Gap junctions in the cardiovascular and immune systems

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Abstract

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Gap junctions are clusters of intercellular channels directly connecting the cytoplasm of adjacent cells. These channels are formed by proteins named connexins and are present in all metazoan organisms where they serve diverse functions ranging from control of cell growth and differentiation to electric conduction in excitable tissues. In this overview we describe the presence of connexins in the cardiovascular and lympho-hematopoietic systems giving the reader a summary of the topics to be covered throughout this edition and a historical perspective of the discovery of gap junctions in the immune system.

Key words
- Gap junctions
- Cardiovascular system
- Immune system
- Connexins

Gap junction channels: A simple overview

Gap junctions are clusters of intercellular channels providing conduits for diffusional exchange of ions and small molecules directly from one cell to another (for details, see Ref. 1). These channels span two plasma membranes and result from the association of two half channels, termed connexons, contributed separately by each cell of a pair (see Figure 1). Connexins are expressed in an overlapping pattern of tissue distribution and more than one connexin type is generally expressed by individual cells. In addition, the hemichannels or connexons contributed by each cell may have selective affinities for one another either within or between connexins, forming homotypic, heterotypic and heteromeric junctions (Figure 1; see also Brink et al. (p. 415), Verselis et al. (p. 379)

and Beyer et al. (p. 391) in this edition). A homotypic gap junction channel is composed of 12 identical connexin subunits; in heterotypic but homomeric channels all the connexins in the connexon of one cell are identical but are different from the connexins of the adjacent cell. In heteromeric gap junction channels, the connexins comprising the connexon are not identical. The expression of numerous connexin types in the cardiovascular and immune system provides an enormous diversity of possible channel configurations; whether some are favored for some functions (as might be the case for homomeric heterotypic junctions at compartmental boundaries) remains to be established.

Characterization of the properties of gap junctions formed by individual connexin types (and how these properties are changed in heterotypic pairings) has been a major effort of several laboratories. Although the

biophysical and biochemical characterization of these channels lags behind the understanding of other voltage- or ligand-gated ion channel complexes, recent application of molecular genetic approaches has had a major impact. In this edition, for example, Nicholson et al. (p. 369) discuss the molecular basis of selective permeability between gap junction channels composed of different connexins and Yahuaca et al. (p. 399) present their results on phosphorylation of Cx43.

Gap junctions in the heart and vasculature

Gap junctions in the heart mediate the intercellular transmission of electrical impulses that coordinate the generation of the cardiac rhythm, conduct the pacemaking signal to muscular regions and synchronize contractions of the atrium and ventricles. The three most abundant cardiac gap junction proteins are Cx43, Cx40, and Cx45, which are expressed within each region of the heart to different extents and in various combinations. For example, Cx43 is most abundant in the ventricle, Cx40 in the atrium, and Cx40 and Cx45 in the conduction system (2).

Because the major role of gap junctions in the normal heart is the generation and maintenance of rhythmic contractions, considerable recent work has been directed at understanding how altered gap junction distribution and function may lead to disturbed cardiac rhythmicity, including reentry. For example, in aging and post-infarct myocardium, gap junction disappearance and remodeling accentuate conduction discontinuities, as summarized in the manuscript in this issue by Jeff Saffitz (p. 407). Moreover, ventricular conduction in Cx43 heterozygous mice (lacking one copy of the Cx43 gene) is significantly slowed (3); atrial conduction time is prolonged (and supraventricular arrhythmias occasionally appear) in Cx40 knockout mice (4,5).

A disease of major importance to Brazil and other countries in Latin America is chagasic cardiomyopathy, which is characterized by both acute and chronic phases; it affects the cardiac system (Figure 2), the smooth muscles and the nervous system. Of particular interest, acute infection of cardiac myocytes and CNS glia results in loss of intercellular communication (Figure 2) without affecting expression levels or post-translational processing of gap junction proteins (6,7). It appears as if the changes in gap junction distribution result from altered trafficking of the junctional proteins to or from the plasma membrane, but the mechanisms remain to be resolved. Masuda et al. (p. 439) in their paper in this collection present a novel set of findings in this area, showing that sera from chagasic patients block inter-

Figure 1 - Possible configurations of gap junction channels which are formed by the end to end (here, top to bottom) alignment of two connexons; two differently shaded components of connexons represent the two connexin subunits.

Homotypic
Homometric

Heterotypic
Homomeric

Homotypic
Heteromeric

Heterotypic
Heteromeric
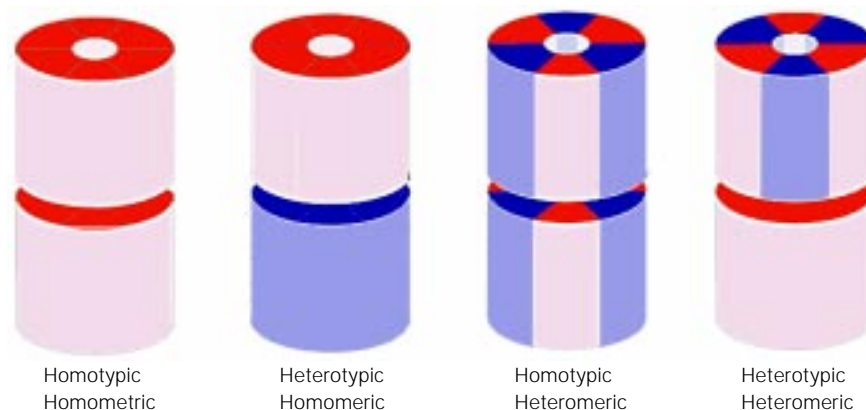
cellular communication and atrioventricular conduction in cardiac myocytes. Thus, perhaps the protozoan parasite appears to exert both local and long-range disorganization of Cx43, generating arrhythmias and thereby disturbing cardiac function.

