Role of ion channels in regulating Ca\(^{2+}\) homeostasis during the interplay between immune and cancer cells

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Ion channels are abundantly expressed in both excitable and non-excitable cells, thereby regulating the Ca\(^{2+}\) influx and downstream signaling pathways of physiological processes. The immune system is specialized in the process of cancer cell recognition and elimination, and is regulated by different ion channels. In comparison with the immune cells, ion channels behave differently in cancer cells by making the tumor cells more hyperpolarized and influence cancer cell proliferation and metastasis. Therefore, ion channels comprise an important therapeutic target in anti-cancer treatment. In this review, we discuss the implication of ion channels in regulation of Ca\(^{2+}\) homeostasis during the crosstalk between immune and cancer cell as well as their role in cancer progression.

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Facts

- Ion channels regulate Ca\(^{2+}\) influx and downstream signaling pathways in immune and cancer cells.
- Altered regulation of ion channels is implicated in carcinogenesis.
- Cytotoxicity of immune cells against cancer cells depends highly on Ca\(^{2+}\) signaling.
- Ion channels comprise an attractive tool for targeted therapy for cancer.

Open Questions

- Are blockers of K\(^{+}\) and CRAC channels able to inhibit cancer progression?
- What is the role of immune cell-specific ion channels in cancer therapy?
- What cancer-specific ion channels are involved in neoplastic transformation in vivo?

Physiological processes depend on the continued flow of ions into and out of cells defeating a barrier impermeable to ions such as plasma membrane, which is built in a form of phospholipid bilayer. Thus, the hydrophobic membrane acts as a serious energy barrier for transporting ions. Ions are charged molecules that have low solubility in the hydrocarbon core of lipid bilayer, thereby having low permeability coefficients across the bilayer. There is a large difference in the electric potential between the two sides of a biological membrane. In order to transfer ions across the membrane and equilibrate both sides of the membrane, eukaryotic cells are equipped in the integrally embedded pore-forming membrane proteins (ion channels) and biological pumps. Such structure allows for the passage of ions through the channel. Opening and closing of the ion channel is usually controlled chemically or mechanically. Depending on the type of ion channel, its conformational change may occur because of changes in the membrane potential (voltage-gated channels), ligand binding (chemical activation) or ligand-driven stretching of the membrane (stretch-activated ion channels). Body response to the external stimuli can be linked to the regulation of ion channel activity. Ion channels play a crucial role in various physiological processes including flow of nerve impulses, muscle contraction, cell division and hormone secretion.\(^{1}\) The intracellular concentration of the key signaling ion such as calcium (Ca\(^{2+}\)) depends on electrical gradients driven in turn by sodium (Na\(^{+}\)) and potassium (K\(^{+}\)) channels. The role of ion channels in pathogenesis of various diseases including cancer and its treatment has been extensively studied. The prime function of an immune cell is to remove...
cancer cells from the body by cytotoxic T lymphocytes (CTL or CD8+ cells) and natural killer (NK) cells through polarized discharge of the contents of cytotoxic granules towards the target cells. The effector function of CTL and NK cells as well as their proliferation and apoptosis of cancer cells largely depend on Ca2+ signaling. The role of ion channels in the regulation of intracellular Ca2+ concentration is well described in the literature. Alterations in Ca2+ homeostasis due to ion channel dysfunction contribute to the common traits of neoplastic transformation, which are known as hallmarks of cancer. These hallmarks include different stages of tumor development like unlimited replication, tissue invasion and metastasis, evasion of apoptosis, sustained angiogenesis, self-sufficiency in growth signals and insensitivity to anti-growth signals. Additionally, modulation of ion-channel-mediated Ca2+ concentration in CTLs regulates their antitumor action.

Regulation of Intracellular Ca2+ Concentration

Na+ and K+ are the most abundant cations in biological systems. Na+ ions are present at high concentrations outside the cell, unlike K+ ions that are present at high concentrations inside the cell. Gradients for these ions across the cell membrane provide the energy source for action potentials generated by opening of Na+ and K+ channels and for transporting solutes and other ions across the cell membrane via coupled transporters. Among several ions, the gradient for Ca2+ ions is the largest. The cytosol is surrounded by two big Ca2+ stores: the extracellular space, where the Ca2+ concentration is ~1.8 mM, and the sarco-endoplasmic reticulum, where the Ca2+ concentration varies from 300 μM to 2 mM. In immune cells, the intracellular Ca2+ concentration is ~0.1 μM in the resting state, but it is significantly increased (~10-fold) when the cells are activated. Plasma membrane Ca2+ channels and Ca2+ influx are particularly important at different steps of the cell-cycle progression and proliferation of immune cells. The molecular features of Ca2+ channels are well defined, which allows for the distinction of four main types of these channels including voltage-activated, receptor-activated, store-operated and second messenger-operated channels. Voltage-activated, receptor-activated, store-operated and second messenger-operated channels are ubiquitous, whereas voltage-activated calcium channels are specific for excitable cells. Voltage-activated calcium channels (e.g., L-, T-, N-, P-, Q-type Ca2+ channels) open when the plasma membrane is depolarized. Receptor-activated calcium channels (e.g., P2X purinergic receptors) open when a ligand binds to the channel, whereas store-operated calcium channels (e.g., transient receptor potential (TRP)) and archetype calcium release-activated channels (CRAC) are activated when the level of Ca2+ within the lumen of the ER decreases below a threshold level. Another type, second messenger-operated channels (e.g., arachidonic acid-regulated Ca2+ current) are activated by intracellular second messengers like arachidonic acid. The role of CRAC, TRPM4 and P2X channels are important in case of immune cells in the continuous effort to keep Ca2+ at an optimal level in order to maintain the cellular functions in parallel with ion pumps like Na+/K+ pumps. In non-excitable cells including immune cells, the membrane potential plays an important role in setting the electrical driving force for Ca2+ entry. In cells where voltage-independent Ca2+ channels like TRPM4 and two-pore K+ channels (K2p) are present, Ca2+ influx only depends on the electrochemical gradient over the membrane and intensifies when the membrane potential is more negative (hyperpolarized).

Among different ion channels involved in the regulation of Ca2+ homeostasis, CRAC channels are the most important. CRAC channels have been widely characterized and are known because of their high ion selectivity for Ca2+ and low conductance. CRAC channels are activated through the binding of the endoplasmic Ca2+ depletion sensor, known as stromal interaction molecule 1 (STIM1) and STIM2 to the CRAC channel units ORAI1-3 (also known as CRACM1-3). ORAI1 is a widely expressed surface glycoprotein with four predicted transmembrane domains, intracellular amino- and carboxyl-termini and no sequence homology to other ion channels except for its homologues ORAI2 and ORAI3. The activation of ORAI/CRAC channels involves a complex series of coordinated steps, during which STIM proteins sense the depletion of ER Ca2+ stores and pass on this store depletion to the CRAC channels. In resting cells with filled Ca2+ stores, STIM proteins are diffusely distributed all over the ER membrane. Following the depletion of Ca2+ stores, STIM proteins get activated, oligomerize and redistribute into puncta within junctional ER sites, which are in close proximity to the plasma membrane.

