Chapter 11
Transient Receptor Potential Canonical 7 (TRPC7), a Calcium (Ca^{2+}) Permeable Non-selective Cation Channel

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Abstract Transient receptor potential canonical subfamily, member 7 (TRPC7) is the most recently identified member of the TRPC family of Ca^{2+}-permeable non-selective cation channels. The gene encoding the TRPC7 channel plasma membrane protein was first cloned from mouse brain. TRPC7 mRNA and protein have been detected in cell types derived from multiple organ systems from various species including humans. G_q-coupled protein receptor activation is the predominant mode of TRPC7 activation. Lipid metabolites involved in the phospholipase C (PLC) signaling pathway, including diacylglycerol (DAG) and its precursor the phosphatidylinositol-4,5-bisphosphate ($PIP_{2)}$, have been shown to be direct regulators of TRPC7 channel. TRPC7 channels have been linked to the regulation of various cellular functions however, the depth of our understanding of TRPC7 channel function and regulation is limited in comparison to other TRP channel family members. This review takes a historical look at our current knowledge of TRPC7 mechanisms of activation and its role in cellular physiology and pathophysiology.

Keywords TRPC7 • Non-selective cation channel • Diacylglycerol • PIP_2 • Phospholipase C • Ca^{2+} signaling • Non-excitable cells

11.1 Gene and Expression

Transient receptor potential canonical subfamily (TRPC) channels are a family of non-selective cation plasma membrane channel proteins. Seven members have currently been identified in mammals (TRPC1-7) [1] while 6 TRPC channel proteins have been identified in humans (TRPC1, TRPC3-7) [2]. The most recently identified member of this family is TRPC7. Below is a brief timeline and discussion of the species specific characterization of the TRPC7 gene and expression patterns of this cation channel (Table 11.1) [3].

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Table 11.1 Species specific TRPC7 expression

Species	mRNA/protein	Date of characterization
Mouse	**mRNA**:	
	Heart, lung, eye, brain, spleen, and testis (1)	1999
	Smooth muscle cells (2)	2001
	MC3T3 osteoblasts (3)	2009
	Embryonic brain and cortex of E13 C57BL6/J mice (4)	2009
	Melanopsin-expressing ganglion cells (5)	2011
	Brain, testis, lung, liver, heart, kidney, and DRG (6)	2012
	MnPO Glutamatergic Neurons (7)	2012
	Protein:	
	Retina (8)	2007
	Cerebellum and ventral respiratory group island (9)	2010
Rat	**mRNA**:	
	Hippocampal H19-7 cells (10, 11)	2004
	A7r5 vascular smooth muscle cells (12)	2006
	Failing myocardium of Dahl salt-sensitive rats (13)	2007
	Cultured rat microglia (14)	2009
	Hypocretin/orexin neurons (15)	2010
	Protein:	
	Brain synaptosomes (16)	2002
	Ganglion neurons (17)	2003
	H19-7 cells (10, 11)	2004
	GH4C1 pituitary cells (18)	2008
	A7r5 vascular smooth muscle cells (12)	2006
Human	**mRNA**:	
	Central nervous system and pituitary gland, kidney, intestine, prostate and cartilage (19)	2002
	Pregnant women myometrium and myometrial cell lines (20, 21)	2002
	Coronary artery endothelial cells (22)	2004
	HEK293 cells (23)	2005
	Undifferentiated gingival keratinocytes (24)	2005
	Differentiated IMR-32 neuroblastoma cells (25)	2006
	HaCaT keratinocyte (26)	2006
	Human breast cancer cell line (ZR-75-1, MCF7 and MDA-MB-231) and human breast epithelial cell line (hTERT-HME1) (27)	2014
	Lung tissue (28)	2010
	Protein:	
	HEK293 cells (29)	2005

