali, D. Ottolini, E. Carafoli, The plasma membrane calcium pump in health and disease, FEBS J. 280 (2013) 5385–5397.

[42] R. Lopreiato, M. Giacomello, E. Carafoli, The plasma membrane calcium pump: new ways to look at an old enzyme, J. Biol. Chem. 289 (2014) 10261–10268.

[43] P. James, M. Maeda, R. Fischer, A.K. Verma, J. Krebs, J.T. Penniston, E. Carafoli, Identification and primary structure of a calmodulin binding domain of the Ca2+ pump of human erythrocytes, J. Biol. Chem. 263 (1988) 2905–2910.

[44] A.K. Verma, et al., Complete primary structure of a human plasma membrane Ca2+ pump, J. Biol. Chem. 263 (1988) 14152–14159.

[45] G.E. Shull, J. Greeb, Molecular cloning of two isoforms of the plasma membrane Ca2+−transporting ATPase from rat brain. Structural and functional domains exhibit similarity to Na+, K+− and other cation transport ATPases, J. Biol. Chem. 263 (1988) 8646–8657.

[46] J. Krebs, The plethora of PMCA isoforms: alternative splicing and differential expression, Biochim. Biophys. Acta 1853 (2015) 2018–2024.

[47] E.E. Strehler, M.-A. Strehler-Page, G. Vogel, E. Carafoli, mRNAs for plasma membrane calcium pump isoforms differing in their regulatory domain are generated by alternative splicing that involves two internal donor sites in a single exon, Proc. Natl. Acad. Sci. U. S. A. 86 (1989) 6908–6912.

[48] E.E. Strehler, D.A. Zacharias, Role of alternative splicing in generating isoform diversity among plasma membrane calcium pumps, Physiol. Rev. 81 (2001) 21–50.

[49] T.P. Stauffer, H. Hilfiker, E. Carafoli, E.E. Strehler, Quantitative analysis of alternative splicing options of human plasma membrane calcium pump genes, J. Biol. Chem. 268 (1993) 25993–26003.

[50] G.W. Okunade, M.L. Miller, G.J. Pyne, R.L. Sutliff, K.T. O'Connor, J.C. Neumann, A. Andringa, D.A. Miller, V. Prasad, T. Doetschman, R.J. Paul, G.E. Shull, Targeted ablation of plasma membrane Ca2+−ATPase (PMCA) 1 and 4 indicates a major housekeeping function for PMCA1and a critical role in hyperactivated sperm motility and male fertility for PMCA4, J. Biol. Chem. 279 (2004) 33742–33750.

[51] H. Hilfiker, M.-A. Strehler-Page, T.P. Stauffer, E. Carafoli, E.E. Strehler, Structure of the gene encoding the human plasma membrane calcium pump isoform 1, J. Biol. Chem. 268 (1993) 19717–19725.

[52] P. Brandt, R.L. Neve, Expression of plasma membrane calcium-pumping ATPase mRNAs in developing rat brain and adult brain subregions: evidence for stage-specific expression, J. Neurochem. 59 (1992) 1566–1569.

[53] A.G. Filoteo, N.L. Elwess, A. Enyedi, A. Caride, H.H. Aung, J.T. Penniston, Plasma membrane Ca2+ pump in rat brain. Patterns of alternative splices seen by isoform specific antibodies, J. Biol. Chem. 272 (1997) 23741–23747.

[54] K.A. Kenyon, E.A. Bushong, A.S. Mauer, E.E. Strehler, R.J. Weinberg, A.C. Burette, Cellular and subcellular localization of the neuron-specific plasma membrane calcium ATPase PMCA1a in the rat brain, J. Comp. Neurol. 518 (2010) 3169–3183.

[55] S.N. Kip, N.W. Gray, A. Burette, A. Canbay, R.J. Weinberg, E.E. Strehler, Changes in the expression of plasma membrane calcium extrusion systems during the maturation of hippocampal neurons, Hippocampus 16 (2006) 20–34.

[56] A.J. Caride, A.G. Filoteo, J.T. Penniston, E.E. Strehler, The plasma membrane Ca2+ pump isoform 4a differs from isoform 4b in the mechanism of calmodulin binding and activation kinetics. Implications for Ca2+ signaling, J. Biol. Chem. 282 (2007) 25640–25648.

[57] J. Xie, D.L. Black, A CaMKIV responsive RNA element mediates depolarization-induced alternative splicing in ion channels, Nature 410 (2001) 936–939.

[58] J. Xie, C. Jan, P. Stoilov, J. Park, D.L. Black, A consensus CaMKIV-responsive RNA sequence mediates regulation of alternative exons in neurons, RNA 11 (2005) 1825–1834.

[59] J.A. Lee, Y. Xing, D. Nguyen, J. Xie, C.J. Lee, D.L. Black, Depolarization and CaMKinase IV modulate NMDA receptor splicing through two essential RNA elements, PLoS Biol. 5 (2007) e40, http://dx.doi.org/10.1371/journal.pbio.0050040.