Role of Ion Channels in Maintaining the Normal Membrane Potential

The resting potential of a lymphocyte membrane is ~50 mV. Membrane potential alterations mainly occur when lymphocytes get activated. TCR engagement activates PLCγ1, which catalyzes the hydrolysis of phosphatidylinositol 4,5-biphosphate (PIP2) into inositol trisphosphate (IP3) and di-acetyl glycerol. IP3 stimulates the release of Ca2+ from intracellular ER stores, which triggers the opening of plasma membrane CRAC channels. It is the resulting influx of extracellular Ca2+ that is responsible for the sustained rise in cytoplasmic Ca2+ after TCR stimulation. Ca2+ binds to the cytoplasmic Ca2+-dependent protein calmodulin, which then activates the phosphatase calcineurin. This phosphatase dephosphorylates and activates the nuclear factor of transcription of activated T cells (NFAT), which enters the nucleus and helps to initiate interleukin-2 (IL-2) gene transcription. During the activation of immune cells, opening of CRAC channels raises the intracellular Ca2+ level. To maintain the balance in membrane conductance, KCa channels get opened to hyperpolarize the membrane, which results in Ca2+ efflux. A negative feedback loop is established when the level of Ca2+ inside the cell is high enough to inhibit CRAC channels. Beside the Ca2+-dependent activation of TRPM4 channels in T cells, there is also involvement of K1.3 channels in order to repolarize the membrane (Figure 1). Along with these conventional ion channels, the K2p TWIK-related acid-sensitive K+ channels 1 and 3 (TASK-1/K2p3.1 and
Ion Channels in Immune Cells

Activation and the effector role of immune cells is dependent on Ca\(^{2+}\) influx, which is regulated by a group of ion channels located in the plasma membrane of the cell. The detailed characteristics of certain ion channels and their implication in the cellular functions became possible with the help of 'gold standard' patch-clamp technique. The role of individual types of ion channels in the physiology of immune cells is briefly presented.

K\(^{+}\) channels. K\(^{+}\) channels comprise the major ion channel family expressed in immune cells that regulate important cellular processes including Ca\(^{2+}\)-mediated cellular proliferation, migration and finally controlling cell volume. They regulate membrane potential by driving K\(^{+}\) efflux resulting in membrane hyperpolarization. From the superfamily of K\(^{+}\) channels, immune cells express voltage-gated (K\(_{v}\)), calcium-activated (K\(_{Ca}\)), inwardly rectifying potassium (Kir) and two-pore gated channels (K\(_{2P}\)). In regard to the structural diversity of the channels, there are several types like six transmembrane one pore (K\(_{v}\)) or transmembrane two pore (K\(_{2P}\)). K\(_{v}\) channels are further subdivided into three conserved gene families: Kv (shaker-like), Ether-a-go-go (EAG) and KCNQ (K\(_{v,7}\)). In addition, K\(_{Ca}\) channels are grouped into big-conductance calcium-activated channels (BK\(_{Ca}\) (K\(_{Ca,1.1}\))), intermediate-conductance calcium-activated channels (IK\(_{Ca}\) (K\(_{Ca,3.1}\))) and small-conductance calcium-activated channels (SK\(_{Ca}\) (K\(_{Ca,2.1}\), K\(_{Ca,2.2}\), K\(_{Ca,2.3}\))).

The role of K\(_{v,1.3}\) and K\(_{Ca,3.1}\) in mediating the efflux of K\(^{+}\) in order to maintain the hyperpolarization of the cell membrane (Figure 1) is well explained in the literature. K\(^{+}\) channels are differently expressed in various subsets of lymphocytes followed by their activation. For example, naïve and regulatory human T cells mainly express K\(_{v,1.3}\), whereas the expression of K\(_{Ca,3.1}\) is upregulated upon activation by cognate antigen. Interestingly, a recent study has shown that K\(_{v,1.3}\) channels are indispensable for the differentiation of CD8\(^{+}\) T cells into effector cells with cytotoxic ability. Moreover, K\(_{v,1.3}\) channels accumulate specifically at the immune synapse (IS) between cytotoxic and target cells in order to modulate the killing process mediated by CTL and NK cells. In addition, blocking of K\(_{Ca,3.1}\) in NK cells increases their tumor cell killing ability and comprises an excellent target for cancer immunotherapy.

K\(_{v}\) channels are responsible for stabilization of the resting membrane potential near to the K\(^{+}\) equilibrium potential by passing positive charge mostly into the cell (inward direction) rather than in the opposite direction. This type of channels is present in a significant amount in macrophages, dendritic cells and microglia. Studies have shown that K\(_{v,2.0}\) and K\(_{v,4.0}\) family members interact with NIL-16, neuronal variant of interleukin 16 (IL-16). As the cytokine IL-16 has been characterized mostly in the immune system, the identification...
of NIL-16 emphasizes the connection of K<sub>i</sub>P channels with the immune and nervous system. On the basis of the observation that memantine inhibits the amplitude of inwardly rectifying K<sup>+</sup> current though the K<sub>i</sub>P channels in macrophages and microglial cells, it is postulated that blocking the K<sub>i</sub>P channels may influence the functional activity of macrophages. K<sub>i</sub>P (KCNK), better known as 'leak channels' are important for setting the resting membrane potential. Furthermore, their action is mainly voltage-independent and can be regulated via various stimuli including mechanical stimulation, lipids, G<sub>q</sub> proteins or muscarine. TASK-1/K<sub>q</sub>P3.1 and TASK-3/K<sub>q</sub>P9.1, the two functional members of the K<sub>q</sub>P family are expressed in T lymphocytes and contribute to the modulation of T-cell effector function including interferon-γ (IFN-γ) and IL-2 secretion as well as T-cell proliferation. Selective blockade of TASK channels present on T lymphocytes leads to improvement of the experimental autoimmune encephalomyelitis course, a model of multiple sclerosis.

**Transient receptor potential (TRP) channel.** Among the superfamily of 28 TRP cation channels, immune cells mainly express TRPM2 and TRPM subfamilies like TRPC-1, 3, 5 and TRPM2, 4, 7. These channels have biological properties to be non-selective and permeable to several cations like Ca<sup>2+</sup> and Na<sup>+</sup>. Regulation of intracellular Ca<sup>2+</sup> concentration is indispensable for lymphocyte activation, and TRP channels may both increase Ca<sup>2+</sup> influx (TRPC3) or decrease Ca<sup>2+</sup> influx through membrane depolarization (TRPM4). The function of TRPM4 channel is well documented in maintaining the normal membrane potential of an immune cell and controlling the Ca<sup>2+</sup> flux mechanism. Interestingly, TRPM4 channel mainly conducts Na<sup>+</sup> and K<sup>+</sup> cations. Activation of TRPM4 channels occurs in response to the increase in intracellular Ca<sup>2+</sup> concentration resulting in Na<sup>+</sup> influx, membrane depolarization and a reduction in electrical driving force for Ca<sup>2+</sup> influx (Figure 1). Therefore, TRPM4 channel acts as a negative feedback mechanism for the regulation of store-operated Ca<sup>2+</sup> entry by CRAC/ORAI as thereby preventing the cellular Ca<sup>2+</sup> overload.

**Purinergic receptors.** P2X receptors are membrane ion channels with the ability to influx several non-selective cations like Na<sup>+</sup> and Ca<sup>2+</sup>, and are activated by extracellular adenosine 5'-triphosphate (ATP). P2X receptors belong to the class of ligand-activated ion channels and there are three P2X receptors expressed in human T cells: P2X1, 4, 7. Among these three, principally P2X7 is abundantly expressed in immune cells and regulates Ca<sup>2+</sup> influx process resulting in the activation of downstream signaling mediators and T-cell proliferation.

