REFERENCES

Adamian, L. and Liang, J. Helix-helix packing and interfacial pairwise interactions of residues in membrane proteins J. Mol. Biol. 311: 891-907, 2001.

Ahmad, S., Diez, J.A., George, C.H., and Evans, W.H. Synthesis and assembly of connexins in vitro into homomeric and heteromeric functional gap junction hemichannels. Biochem. J. 339: 247-253, 1999.

Baldwin, E.P., Hajiseyedjavadi, O., Baase, W.A., and Matthews, B.W. The role of backbone flexibility in the accommodation of variants that repack the core of T4 lysozyme. Science 262: 1715-1718, 1993

Bao, X., Altenberg, G. A., and Reuss, L. Mechanism of regulation of the gap-junction protein connexin 43 by protein kinase C-mediated phosphorylation. Am. J. Physiol. Cell Physiol. 286: 647-654, 2004a.

Bao, X., Reuss, L., and Altenberg, G. A. Regulation of purified and reconstituted connexin 43 hemichannels by protein kinase C-mediated phosphorylation of Serine 368. J. Biol. Chem. 279: 20058-20066, 2004b.

Bao, X., Chen, Y., Reuss, L., and Altenberg, G. A. Functional expression in *Xenopus* oocytes of gap-junctional hemichannels formed by a cysteine-less connexin 43. J. Biol. Chem. 279: 9689-9692, 2004c.

Bao, X., Chen, Y., Lee, S. H., Lee, S. C., Reuss, L., and Altenberg, G. A. Membrane transport proteins with complete replacement of transmembrane helices with polyalanine sequences remain functional. J. Biol. Chem. 280: 8647-8650, 2005.

Bao, X., Lee, S. C., Reuss, L., and Altenberg, G. A. Change in permeant size selectivity by phosphorylation of connexin 43 gap-junctional hemichannels by PKC. Proc. Natl. Acad. Sci. USA. 104: 4919-4924, 2007.

Beltramello, M., Piazza, V., Bukauskas, F. F., Pozzan, T. and Mammano, F. Impaired permeability to Ins(1,4,5)P3 in a mutant connexin underlies recessive hereditary deafness. Nat. Cell Biol. 7**:** 63-69, 2005.

Bennett, M.V.L., Contreras, J.E., Bukauskas, F.F., and Sáez, J.C. New roles for astrocytes: gap junction hemichannels have something to communicate. Trends Neurosci. 26: 610-7, 2003.

Berthoud, V. M., Ledbetter, M. L. S., Herzberg, E. L., and Sáez, J.C. Connexin43 in MDCK cells: regulation by a tumor-promoting phorbol ester and Ca^{2+}. Eur. J. Cell Biol. 57: 40-50, 1992.

Bevans, C. G., Kordel, M., Rhee, S. K., and Harris, A. L. Isoform composition of connexin channels determines selectivity among second messengers and uncharged molecules. J. Biol. Chem. 273: 2808-2816, 1998.

Bevans C. G., and Harris A. L. Regulation of connexin channels by pH. Direct action of the protonated form of taurine and other aminosulfonates. J. Biol. Chem. 274: 3711-3719, 1999.

Beyer, E. C., Lampe P. D., and Goodenough, D. A. Connexin-43: a protein from rat heart homologous to a gap junction protein from liver. J. Cell Biol. 105: 2621-2629, 1987.

Armstrong, C. M., and Bezanilla, F. Currents related to movement of the gating particles of the sodium channels. Nature 242:459-61, 1973.

Button, B., Reuss, L., and Altenberg, G. A. PKC-mediated stimulation of amphibian CFTR depends on a single phosphorylation consensus site. insertion of this site confers PKC sensitivity to human CFTR. J. Gen. Physiol. 117: 457-468,2001.

Cascio, M., Kumar, N. M., Safarik, R., and Gilula, N. B. Physical characterization of gap junction membrane connexons (hemi-channels) isolated from rat liver. J. Biol. Chem. 270: 18643-18648, 1995.

Cha, A., and Bezanilla, F. Structural implications of fluorescence quenching in the Shaker K^+ channel. J. Gen. Physiol. 112: 391-408, 1998.