(1) Okada et al. [4], (2) Walker et al. [5], (3) Abed et al. [10], (4) Boisseau et al. [7], (5) Perez-Leighton et al. [11], (6) Jang et al. [9], (7) Tabarean [12], (8) Sekaran et al. [6], (9) Ben-Mabrouk and Tryba [13], (10) Wu et al. [22], (11) Numaga et al. [21], (12) Maruyama et al. [23], (13) Satoh et al. [18], (14) Ohana et al. [19], (15) Cvetkovic-Lopes et al. [20], (16) Goel et al. [14], (17) Buniel et al. [15], (18) Lavender et al. [24], (19) Riccio et al. [29], (20) Yang et al. [31], (21) Dalrymple et al. [30], (22) Yip et al. [32], (23) Zagranichnaya et al. [34], (24) Cai et al. [33], (25) Nasman et al. [36], (26) Beck et al. [35], (27) Gogebakan et al. [38], (28) Finney-Hayward et al. [37], (29) Zagranichnaya et al. [34]

Mouse The gene encoding the TRPC7 channel protein was first isolated from mouse brain by Okada et al. in 1999. This gene is made up of 12 exons, and is located at chromosomal region 13 B2. TRPC7 mRNA is highly expressed in mouse eye, lung, and heart; expression was also detected in the brain, spleen and testis, although to a lesser extent [4]. In 2001, TRPC7 transcripts and splice variants were detected in smooth muscle derived from murine colon and jejunum [5]. In 2007, TRPC7 was identified in the mouse retina [6]. In 2009, Boisseau et al. examined the expression patterns of TRPC channels in the embryonic forebrain of C57BL6/J mice; at E13 TRPC7 mRNA expression was detected although to a lesser extent than other TRPC family members [7]. No TRPC7 mRNA expression was detected in skeletal muscle and inner ear organs from embryonic and early postnatal Swiss Webster mice [8, 9]; these studies highlight a development-dependent pattern of expression. TRPC7 mRNA expression was also observed in the mouse osteoblast cell line, MC3T3 in a 2009 study [10]. Recently major focus has centered on characterizing TRPC7 in neuronal cells after TRPC7 was identified in photosensitive retinal ganglion cells in 2011 [11] and glutamatergic preoptic neurons in 2012 [12]. In 2010, examination of mouse brain stem slices containing the respiratory-regulating neuronal bundle, the pre-Bötzinger complex, detected TRPC7 protein expression localized to this regulatory region [13].

Rat Rat TRPC7 genes are also made up of 12 exons, and have been mapped to chromosomal region 17p14. In rat, TRPC7 is predominantly expressed in the nervous system. In 2002, TRPC7 protein was isolated from brain synaptosomes [14]. In 2003, TRPC7 mRNA expression was detected in neurons throughout the ganglia [15]. TRPC7 protein was observed in rat striatal cholinergic interneurons [16], and had a strikingly high level of expression in the neuropil in the rat globus pallidus [17]. In 2007, TRPC7 expression was found to be up-regulated in the failing myocardium of Dahl salt-sensitive rats [18]. Characterization of TRPC7 mRNA expression has also been analyzed in various rat cell lines, including cultured rat microglia where expression was detected in 2009 [19] and in hypocretin/orexin neurons in 2010 [20]. In rat hippocampal H19-7 cells, the levels of mRNA and protein for TRPC7 are high in proliferating cells and decline upon differentiation [21, 22]. In rat vascular smooth muscle cell line A7r5 [23] and pituitary cell line GH_4C_1 [24] TRPC7 transcripts were also detected. TRPC7 mRNA was not detected in freshly isolated rat renal resistance vessels, glomeruli, and aorta [25], nor was it found in rat distal pulmonary arterial smooth muscle [26], and rat dorsal root ganglia (DRG) neurons [27].

