

## Product Information

### **ANTI-CONNEXIN 32 (106-124)** **Developed in Rabbit, Affinity Isolated Antibody**

Product Number **C3595**

#### **Product Description**

Anti-Connexin 32 is developed in rabbit using a synthetic peptide Leu-Arg-Leu-Glu-Gly-His-Gly-Asp-Pro-Leu-His-Leu-Glu-Glu-Val-Lys-Arg-His-Lys conjugated to KLH with glutaraldehyde as immunogen. The peptide corresponds to a segment of the cytoplasmic loop domain of human, mouse and rat connexin 32, amino acid residues 106-124. Affinity isolated antigen specific antibody is obtained by immunospecific purification which removes essentially all rabbit serum proteins, including immunoglobulins, which do not specifically bind to connexin 32.

Anti-Connexin 32 reacts specifically with connexin 32. By immunoblotting the antibody detects a single band (~ 27kD). Staining of the connexin 32 band by immunoblotting is specifically inhibited with the connexin 32 peptide (amino acid residues 106-124). The antibody also detects connexin 32 by immunohistochemistry (frozen sections). Reactivity has been observed with rat and human connexin 32.

Gap junctions<sup>1</sup> are specialized cell membrane domains consisting of aggregations of intercellular channels that directly connect the cytoplasm of adjacent cells. Gap junctions coordinate cellular and organ function in tissues and are involved in metabolic cooperation between cells, electrical coupling, synchronization of cellular physiological activities, growth control, and developmental regulation. The gap junction channels allow intercellular exchange of ions, nucleotides and small molecules between adjacent cells. Unlike other membrane channels, intercellular channels span two apposed plasma membranes and require the contribution of hemi-channels, called connexons, from both participating cells. These channels are permeable to molecules as large as 1 kD, and they have been reported in most mammalian cell types.<sup>2</sup> Two connexons interact in the extracellular space to form the complete intercellular channel. Each connexon is composed of six similar or identical proteins, which have been termed connexins. Connexins (Cx) are a multi-gene family of highly related proteins with molecu-

lar weights ranging from 26 to 70 kD. At least a dozen distinct connexin genes have been identified in mammals, many expressed in a diverse tissue and cell specific pattern.<sup>2</sup> Two distinct lineages have been identified in mammals, one termed class I or  $\beta$  group, in which Cx26, Cx30, Cx31, Cx31.1 and Cx32 fall, and the other termed class II or  $\alpha$  group, represented by Cx33, Cx37, Cx40, Cx43 and Cx46.<sup>2</sup> All connexins share a common membrane topology, but differ in their unitary conductance and channel gating properties.<sup>3-5</sup> The structure of connexin molecules includes a cytoplasmic N-terminal region, four transmembrane domains, two extracellular loops, and a C-terminal cytoplasmic tail of varying length. The various connexins are highly conserved in the transmembrane and extracellular regions, but they differ in both sequence and length in their cytoplasmic domain. The 27kD connexin protein (connexin 32, Cx32) is expressed in several tissues. The pattern of expression may differ in various cell types and in various tissues in different species. For example, in the rodent brain, it is found in oligodendrocytes and certain neurons, but not in astrocytes, ependyma, leptomeninges and pinealocytes; or in the liver, it is present in hepatocytes. but not in Ito cells. Gap junction protein levels change in response to disruption of tissue architecture.<sup>6</sup> For instance, a decreased expression of Cx32 plasma membrane mRNA and protein levels was found in rats with common bile duct ligation (CBDL) induced hepatic injury.<sup>7</sup> Similar results were reported for partially hepatectomized mice.<sup>8</sup> Interestingly, a combination of myelin disruption and axonal degeneration has been shown to occur with Cx32 mutations in the X-linked Charcot-Marie-Tooth (CMTX) disease.<sup>9</sup> Antibodies reacting specifically with Cx32, may be used in diverse cellular and molecular approaches to the study of gap junctions and their properties, and to correlate their expression pattern in a variety of cell types and tissues with physiological functions or pathological conditions.

#### **Reagents**

The product is supplied as an affinity isolated antibody in 0.01M phosphate buffered saline, pH 7.4, containing

1% BSA and 15 mM sodium azide (see MSDS)\* as a preservative.

Protein concentration is approximately 1 mg/ml by E<sub>280</sub> prior to the addition of BSA.

#### **Precautions and Disclaimer**

\* Due to the sodium azide content a material safety data sheet (MSDS) for this product has been sent to the attention of the safety officer of your institution. Consult the MSDS for information regarding hazardous and safe handling practices.

#### **Storage/Stability**

For continuous use, store at 2-8°C for up to one month. For extended storage freeze in working aliquots. Repeated freezing and thawing is not recommended. Storage in "frost-free" freezers is not recommended. If slight turbidity occurs upon prolonged storage, clarify the solution by centrifugation before use.

#### **Product Profile**

A minimum working dilution of 1:400 is determined by immunoblotting using a liver membrane preparation from rat.

A minimum working dilution of 1:400 is determined by indirect immunoperoxidase staining of acetone-fixed and frozen sections of rat and human tissue.

Note: In order to obtain best results and assay sensitivity in different techniques and preparations we recommend determining optimal working dilutions by titration test.

#### **References**

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