Chee, K. N., Ormonde, S., Petsoglou, C., Goold, L., C. and McGhee, N. J., Sherwin, T., and Green, C. R. Non-healing corneal recovery triggered by regulation of direct cell-cell communication. The ASCB 47th Annual meeting regular abstracts, p. 251, 2007.

Chen, J., and Selvin, P. R. Thiol-reactive luminescent chelates of terbium and europium Bioconjug. Chem. 10: 311-315, 1999.

Contreras, J. E., Sánchez, H. A., Eugenin, E. A., Speidel, D., Theis, M., Willecke, K., Bukauskas, F. F., Bennett, M. V. L., and Sáez, J. C. Metabolic inhibition induces opening of unapposed connexin 43 gap junction hemichannels and decreases gap junctional communication in cortical astrocytes in culture. Proc. Natl. Acad. Sci. USA 99: 495-500, 2002.

Contreras, J. E., Sáez J. C., Bukauskas, F. F., and Bennett, M. V. L. Gating and regulation of connexin 43 (Cx43) hemichannels. Proc. Natl. Acad. Sci. USA 100: 11388-11393, 2003.

Cunningham, B. C. and Wells, J. A. High-resolution epitope mapping of hGH-receptor interactions by alanine-scanning mutagenesis. Science 244: 1081-1085, 1989.

Curran, A. R., and Engelman, D. M. Sequence motifs, polar interactions and conformational changes in helical membrane proteins. Curr. Opin. Struct. Biol. 13: 412-417, 2003.

Eilers, M., Shekar, S. C., Shieh, T., Smith, S. O., and Fleming, P. J. Internal packing of helical membrane proteins. Proc. Natl. Acad. Sci. USA 97: 5796-5801, 2000.

Escobal, A. Personal communication, 2008

Falk, M. M., Buehler, L. K., Kumar, N. M., and Gilula, N. B. Cell-free synthesis and assembly of connexins into functional gap junction membrane channels. EMBO J. 16: 2703-2016, 1997.

Fleishman, S. J., Unger, V. M., Yeager, M., and Ben-Tal, N. A. Calpha model for the transmembrane alpha helices of gap junction intercellular channels. Mol. Cell. 15: 879-888, 2004.

Foote, C. I., Zhou L., Zhu, X., and Nicholson, B. J. The pattern of disulfide linkages in the extracellular loop regions of connexin 32 suggests a model for the docking interface of gap junctions. J. Cell. Biol. 140:1187-97, 1998.

Francis, D., Stergiopoulos, K., Ek-Vitorin, J. F., Cao, F. L., Taffet, S. M., and Delmar, M. Connexin diversity and gap junction regulation by pHi. Dev. Genet. 24: 123-136, 1999.

Frillingos, S., Sahin-Toth, M., Wu, J., and Kaback, H. R. Cys-scanning mutagenesis: a novel approach to structure function relationships in polytopic membrane proteins. FASEB J. 12: 1281-1299, 1998.

Gong, X. Q., and Nicholson, B. J. Size selectivity between gap junction channels composed of different connexins. Cell Commun. Adhes. 8: 187-192, 2001.

Harris, A. L. Emerging issues of connexin channels: biophysics fills the gap. Q. Rev. Biophys. 34: 325-472, 2001.

Harris, A. L. Connexin channels permeability to cytoplasmic moledules. Prog. Biophys. Mol. Biol. 94: 120-143, 2007.

He, M. M., Sun, J., and Kaback, H. R. Cysteine-scanning mutagenesis of transmembrane domain XII and the flanking periplasmic loop in the lactose permease of Escherichia coli. Biochemistry. 35: 12909-12914, 1996.

Heyduk, T., and Heyduk, E. Luminescence energy transfer with lanthanide chelates: interpretation of sensitized acceptor decay amplitudes. Anal. Biochem. 289: 60-67, 2001.

Heyduk, T. Measuring protein conformational changes by FRET/LRET. Curr. Opin. Biotechnol. 13: 292-296, 2002.

Heyman N. S., and Burt J. M. Hindered diffusion through an aqueous pore describes invariant dye selectivity of Cx43 junctions. Biophys. J. 94: 840-854, 2008.

Hille, B. Ion Channels of Excitable Membranes. Sinauer Associates, Inc., Sunderland, MA, USA, chapters 14 and 15, 2001.