Human In 2002, Riccio et al. cloned human TRPC7 gene from brain [28]. The gene encoding human TRPC7 is located at the chromosomal region 5q31.1, and similarly to the mouse and rat gene, has 12 exons. Both human and mouse TRPC7 genes consist of an open reading frame of 2,589 bp yielding a protein of 862 amino acids [4, 28]. Human TRPC7 has 98 % sequence homology with mouse TRPC7 [21]. Human TRPC7 mRNA expression is mostly restricted to the central nervous system however expression has been detected in some peripheral tissues including

cartilage, intestine, kidney, pituitary gland and prostate [29]. TRPC7 expression has also been examined in various human cell lines. In 2002, human myometrial cells were found to express TRPC7 mRNA, and its expression was upregulated in myometrium obtained during active labor from full term pregnant women [30, 31]. In 2004, TRPC7 expression was found in human coronary artery endothelial cells, although not in vascular smooth muscle cells [32]. In 2005, TRPC7 mRNA was also detected in undifferentiated human gingival keratinocytes [33] and protein expression was observed in the human embryonic kidney (HEK) 293 cell line [34]. In 2006, detection was observed in human differentiated keratinocytes [35] and differentiated IMR-32 neuroblastoma cells [36]. In 2009, studies failed to detect TRPC7 mRNA in human osteoblast cell lines MG-63, SaOS, and U2 OS [10], while in 2010 expression was observed in lung tissue [37]. Recently TRPC7 expression was characterized in various breast cancer cell lines including ZR-75-1, MCF7 and MDA-MB-231 and the human breast epithelial cell line hTERT-HME1 [38].

11.2 Protein Structure and Channel Configuration

TRPC7 is composed of 862 amino acid residues [4]. TRPC channel proteins including TRPC7 possess a similar basic domain structures. These defining structures include a cytosolic N-terminal domain containing four ankyrin repeats, six transmembrane spanning domains, within transmembrane domain 5 and 6 lies a highly conserved hydrophobic α helix pore-loop motif and finally a cytosolic C-terminal region [39]. This carboxy terminal region is made up of the TRP domain containing an EWKFAR and proline rich domain [40]. In 2001, Tang et al. further characterized the TRPC family C- terminal domain by identifying a calmodulin and Inositol1,4,5-Trisphosphate (IP_3) receptor binding (CIRB) region [41]. Variable regions between TRPC family members exist within this C-terminal domain contributing to their differential modes of regulation [42].

TRPC family proteins are believed to assemble into tetramers. The stoichiometry of this assembly varies depending on both TRPC family member and cell type, as homo- and heterotetrameric assembly has been proposed [43]. Specifically, TRPC7 has been shown to selectively interact with TRPC3 and TRPC6. TRPC3/6/7 channels share 70–80 % structural homology and are members of a sub-family within the TRPC family [4, 39, 43]. Co-expression of these channels has been observed in various smooth muscle tissues [44] suggesting possible heterotertrameric assemblies. However, existence of cell specific expression of different TRPC channel isoforms does not necessarily mean heteromultimeric assembly.

A study examined native TRPC7 and TRPC6 function highlighting that both the assembly and function of these channels is cell type specific. Biochemical analysis in rabbit portal vein myocytes determined strong association between TRPC6 and TRPC7 and single channel patch recordings in cells treated with anti-TRPC6 and anti-TRPC7 antibodies concluded both of these subunits were necessary for channel activity. However, examination of myocytes from rabbit mesenteric arteries revealed

that the biochemical and functional characteristics of these channel proteins were not the same [45]. More studies assessing native TRPC7 expression and function and the use of TRPC7 deficient animals are necessary to further understand in vivo the native assembly and function of these tetrameric channels [39]. While TRPC7 has been well characterized as a plasma membrane channel, one overexpression study examining localization of this channel protein in COS-7 cells observed a pool of intracellular TRPC7 protein that co-localized with the golgi network. Similar patterns of endogenous TRPC7 distribution were observed in the rat pituitary cell line, GH_4C_1. This localization within the golgi stack was thought to enhance cellular secretion either by mediating cargo transport or plasma membrane fusion [24].