**Store-operated calcium channels (SOCs),** CRAC is the major store-operated Ca<sup>2+</sup> channel of immune cells with the biophysical properties of higher Ca<sup>2+</sup> dependence and low conductivity in the range of 0.024–0.4 ps. CRAC channels get opened with the signal of depleting endoplasmic reticulum (ER) Ca<sup>2+</sup> pool. This signal in ER is mainly mediated by ER Ca<sup>2+</sup> sensors stromal interaction molecule (STIM) 1 and STIM2 and transferred to the pore-forming subunits of the CRAC channel, mainly ORAI1–3. This results in the activation of the CRAC channel. Lymphocytes express two STIM isoforms, STIM1 and STIM2, which mediate store-operated Ca<sup>2+</sup> entry in B and T cells. CD4<sup>+</sup> and CD8<sup>+</sup> T cells from ORAI1- and STIM1-deficient patients exhibit defective production of various cytokines, including IL-2, IL-17, IFN-γ and tumor necrosis factor (TNF). Furthermore, store-operated calcium entry is indispensable for the cytotoxic action of CTLs. STIM1- and STIM2-mediated store-operated calcium entry in CD8<sup>+</sup> T cells is crucial for anti-tumor immunity.

**Anti-tumor Action of Immune Cells**

Human immune system has the great potential to destroy cancer cells either by CTL or NK cells without being toxic to the healthy tissue and organs. These distinct immune cells are able to recognize cancer cell by forming a Ca<sup>2+</sup>-dependent cytotoxic IS with the cancer cell and perform a killing mechanism either through the release of lytic granules and granzymes, or by the activation of Fas-FasLigand receptors (known as death receptors). Efficient CRAC channels and the resulting increase in the cytosolic Ca<sup>2+</sup> concentration are necessary for adherence to the target cell as well as its recognition. The adhesion molecule, particularly lymphocyte function-associated antigen 1 (LFA-1) integrin is essential for this process and interacts with Ca<sup>2+</sup> in diverse ways. This includes inside-out (transmission of the regulatory signals originating within the cytoplasm to the external ligand-binding domain of the receptor) signaling-based LFA-1 activation or outside-in (transmission of chemical signals into the cell) signaling via LFA-1. Interaction between CTL and epithelial tumor cell is integrin-dependent and promotes maturation of the cytotoxic IS and modulates anti-tumor CTL response. Additionally, LFA-1 activation is implicated in mitochondria positioning at the IS in order to control Ca<sup>2+</sup>-influx through CRAC/ORAI Ca<sup>2+</sup> channels. It has recently been shown that store-operated Ca<sup>2+</sup> release driven by ORAI1 is crucial for lytic granule exocytosis in NK cells and CTLs as well as production of cytokines (TNF-α and IFN-γ) by NK cells. Furthermore, delineation of the accurate STIM/ORAI1 ratio could be a feature of the killing efficiency of CTL and NK cells. Ca<sup>2+</sup> does not directly play a role in the formation of the IS, but it has enormous effect in controlling the duration and kinetics of the cytotoxic IS between killer immune and cancer cell. Along with the depolarizing nature of cancer cells, Ca<sup>2+</sup> concentration can also be a marker of the action of a killer T cell. Small fluctuations from the external Ca<sup>2+</sup> (~1.2 mM) range of a cancerous tissue can indicate the influence of cancer cell killing by CTL or NK cells.

**Ion Channels in Cancer**

Ion channels comprise an important factor influencing the formation and development of tumors. Such malignant transformation leads to enhanced proliferation, abnormal differentiation, impaired apoptosis, and finally uncontrolled migration and
invasion (Table 1). This is often associated with altered levels of ion channel expression as well as their activity in the mutated cancer cells. The role of ion channels in pathogenesis of various diseases including cancer and its treatment has been extensively studied. The major types of ion channels implicated in carcinogenesis are presented below.

**Voltage-gated K⁺ channels**

*Shaker-like:* Shaker-type of voltage-gated K⁺ channels regulate cell cycle progression by four mechanisms such as controlling membrane potential oscillations, controlling cell volume dynamics, controlling calcium signaling and promoting malignant growth through the migratory pathway. Influence of voltage-dependent K⁺ channels in the early stages of cancer development confirms the evidence for the overexpression of these channel proteins in cells exposed to chemical carcinogens. It has been shown that voltage-gated K⁺ channels affect tumor cell proliferation through the regulation of the membrane potential. As an example, overexpression of Kᵥ1.1 and Kᵥ1.3 are found in glioma, lymphoma, breast, lung, pancreas and prostate cancer. Furthermore, Kᵥ1.3 channel overexpression is also linked

| Ion channels                        | Expression profile                                      | Cancer type                                      | References |
|-----------------------------------|--------------------------------------------------------|-------------------------------------------------|------------|
| **Proliferation of cancer cells**  |                                                        |                                                 |            |
| Shaker-like K⁺ channels (Kᵥ1.1, Kᵥ1.3, Kᵥ1.5) | Gene and protein upregulation                          | Glioma, breast cancer, lung cancer, pancreas cancer, prostate cancer, lymphoma | 64,123     |
| EAG K⁺ channels (EAG1, EAG2)      | Gene and protein upregulation                          | Medulloblastoma, breast cancer, head and neck cancer, melanoma, gastrointestinal tract cancer | 65–67      |
| EAG-related K⁺ channels (HERG/Kᵥ11.1) | Gene and protein upregulation                          | Melanoma, neuroblastoma, breast cancer          | 68         |
| Ca²⁺-activated K⁺ channels (Kᵥ3.1) | Gene and protein upregulation                          | Glioma, breast cancer, ovarian cancer, prostate cancer, melanoma | 124–127    |
| TRP (TRPC6, TRPV6, TRPM7, TRPM8)  | Gene and protein upregulation                          | Breast cancer, prostate cancer, head and neck cancer, human glioblastoma cell line | 89,95–97,128,129 |
| P2Y (P2Y2), P2X (P2X7), P2U       | Gene and protein upregulation                          | Melanoma, colorectal cancer cells, lung cancer cells | 101,130,131 |
| SOCs (ORAI1/STIM1)                | Gene and protein upregulation                          | Lung cancer cells, cervical cancer               | 113,132    |
| SOCs (ORAI1/STIM1)                | Gene and protein upregulation                          | Cervical cancer, glioblastoma cells              | 113,133    |

**Cell migration and metastasis**

| Ion channels                        | Expression profile                                      | Cancer type                                      | References |
|-----------------------------------|--------------------------------------------------------|-------------------------------------------------|------------|
| EAG K⁺ channels (EAG1/ Kᵥ10.1)    | Gene and protein upregulation                          | Migration of breast cancer cells                | 134        |
| Ca²⁺-activated K⁺ channels (KCNA1, SK3/ORAI1, Kᵥca3.1, Kᵥca3.1) | Gene and protein upregulation                          | Breast cancer → metastasis to brain             | 75–78,135  |
| Kᵥ channels (Kᵥ3.1/GIRK1)         | Gene and protein upregulation                          | Breast cancer → bone metastasis                 | 75–78,135  |
| TRP (TRPM7, TRPM8, TRPV1, TRPV6)  | Gene and protein upregulation                          | Migration of glioma cells, transformed renal epithelial cells and breast cancer cells | 81         |
| P2X (P2X7)                        | Gene and protein upregulation                          | Primary breast cancer → axillary lymph node metastasis | 90,91,97,136–138 |
| SOCs (ORAI1/STIM1)                | Gene and protein upregulation                          | Lung cancer cells, primary breast cancer, prostate cancer cells, squamos carcinoma, hepatoblastoma | 139        |

**Tumor angiogenesis**

| Ion channels                        | Expression profile                                      | Cancer type                                      | References |
|-----------------------------------|--------------------------------------------------------|-------------------------------------------------|------------|
| EAG K⁺ channels (EAG1)            | Gene and protein upregulation                          | Breast cancer and other solid tumors            | 65,66      |
| TRP (TRPC6)                       | Gene and protein upregulation                          | Human glioblastoma cell line                    | 88,94,141  |
| SOCs (ORAI1/STIM1)                | siRNA- or dominant-negative mutant-mediated knockdown  | VEGF-induced angiogenesis observed in tumors     | 141,142    |

**Apoptosis resistance**

| Ion channels                        | Expression profile                                      | Cancer type                                      | References |
|-----------------------------------|--------------------------------------------------------|-------------------------------------------------|------------|
| Shaker-like K⁺ channels (Kᵥ1.3)    | Gene and protein upregulation                          | Large B-cell lymphoma, glioma                    | 64         |
| TRP (TRPA1)                       | Gene and protein upregulation                          | Lung cancer cell line                            | 143        |
| P2X (P2X7)                        | Gene and protein downregulation                        | Breast cancer, melanoma                          | 104        |
| SOCs (ORAI1)                      | siRNA-mediated knockdown                               | Prostate cancer cell line                        | 109,144    |

Table 1 The role of distinct ion channels in cancer development and progression
with resistance to apoptosis as shown by the upregulation of K<sub>1.3</sub> expression in diffuse large B-cell lymphoma and glioma.