Homma, N., Alvarado, J. L., Coombs, W., Stergiopoulos, K., Taffet, S. M., Lau, A.F., and Delmar, M. A particle-receptor model for the insulin-induced closure of connexin43 channels. Circ. Res. 83: 27-32, 1998.

Hua, V. B., Chang, A. B., Tchieu, J. H., Kumar, N. M., Nielsen, P. A., and Saier, M. H. Jr. Sequence and phylogenetic analyses of 4 TMS junctional proteins of animals: connexins, innexins, claudins and occludins. J. Membr. Biol. 194: 59-76, 2003.

John, S. A., Kondo, R., Wang, S. Y., Goldhaber, J. I., and Weiss, J. N. Connexin-43 hemi-channels opened by metabolic inhibition. J. Biol. Chem. 274: 236-240, 1999.

Karlin, A. SCAM feels the pinch. J. Gen. Physiol. 117:235-237, 2001.

Kistler, J., Goldie, K., Donaldson, P. and Engel, A. Reconstitution of native-type noncrystalline lens fiber gap junctions from isolated hemichannels. J. Cell. Biol. 126, 1047-1058, 1994.

Knauf, P. A., and Pal, P. Use of luminescence resonance energy transfer to measure distances in the AE1 anion exchange protein dimer. Blood Cells Mol. Dis. 32: 360-365, 2004.

Kronengold, J., Trexler, E. B., Bukauskas, F. F., Bargiello, T. A., and Verselis, V. K. Single-channel SCAM identifies pore-lining residues in the first extracellular loop and first transmembrane domains of Cx46 hemichannels. J. Gen. Physiol. 122: 389-405, 2003.

Kondo, R. P., Wang, S. Y., John, S. A., Weiss, J. N., and Weiss, J. N. Metabolic inhibition activates a non-selective current through connexin hemichannels in isolated ventricular myocytes. J. Mol. Cell Cardiol. 32: 1859-1872, 2000.

Kovacs, J. A., Baker, K. A., Altenberg, G. A., Abagyan, R., and Yeager, M. Molecular modeling and mutagenesis of gap junction channels. Prog. Biophys. Mol. Biol. 94:15-28, 2007.

Kwak, B. R., van Veen, T. A., Analbers, L. J. and Jongsma, H. J. TPA increases conductance but decreases permeability in neonatal rat cardiomyocyte gap junction channels. Exp. Cell Res. 220: 456-463, 1995.

Lampe, P. D., TenBroek, E. M., Burt, J. M., Kurata, W. E., Johnson, R. G., and Lau, A. F. Phosphorylation of connexin43 on serine368 by protein kinase C regulates gap junctional communication. J. Cell Biol. 149: 1503-1512, 2000a.

Lampe, P. D. and Lau, A. F. Regulation of gap junctions by phosphorylation of connexins. Archiv. Biochem. Biophys. 384: 205-215, 2000b.

Li, H., Liu, T. F., Lazrak, A., Peracchia, C., Goldberg, G. S., Lampe, P. D., and Johnson, R. G. Properties and regulation of gap junctional hemichannels in the plasma membranes of cultured cells. J. Cell Biol. 134: 1019-1030, 1996.

Li, W. E., and Nagy, J. I. Connexin43 phosphorylation state and intercellular communication in cultured astrocytes following hypoxia and protein phosphatase inhibition. Eur. J. Neurosci. 12: 2644-2650, 2000.

Little, T. L., Beyer, E. C., and Duling, B. R. Connexin 43 and connexin 40 gap junctional proteins are present in arteriolar smooth muscle and endothelium in vivo. Am. J. Physiol. 268: 729-739, 1995.

Lurtz, M.M. and Louis, C.F. Intracellular calcium regulation of connexin43. Am. J. Physiol. Cell Physiol. 293:C1806-13, 2007.

Manthey, D., Banach, K., Desplantez, T., Lee, C.G., Kozak, C.A., Traub, O., Weingart, R., and Willecke, K. Intracellular domains of mouse connexin26 and -30 affect diffusional and electrical properties of gap junction channels. J Membr. Biol. 181: 137-48, 2001.