[60] A. Razanau, J. Xie, Emerging mechanisms and consequences of calcium regulation of alternative splicing in neurons and endocrine cells, Cell. Mol. Life Sci.: CMLS 70 (2013) 4527–4536.

[61] A. Busch, K.J. Hertel, Evolution of SR protein and hnRNP splicing regulatory factors, 3, Wiley Interdiscip. Rev. RNA, 2012 1–12.

[62] J. Yu, Y. Hai, G. Liu, T. Fang, S.K.P. Kung, J. Xie, The heterogenous nuclear ribonucleoprotein L is an essential component in the Ca2+/calmodulin-dependent protein kinase IV-regulated alternative splicing through cytidine-adenosine repeats, J. Biol. Chem. 284 (2009) 1505–1513.

[63] G. Liu, A. Razanau, Y. Hai, J. Yu, M. Sohail, V.G. Lobo, J. Chu, S.K. Kung, J. Xie, A conserved serine of heterogeneous nuclear ribonucleoprotein L (hnRNP L) mediates depolarization-regulated alternative splicing of potassium channels, J. Biol. Chem. 287 (2012) 22709–22716.

[64] P.J. Shepard, K.J. Hertel, Conserved RNA secondary structures promote alternative splicing, RNA 14 (2008) 1463–1469.

[65] D. Guerini, E. Garcia-Martin, A. Gerber, C. Volbracht, M. Leist, C. Gutiérrez-Merino, E. Carafoli, The expression of plasma membrane Ca2+ pump isoforms in cerebellar granule neurons is modulated by Ca2+, J. Biol. Chem. 274 (1999) 1667–1676.

[66] H. Hu, J. Gian, P. Jonas, Fast-spiking, parvalbumin GABAergic interneurons: from cellular design to microcircuit function, Science 345 (2014) 529–542.

[67] B. Schwaller, Cytosolic Ca2+ buffers, Cold Spring Harb. Perspect. Biol. 2 (2010) a004051, http://dx.doi.org/10.1101/cshperspect.a004051.

[68] G. Morreale de Escobar, R. Calvo, M. Obregon, F. Escobar del Rey, Contribution of maternal thyroxine to fetal thyroxine pools in normal rats near term, Endocrinology 126 (1990) 2765–2767.

[69] F. Wei, C.-S. Qiu, J. Liauw, D.A. Robinson, N. Ho, T. Chatila, M. Zhuo, Calcium-calmodulin-dependent protein kinase IV is required for fear memory, Nat. Neurosci. 5 (2002) 573–579.

[70] N. Ho, et al., Impaired synaptic plasticity and cAMP response element-binding protein activation in Ca2+/calmodulin-dependent protein kinase type IV/Gr-deficient mice, J. Neurosci. 20 (2000) 6459–6472.

[71] T.J. Ribar, R.M. Rodriguiz, L. Khiroug, W.C. Wetsel, G.J. Augustine, A.R. Means, Cerebellar defects in Ca2 +/calmodulin kinase IV-deficient mice, J. Neurosci. 20 (2000) RC107.

[72] M. Kokubo, M. Nishio, T.J. Ribar, K.A. Anderson, A.E. West, A.R. Means, BDNF-mediated cerebellar granule cell development is impaired in mice null for CaMKK2 or CaMKIV, J. Neurosci. 29 (2009) 8901–8913.

[73] K. Deisseroth, H. Bito, R.W. Tsien, Signaling from synapse to nucleus: postsynaptic CREB phosphorylation during multiple forms of hippocampal synaptic plasticity, Neuron 16 (1996) 89–101.

[74] H. Kang, L.D. Sun, C.M. Atkins, T.R. Soderling, M.A. Wilson, S. Tonegawa, An important role of neuronal-dependent CaMKIV signaling in the consolidation of long-term memory, Cell 106 (2001) 771–783.

[75] S.M. Cohen, H. Ma, K.V. Kuchibhotla, B.O. Watson, G. Buzsaki, R.C. Froemke, R.W. Tsien, Excitation-transcription coupling in parvalbumin-positive interneurons employs a novel CaM kinase-dependent pathway distinct from excitatory neurons, Neuron 90 (2016) 292–307.

[76] C. Buetfering, K. Allen, H. Monyer, Parvalbumin interneurons provide grid cell-driven recurrent inhibition in the medial entorhinal cortex, Nat. Neurosci. 17 (2014) 710–718.

[77] H. Bading, Nuclear calcium signalling in the regulation of brain function, Nat. Rev. Neurosci. 14 (2013) 593–608.

[78] J. Xie, Control of alternative pre-mRNA splicing by Ca(++) signals, Biochim. Biophys. Acta 1779 (2008) 438–452.