11.3 Biophysical Properties of TRPC7 Channel

Okada et al. studied the permeability of TRPC7 channel using patch clamp recordings from TRPC7-expressing cells and showed constitutively activated and ATP-enhanced inward cation currents mediated by TRPC7 with permeability ratios P_{Cs}:P_{Na}:P_{Ca}:P_{Ba} of 1:1:1.9:3.5 and 1:1.1:5.9:5.0, respectively [4]. Lievremont et al. used Fura2 imaging and observed that diacylglycerol analog OAG- or muscarinic receptor stimulation-induced Ba^{2+} entry that was significantly reduced in TRPC7-deficient DT40 cell, further confirming that TRPC7 channel permeates Ba^{2+} ions [46]. Shi et al. reported that the current-voltage (I-V) relationship obtained from HEK293 cells transfected with mouse or human TRPC7 gene is almost linear with a slight flattening around the reversal potentials; mouse TRPC7 single-channel conductance is 24.3 pS and 24.8 pS under bath solutions containing 10 mM Ca^{2+} and 1 mM Ca^{2+}, respectively. However, when cells were bathed in Ca^{2+} free solution, single-channel conductance was increased to 49.3 pS [42]. Okada et al. also showed that heterologously expressed mouse TRPC7 behaves as receptor-operated DAG-activated cation channel that is insensitive to store depletion. Subsequent studies on native TRPC7 in DT40 cells and on TRPC7 ectopically expressed in HEK293 cells further suggested that TRPC7 forms a store-independent receptor-operated channel [4, 46, 47]. Thus, TRPC7 is a receptor-regulated, store-independent, non-selective cation channel activated through phospholipase C (PLC)-mediated metabolism of phosphatidylinositol 4,5-bisphosphate (PIP_2) and production of DAG [4, 48]. Regarding the pharmacological profile of TRPC7, few inhibitors are known to interfere with TRPC7 activation and none of them are specific to TRPC7. Okada et al. showed that ATP-activated mouse TRPC7 ectopically expressed HEK293 cells is more efficiently inhibited with 100 μM lanthanum (La^{3+}) and 25 μM SKF96365 than 100 μM gadolinium (Gd^{3+}) [4]. Riccio et al. used 250 μM Gd^{3+}, 250 μM La^{3+}, and 25 μM SKF96365 to completely block the Ca^{2+} entry in HEK293 cells expressing human TRPC7 [28]. 300 μM Amiloride also inhibited constitutively activated TRPC7 currents, and 2-aminoethoxydiphenyl borate (2-APB) at concentrations of 10–100 μM partially inhibited TRPC3, TRPC6, and TRPC7 channels by a mechanism not involving the known inhibitory effects of 2-APB on IP_3 receptors [4, 49].

The Schaefer group has screened a chembionet library and found three compounds, 6228-0353, 6228-0473 and 2910-0498 showing strong inhibition towards TRPC7 channels [50].

11.4 TRPC7 Channel Regulation and Function

The exact regulatory mechanisms that control TRPC7 channel activity have been the subject of contention since TRPC7 was first cloned in 1999 with many discrepancies between different groups. TRPC7 channel activity has been shown to vary depending on the molecular reagents used and the expression levels of this channel protein in heterologous model systems [51]. The use of cell specific TRPC7 gene deficient transgenic animals will likely generate more accurate and reproducible characterization of TRPC7 mediated channel activity. The general consensus is that TRPC7 is a receptor-activated channel that depends on DAG production through the catalytic activity of PLC. Below is a brief description of the current understanding of the mediators that regulate TRPC7 channel activity.