Marheineke, K., Grünewald, S., Christie, W., and Reiländer, H. Lipid composition of Spodoptera frugiperda (Sf9) and Trichoplusia ni (Tn) insect cells used for baculovirus infection. FEBS Lett. 441: 49-52, 1998

McLachlan, E., Manias, J. L., Gong, X. Q., Lounsbury, C. S., Shao, Q., Bernier, S. M., Bai, D., and Laird, D. W. Functional characterization of oculodentodigital dysplasia-associated Cx43 mutants. Cell Commun. Adhes. 12: 279-292, 2005.

Miller, J. S., Kennedy, R. J. and Kemp, D. S. Short, solubilized polyalanines are conformational chameleons: exceptionally helical if N- and C-capped with helix stabilizers, weakly to moderately helical if capped with rigid spacers. Biochemistry 40: 305-309, 2001.

Muller, D. J., Hand, G. M., Engel, A., and Sosinsky, G. E. Conformational changes in surface structures of isolated connexin 26 gap junctions. EMBO J. 21: 3598-3607, 2002.

Neveu, M. J., Hully, J. R., Babcock, K. L., Vaughan, J., Hertzberg, E. L., Nicholson, B. J., Lampe P. D., and Pitot, H. C. Proliferation-associated differences in the spatial and temporal expression of gap junction genes in rat liver. Hepatology 22: 202-212, 1995.

Nnamani, C., Godwin, A., Ducsay, C. A., Longo, L. D., and Fletcher, W. H. Regulation of cell-cell communication mediated by connexin 43 in rabbit myometrial cells. Biol. Reprod. 50: 377-389, 1994.

Nightingale, E. R., Jr. Viscosity of aqueous sodium perchlorate solutions. J. Phys. Chem. 63: 1381-1387, 1959.

Oshima, A., Tani, K., Hiroaki, Y., Fujiyoshi, Y., and Sosinsky, G. E. Three-dimensional structure of a human connexin26 gap junction channel reveals a plug in the vestibule. Proc. Natl. Acad. Sci. USA 104: 10034-10039, 2007.

Pal, P., Holmberg, B. E., and Knauf, P. A. Conformational changes in the cytoplasmic domain of human anion exchanger 1 revealed by luminescence resonance energy transfer. Biochemistry 44: 13638-13649, 2005.

Pfahnl, A. and Dahl, G. Localization of a voltage gate in connexin46 gap junction hemichannels. Biophys. J. 75: 2323-2331, 1998.

Pepper, M. S., and Meda, P. Basic fibroblast growth factor increases junctional communication and connexin43 expression in microvascular endothelial cells. J. Cell Physiol. 153: 196-205, 1992.

Peracchia C. Increase in gap junction resistance with acidification in crayfish septate axons is closely related to changes in intracellular calcium but not hydrogen ion concentration. J. Membr. Biol. 113: 75-92, 1990

Reaume, A. G., de Sousa, P. A., Kulkarni, S., Langille, B. L., Zhu, D., Davies, T. C., Juneja, S. C., Kidder, G. M., and Rossant, J. Cardiac malformation in neonatal mice lacking connexin43. Science 267:1831-1834, 1995.

Retamal, M. A., Cortes, C. J., Reuss, L., Bennett, M. V. and Sáez, J. C. Opening of connexin 43 hemichannels is increased by lowering intracellular redox potential. Proc. Natl. Acad. Sci. USA 103: 4475-4480, 2006.

Rosenbaum, D. M., Cherezov, V., Hanson, M. A., Rasmussen, S. G., Thian, F. S., Kobilka, T. S., Choi, H. J., Yao, X. J., Weis, W. I., Stevens, R. C., and Kobilka, B. K. GPCR engineering yields high-resolution structural insights into beta2-adrenergic receptor function. Science 318:1266-7123, 2007.

Roell, W., Lewalter, T., Sasse, P., Tallini, Y. N., Choi, B. R., Breitbach, M., Doran, R., Becher, U. M., Hwang, S. M., Bostani, T., von Maltzahn, J., Hofmann, A., Reining, S., Eiberger, B., Gabris, B., Pfeifer, A., Welz, A., Willecke, K., Salama, G., Schrickel, J. W., Kotlikoff, M. I., and Fleischmann, B. K. Engraftment of

connexin 43-expressing cells prevents post-infarct arrhythmia. Nature 450: 819-824, 2007.