**EAG channels:** The EAG subfamily of voltage-gated K<sup>+</sup> channels is divided into three distinct groups including EAG (EAG1/K<sub>10.1</sub>; EAG2/K<sub>10.2</sub>), EAG-like K<sup>+</sup> (ELK) and EAG-related (HERG/K<sub>11.1</sub>). EAG1 overexpression has showed tumorigenic potential and poor overall patient survival in multiple cancer types. Additionally, EAG1 plays a significant role in cell proliferation and tumor angiogenesis. Another member of the EAG subfamily of voltage-gated K<sup>+</sup> channels, particularly EAG2, regulates cell volume dynamics important for cell cycle progression and cell proliferation in medulloblastoma. Similar to EAG1, HERG overexpression is found in brain, breast, gastrointestinal tract, head and neck, kidney, lung, melanoma, ovary, and thyroid cancers. Moreover, HERG expression correlates with TNF-mediated tumor cell proliferation.

**K<sub>ir</sub> channels:** As mentioned above, K<sub>ir</sub> channels allow for easy movement of K<sup>+</sup> into the cell. They are activated by PIP<sub>2</sub>, but they can also be modulated by other regulatory factors such as ATP (ATP-sensitive K<sup>+</sup> channels) and G-proteins (G protein-gated K<sub>ir</sub> channels) or by some non-specific regulators including polyamines, kinases, pH and Na<sup>+</sup> ions.

The mRNA upregulation of the G-protein regulated inward-rectifier K<sup>+</sup> (GIRK) channel called K<sub>ir3.1</sub> (GIRK1) has been shown in invasive breast cancer and non-small-cell lung cancer. Additionally, overexpression of GIRK1 in both types of tumors was correlated with poor prognosis for the patients.

**TRP channels.** TRP cation channels have been implicated in various pathological states including cancer due to their role as intracellular Ca<sub>2+</sub>-release channels. Recent studies have shown the association of TRP channels with various cancer types such as melanoma, prostate cancer, glioblastoma, hepatoblastoma (TRPV1 and TRPC6). Besides the roles of volume control and motility, TRP channels are considered a good pharmaceutical target for cancer therapy.

**Purinergic Receptors.**

The ATP-dependent activity of P2X7 channel is associated with various physiological functions including cell proliferation, cell death and cytokine secretion. Recent studies have implicated the role of P2X and P2Y receptors in B cell leukemia, melanoma and colorectal cancer. Targeting the P2X7 receptor by selective P2X7 agonists as well as P2X7 antagonists in cancer has shown antitumor effect. Furthermore, the effect of ATP infusion in patients with advanced lung cancer has proven the potential of ATP, which might become an anti-cancer agent in the future. However, larger studies are required in order to verify these findings.

**Store-operated calcium channels (SOCs).** SOC-mediated sustained increase in the cytosolic Ca<sub>2+</sub> has shown to trigger apoptosis in tumor cells. STIM1-ORAI1 driven store-operated calcium entry seems to be indispensable for migration and metastasis of breast cancer, cervical cancer and hepatocarcinoma, which was potently blocked by the
store-operated calcium entry inhibitor.\textsuperscript{110–113} Moreover, CRAC channels are implicated in VEGF-activated Ca\textsuperscript{2+} influx promoting angiogenesis, which might be crucial for cancer progression.\textsuperscript{111}

Ion Channel Modulators

Ion channels are often overexpressed in numerous types of tumors and their altered activity plays a significant role in apoptosis resistance, proliferation and metastasis of cancer cells. Thus, blocking the activity of ion channels seems to be an obvious strategy to impair cancer growth. However, such treatment is not as straightforward as it may look. When targeting ion channels, we aim at efficient killing of cancer cells without causing toxic effects in other tissues expressing the same or related channels. A vast amount of known ion channels blockers are used to treat cardiac arrhythmias or epilepsy (anticonvulsants);\textsuperscript{114} thus, incorporating them into oncology is accompanied by the risk of heart or nervous system disorders.

Unspecificity of ion channel blockers is still a big challenge that needs to be overwhelmed to avoid serious side effects during oncological treatment. Specific inhibition can be obtained by developing monoclonal blocking antibodies, antisense oligonucleotides, small interfering RNAs, peptide toxins and novel small organic compounds.\textsuperscript{115} As discussed by Arcangeli and Becchetti, to improve the efficiency of ion channels targeting cancer, one should also focus on finding inhibitors recognizing conformational changes in ion channels (e.g., open channel versus close channel). So far, such an approach was found to be possible in a case of lamotrigine and lidocaine that preferentially target open and inactivated voltage-gated Na\textsuperscript{+} channels, without distinguishing other conformational states.\textsuperscript{116} Similar property exhibits in R-roscovitine recognizing open HERG channel.\textsuperscript{117}

Interesting alternative for conventional ways of targeting ion channels in cancer treatment are some dietary compounds.\textsuperscript{118} Curcumin, resveratrol (grape polyphenol), docosahexaenoic acid (omega-3) and epigallocatechin gallate (catechin from green tea) extract were shown to modulate ion channels activity and suppress migration and growth of breast and ovarian cancer cells.\textsuperscript{119–122} Other examples of targeting ion channels in cancer and immune cells are presented in Table 2.

### Table 2 Ion channel blockers in immune and cancer cells

| Ion channel blocker | Ion channel | Cell type | Comments | References |
|---------------------|-------------|-----------|----------|------------|
| Margatoxin (MgTX)   | Kv1.3       | T lymphocytes, Jurkat cells, NK cells, leukemia cells | Antiproliferative effect in T-lymphocytes, regulation of immunoresponsiveness | 145,146 |
| Charybdotoxin (CTX) | Kv1.3, Kv3.1 | Leukemia | Inhibition of KCa3.1 increased the degranulation of adherent NK cells and their ability to kill K562 leukemia cells | 147 |
| TRAM-34, NS6180, ShK-186 | Kv1.3, Kv2.1, Kv4.2, HERG (Kv11.1) | | | |
| R-roscovitine       | Kv1.3, Kv2.1, Kv4.2, HERG (Kv11.1) | Pancreas carcinoma, breast cancer | Roscovitine is well known cyclin-dependent kinase inhibitor, inhibited tumor cell growth both in vivo and in vitro | 148,149 |
| Way 123,398         | HERG (Kv11.1) | Colorectal cancer | Reduced cell migration of H630, HCT and HCT8 cells; unaffected growth of HEK 293 cells | 150 |
| Way 123,398; CsCl; E4031 | HERG (Kv11.1) | Acute myeloid leukemia, Gastric cancer | Impaired cell proliferation, increased survival rate for patients treated with verapamil + chemotherapy | 151,153 |
| Cisapride           | | | | |
| Verapamil           | HERG (Kv11.1) | Lung cancer, melanoma, colon cancer | Decrease in intracellular ATP concentration leads to autophagy in glioma cells | 152,153 |
| UNBS0 (Cardenolide) | Na\textsuperscript{+}/K\textsuperscript{+} ATPase | Glioblastoma | Decrease in intracellular ATP concentration leads to autophagy in glioma cells | 154 |
| Tetrodotoxin (TTX)  | Nav1.5, Nav1.6 | Human melanoma, macrophages, breast cancer | Decrease in intracellular ATP concentration leads to autophagy in glioma cells | 155,156 |
| Charybdotoxin (CTX) | KC\textsubscript{r} (IK1) | Human melanoma | Reduced migration of melanoma cells treated with CTX | 157 |
| Zinc, methanandamide | K\textsubscript{a}2p.9.1 (TASK-3) | Ovarian cancer | Reduced cell proliferation and increase in apoptosis | 158,159 |