11.4.1 Metabolites of the PLC Pathway as Regulators of TRPC7 Channel

G_q-coupled receptor mediated activation is the predominant mode of regulation of the TRPC7 channel. Receptor mediated activation of PLC leads to hydrolysis of PIP_2 and the production of the signaling molecules IP_3 and DAG. TRPC7 channel function was first characterized by ectopically expressing this protein in cultured HEK293 cells [4]; this study proposed DAG as the activator of TRPC7 channels based in the use of exogenous OAG. Addition of OAG to the bath solution during whole-cell patch clamp recordings activated TRPC7 currents in a PKC-independent manner. Subsequently, Beck et al. reported that in human keratinocytes, OAG evoked a TRPC7 channel cation current, and this OAG-induced current was decreased by TRPC7 knock down [35]. Itsuki et al. identified TRPC7 activity being mediated by DAG produced through the hydrolysis of PIP_2 by PLC and reported a requirement for PIP_2 in channel activation as TRPC7 current subsequently inactivated upon hydrolysis of PIP_2. Further, PIP_2 depletion by a heterologously expressed phosphatase inhibited TRPC7 activity independently of DAG [52]. Patch clamp recordings on TRPC7-expressing HEK293 cells using the cell-attached mode detected TRPC7 currents after addition of OAG, but OAG failed to activate TRPC7 channels in excised patches. This finding suggests that DAG does activate TRPC7 channels rather indirectly perhaps through cytosolic proteins or factors that are lost in excised patches [53].

PIP_2 has been shown to directly regulate several ion channels, including the broader TRP family of channels [3, 53]. The ability of PIP_2 to directly activate TRPC7 as well as members of its subfamily TRPC3 and TRPC6 was first observed in 1998 using the phosphoinositide lipid kinase inhibitor, LY294002 which causes PIP_2 depletion [54]. Treatment of HEK293 cells overexpressing TRPC7 with this drug prevented receptor- and OAG-mediated TRPC7 channel activation. In excised inside – out patches, direct application of either PIP_2 or ATP activated TRPC7 single channels while IP_3 application had no effect [53]. Imai et al. [55] used an ectopic expression system of the voltage-sensing PIP phosphatase (DrVSP) and showed that Carbachol, OAG or RHC80267 (a DAG lipase inhibitor that enhances endogenous DAG and thus activates TRPC7) -mediated channel activation was inhibited by DrVSP activation. The extent of phosphatase inhibition was TRPC isoform specific as TRPC7 currents were attenuated to a larger extent than either TRPC6 or TRPC3 currents. Ju et al. used antibodies to inhibit TRPC isoforms in inside-out patches and concluded that noradrenaline-activated native cationic currents in portal vein myocytes are mediated by TRPC6/TRPC7 heteromultimers. These authors reported that PIP_2 inhibited this OAG-activated TRPC6/TRPC7 channel, and that this PIP_2 inhibition is rescued by IP_3 through an IP_3 receptor-independent manner [45]. The reasons for the discrepancy between this study and those reporting PIP_2 requirement for TRPC7 channel activation are unclear.

11.4.2 TRPC7 and Store Operated Ca^{2+} Entry

TRPC7 along with other TRPC channels have been proposed to constitute the pore forming unit of store-operated calcium entry (SOCE) channels [56]; this notion however remains highly controversial. In stably expressing TRPC7 HEK-293 cells either pharmacological Ca^{2+} store depletion with thapsigargin or activation of the PLC signaling pathway was able to promote TRPC7 channel activation [57]. However, in a transient expression system only stimulation of PLC-coupled receptors induced TRPC7 activation. STIM1, the ER residing Ca^{2+} sensor [58, 59] and Orai1, the highly Ca^{2+}-selective plasma membrane ion channel subunit have been established as the *bona fide* molecular components of SOCE and the calcium release-activated calcium (CRAC) current [60]. A number of studies have examined the role of STIM1 and Orai1 in regulating TRPC7-mediated calcium entry. In stably expressing TRPC7 HEK293 cells, silencing of STIM1 and Orai1 proteins by RNA interference did not affect TRPC7 activity nor did the expression of a constitutively active STIM1 mutant [47]. Studies from one group however suggested that STIM1 is capable of regulating specific isoforms of TRPC channels. Huang et al. studied the interaction between STIM1 and TRPC channels, concluding that STIM1 ERM domain mediates the selective binding of STIM1 to TRPC1, TRPC2 and TRPC4, but not to TRPC3, TRPC6 or TRPC7 [61]. Subsequently, the same group reported that STIM1 binds TRPC1, TRPC4 and TRPC5 allowing them to function as

store-operated channels. The authors also demonstrated that STIM1 indirectly regulates TRPC3 and TRPC6 channels by mediating the heteromultimerization of TRPC3 with TRPC1 and TRPC6 with TRPC4. In these studies, TRPC7 was the only TRPC family member that was not regulated by STIM1 [62].