Posson, D. J., Ge, P., Miller, C., Bezanilla, F., and Selvin, P. R. Small vertical movement of a K^+ channel voltage sensor measured with luminescence energy transfer. Nature 436: 848-851, 2005.

Pruvot, E. J., Katra, R. P., Rosenbaum, D. S., and Laurita, K. R. Role of calcium cycling versus restitution in the mechanism of repolarization alternans. Circ. Res. 94: 1083-1090, 2004.

Sáez J. C., Retamal M. A., Basilio D., Bukauskas F. F., and Bennett M. V. L. Connexin-based gap junction hemichannels: gating mechanisms. Biochim. Biophys. Acta. 1711: 215-24, 2005.

Sainio, K., Gilbert, S. F., Lehtonen, E., Nishi, M., Kumar, N. M., Gilula, N. B., and Saxen, L. Differential expression of gap junction mRNAs and proteins in the developing murine kidney and in experimentally induced nephric mesenchymes. Development 115: 827-837, 1992.

Segretain, D., and Falk, M. M. Regulation of connexin biosynthesis, assembly, gap junction formation, and removal. Biochim. Biophys. Acta. 1662: 3-21, 2004.

Selvin, P. R. Principles and biophysical applications of lanthanide-based probes. Annu. Rev. Biophys. Biomol. Struct. 31: 275-302, 2002.

Sharon, D., Vorobiov, D., and Dascal, N. Positive and negative coupling of the metabotropic glutamate receptors to a G protein-activated K+ channel, GIRK, in Xenopus oocytes. J. Gen. Physiol. 109: 477-490, 1997.

Skerrett, I. M., Aronowitz, J., Shin, J. H., Cymes, G., Kasperek, E., Cao, F. L., and Nicholson, B. J. Identification of amino acid residues lining the pore of a gap junction channel. J. Cell Biol. 159: 349-360, 2002.

Solan, J. L. and Lampe, P. D. Connexin phosphorylation as a regulatory event linked to gap junction channel assembly. Biochim. Biophys. Acta 1711: 154-163, 2005.

Spray, D. C., Harris, A. L. and Bennett, M. V. L. Kinetic properties of a voltage-dependent junctional conductance. J. Gen. Physiol. 77: 77-93, 1981

Stauffer, K. A. The gap junction proteins beta 1-connexin (connexin-32) and beta 2-connexin (connexin-26) can form heteromeric hemichannels. J. Biol. Chem. 270: 6768-72, 1995.

Stauffer, K. A., Kumar, N. A., Gilula, N. B., and Unwin, P. N. Isolation and purification of gap junction channels. J. Cell Biol. 115: 141-150, 1991.

Thimm, J., Mechler, A., Lin, H., Rhee, S., and Lal, R. Calcium-dependent open/closed conformations and interfacial energy maps of reconstituted hemichannels. J. Biol. Chem. 280: 10646-10654, 2005.

Ulmschneider, M. B. and Sansom, M. S. Amino acid distributions in integral membrane protein structures. Biochim. Biophys. Acta 1512: 1-14, 2001

Unger, V. M., Kumar, N. M., Gilula, N. B., and Yeager, M. Three-dimensional structure of a recombinant gap junction membrane channel. Science 283: 1176-1180, 1999.

Vazquez-Ibar, J. L., Weinglass, A. B., and Kaback, H. R. Engineering a terbium-binding site into an integral membrane protein for luminescence energy transfer. Proc. Natl. Acad. Sci. USA 99: 3487-3492, 2002.

Venance, L., Premont, J., Glowinski, J., and Giaume, C. Gap junctional communication and pharmacological heterogeneity in astrocytes cultured from the rat striatum. J. Physiol. 510: 429-440, 1998.

Vergara, L., Bao, X., Cooper, M., Bello-Reuss, E., and Reuss, L. Gap-junctional hemichannels are activated by ATP depletion in human renal proximal tubule cells. J. Membr. Biol. 196: 173-184, 2003.

Watkins, R. E., Wisely, G. B., Moore, L. B., Collins, J. L., Lambert, M. H., Williams, S. P., Willson, T. M., Kliewer, S. A. and Redinbo, M. R. The human nuclear xenobiotic receptor PXR: structural determinants of directed promiscuity. Science 292: 2329-2333, 2001.