Conclusions and Future Perspectives

The main task of the immune system is to defend against attacks by foreign invaders including bacteria, viruses, fungi, parasites and other microorganisms. It has been shown by the researchers from both immunology and oncology fields that cancer cells are also recognized by the immune system, and their proliferation can be controlled immunologically. Alterations in ion channel-based Ca\textsuperscript{2+} signaling are linked to the behavior of cancer cells. Recent studies indicate the significance of ion channels and Ca\textsuperscript{2+} signaling in activation of cancer killing immune cells as well as cancer progression. Generation of an appropriate Ca\textsuperscript{2+} response, which is induced by recognition of a tumor antigen is driven by above-described ion channels (Figure 2). Regulation of certain features of cancer cells by decreasing the activity of ion channel proteins is still under investigation. The market success of Ambien (GABAA receptor inhibitor for the treatment of insomnia) and
Norvasc (Ca\(^{2+}\) channel blocker used to lower blood pressure and to treat angina pectoris) have energized the drug market to explore more the ion channel field searching for new therapeutics including cancer therapy. Nevertheless, the ion channel-based treatment comprises still far unused anti-cancer strategy. Thus, future research will focus on ion channels as therapeutic target in order to inhibit proliferation of cancer cells and promote their apoptosis together with modulation of cancer-specific cytotoxicity of immune cells. Furthermore, studies involving mutating ion channels in cancer using animal models should uncover novel insights into the ion channel function in tumorigenesis.

Conflict of Interest
The authors declare no conflict of interest.

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1. Berdichevsky D, Lipp P, Bootman MD. The versatility and universality of calcium signalling. Nat Rev Mol Cell Biol 2000; 1: 11–21.
2. Dustin ML, Long EO. Cytotoxic immunological synapses. Immunol Rev 2010; 235: 24–34.
3. Prevarskaya N, Skryma R, Shuba Y. Ion channels and the hallmarks of cancer. Trends Mol Med 2010; 16: 107–121.
4. Hanahan D, Weinberg RA. The hallmarks of cancer. Cell 2000; 100: 57–70.
5. Schwarz EC, Qu B, Hoth M. Calcium, cancer and killing: the role of calcium in killing cancer cells by cytotoxic T lymphocytes and natural killer cells. Biochim Biophys Acta 2013; 1833: 1603–1611.
6. Weidinger C, Shaw PJ, Feske S. STIM1 and STIM2-mediated Ca\(^{2+}\) influx regulates antitumor immunity by CD8(+) T cells. EMBIO Mol Biol Med 2013; 5: 1311–21.
7. Meier T, Poizier P, Diederichs K, Welte W, Dimroth P. Structure of the rotor ring of F-Type Na+-ATPase from Enterococcus hirae. Science 2005; 308: 659–662.
8. Murata T, Yamato I, Kakimura Y, Leslie AG, Walker JE. Structure of the rotor of the V-Type Na+-ATPase from Enterococcus hirae. Science 2005; 308: 654–659.
9. Hamaire-Merah Z, Combettes L, Coquil JP, Swillens S, Mauger JP, Claret M, Champel P. Characterization of the co agonist effects of strontium and calcium on myo inositol trisphosphate-dependent ion fluxes in cerebellar microsomes. Cell Calcium 1995; 18: 390–399.
10. Feske S, Skolnik EY, Prakriya M. Ion channels and transporters in lymphocyte function and immunity. Nat Rev Immunol 2012; 12: 532–547.