11.4.3 Ca^{2+} and Calmodulin

Most Ca^{2+}-permeable channels including TRPC7 channels are themselves regulated by Ca^{2+}. Specifically studies have shown that TRPC7 channels can be negatively regulated by Ca^{2+} both directly and indirectly. Lemonnier et al. identified a negative feedback mechanism of TRPC7 channels in HEK293 cells, whereby Ca^{2+} entry through TRPC7 channel negatively regulates its own activity [63]. This Ca^{2+}-mediated negative feedback at the mouth of TRPC7 channels is more pronounced when the SERCA pump is inhibited by thapsigargin, suggesting that physiologically SERCA pumps that are closely associated with TRPC7 channels attenuate this negative feedback by buffering calcium into the endoplasmic reticulum. Indeed, application of the SERCA pump inhibitor thapsigargin or CPA prevented OAG-activated TRPC7 channel activation. The inhibitory effect of thapsigargin was reversed by inhibition of calmodulin and was recapitulated by pharmacological disruption of the actin cytoskeleton. Shi et al. used whole-cell and single-channel recordings to reveal that voltage-dependent inhibitory actions of extracellular Ca^{2+} on TRPC7 channel currents are likely mediated through Ca^{2+} interaction with an extracellular site capable of sensing the membrane potential. This group also reported a concentration-dependent inhibitory effect of intracellular Ca^{2+} on TRPC7 channel currents, which they suggested was mediated by calmodulin [42].

The variable C-terminal domain distinguishes members of the TRPC family from one another [42]. TRPC7 has the CIRB domain within this variable region that contains binding sequences for both calmodulin and the IP_3R. Direct interactions between these proteins and TRPC7 have been shown to regulate TRPC7 channel activity. As discussed above, Shi et al. used cell-attached mode single-channel recordings to show that cytosolic Ca^{2+}-mediated inhibition of TRPC7 channel activity was strongly attenuated by pretreatment with pharmacological agent calmidazolium (CMZ), a potent calmodulin (CaM) antagonist or coexpression of the calmodulin mutant, mutCaM. These findings suggests binding of calmodulin to the TRPC7 C-terminal CIRB domain inhibits channel activity [42]. In the avian B-cell line, DT40, using the cell-attached mode of patch clamp, OAG activated single channel activity was not observed in $TRPC7^{-/-}$ cells, nor in $IP_3R^{-/-}$ cells. Interestingly, exogenous expression of either TRPC7 in the $TRPC7^{-/-}$ cells or IP_3R in the $IP_3R^{-/-}$ cells rescued OAG-activated single channel activity to the same extent as wild type cells [51]. These findings support a native role for the TRPC7 channel protein in mediating DAG-activated currents that are dependent on IP_3R.

