UNIVERSITÀ DEGLI STUDI DI TORINO

EDITORIAL

ION FLUXES AND CANCER

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The cell is an open thermodynamic system that constitutively exhanges energy and matter with the surrounding microenvironment through the plasma membrane. All cell functions, e.g. survival, proliferation, differentiation, migration, secretion, and excitability, rely on the maintenance of the physiological membrane potential and of driving forces that sustain membrane fluxes of Ca^{2+}, K^+, Na^+, Cl^- and H^+, i.e. the main inorganic components of biological fluids. Consequently, the activity of ion channels and transporters, required for transmembrane ion currents, is tighly coupled to cell metabolism. Such a reciprocal interaction between ion fluxes and cell biochemistry occurs through a variety of regulatory pathways and is better evidenced in diseases. On one hand, disfunctions of channels and transporters can impair cell functions; conversely, altered cell biochemical pathways modify ion fluxes through post-translational modifications (such as phosphorylation and nitrosylation) as well as through interactions with other proteins, lipids and intracellular messengers.

Human genome contains 235 genes encoding ion channels, and functional membrane pores enhance their diversity through heteromeric clusterization of different protein subunits [1].

For many decades, the interest on the role of ion channels in the progression of human diseases was mainly focused on alterations of excitability due to channellopaties (diseases linked to channel mutations) of voltage-operated channels (VOCs) [2]. VOCs have been extensively studied by the use of electrophysiological and biochemical approaches, that provided powerful pharmacological and molecular tools to selectively interfere with their activity. Nonetheless, now the roles of VOCs extend far beyond their established actions on excitable tissues, being involved in virtually all cell functions, even in nonexcitable tissues. On the other hand, our knowledge on the structure and roles of voltage-independent channels has been significantly improved, although selective pharmacological compounds are still lacking for many of them. A striking example is provided by the discovery and characterization of the superfamily of transient receptor potential (TRP) channels that led to a drastic revision of cellular physiology and physiopathology.

Only recently, the involvement of ion channels in the progression of neoplastic diseases was identified [3]. Ion fluxes regulate a broad range of cellular processes strictly involved in tumoral progression. Changes in intracellular calcium concentration due to calcium currents represent a chemical signal controlling cell proliferation, motility, death and metabolism through the involvement of intracellular calcium-sensors and

calcium-sensitive enzymes. But calcium is not only a chemical messenger: calcium entry directly promotes membrane depolarization and exerts indirect effects affecting the activity of a number of K^+ and Cl^- channels. On the other hand, fluxes of Na^+, K^+ and Cl^- are the main modulators of membrane potential in many cell types, concurring to their excitability and related functions (including secretion and motility). Furthermore, they finely tune the driving force for calcium fluxes. Finally, H^+ currents, in addition to their electrical relevance, regulate intracellular pH and consequently a great number of biochemical reactions in all tissues, including cancer cells. Moreover, similarly to Ca^{2+}, H^+ modulates the activity of several different ion channels. For these reasons, it is not surprising that cancer growth, vascularization and metastasis are often associated to altered structure, expression or activity of Ca^{2+} channels (voltage-dependent as well as voltage-independent, such TRPs and Orai), voltage-gated K^+ and Na^+ channels, ligand-gated cationic channels and carriers [3, 4]. Experimental reports and review articles focused on this topic are now published by journals not only in the field of Biophysics, but also of experimental oncology and cancer research.

