**REVIEW —Current Perspective—**

Dissecting Receptor-Mediated Ca\(^{2+}\) Influx Pathways: TRP Channels and Their Native Counterparts

Yasuo Mori\(^{1,2,4,*}\), Ryuji Inoue\(^{5}\), Masakazu Ishii\(^{1,2}\), Yuji Hara\(^{1,3,4}\) and Keiji Imoto\(^{3,4}\)

\(^{1}\)Center for Integrative Bioscience, Okazaki National Research Institutes, Okazaki 444-8585, Japan
\(^{2}\)Department of Cell Physiology, \(^{3}\)Department of Information Physiology, National Institute for Physiological Sciences, Okazaki National Research Institutes, Okazaki 444-8585, Japan
\(^{4}\)School of Life Science, The Graduate University for Advanced Studies, Okazaki 444-8585, Japan
\(^{5}\)Department of Pharmacology, Faculty of Medicine, Kyushu University, Fukuoka 812-8582, Japan

Received September 5, 2001

ABSTRACT—Cellular stimulation from the surrounding extracellular environment via receptors and other pathways evoke activation of Ca\(^{2+}\)-permeable cation channels that form essential signaling pathways in controlling biological responses. An important clue to understand the molecular mechanisms underlying these cation channels (tentatively termed as receptor-mediated cation channels (RMCC)) was first provided through molecular studies of the transient receptor potential (trp) protein (TRP), which controls light-induced depolarization in *Drosophila* photoreceptor cells. Use of the genetic information and recombinant expression technique lead to the discovery of numerous mammalian TRP homologues revealing novel RMCCs. In this review, we focus on the dramatic progress in the molecular investigation of RMCC in mammalian systems. The recent findings should provide powerful tools for the development of novel pharmaceutical targets.

**Keywords:** Receptor-mediated Ca\(^{2+}\) channel, TRP, Ca\(^{2+}\)-permeable cation channel

**Introduction**

Calcium (Ca\(^{2+}\)) influx across the plasma membrane plays a vital role in the regulation of diverse cellular processes, ranging from ubiquitous activities like gene expression to tissue-specific functions such as neurotransmitter release and muscle contraction, by controlling the cytosolic free Ca\(^{2+}\) concentration ([Ca\(^{2+}\)]\(_i\)). In addition to the well characterized modes of Ca\(^{2+}\) entry through voltage-dependent Ca\(^{2+}\) channels and ligand-gated cation channels, receptor-mediated, Ca\(^{2+}\)-permeable cation channels (RMCC) have been recognized for their physiological significance.

RMCCs are a group of diverse Ca\(^{2+}\)-permeable cation channels evoked by stimulation with various hormones, neurotransmitters, autacoids, Ca\(^{2+}\) and antigens (Fig. 1) (1). It has been recognized that a group of RMCCs are responsible for the Ca\(^{2+}\) influx triggered in response to receptor activation that leads to phospholipase C (PLC) activation and subsequent hydrolysis of phosphoinositides (PI) via Gq protein or protein kinases (1). Among members of the group, recent attention was particularly directed to store-operated channels (SOCs; in other words, Ca\(^{2+}\) release-activated current (I\(_{\text{CRAC}}\)) or capacitative Ca\(^{2+}\) entry (CCE)) that is activated through Ca\(^{2+}\) release from the intracellular Ca\(^{2+}\) store, endoplasmic reticulum (ER), induced by inositol 1,4,5-trisphosphate (IP\(_3\)) and consequent depletion of Ca\(^{2+}\) from the store (1 – 5). In addition, other plasma membrane ion channels directly activated by second messengers such as Ca\(^{2+}\), IP\(_3\), inositol 1,3,4,5-tetraphosphate (IP\(_4\)) and arachidonic acid metabolites are also important members of this RMCC group (1).

Because of the lack of pharmacological agents that selectively modulate specific RMCC subtypes, it was extremely difficult to tell whether those RMCCs identified in different preparations really represent different RMCC subtypes and to relate RMCCs with particular physiological responses. Furthermore, it was essentially impossible to biochemically characterize them as receptors of certain pharmacological agents. Thus, a powerful approach to aid development of selective pharmacological tools was a long-awaited for the molecular/physiological studies of RMCCs. In this review, we attempt to overview recent progress in RMCC research,
Y. Mori et al. highlighting the studies on mammalian homologues of the transient receptor potential (trp) protein (TRP) (6) that has been a likely candidate for the molecular entity of RMCCs.