11.4.4 N-Terminal Binding Proteins

The N-terminal domain of TRPC7 is made up of four ankyrin-like repeats. These protein-protein interacting domains have been shown to regulate TRPC7 channel activity and localization. Co-immunoprecipitation studies revealed that the cytosolic cGMP/cGMP-dependent protein kinase (cGK) isoform, cGK-I α binds to the N-terminal ankyrin-like repeat domain of TRPC7. Yuasa et al. also observed cGK-I α phosphorylates TRPC7 on its threonine 15 site without any effect on TRPC3. This TRPC7 specific phosphorylation reduced carbachol-activated calcium signaling and phosphorylation of the transcription factor, CREB [64]. Lussier and colleagues used protein-protein interaction assays, including yeast two-hybrid screen, GST pull-down, and co-immunoprecipitation and demonstrated that MxA which is a member of the dynamin GTPase superfamily that interacts with the N-terminal second ankyrin-like repeat domain of TRPC1, TRPC3, TRPC4, TRPC5, TRPC6, and TRPC7 [65]. It was proposed that MxA enhances channel activity by regulating trafficking of the channel proteins to the plasma membrane. Although this functional role has not been tested for TRPC7, mutation studies involving subfamily members TRPC3 and TRPC6 have shown deletion of the ankyrin-like repeat domain leads to intracellular retention of channel protein, inhibition of plasma membrane localization and overall channel dysfunction [43, 66]. Future studies looking at TRPC7 channel specific protein trafficking are necessary to gain insights into TRPC7 regulation through membrane trafficking.

11.5 TRPC7 in Pathophysiology of Disease

Altered TRPC7 channel expression and activity are observed in various diseases including malignant breast cancer tumors. Regulation of multiple TRPC genes has been shown to be mediated by the Rho kinase pathway [67]. Rho kinase activity has been associated with metastasis of esophageal cancer cell lines and inhibition of this pathway using the pharmacological inhibitor Y-27632 prevented the growth and invasiveness of these cancer cells [68]. In human breast cancer cell lines ZR-75-1, MCF7, and MDA-MB-231 and the human breast cancer epithelial cell line hTERT-HME1 inhibition of the Rho-kinase pathway with Y-27632 increased the expression of TRPC7 in all cell lines as detected by quantitative real time PCR [38]. Interestingly the Rho-kinase inhibitors attenuated expression of TRPC1 and another TRP family member TRPV2. This study suggests a potential protective role of TRPC7 in cancer cell growth and progression.

TRPC7 expression has also been shown to be up-regulated in animal models of disease. In Dahl salt sensitive rats, TRPC7 mRNA was increased in the failing myocardium of these animals. This increased expression correlated with an increased level of apoptosis as assessed by TUNEL staining. Heart failure in these mice was

suggested to be contributed by angiotensin II-induced Ca^{2+} entry through activation of TRPC7 and subsequent myocardial apoptosis [18].

In a mouse model of pilocarpine induced status epilepticus, TRPC7 was shown to mediate the initiation of acute seizures, as observed by the reduction in pilocarpine-induced gamma wave activity in TRPC7 knockout animals [69]. The mechanism by which TRPC7 mediates seizure induction was characterized in brain slices derived from the hippocampal region cornu ammonis CA3, a central region involved in seizure generation. TRPC7 was shown to generate spontaneous epileptiform burst firing, these signals of seizure induction were initiated at the synaptic level where TRPC7 was shown to be involved in the potentiation of these signals in the CA3 synapses and Schaffer collateral-CA1 synapses. This study using TRPC7 knockout animals was one of the first studies to provide insight into the in vivo function of this enigmatic channel protein.

11.6 Conclusion

Since the initial characterization of the mouse TRPC7 gene in 1999 [4], the following two decades of research studying this channel protein have shown that: (i) TRPC7 expression is present in both excitable and non-excitable cells; (ii) Biophysical and functional studies have described TRPC7 as a store-independent DAG-activated Ca^{2+}-permeable non-selective cation channels [47, 49]; (iii) The mode of TRPC7 channel regulation is likely complex, attracting much debate, including the role of PIP_2 in regulating TRPC7 function. Channel heteromultimerization with other TRPC isoforms and isoforms of the broader TRP superfamily are likely to enhance the diversity of signaling mechanisms through TRPC7. Thus far, TRPC7-associated diseases have rarely been reported. With increasing number of studies employing transgenic TRPC7 knockout animals, understanding of the various physiological functions controlled by this dynamic Ca^{2+}-permeable non-selective cation channel is likely increase.

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