Wei, C. J., Xu, X., and Lo, C. W. Connexins and cell signaling in development and disease. Annu. Rev. Cell Dev. Biol. 20: 811-838, 2004.

Weber, P. A., Chang, H. C., Spaeth, K. E., Nitsche, J. M. and Nicholson, B. J. The permeability of gap junction channels to probes of different size is dependent on connexin composition and permeant-pore affinities. Biophys. J. 87: 958-973, 2004.

Willecke, K., Kirchhoff, S., Plum, A., Temme, A., Thonnissen, E., and Ott, T. Biological functions of connexin genes revealed by human genetic defects, dominant negative approaches and targeted deletions in the mouse. Novartis Found. Symp. 219: 76-88, 1999.

Willecke, K., Eiberger, J., Degen, J., Eckardt, D., Romualdi, A., Güldenagel, M., Deutsch, U., and Söhl, G. Structural and functional diversity of connexin genes in the mouse and human genome. Biol. Chem. 383: 725-37, 2002.

Yeager, M. J., and Gilula, N. B. Membrane topology and quaternary structure of cardiac gap junction ion channels. J. Mol. Biol. 223: 929-948, 1992.

Yu, J., Bippes, C. A., Hand, G. M., Muller, D. J., and Sosinsky, G. E. Aminosulfonate modulated pH-induced conformational changes in connexin26 hemichannels. J Biol Chem. 282:8895-904, 2007.

Zhou, X. W., Pfahnl, A., Werner, R., Hudder, A., Llanes, A., Luebke, A., and Dahl, G. Identification of a pore lining segment in gap junction hemichannels. Biophys J. 72: 1946-1953, 1997.

Zhou, Y., Yang, W., Lurtz, M.M., Ye, Y., Huang, Y., Lee, H.W., Chen, Y., Louis, C.F., and Yang, J.J. Identification of the calmodulin binding domain of connexin 43. J. Biol. Chem. 282:35005-17, 2007

Vita

Sung-Chang Lee

Sung-Chang Lee was born and raised in Seoul, Republic of Korea, the son of Kwan-Kook Lee and Sung-Ju Bae. In 2002, Sung-Chang married with Seung-Hee Choi and is a proud father to Daniel Taeho Lee. After completing Jemulpo High School in Incheon, and then attended Woosuk University, Jeollabuk-do, Korea and transferred to Suwon University, Gyeonggi-do, Korea and he received his Bachelor degree in Biology in 1998. Following his undergraduate studies, Sung-Chang joined the graduate program at Hanyang University, Seoul, Korea and awarded his M. M. Sc. degree from the Department of Biochemistry at Hanyang University in 2002. Then he worked for several research institutes, such as Inha University, Samsung Medical Center, Duksung Women's University, and the University of Texas Medical Branch until he was admitted to the Ph.D. program at the University of Texas Medical Branch in 2005. Sung-Chang entered graduate school in the Cell Physiology and Molecular Biophysics at the University of Texas Medical Branch where he is pursuing Ph.D. degree in the field of membrane protein structural biology.

During his graduate study at UTMB, Sung-Chang has been awarded a number of competitive awards. He was the recipient (2006, ASBMB Best Graduate Student Poster Presentation Honor) from the American Society for Biochemistry and Molecular Biology, He also received three scholarships (2006 GSBS Associates Scholarship, 2007 Mason Guest Scholar Award, and 2007 Arthur V. Simmang Academic Scholarship) from the University of Texas Medical Branch Graduate School of Biomedical Sciences, and one travel award (2007 National Graduate Student Research Festival Travel Award) from National Institutes of Health.

Education

B.S., 1999, University of Suwon, Suwon, Republic of Korea
M.S., 2002, Hanyang University, Seoul, Republic of Korea

Publications

A. Articles in peer-reviewed journals

1. Oh, K. J., Lee, S. C., Choi, H. J., Oh, D. Y., Kim, S. C., Min, D. S., Kim J. M., Lee, K. S., and Han, J. S. Role of phospholipase D2 in anti-apoptotic signaling through increased expressions of Bcl-2 and Bcl-xL. J. Cell Biochem. 101:1409-1422, 2007.

2. Bao, X.*, Lee, S. C.*, Reuss, L., and Altenberg, G. A. Change in permeant size-selectivity by phosphorylation of connexin 43 gap-junctional hemichannels by PKC. Proc. Natl. Acad. Sci. USA 104: 4919-4924, 2007 *These authors contributed equally to this work.