11. Takewta N, Iwamoto A, Kamada M, Yamashita K, Takewta Y. Role of Ca\(^{2+}\) influx in bombesin-induced mitogenesis in Swiss 3T3 fibroblasts. J Biol Chem 1991; 266: 1403–1409.
12. Nordstrom T, Nevanlinna HA, Andersson LC. Mitasia-arresting effect of the calcium channel inhibitor SK&F 069665 on human leukemia cells. Exp Cell Res 1999; 202: 487–494.
13. Takewta N, Zhou W, Kamada M, Takewta Y. Ca\(^{2+}\)/calmodulin is involved in growth factor-induced retinoblastoma gene product phosphorylation in human vascular endothelial cells. FEBS Lett 1992; 306: 173–175.
14. Markenkaei AB, Surprenant A, North RA. Functional and molecular diversity of purinergic ion channel receptors. Ann NY Acad Sci 1999; 885: 716–729.
15. Putney JW Jr. Broad LM, Braun FF, Lievermeent JP, Bird GS. Mechanisms of capacitative calcium entry. J Cell Sci 2001; 114: 9.
16. Putney JW Jr. A model for receptor-regulated calcium entry. Cell Calcium 1986; 7: 1–12.
17. Shuttlesworth TJ. Arachidonic acid activates the noncapacitative entry of Ca\(^{2+}\) during [Ca\(^{2+}\)]
oscillations. J Biol Chem 1996; 271: 21720–21725.
18. Letrang F, Kiss R. The sodium pump alpha subunit as a potential target to combat apoptosis-resistant glioblastomas. Neoplasia 2008; 10: 198–206.
19. Maitovic R, Roland I, Van Quaquebeke E, Nilsson B, Mathieu A, Van Vondt F et al. The alpha subunit of the sodium pump could represent a novel target to combat non-small cell lung cancers. J Pathol 2007; 212: 170–179.
20. Gao YD, Hanley PJ, Rinne S, Zuzarte M, Daut J. Calcium-activated K+ channel (KCa 3.1) activity during Ca\(^{2+}\)/ store depletion and store-operated Ca\(^{2+}\)/ entry in human macrophages. Cell Calcium 2010; 48: 19–27.
21. Zweeluck A, Lewis RS. Mitogen-regulated Ca\(^{2+}\) current of T lymphocytes is activated by depletion of intracellular Ca\(^{2+}\) stores. Proc Natl Acad Sci USA 1993; 90: 6295–6299.
22. Lis A, Peinelt C, Beck A, Parvez S, Monteilh-Zoller M, Fleig A et al. CRACM1 CRACM2, and CRACM3 are store-operated Ca\(^{2+}\)/ channels with distinct functional properties. Curr Biol 2007; 17: 794–800.
23. Dinh-Van N, Smyth JY, Boyle RS, Putney JW Jr. Calcium inhibition and calcium potentiation of Orai 1, Orai 2, and Orai 3 calcium release-activated calcium channels. J Biol Chem 2007; 282: 17548–17556.
24. Lou J, Kim ML, Heo WD, Jones JT, Myers JW, Ferrell JR Jr et al. STIM is a Ca\(^{2+}\)-sensor essential for Ca\(^{2+}\)/store-depletion-triggered Ca\(^{2+}\)/ influx. Curr Biol 2005; 15: 1235–1241.
25. Ross J, DiGregorio PJ, Yeromin AV, Ohlsen K, Lidouy M, Zhang S et al. STIM1 an essential and conserved component of store-operated Ca\(^{2+}\)/ channel function. J Cell Biol 2005; 169: 435–445.
26. Liuk RM, Wang B, Prakriya M, Wu MM, Lewis RS. Oligomerization of STIM1 couples ER calcium depletion to CRAC channel activation. Nature 2008; 454: 538–542.
27. Meuth SG, Bittner S, Meuth P, Simon OJ, Budde T, Wiendl H. TWIK-related acid-sensitive K+ channel 1 (TASK1) and TASK3 critically influence T lymphocyte effector functions. J Biol Chem 2008; 283: 14559–14570.
28. Wulf H, Castle NA, Pardo LA. Voltage-gated potassium channels as therapeutic targets. Nat Rev Drug Discov 2009; 8: 982–1001.
29. Harmar AJ, Hills RA, Rossner EM, Jones M, Buneman OP, Dunbar DR et al. IUPHAR database of G protein-coupled receptors and ion channels. Nucleic Acids Res 2009; 37: D680–D685.
30. Velden S. The voltage-gated potassium channels and their relatives. Nature 2002; 419: 35–42.
31. Judge SI, Lee JM, Biver J, CT, Hoffman PM. Voltage-gated potassium channels in multiple sclerosis: overview and new implications for treatment of central nervous system inflammation and degeneration. J Rehabil Res Dev 2006; 43: 111.
32. Leonard RJ, Garcia ML, Slaughter RS, Reuben JP. Selective blockers of voltage-gated K+ channels depolarize human T lymphocytes: mechanism of the antiproliferative effect of charybdotoxin. Proc Natl Acad Sci USA 1992; 89: 10094–10098.
33. Ghanisian S, Wulf H, Miller MJ, Rohm H, Neien A, Gutman GA et al. Up-regulation of the KCa1 potassium channel during T-cell activation. Molecular mechanism and functional consequences. J Biol Chem 2005; 279: 37137–37149.
34. Di L, Srivastava S, Zhanova D, Ding Y, Li Z, Wulf H et al. Inhibition of the K+ channel KCa3.1 ameliorates T-cell-mediated colitis. Proc Natl Acad Sci USA 2010; 107: 1541–1546.
35. Hu L, Wang T, Gooke AR, Nath A, Zhang H, Margolick JB et al. Blockade of K+V3.1 potassium channels inhibits differentiation and granulocyte B secretion of human CD8\(^{+}\)/ T effector memory lymphocytes. PLoS One 2013; 8: e54267.
36. Panipy G, Vamoni G, Bascio Z, Bagdany M, Bodnar A, Varga Z et al. Kv1.3 potassium channels are localized in the immunologcal synapse formed between cytotoxic and target cells. Proc Natl Acad Sci U S A 2004; 101: 1285–1290.
37. Kochs S, Wu D, Hu X, Tahya RB, Hup R, Khan FS et al. Blocking KCa3.1 channels increases tumor cell killing by a subpopulation of human natural killer lymphocytes. PLoS One 2013; 8: e67640.
38. Perler F, Radke CM, Vanderberg CA. Primary structure and characterization of a small-conductance inwardly rectifying potassium channel from human hippocampus. Proc Natl Acad Sci USA 1994; 91: 6240–6244.
39. Judge SI, Lee JM, Biver J, CT, Hoffman PM. Voltage-gated potassium channels in multiple sclerosis: Overview and new implications for treatment of central nervous system inflammation and degeneration. J Rehabil Res Dev 2006; 43: 111.
40. Kurschner C, Yuzaki M. Neuronal interlinkein-16 (NIL-16): a dual function PDZ domain protein. J Neurosci 1999; 19: 7770–7780.
41. Tsai KL, Chang HF, Wu SN. The inhibition of inwardly rectifying K+ channels by mentamidine in macrophages and microglial cells. Cell Physiol Biochem 2013; 31: 938–951.
42. Nistvasta R, Asiam M, Kalluri SR, Schimmer L, Buck D, Tackenberg B et al. Potassium channel Kir3.1 as an immune target in multiple sclerosis. N Engl J Med 2012; 367: 115–23.
43. Goldstein SA, Bockenhauer D, O'Dell L, Zilberberg N. Potassium leak channels and the KCNA family of two-P-domain subunits. Nat Rev Neurosci 2002; 3: 175–183.
44. Verkman AS, Xie Y, Rhee JS. TRP channel. Annu Rev Biochem 2007; 76: 387–417.
45. Wenning AS, Nebeling K, Strauss B, Wolfs MJ, Sappok A, Hoth M et al. TRP expression pattern and the functional importance of TRP channels in primary human T cells. Biochim Biophys Acta 2011; 1813: 412–423.
46. Vennekens R, Nilius B. Insights into TRPM4 function, regulation and physiological role. Immun Exp Allergy 2007; 39: 285–298.
47. Launay P, Cheng H, Sivastava S, Penner R, Fleig A, Kinet JP. TRPM4 regulates calcium oscillations after T cell activation. Science 2004; 306: 1374–1377.
48. Yip L, Woehrle T, Corriden R, Hirsh M, Chen Y, Inoue Y et al. Autoimmune regulation of T-cell activation by ATP release and P2X7 receptors. FASEB J 2009; 23: 1635–1639.
49. Dieterich T, Yip L, Elhai A, Sumi Y, Chen Y, Yao Y et al. Pannexin-1 channel-mediated ATP release together with P2X1 and P2X4 receptors regulate T-cell activation at the immune synapse. Blood 2010; 115: 3475–3484.
50. Paddeh S, Cohen A, Rolfman CM. ATP-induced activation of human B lymphocytes via P2-purinoceptors. J Immunol 1991; 146: 1628–1632.
51. Barcouda OR, Drir M, Melchiorri L, Chiozzi P, Hanau S, Chiari E et al. An ATP-evoked channel is involved in mitogenic stimulation of human T lymphocytes. Blood 1996; 87: 682–690.
52. Adinolfi E, Callegari MG, Ferrari D, Bologna C, Minelli M, Wielocki MR et al. Basal activation of the P2X7 ATP receptor elevates mitochondrial calcium and potential, increases cellular ATP levels, and promotes serum-independent growth. Mol Biol Cell 2005; 16: 3260–3272.
53. Oh-Hora M, Yamashita M, Hogan PG, Sharma S, Lamper E, Chung W et al. Dual functions for the endoplasmic reticulum calcium sensors STIM1 and STIM2 in T cell activation and tolerance. Nat Immunol 2009; 10: 432–440.
54. Matusiak M, Fuji Y, Baba A, Hikido M, Kurosaki T, Baba Y. The calcium sensors STIM1 and STIM2 control B cell regulatory function through interleukin-10 production. Immunity 2011; 34: 703–714.
55. Feske S. STIM1 deficiency in human and mice: roles of store-operated Ca2+ entry in the immune synapse and beyond. Immunol Rev 2009; 231: 169–209.
56. Franciszkiewicz K, Le Floc’h A, Boutet M, Vergnon I, Schmitt A, Mami-Chouaib F. CD103 or CD101 and STIM2 control B cell regulatory function through interleukin-10 production. Immunity 2011; 70: 1225–1235.
57. Wang H, Zhang Y, Cao L, Han H, Wang J, Yang B et al. HERG K+ channel, a regulator of tumor cell apoptosis and proliferation. Cancer Res 2002; 62: 4843–4848.
58. Barriere H, Belford R, Rubera I, Tauc M, Lesage F, Poujol C et al. Role of TASK2 potassium channels regarding volume regulation in primary cultures of mouse proximal tubules. J Gen Physiol 2003; 120: 177–190.
59. Williams S, Bateman A, O’Kelly I. Altered expression of two-pore domain potassium (K2P) channels in cancer. PLoS One 2013; 8: e74569.
60. Mu D, Chen L, Zhang X, See LH, Koch CM, Yen C et al. Genomic amplification and oncogenic properties of the KCNQ9 potassium channel gene. Cancer Cell 2003; 3: 297–302.
61. Yang M, Blackenbury WJ. Membrane potential and cancer progression. Front Physiol 2013; 4: 185.
62. Morokuma J, Blackiston D, Adams DS, Seebohm G, Trimmer B, Levin M. Modulation of potassium channel function confers a hyperproliferative invasive phenotype on embryonic stem cells. Proc Natl Acad Sci USA 2008; 105: 16608–16613.
63. Hanahan D, Weinberg RA. Hallmarks of cancer: the next generation. Cell 2011; 144: 648–674.
64. Schwab A, Fabian A, Henley PJ, Stock C. Role of ion channels and transporters in cancer migration. Physiol Rev 2012; 92: 1865–1913.
65. Khanal D, Sarkalut UT, Wisker B, Meister EA, Romero IA, Couraud PO et al. Role of KCNMA1 gene in breast cancer invasion and metastasis to brain. BMC Cancer 2009; 9: 258.
66. Chantome A, Potier-Cerneau M, Clayeys L, Fromont G, Marionneau-Lambot S, Gueguinou M et al. Pivotal role of the lipid Raft SK3-Orai1 complex in human cancer cell migration and bone metastases. Cancer Res 2013; 73: 4852–4861.
67. Schwab A, Schuhrot B, Seeberg P, Reinhardt J, Dartich PC. Migration of transformed rat epidermal cells is regulated by K+ channel modulation of actin cytoskeleton and cell volume. Pfugers Arch 1999; 438: 330–337.
68. Kraft R, Krause P, Jung S, Basar D, Liebl M, Botz J et al. BK channel opens inhibit migration of human glioma cells. Pfugers Arch 2003; 448: 248–255.
69. Xie LH, John SA, Ribalet B, Weiss JA. Activation of inwardly rectifying potassium (Kir) channels by phosphodiesterase-4A-bisphosphate (PPI2) interacts with the hippocampal ligand. Preg Biol Mol Biol 2007; 9: 320–335.
70. Stringer BK, Cooper AG, Shepard SB. Overexpression of the G-protein inwardly rectifying potassium channel 1 (GIRK1) in primary breast carcinomas correlates with axillary lymph node metastasis. Cancer Res 2001; 61: 582–586.
71. Takamai I, Inoue Y, Gika M. G-protein inwardly rectifying potassium channel 1 (GIRK1) gene expression correlates with tumor progression in non-small cell lung cancer. BMC Cancer 2004; 4: 79.
72. Williams S, Bateman A, O’Kelly I. Altered expression of two-pore domain potassium (K2P) channels in cancer. PLoS One 2013; 8: e74569.
73. Lehen’kyi V, Flourakis M, Skryma R, Prevarskaya N. TRPV6 channel controls prostate cancer cell migration and bone metastases. PLoS ONE 2011; 6: 387–392.
74. Chon K, Lee SJ, Yi TK, Park JH, Kim JS, Park JH et al. Essential role of TRPC6 channels in G2/M transition and migration of human glioma cells. Pflugers Arch 2007; 456: 1022–1030.
75. Chon K, Lee SJ, Yi TK, Park JH, Kim JS, Park JH et al. Essential role of TRPC6 channels in G2/M transition and migration of human glioma cells. Pflugers Arch 2007; 456: 1022–1030.
76. Gao H, Chen X, Du X, Guan B, Liu Y, Zhang H. EGF enhances the migration of cancer cells by up-regulation of TRPM7. Cell Calcium 2011; 50: 559–568.
77. Mihalikov AG, Lebedev EV, Itoh T, Kouchakji M, Eisenberg E, von Harnasch P et al. TRPM7 is required for breast tumor cell metastasis. Cancer Res 2012; 72: 4320–4321.
78. Bates DO, Hilman NJ, Williams B, Neal GR, Pickow TM, Regulation of microvascular permeability by vascular endothelial growth factors. J Anat 2007; 210: 417–428.
79. Chon K, Lee SJ, Yi TK, Park JH, Kim JS, Park JH et al. Essential role of TRPC6 channels in G2/M transition and migration of human glioma cells. Pflugers Arch 2007; 456: 1022–1030.
80. Lehen’kyi V, Flourakis M, Skryma R, Prevarskaya N. TRPV6 channel controls prostate cancer cell migration and bone metastases. PLoS ONE 2011; 6: 387–392.
104. Roger S, Pelegrin P. P2X7 receptor antagonism in the treatment of cancers. Expert Opin Investig Drugs 2011; 20: 875–880.