**Activation mechanism and ion permeation properties of mammalian TRP channels**

An important clue for understanding the molecular basis of RMCCs was first attained through the finding of a Drosophila mutant with visual transduction mutation trp, whose photoreceptors fail to generate the Ca$^{2+}$-dependent sustained phase of receptor potential and to induce subsequent Ca$^{2+}$-dependent adaptation to light (7, 8). Since the gene products of the trp and trp-like (trpl) gene (TRP and TRPL) comprise the light-activated, PI-dependent Ca$^{2+}$ conductance in Drosophila photoreceptors (8–10), the original hypothesis that the counterparts of TRP and TRPL direct formation of SOCs in vertebrate cells, was based upon the analogy between the phototransduction mechanism in Drosophila and the PI-dependent signal transduction processes in vertebrates. The first mammalian homologues were discovered by searching the amino acid sequences similar to TRP and TRPL in the Expressed Sequence Tag data base or by using PCR primers designed to anneal conservative regions (11–13). Seven mammalian TRP homologues, TRP1–7, have been so far identified (Table 1).

Some lines of supportive evidence for the hypothesis that TRP homologue genes encode SOCs were presented from cDNA expression experiments on different TRP subtypes (14–22). Among different models that have been proposed to explain how information is transmitted from Ca$^{2+}$-released/depleted ER to the plasma membrane (1–3, 5), recent reports are somewhat in favor of a direct coupling between TRP proteins and IP$_3$ receptors (IP$_3$Rs) (22–24). However, store-independent activation of Ca$^{2+}$ influx and cation currents mediated by TRP members were also reported (25–34). As for Drosophila TRP and TRPL, polyunsaturated fatty acids, such as arachidonic acid and linolenic acid, were shown to activate them in both native and recombinant systems (30). This store-independent activation of TRP and TRPL is consistent with the observation that IP$_3$R is not essential for receptor potentials generated by TRP and TRPL in Drosophila photoreceptors (35). As for vertebrate TRP homologues (Table 1), TRP2, TRP3, TRP5, TRP6 and TRP7 have been reported to exhibit store-independent activities in recombinant expression systems (25, 26, 28, 29, 31–33). Intracellular Ca$^{2+}$ plays an essential role in activation of TRP3, which suggests that TRP3 codes for Ca$^{2+}$-activated Ca$^{2+}$-permeable cation channels (28), while TRP5 channel activity depends on extracellular Ca$^{2+}$ (Y. Mori and M. Wakamori, unpublished results). Involvement of calmodulin (10, 32, 36) or Ca$^{2+}$-calmodulin-dependent kinases such as myosin light chain kinase is suggested in TRP activation (Y. Mori and S. Shimizu, unpublished results). Most recently, activation of TRP3, TRP6 and TRP7 by diacylglycerol (DAG) has been demonstrated (31, 32), although the exact mechanism of activation is yet to be resolved.

TRP1-7 can all be categorized as non-selective cation channels. The permeability ratio between Ca$^{2+}$ and Na$^+$ Fig. 1. Activation mechanisms of receptor-mediated Ca$^{2+}$-permeable cation channels.
Table 1. Characteristics of ‘classical’ TRP homologues

| TRP homologues | Expression                        | Trigger                  | Permeation              | Conductance            |
|----------------|----------------------------------|--------------------------|-------------------------|------------------------|
| TRP1 (human, mouse) | ubiquitous                        | depletion of Ca\(^{2+}\) stores (thapsigargin, IP\(_3\)) | Ca\(^{2+}\) = Na\(^+\) = Ba\(^{2+}\) | 16 pS (Ca\(^{2+}\) & Na\(^+\))
| TRP2 (human, mouse) | brain, vomeronasal organ (terrestrial vertebrates), sperm | ? (depletion of Ca\(^{2+}\) stores or Ca\(^{2+}\)), diacylglycerol | Ca\(^{2+}\), Na\(^+\) | 20 pS (Ca\(^{2+}\) & Na\(^+\)) | 66 pS (Na\(^+\))
| TRP3 (human, mouse) | brain (cerebellum, Purkinje cells) | depletion of Ca\(^{2+}\) stores (thapsigargin, IP\(_3\)) | Ba\(^{2+}\) > Ca\(^{2+}\) > Na\(^+\) = Cs\(^+\) | 28 pS (Ca\(^{2+}\) & Na\(^+\)) |
| TRP4 (bovine, mouse) | brain (cerebrum), adrenal gland vascular smooth muscle | receptor stimulation and Ca\(^{2+}\) receptor stimulation diacylglycerol | Ca\(^{2+}\) > Na\(^+\), Ba\(^{2+}\), Mn\(^{2+}\) | 48 pS (Na\(^+\)) |
| TRP5 (mouse) | brain | constitutively activated, receptor stimulation, Ca\(^{2+}\) and diacylglycerol | Ca\(^{2+}\) > Na\(^+\) = Cs\(^+\) | Na\(^+\); unitary current. |
| TRP6 (mouse) | brain, lung (epithelial cells in respiratory bronchiole), spleen, vascular smooth muscle | constitutively activated (?) G\(_{q}\) or polyunsaturated fatty acid | Ca\(^{2+}\) > Na\(^+\) | 64 pS (Na\(^+\)) |
| Drosophila TRP | photoreceptor cells | depletion of Ca\(^{2+}\) stores ? (recombinant) or polyunsaturated fatty acid | Ca\(^{2+}\) > Na\(^+\) | Na\(^+\); unitary current. |
| Drosophila TRPL | photoreceptor cells | constitutively active (?) G\(_{q}\) or polyunsaturated fatty acid | Ca\(^{2+}\) > Na\(^+\), Ba\(^{2+}\) | 64 pS (Na\(^+\)) |