Gap junctions in the vessel wall play critical roles in maintenance and modulation of vascular tone. In this issue, George Christ and colleagues (p. 423) summarize the evidence for this role of gap junctions between smooth muscle cells, indicating that these channels provide the critical pathway for second messenger diffusion in this process. Endothelial cells are also interconnected by gap junctions, and this pathway presumably serves to coordinate vascular tone through coordinated nitric oxide production and distribution. In the vessels of hypertensive rats, Haefliger and Meda (p. 431) show that chronic hypertension alters the expression of Cx43 in muscle cells; they suggest that the cell-cell communication mediated by Cx43 may contribute to regulate the elasticity of the vascular wall.

Immune response

The roles of gap junction channels in the immune system is increasingly appreciated. As early as 30 years ago electron microscopy and functional studies revealed close contacts between lymphoreticular cells in rheumatoid synovial membrane and suggested the expression of gap junction channels among activated macrophages (8-10).

Although the argument can be traced earlier, the modern portion of the story begins with Gilberto M. Oliveira Castro and Marcelo A. Barcinski, at that time Associate Professors at the Institute of Biophysics "Carlos Chagas Filho" at the Federal University of Rio de Janeiro. Of special interest, Oliveira Castro collaborated with and was highly influenced by the work of Werner R. Loewenstein (11) in their initial studies of the effects of divalent cations on junctional membrane

permeability. These studies were further developed in Brazil and, together with the work by Marcelo A. Barcinski, they showed the existence of intercellular communication in stimulated human lymphocytes (12) and suggested that calcium could mediate uncoupling of these cells (13). Such collaborations were very fruitful in the field of immunology and their major results are well known to everyone in this field. Furthermore, this work impacted on different generations of electrophysiologists at the Institute of Biophysics, among them Antonio C. Campos de Carvalho, Pedro Persechini and George A. Dos Reis, who have characterized, among other issues, the ATP-induced currents in macrophages and electrophysiological properties of phagocytic membranes in activated macrophages (14-17).
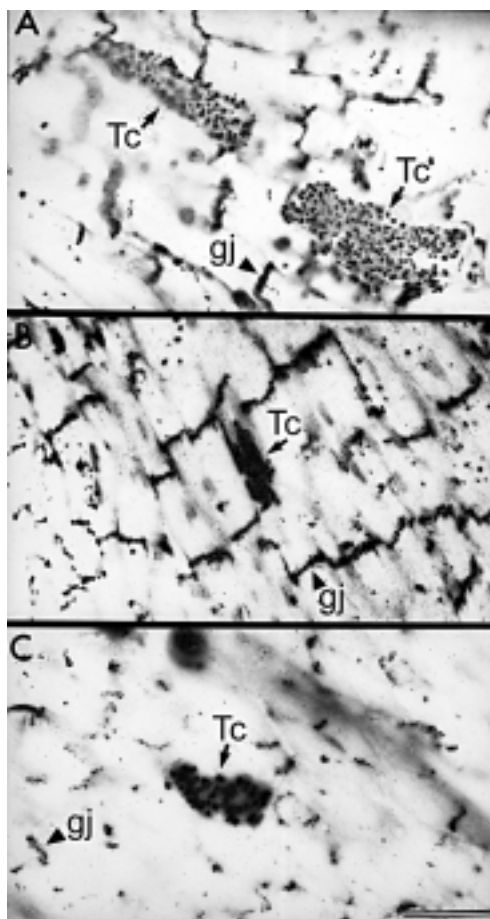
The occurrence of gap junctions in mi-

Figure 2 - Disruption of intercalated disks in hearts of mice infected with Trypansoma cruzi, the protozoan parasite that causes Chagas' disease. A, B and C illustrate sections of T. cruzi-infected mouse heart immunostained with antibody specific for Cx43. Note the presence of normal-appearing intercalated disks (gap junctions: gj, arrows) in uninfected myocytes and their disruption in cells infected with T. cruzi (Tc, arrows). Frozen sections and immunostaining performed by C. Roy; infected mouse hearts obtained from Dr. H.B. Tanowitz. Bar = 100 µm.

gratory cells (i.e., macrophages) of the immune system has been a controversial issue. Nevertheless, gap junctions composed of Cx43 have been identified in blood-forming tissue and in hemopoietic cultured cells (e.g., lymphocytes). At the Rio de Janeiro meeting, Rosendall presented data on the functional coupling between follicular dendritic cells and lymphocytes and suggested that metabolic cooperation between these cells is important to time germinal center events. Furthermore, Rosendall suggested that second messenger permeation through gap junction channels may be involved in the rescue

of B cells from apoptotic cell death. In this edition, Sáez et al. (p. 447) and Alves et al. (p. 457) discuss the involvement of gap junction channels in the response of the immune system and the effects of different neuropeptides and hormones on the modulation of intercellular communication in thymic cells.

Acknowledgments

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