105. Haskell CM, Mendoza E, Pisters KM, Fossella FV, Figlin RA. Phase II study of intravenous adenosine 5'-triphosphate in patients with previously untreated stage IIIB and stage IV non-small cell lung cancer. Am J Respir Crit Care Med 2010; 181: 2331–2337.

106. Haskell CM, Wong M, Williams A, Lee LY. Phase I trial of extracellular adenosine 5'-triphosphate in patients with advanced cancer. Anticancer Drugs 2003; 14: 639–644.

107. Haskell CM, Mendoza E, Pisters KM, Fossella FV, Figlin RA. Phase I trial of intravenous adenosine 5'-triphosphate in patients with previously untreated stage IIIIB and stage IV non-small cell lung cancer. Invest New Drugs 1998; 16: 81–85.

108. Beijer S, Hupperets PS, van den Borne BE, Eussen SR, van Henten AM, van den Beukel-Edenrogen M et al. Effect of adenosine 5'-triphosphate infusion on the nutritional status and survival of pretreatment cancer patients. Anti-cancer Drugs 2009; 20: 625–633.

109. Flourakis M, Lehen'kyi V, Baroja-Mazo A, Cayuela ML, Pelegrin P et al. P2X7 receptor activation may play a role in tumorigenesis. Cancer Res 2007; 67: 11913–11920.

110. Beijer S, Hupperets PS, van den Borne BE, Eussen SR, van Henten AM, van den Beukel-Edenrogen M et al. Effect of adenosine 5'-triphosphate infusion on the nutritional status and survival of pretreatment cancer patients. Anti-cancer Drugs 2009; 20: 625–633.

111. Beijer S, Hupperets PS, van den Borne BE, Eussen SR, van Henten AM, van den Beukel-Edenrogen M et al. Effect of adenosine 5'-triphosphate infusion on the nutritional status and survival of pretreatment cancer patients. Anti-cancer Drugs 2009; 20: 625–633.

112. Beijer S, Hupperets PS, van den Borne BE, Eussen SR, van Henten AM, van den Beukel-Edenrogen M et al. Effect of adenosine 5'-triphosphate infusion on the nutritional status and survival of pretreatment cancer patients. Anti-cancer Drugs 2009; 20: 625–633.

113. Beijer S, Hupperets PS, van den Borne BE, Eussen SR, van Henten AM, van den Beukel-Edenrogen M et al. Effect of adenosine 5'-triphosphate infusion on the nutritional status and survival of pretreatment cancer patients. Anti-cancer Drugs 2009; 20: 625–633.

114. Beijer S, Hupperets PS, van den Borne BE, Eussen SR, van Henten AM, van den Beukel-Edenrogen M et al. Effect of adenosine 5'-triphosphate infusion on the nutritional status and survival of pretreatment cancer patients. Anti-cancer Drugs 2009; 20: 625–633.