NA: noise analysis, U: unitary current.

PCa/PNa, that indicates selectivity of TRPs to Ca\(^{2+}\) over Na\(^+\), are in the range 1 to 10 (Table 1). However, acute response to extracellular application of Ca\(^{2+}\) of TRP monovalent currents is different: TRP5 (26) and TRP6 currents (34) were facilitated, TRP7 (32) currents were suppressed, but TRP1 (14) and TRP3 currents (28) were essentially unaffected. This, together with the variability of TRP homologues in conductance at unitary levels (Table 1), suggests that the fine structure of ion channel pore varies among TRP channels in interaction with conducting ions.

Physiological function of TRP channels

As discussed above, because of the unavailability of pharmacological agents that selectively recognize specific RMCC subtypes, it was extremely difficult to functionally distinguish RMCC subtypes in different preparations and to establish physiological roles of respective RMCCs. However, there is gradually accumulating evidence that the TRP proteins are a requisite component of native Ca\(^{2+}\)-permeable cation channels activated by plasma membrane receptors and other stimuli. In the vascular system, the \(\alpha_1\)-adrenoceptor (\(\alpha_1\)-AR) is distributed widely and plays a central role in control of systemic blood pressure via sympathetic nerves. Stimulation of the \(\alpha_1\)-AR leads to activation of G-protein (G\(_{q/11}\))-coupled PLC\(\beta\) and formation of IP\(_3\) and DAG, thereby causing a release of stored Ca\(^{2+}\) and an accompanying sustained Ca\(^{2+}\) entry. The \(\alpha_1\)-AR-activated non-selective cation channels is thought to contribute to this Ca\(^{2+}\) entry in both direct and indirect ways, since it is activated by DAG and allows preferential movement of divalent cations, and secondarily evokes Ca\(^{2+}\) entry through the voltage-dependent pathway by depolarizing the membrane. Despite this potential physiological importance, no clues elucidating the molecular entity of the \(\alpha_1\)-AR-activated cation channels had been obtained. We have recently found that heterologous expression of murine TRP6 in HEK293 cells reproduces almost exactly the essential biophysical and pharmacological properties of \(\alpha_1\)-AR-activated cation channels previously identified in rabbit portal vein smooth muscle (34). Such properties include activation by DAG, ‘S-shaped’ current-voltage relationship, high divalent cation permeability (Ca\(^{2+}\), Ba\(^{2+}\) > Na\(^+\)), unitary conductance of 25 – 30 pS and augmentation by fluafenamate and Ca\(^{2+}\) and inhibition by inorganic and organic blockers. The level of TRP6 mRNA expression was remarkably high in portal vein smooth muscles as compared with other TRP subtypes, and the TRP6 protein was localized near the sarcolemmal region of single portal vein myocytes. Furthermore, treatment of primary cultured portal vein myocytes with TRP6 antisense oligonucleotides resulted in marked inhibition of TRP6 protein immunoreactivity as well as selective suppression of \(\alpha_1\)-AR-activated, store depletion-independent cation current and Ba\(^{2+}\) influx.
These results strongly indicate that TRP6 is the essential component of the $\alpha_\text{1R}$-AR-activated cation channel, which may serve as a store-depletion-independent $\text{Ca}^{2+}$ entry pathway during increased sympathetic activity. From an electrophysiological point of view, in other native systems, there are groups of nonselective cation channels activated independently of store depletion that show considerable resemblance to the $\alpha_\text{1R}$-AR-activated cation channel. These groups include muscarinic cation channels ubiquitously identified in gastrointestinal smooth muscle (37–39) and some of the $\text{Ca}^{2+}$-activated nonselective cation channels in cardiac and epithelial tissues (40–42).