Among calcium channels, TRPs attracted high interest in the last decades for their involvement in pathophysiological processes, and recently in cancer [5-7]. Human TRPs are a superfamily of proteins encoded by 26 genes, widely distributed in all tissues, and many of them are regulated by multiple mechanisms (chemical, thermal, mechanical stimula) [8]. In addition to their well described involvement in sensory transduction, some TRPs mediate calcium fluxes that control cell proliferation, migration and differentiation in many tissues. Moreover, their expression is altered in some cancer cells and in tumor-derived endothelial cells, through which they contribute to the regulation of angiogenic progression: recently, some TRP members have been included in the oncogenic and tumor suppressor protein family [7]. TRPM1 and TRPV1 are tumor suppressors, respectively for localized malignant melanoma and bladder carcinoma, while TRPM8 and TRPV6 are oncogenes in prostate cancer and their overexpression may be used as a diagnostic marker. TRPV1, TRPC1, TRPC6, TRPM4, and TRPM5 are also overexpressed in some cancers [7]. In addition, a number of TRPs play a role in tumor vascularization, required for tumor growth and metastasis. They include some TRPCs and TRPVs (TRPV1 and TRPV4), even if only for the latter a specific role in tumor angiogenesis has been shown [9-11].

The features of TRP channels make them potentially very powerful as molecular targets for therapy as well as markers for diagnostics: however, the specificity of pharmacological tools is often weak, and side-effects due to their broad distribution in normal tissues limits their clinical application.

While Ca^{2+} channels mediate chemical and electrical signals, K^+ currents are universal and key regulators of cell membrane potential as well as, indirectly, of the driving force for Ca^{2+} fluxes. K^+ channels form the largest ion channel family in humans, including 80 members encoded by more than 50 genes [12]. In addition to their role in nerve and cardiac action potentials, these proteins are involved in a number of physiological processes, including cell volume regulation, apoptosis, immunomodulation, differentiation and proliferation. All these events are strictly related to tumor progression. Similarly to Ca^{2+} channels, K^+ channel expression is altered in different cancer cells and affects a number of tumoral features, from cell proliferation, to resistance to apoptotic cell death, to the regulation of tumor angiogenesis and invasiveness [13]. Because Kv1.3 and Kv1.5 channels modulate the proliferation of different mammalian cells, these proteins have been analyzed in a number of tumors and cancer cells. In most cancers, their expression is remodeled, and in some cases correlated with the grade of tumor malignancy. Human ether à go-go (hEAG) potassium channels are primarily expressed in brain but also frequently overexpressed in solid tumors [14]. Human Ether-à-go-go-Related (hERG) K^+ channels are also expressed in a variety of cancer cells where they control cell proliferation and apoptosis [15, 16].

In addition to Ca^{2+}- and K^+-permeable channels, also voltage-dependent Na^+ channels (VGSCs) are involved in tumor progression. Functional VGSCs are upregulated in some human prostate, breast, lung, cervix and colon carcinomas [17]. Blocking VGSC activity with tetrodotoxin (TTX) in rat and human prostate cancer cells impaired cell functions related to the metastatic process. Surprisingly for nonexcitable cells, some members of VGSCs (Nav1.5 and Nav1.7) have been recently detected in human endothelial cells (HUVECs) where they regulate multiple angiogenic functions and VEGF signaling [18]. Targeting VGSC expression or activity could be a novel strategy for controlling altered angiogenesis.

Ion channels are not the only mediators of transmembrane ion fluxes that are involved in tumor progression. Neoplastic cell transformation is associated to the dysregulation of intracellular pH, an early event in carcinogenesis and a hallmark of tumors [19, 20]. Cell growth is dependent on pH and all cancer

cells exhibit an aberrant regulation of H^+ dynamics leading to a reversal of the intracellular to extracellular pH gradient as compared to normal tissues. The best studied regulators of both pH_i and pH_e in tumors, Na^+/H^+ exchanger isoform 1 (NHE1) and proton pumps, have been directly associated with cellular transformation, invasion and metastasis [21, 22]. NHE1 is activated during oncogene-dependent transformation resulting in cytosolic alkalinization. Pharmacological approaches targeting the mechanisms responsible for the reversed pH gradient of cancer cells may be a powerful strategy for treatments against cancer.

A number of patents cover the association of ion channels and transporters with cancer growth, metastasis and angiogenesis [23-50].

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