115. Beijer S, Hupperets PS, van den Borne BE, Eussen SR, van Henten AM, van den Beukel-Edenrogen M et al. Effect of adenosine 5'-triphosphate infusion on the nutritional status and survival of pretreatment cancer patients. Anti-cancer Drugs 2009; 20: 625–633.

116. Beijer S, Hupperets PS, van den Borne BE, Eussen SR, van Henten AM, van den Beukel-Edenrogen M et al. Effect of adenosine 5'-triphosphate infusion on the nutritional status and survival of pretreatment cancer patients. Anti-cancer Drugs 2009; 20: 625–633.

117. Ganapathi SB, Kester M, Elmslie KS. State-dependent block of HERG potassium channels by ATP and G1035. J Gen Physiol 2008; 132: 327–338.

118. Hammadi M, Chopin V, Mattiat F, Derrin-Duthille I, Chassaund A, Sevestre H et al. Human ether-a-go-goRelated channel 1 (ERG1) regulates MDA-MB-231 breast cancer cell migration. J Cell Physiol 2012; 227: 3837–3846.

119. Jelassi B, Chantome A, Alcaraz-Perez F, Baroja-Mazo A, Cayuela ML, Pelegrin P. P2X7 receptor activation induces calcium entry and cell migration in glioblastoma derived human glioma cells. Cell Calcium 2013; 53: 165–173.

120. Lee MH, Choi BY, Kundu JK, Shin YK, Na HK, Surh YJ. Resveratrol suppresses growth of glioma BK, a novel BK channel isoform highly expressed in human glioma cells. J Neurosci 2002; 22: 1840–1849.

121. Ling C, Ao J, Li Y, Ma X, Hu M, Shi Y et al. 4-Ethoxy-2-[6-(2-furfurylidene)-4H-1,2,4-triazin-3-yl]-5,6-dihydro-2H-pyran-3-carboxylic acid inhibits proliferation of cervical cancer cells. J Med Food 2013; 16: 2207–2213.

122. Li J, Xiong B, Wang J, Zhang J, Wu X, Hou Y et al. Roscovitine differentially affects CaV2 and Kv channels: a potential therapeutic strategy for glioblastoma. Biochem Biophys Res Commun 2013; 433: 213–218.

123. Comes N, Bielanska J, Vallejo-Gracia A, Serrano-Albarras A, Marruecos L, Gomez D et al. Blockade of intracellular calcium influx mediated signaling pathway in A549 lung cancer cells. Biochim Biophys Acta 2011; 1810: 8–84.

124. Liu H, Hughes JD, Rolins S, Chen B, Perkins E. Calcium entry via ORAI1 regulates glioblastoma cell proliferation and apoptosis. Exp Mol Pathol 2011; 91: 753–760.

125. Pardo LA, Stuhmer W. The roles of K(+) channels in cancer. Cell Tissue Res 2005; 321: 411–418.

126. Cheng PN, Leung YC, Lo WH, Tsui SM, Lam KC. Remission of hepatocellular carcinoma with arginine deplletion induced by systemic release of endogenous hepatic arginase due to transhepatic arterial embolisation, augmented by high-dose isulin: arginase as a potential drug candidate for hepatocellular carcinoma. Cancer Lett 2005; 224: 67–70.

127. Costinio-Silva R, Stal L, Cheung KK, de Campos NE, de Oliveira Souza C, Otupa DM et al. P2X7 and P2Y2 purinergic receptors on human intestinal epithelial carcinoma cells: effects of extracellular nucleotides on apoptosis and cell proliferation. J Am Physiol Gastrointest Liver Physiol 2005; 288: G1024–G1035.

128. Roger S, Pelegrin P. P2X7 receptor antagonism in the treatment of cancers. Expert Opin Investig Drugs 2011; 20: 875–880.

129. Ageresch HG, Burgera SP, van der Gaast A, Wilson JH, Dagleicie PC. Randomized clinical trial of adenosine S-triphosphate on tumor growth and survival in advanced lung cancer patients. Anticancer Drugs 2003; 14: 639–644.

130. Hupperets PS, van den Borne BE, Eussen SR, van Henten AM, van den Beukel-Edenrogen M et al. Effect of adenosine 5'-triphosphate infusion on the nutritional status and survival of pretreatment cancer patients. Anti-cancer Drugs 2009; 20: 625–633.

131. Roger S, Pelegrin P. P2X7 receptor antagonism in the treatment of cancers. Expert Opin Investig Drugs 2011; 20: 875–880.

132. Hou MF, Kuo HC, Li JH, Wang YS, Chang CC, Chen KC et al. ORAI1/CRACM1 overexpression suppresses cell proliferation via attenuation of the store-operated calcium influx-mediated signalling pathway in A549 lung cancer cells. Biochim Biophys Acta 2011; 1810: 8–84.

133. Beijer S, Hupperets PS, van den Borne BE, Eussen SR, van Henten AM, van den Beukel-Edenrogen M et al. Effect of adenosine 5'-triphosphate infusion on the nutritional status and survival of pretreatment cancer patients. Anti-cancer Drugs 2009; 20: 625–633.
1. Pilozzi S, Brizzi MF, Bernabei PA, Bartolozzi B, Caporale R, Basile V et al. VEGFR-1 (FL3-1), beta1 integrin, and hERG K+ channel for a macromolecular signaling complex in acute myeloid leukemia: role in cell migration and clinical outcome. Blood 2007; 110: 8–0.

2. Shao XD, Wu KC, Hao ZM, Hong L, Zhang J, Fan DM. The potent inhibitory effects of cisapride, a specific blocker for human ether-a-go-go-related gene (HERG) channel, on gastric cancer cells. Cancer Biol Ther 2005; 4: 295–301.

3. Millward MJ, Cantwell BM, Munro NC, Robinson A, Corris PA, Harris AL. Oral verapamil with chemotherapy for advanced non-small cell lung cancer: a randomised study. Br J Cancer 1993; 67: 1031–1035.

4. Yokem KH, Clothier JL, Montague SL, Geary RJ, Winters AL 3rd, Hendrix MJ et al. Inhibition of tumor cell invasion by verapamil. Pigment Cell Res 1991; 4: 225–233.

5. Van Quaquebeke E, Simon G, Andre A, Dewelle J, El Yazzidi M, Bruyneel F et al. Identification of a novel cardenolide (2′-oxovoruscharin) from Calotropis procera and the hemisynthesis of novel derivatives displaying potent in vitro antitumor activities and high in vivo tolerance: structure-activity relationship analyses. J Med Chem 2005; 48: 849–856.

6. Cannithers MD. Chatterjee G, Cannithers LM, Ofoha R, Ihagwara U, Rahner C et al. Regulation of podosome formation in macrophages by a splice variant of the sodium channel SCN8A. J Biol Chem 2009; 284: 8114–8118.

7. Fraser SP, Diss JK, Chioni AM, Mycielska ME, Pan H, Yamaci RF et al. Voltage-gated sodium channel expression and potentiation of human breast cancer metastasis. Clin Cancer Res 2005; 11: 5381–5389.

8. Schwab A, Reinhartd J, Schneider SW, Gaisser B, Schuricht B. K(+) channel-dependent migration of fibroblasts and human melanoma cells. Cell Physiol Biochem 1999; 9: 129–132.

9. Innamaa A, Jackson L, Asher V, Van Shalkwyk G, Warren A, Hay D et al. Expression and prognostic significance of the oncogenic K2P potassium channel KCNK9 (TASK-3) in ovarian carcinoma. Anticancer Res 2013; 33: 1–8.

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