In our recombinant studies, TRP7 channels gave rise to constitutively activated and receptor stimulation-induced cation currents in HEK cells (32). In native systems, time-independent, spontaneous currents such as background currents have been reported (43–60). The constitutive current induced by TRP7 shares a number of functional characteristics with native background currents, including time independence (43–46), susceptibility to suppression by external $\text{Ca}^{2+}$ and $\text{Mg}^{2+}$ (44, 46, 47), selectivity among monovalent cations (44–46), linearity of current-voltage relationship (44, 46, 47), and amiloride sensitivity (38). Indeed, preparations used for the measurement of background currents are from the heart (43, 44, 46), lung (47) and brain (45) that are abundant in TRP7 RNA (32).

Physiological roles of spontaneously active, or background and receptor stimulation-induced non-selective cation currents mediated by TRP7 may be essential for the pacemaking activity in the sino-atrial nodal cells (43, 44), the spontaneous secretion in pace-making endocrine cells (45), the regulation of resting membrane potential (47) and the frequency and pattern modulation of action potentials (49) in smooth muscle cells.

TRP function has been also characterized during sperm-egg interaction, since the sperm acrosome reaction is a $\text{Ca}^{2+}$-dependent secondary event that must be completed before fertilization (50). In mammals, exocytosis is triggered during gamete contact by ZP3, a glycoprotein constituent of the egg’s extracellular matrix, or zona pellucida, that activates trimeric G proteins and PLC and causes a transient $\text{Ca}^{2+}$ influx into sperm through T-type $\text{Ca}^{2+}$ channels. Using the neutralizing anti-TRP2 antibody, it was shown that TRP2 is essential for the activation of second sustained $\text{Ca}^{2+}$ influx into sperm that drive acrosome reactions.

The idea that genetics is a powerful approach in elucidating physiological significance of proteins that lack selective ligands or modulators is also applicable to $\text{Ca}^{2+}$ channel subtypes that lack selective blockers. Freichel et al. (51) described a SOC current in vascular endothelium and showed that endothelial cells of mice deficient in TRP4 lack this current. Agonist-induced $\text{Ca}^{2+}$ entry and vaso-relaxation was reduced markedly, showing that TRP4 is an indispensable component of store-operated channels in native endothelial cells. TRP4 channels were thus suggested to directly provide an $\text{Ca}^{2+}$ entry pathway, essentially contributing to the regulation of blood vessel tone. Most recently, we found that the TRP1 plays critical roles in the regulation of elementary and coordinated properties of B lymphocyte $\text{Ca}^{2+}$ signaling (Y. Mori and T. Kurosaki, unpublished results). Genetic disruption of TRP1 significantly attenuated SOC activity induced upon the stimulation of B-cell antigen receptors (BCR) and $\text{Ca}^{2+}$ depletion of ER and $\text{Ca}^{2+}$ release-activated $\text{Ca}^{2+}$ currents in avian DT40 B cells. Strikingly, the TRP1-deficient B cells showed significant reduction in rates of IP$_3$R-mediated $\text{Ca}^{2+}$ release from ER independently of the attenuation of SOC activity. Furthermore, disruption of the TRP1 gene diminished BCR-induced $\text{Ca}^{2+}$ oscillation. The results suggest that SOCs, whose formation involves TRP1 as an important component, directly promote activity of IP$_3$R $\text{Ca}^{2+}$ release channels via TRP1 to enhance functional coupling between the ER and plasma membrane in transduction of intracellular $\text{Ca}^{2+}$ signaling.

Pharmacological modulators of RMCCs

The inhibitors of RMCCs have so far been mostly those substances proven to inhibit other channels and enzymes more efficiently (52). SK&F96365 and La$^3+$ are also known as blockers for voltage-dependent $\text{Ca}^{2+}$ channels. Flufenamate, which blocks cyclooxygenase, inhibited the TRP7 channel but potentiated the TRP6 channel (or the $\alpha_\text{1R}$-AR-activated cation channel) (34). Interestingly, 2-aminooctaneboronate, which was originally believed to selectively inhibit IP$_3$R, has been demonstrated to suppress TRP channels or SOCs in the plasma membrane of the IP$_3$R-deficient cells (53–55). The susceptibility of RMCCs to the inhibitors of other channels and enzymes may suggest that RMCCs are closely coupled with other channels and enzymes in activation. Recent studies have disclosed a few promising substances that may turn out to be a selectively modulator for RMCCs. Zhang et al. have demonstrated that LOE908 inhibits non-selective cation channels but not SOCs among RMCCs (56). We have identified series of aniline derivatives that selectively modulate the TRP5 channel (M. Ishii and Y. Mori, unpublished results). Because of this situation for the availability of selective pharmacological agents for RMCCs, search for novel RMCC-subtype-specific modulators is increasingly recognized as a crucial aspect of RMCC research.

TRP superfamily

As demonstrated in Fig. 2, comparison of amino acid sequences obtained from cDNA cloning or deduced from genomic sequences allows us to construct a phylogenetic
tree for the TRP gene superfamily. The superfamily contains a number of physiologically important Ca\textsuperscript{2+}-permeable cation channels such as the capsaicin receptor (VR1), polycystin (PKD1 and PKD2), osmolarity-sensing channel (OSM9) and Ca\textsuperscript{2+}-absorption channel (ECaC). Recent functional studies of recombinant channels produced by the members of the TRP superfamily, particularly by the members of the ‘so-called’ LTRP subfamily (57), revealed novel RMCCs that were not fully identified using native preparations.

Two independent groups have reported that the LTRPC7 (TRP-PLIK) gene encodes ATP-dependent Ca\textsuperscript{2+}-permeable cation channels (58, 59). LTRPC7 phosphorylated itself, and the channel’s kinase activity is essential for channel function (58). Targeted deletion of LTRPC7 in DT-40 B cells was lethal, indicating that LTRPC7 has a fundamental and non-redundant role in cellular physiology (59). It was proposed that LTRPC7, by virtue of its sensitivity to physiological Mg-ATP levels, may be involved in a fundamental process that adjusts plasma membrane divalent cation fluxes according to the metabolic state of the cell (59).

Perraud et al. showed that LTRPC2 functions as a Ca\textsuperscript{2+}-permeable cation channel that is gated by free ADP-ribose (ADPR), a product of nicotinamide adenine dinucleotide (NAD\textsuperscript{+}) hydrolysis and a breakdown product of the calcium-release second messenger cyclic ADPR (60). The NUDT homology domain near the carboxy terminus of the LTRPC2/TrpC7 putative cation channel functioned as a specific ADPR pyrophosphatases. The expression of native LTRPC2 transcripts was detectable in many tissues including the U937 monocyte cell line, in which ADPR induces large cation currents that closely matched those mediated by recombinant LTRPC2, indicating that intracellular ADPR regulates calcium entry into cells that express LTRPC2. Most recently, we have found that LTRPC2, which we designated as ROSC1, is a novel Ca\textsuperscript{2+}-permeable cation channel activated in response to extracellular H\textsubscript{2}O\textsubscript{2} at concentrations of micromolar range and to other agents that generate reactive oxygen and nitrogen species (61), in contrast to other types of Ca\textsuperscript{2+} channels which require much higher H\textsubscript{2}O\textsubscript{2} concentrations. Direct action of β-NAD\textsuperscript{+} through the MutT motif was responsible for the activation of ROSC1 channels. Strikingly, expression of ROSC1 channels conferred susceptibility to cell death on HEK cells, but the ROSC1 antisense oligonucleotide attenuated ROSC1-channel conferred susceptibility to cell death on HEK cells, but the ROSC1 antisense oligonucleotide attenuated cell death in various tissues.

Thus, on the basis of primary structure information deduced from the cDNA and genomic sequences, existence
of numerous TRP-related genes encoding proteins, which can be categorized as “orphan” Ca\(^{2+}\) channels, have been revealed in the genome. Functional characterization of these novel TRP proteins using recombinant expression systems have demonstrated not only novel RMCCs but also novel signaling cascades that trigger physiological responses. Furthermore, studying the physiological functions of the different TRP channels (in relation to the ER, cytoskeleton and other intracellular units of structure and function) should lead to understanding the “specificity” for Ca\(^{2+}\) signaling in evoking cellular responses (62).

Acknowledgments
This work is supported by research grants from the Ministry of Education, Culture, Sports, Science and Technology, Japan and by the Ministry of Health, Labour and Welfare, Japan.

REFERENCES
1. Fasolato C, Innocenti B and Pozzan T: Receptor-activated Ca\(^{2+}\) influx: how many mechanisms for how many channels? Trends Pharmacol Sci 15, 77 – 83 (1994)
2. Irvine RF: Quantal Ca\(^{2+}\) release and the control of Ca\(^{2+}\) entry by inositol phosphates – a possible mechanism. FEBS Lett 263, 5 – 9 (1990)
3. Putney JW Jr and Bird GSJ: The signal for capacitative calcium entry. Cell 75, 199 – 201 (1993)
4. Hoth M and Penner R: Depletion of intracellular calcium stores activates a calcium current in mast cells. Nature 355, 353 – 356 (1992)
5. Berridge MJ: Capacitative calcium entry. Biochem J 312, 1 – 11 (1995)
6. Montell C and Rubin GM: Molecular characterization of the *Drosophila* trp locus: a putative integral membrane protein required for phototransduction. Neuron 2, 1313 – 1323 (1989)
7. Fein A, Payne R, Corson DW, Berridge MJ and Irvine RF: Photoreceptor excitation and adaptation by inositol 1,4,5-trisphosphate. Nature 311, 157 – 160 (1984)
8. Niemeyer BA, Suzuki E, Scott K, Jalink K and Zuker CS: The *Drosophila* light-activated conductance is composed of the two channels TRP and TRPL. Cell 85, 651 – 659 (1996)
9. Hardie RC and Minke B: The trp gene is essential for a light-activated Ca\(^{2+}\) channel in *Drosophila* photoreceptors. Neuron 8, 643 – 651 (1992)
10. Phillips AM, Bull A and Kelly LE: Identification of a *Drosophila* gene encoding a calmodulin-binding protein with homology to the trp phototransduction gene. Neuron 8, 631 – 642 (1992)
11. Petersen CCH, Berridge MJ, Borgese MF and Bennett DL: Putative capacitative calcium entry channels: expression of *Drosophila* trp and evidence for the existence of vertebrate homologues. Biochem J 311, 41 – 44 (1995)
12. Wes PD, Chevesich J, Jeromin A, Rosenberg C, Stetten G and Montell C: TRPC1, a human homolog of a *Drosophila* store-operated channel. Proc Natl Acad Sci USA 92, 9652 – 9656 (1995)
13. Zhu X, Chu PB, Peyton M and Birnbaumer L: Molecular cloning of a widely expressed human homologue for the *Drosophila* trp gene. FEBS Lett 373, 193 – 198 (1995)
14. Zitt C, Zobel A, Obukhov AG, Harteneck C, Kalkbrenner F, Lückhoff A and Schultz G: Cloning and functional expression of a human Ca\(^{2+}\)-permeable cation channel activated by calcium store depletion. Neuron 16, 1189 – 1196 (1996)
15. Zhu X, Jiang M, Peyton M, Boulay G, Hurst R, Stefani E and Birnbaumer L: *trp*, a novel mammalian gene family essential for agonist-activated capacitative Ca\(^{2+}\) entry. Cell 85, 661 – 671 (1996)
16. Philipp S, Cavalié A, Freichel M, Wissenbach U, Zimmer S, Trost C, Marquart A, Murakami M and Flockerzi V: A mammalian capacitative calcium entry channel homologous to *Drosophila* TRP and TRPL. EMBO J 15, 6166 – 6171 (1996)
17. Vaca L, Sinkins WG, Hu Y, Kunze DL and Schilling WP: Activation of recombinant trp by thapsigargin in S9 insect cells. Am J Physiol 267, C1501 – C1505 (1994)
18. Gillo B, Chorna I, Cohen H, Cook B, Manistersky I, Chorev M, Arnon A, Pollock JA, Selinger Z and Minke B: Coexpression of *Drosophila* TRP and TRPL-like proteins in Xenopus oocytes reconstitutes capacitative Ca\(^{2+}\) entry. Proc Natl Acad Sci USA 93, 14146 – 14151 (1996)
19. Philipp S, Hambrecht J, Braslavski L, Schroth G, Freichel M, Murakami M Cavalié A and Flockerzi V: A mammalian capacitative calcium entry channel homologous to *Drosophila* TRP and TRPL. EMBO J 17, 4274 – 4282 (1998)
20. Mizuno N, Kitayama S, Saishin Y, Shimada S, Morita K, Mitsuhashi C, Kurihara H and Dohi T: Molecular cloning and characterization of rat trp homologues from brain. Brain Res Mol Brain Res 64, 41 – 51 (1999)
21. Tomita Y, Kaneko S, Funayama M, Kondo H, Satoh M and Kaneko S: Intracellular Ca\(^{2+}\) store-operated Ca\(^{2+}\) influx of through TRP-R, a rat homolog of TRP, expressed in Xenopus oocytes. Neurosci Lett 248, 195 – 198 (1998)
22. Boulay G, Brown DM, Qin N, Jiang M, Dietrich A, Zhu MX, Chen Z, Birnbaumer M, Mikoshiba K and Birnbaumer L: Modulation of Ca\(^{2+}\) entry by polypeptides of the inositol 1,4,5-trisphosphate receptor (IP\(_R\)) that bind transient receptor potential (TRP): evidence for roles of TRP and IP\(_R\) in store depletion-activated Ca\(^{2+}\) entry. Proc Natl Acad Sci USA 96, 14955 – 14960 (1999)
23. Kisenkov Y, Mignery GA, Zhu MX and Mualem S: The N-terminal domain of the IP\(_R\) receptor gates store-operated hTrp3 channels. Mol Cell 4, 423 – 429 (1999)
24. Ma HT, Patterson RL, van Rossum DB, Birnbaumer L, Mikoshiba K and Gill DL: Requirement of the inositol trisphosphate receptor for activation of store-operated Ca\(^{2+}\) channels. Science 287, 1647 – 1651 (2000)
25. Boulay G, Zhu X, Peyton M, Jiang M, Hurst R, Stefani E and Birnbaumer L: Cloning and expression of a novel mammalian homolog of *Drosophila* transient receptor potential (Trp) involved in calcium entry secondary to activation of receptors coupled by the Gq class of G protein. J Biol Chem 272, 29672 – 29680 (1997)
26. Okada T, Shimizu S, Wakamori M, Maeda A, Kuroskai T, Takada N, Imoto K and Mori Y: Molecular cloning and functional characterization of a novel receptor-activated TRP Ca\(^{2+}\) channel from mouse brain. J Biol Chem 273, 10279 – 10287 (1998)
27. Obukhov AG, Harteneck C, Zobel A, Harhammer R, Kalkbrenner F, Leopold D, Lückhoff A, Nürnberg B and Schultz G: Direct activation of trpl cation channels by G alpha 11 subunits. EMBO J 15, 5833 – 5838 (1996)
Receptor-Mediated TRP Ca\(^{2+}\) Channels

Zitt C, Obukhov AG, Strübing C, Zobel A, Kalkbrenner F, Lückhoff A and Schultz G: Expression of TRPC3 in Chinese hamster ovary cells results in calcium-activated cation currents not related to store depletion. J Cell Biol 138, 1333 – 1341 (1997)

Zhu X, Jiang M and Birnbaumer L: Receptor-activated Ca\(^{2+}\) influx via human Trp3 stably expressed in human embryonic kidney (HEK)293 cells. Evidence for a non-capacitative Ca\(^{2+}\) entry. J Biol Chem 273, 133 – 142 (1998)

Chyb S, Raghu P and Hardie RC: Polyunsaturated fatty acids activate the Drosophila light-sensitive channels TRP and TRPL. Nature 397, 255 – 259 (1999)

Hofmann T, Obukhov AG, Schaefer M, Harteneck C, Güdermann T and Schultz G: Direct activation of human TRPC6 and TRPC3 channels by diacylglycerol. Nature 397, 259 – 263 (1999)

Okada T, Inoue R, Yamazaki K, Maeda A, Kuroasaki T, Yamakuni T, Tanaka I, Shimizu S, Ikenaka K, Imoto K and Mori Y: Molecular and functional characterization of a novel mouse transient receptor potential protein homologue TRP7. Ca\(^{2+}\)-permeable cation channel that is constitutively activated and enhanced by stimulation of G protein-coupled receptor. J Biol Chem 274, 27359 – 27370 (1999)

Vannier B, Peyton M, Boulay G, Brown D, Qin N, Jiang M, Zhu X and Birnbaumer L: Mouse trp2, the homologue of the human trpc2 pseudogene, encodes mTrp2, a store depletion-activated capacitative Ca\(^{2+}\) entry channel. Proc Natl Acad Sci USA 96, 2060 – 2064 (1999)

Inoue R, Okada T, Onoue H, Hara Y, Shimizu S, Naitoh S, Ito Y and Mori Y: The transient receptor potential protein homologue TRP6 is the essential component of vascular \(\alpha_{1}\)-adrenergic receptor-activated Ca\(^{2+}\)-permeable cation channel. Circ Res 88, 325 – 332 (2001)

Acharya JK, Jalink K, Hardy RW, Hartenstein V and Zuker CS: InsP\(_3\), receptor is essential for growth and differentiation but not for vision in Drosophila. Neuron 13, 881 – 887 (1997)

Thorn P and Petersen OH: Nonselective cation channels in exocrine gland cells. In Nonselective Cation Channels, Edited by Siemen D and Heschler J, pp 185 – 200, Birkhäuser, Basel (1993)

Siemen D: Nonselective cation channels. In Nonselective Cation Channels, Edited by Siemen D and Heschler J, pp 3 – 25, Birkhäuser, Basel (1993)

Balzer M, Lintchinger B and Groschner K: Evidence for a role of TRP proteins in the oxidative stress-induced membrane conductances of porcine aortic endothelial cells. Cardiovasc Res 42, 543 – 549 (1999)

Denyer JC and Brown HF: Pacemaking in rabbit isolated sino-atrial node cells during Cs\(^+\) block of the hyperpolarization-activated current if. J Physiol (Lond) 429, 401 – 409 (1990)

Hagiwara N, Irisawa H, Kasanuki H and Hosoda S: Background current in sino-atrial node cells of the rabbit heart. J Physiol (Lond) 448, 53 – 72 (1992)

Sankaranarayanan S and Simasko SM: A role for a background sodium current in spontaneous action potentials and secrettion from rat lactotrophs. Am J Physiol 271, C1927 – C1934 (1996)

Mubagwa K, Stengl M and Flameng W: Extracellular diveralent cations block a cation non-selective conductance unrelated to calcium channels in rat cardiac muscle. J Physiol (Lond) 502, 235 – 247 (1997)

Bae YM, Park MK, Lee SH, Ho W-K and Earm YE: Contribution of Ca\(^{2+}\)-activated K\(^+\) channels and non-selective cation channels to membrane potential of pulmonary arterial smooth muscle cells of the rabbit. J Physiol (Lond) 514, 747 – 758 (1999)

Wang Q and Large WA: Noradrenaline-activated cation conducance recorded with the nystatin whole-cell method in rabbit portal vein cells. J Physiol (Lond) 435, 21 – 39 (1991)

Inoue R and Kuriyama H: Dual regulation of cation-selective channels by muscarinic and alpha 1-adrenergic receptors in the rabbit portal vein. J Physiol (Lond) 465, 427 – 448 (1993)

Jungnickel MK, Marrero H, Birnbaumer L, Lemos JR and Florman HM: Trp2 regulates entry of Ca\(^{2+}\) \(\rightarrow\) mouse sperm triggered by egg ZP3. Nat Cell Biol 3, 499 – 502 (2001)

Freichel M, Suh SH, Pfeifer A, Schweig U, Trost C, Weissgerber P, Biel M, Philipp S, Freise D, Droogmans G, Hofmann F, Flockezer V and Nilius B: Lack of an endothelial store-operated Ca\(^{2+}\) current impairs agonist-dependent vasorelaxation in TRP4\(-/-\) mice. Nat Cell Biol 3, 121 – 127 (2001)

Clementi E and Meldolesi J: Pharmacological and functional properties of voltage-independent Ca\(^{2+}\) channels. Cell Calcium 19, 269 – 279 (1996)

Broad LM, Braun FJ, Lievremont JP, Bird GSJ, Kuroasaki T and Putney JW Jr: Role of the phospholipase C-inositol 1,4,5-trisphosphate pathway in calcium release-activated calcium current and capacitative calcium entry. J Biol Chem 276, 15945 – 15952 (2001)

Ma HT, Venkatachalam K, Li HS, Montell C, Kuroasaki T, Patterson RL and Gill DL: Assessment of the role of the inositol 1,4,5-trisphosphate receptor in the activation of transient receptor potential channels and store-operated Ca\(^{2+}\) entry channels. J Biol Chem 276, 18888 – 18896 (2001)

Iwasaki H, Mori Y, Hara Y, Uchida K, Zhou H and Mikoshiba K: 2-Aminoethoxylidinyl borate (2-APB) inhibits capacitative calcium entry independently of the function of IP\(_3\)Rs. Receptors Channels (in press)

Zhang XF, Iwamuro Y, Enoki T, Okazawa M, Lee K, Komuro T, Minowa T, Okamoto Y, Hasegawa H, Furutani H, Miwa S and Masaki T: Pharmacological characterization of Ca\(^{2+}\) entry channels in endothelin-1-induced contraction of rat aorta using LOE 908 and SK&F 96365. Br J Pharmacol 127, 1388 – 1398 (1999)

Haretneck C, Plant TD and Schultz G: From worm to man: three subfamilies of TRP channels. Trends Neurosci 23, 159 – 166 (2000)

Runnels LW, Yue L and Clapham DE: TRP-PLIK, a bifunctional protein with kinase and ion channel activities. Science 291, 1043 – 1047 (2001)

Nadler MJ, Hermosura MC, Inake K, Perraud AL, Zhu Q, Stokes AJ, Kuroasaki T, Kinet JP, Penner R, Scharenberg AM and
Fleig A: LTRPC7 is a MgATP-regulated divalent cation channel required for cell viability. Nature **411**, 590 – 595 (2001)

Perraud AL, Fleig A, Dunn CA, Bagley LA, Launay P, Schmitz C, Stokes AJ, Zhu Q, Bessman MJ, Penner R, Kinet JP and Scharenberg AM: ADP-ribose gating of the calcium-permeable LTRPC2 channel revealed by Nudix motif homology. Nature **411**, 595 – 599 (2001)

Hara Y, Wakamori M, Ishii M, Maeno E, Yamada H, Shimizu S, Okada Y, Imoto K and Mori Y: LTRPC2 Ca²⁺-permeable channel activated by changes in redox status confers susceptibility to cell death. Mol Cell (in press)

Barritt GJ: Receptor-activated Ca²⁺ inflow in animal cells: a variety of pathways tailored to meet different intracellular Ca²⁺ signalling requirements. Biochem J **15**, 153 – 169 (1999)