3. Bao, X., Chen, Y., Lee, S. H., Lee, S. C., Reuss, L and Altenberg, G. A. Membrane Transport Proteins with Complete Replacement of Transmembrane Helices with Poly-Alanine Sequences Remain Functional. J. Biol. Chem. 280: 8647-8650, 2005.

4. Lee, S.C., Han J. S., and Cha, Y. N. Regulation of cyclooxygenase-2 by phosphatidylcholine-specific phospholipase C and D in lipopolysaccharide stimulated murine RAW264.7 macrophages. Mol. Cells 15: 320-326, 2003.

5. Lim, S. Y., *Lee, S.C.*, Shin, I., and Han, J. S. Differential effects of Fas cross-linking on phospholipase D activation and related lipid metabolism in Fas resistant A20 cells. Exp. Mol. Med. 34: 201-210, 2002.

6. Yoo, J., and Lee, S. C. Elevated level of plasma homocysteine and asymmetric dimethylarginine in elderly patients with stroke. Atherosclerosis 158: 425-430, 2001.

B. Abstracts

1. Lee, S. C., Reuss, L., and Altenberg, G. A. Movement of M3 helices during gating of Cx43 gap-junctional hemichannels by PKC-mediated phosphorylation. Biophysical Society Meeting, Long beach, CA, 2008.

2. Lee, S. C., Reuss, L., and Altenberg, G. A. The decrease in large-solute permeability of connexin 43 gap-junctional hemichannels (Cx43 GJH) by PKC-mediated phosphorylation is a cooperative phenomenon. The American Society for Cell Biology, Washington D.C., 2007.

3. Lee, S. C., Reuss, L., and Altenberg, G. A. Movement of M3 helices during gating of Cx43 gap-junctional hemichannels by PKC-mediated phosphorylation. Gap junction special interest subgroup meeting in the American Society for Cell Biology, Washington D.C., 2007.

4. Lee, S. C., Reuss, L., and Altenberg, G. A. Identification of a pore-lining helix of the gap-junctional protein connexin 43 (Cx43). Biophysical Society Meeting, Baltimore, MD, 2007.

5. Lee, S. C., Bao, X., Reuss, L., and Altenberg, G. A. Change in permeant size-selectivity by phosphorylation of connexin 43 gap-junctional hemichannels by PKC. Meeting of the American Society for Cell Biology, San Diego, CA, 2006.

6. Lee, S. C., Bao, X., Reuss, L., and Altenberg, G. A. Generation of gap-junctional hemichannels of controlled subunit composition to study their regulation by PKC-mediated phosphorylation. Department Neuroscience and Cell Biology Scientific Retreat, Galveston, TX, 2006.

7. Lee, S. C., Bao, X., Reuss, L., and Altenberg, G. A. Generation of gap-junctional hemichannels of controlled subunit composition to study their regulation by PKC-mediated phosphorylation. The 11th Annual Structural Biology Symposium, Galveston, TX, 2006.

8. Lee, S. C., Reuss, L., and Altenberg, G.A. Residues on one face of the third tranmembrane helix of connexin 43 (Cx43) line the gap-junctional hemichannel (GJH) pore. National Student Research Forum, Galveston, TX, 2006.

9. Lee, S. C., Bao, X., Reuss, L., and Altenberg, G.A. Generation of gap-junctional hemichannels of controlled subunit composition to study their regulation by PKC-mediated phosphorylation. Meeting of the American Society for Biochemistry and Molecular Biology, San Francisco, CA, 2006.

10. Lee, S. C., Reuss, L., and Altenberg, G.A. Identification of residues of the third transmembrane helix of connexin 43 (Cx43) that line the gap-junctional hemichannel (GJH) pore. Meeting of the American Society for Cell Biology, San Francisco, CA, 2005.

11. Bao, X., Lee, S. H., Chen, Y., Lee, S. C., Reuss, L., and Altenberg, G.A. Connexin43 mutants with complete transmembrane helix replacements with poly-alanine sequences form functional hemichannels. Biophysical Society Meeting, Long Beach. CA, 2005.

12 Lee, S. C., Yoon, M. S., and Han, J. S. Phosphatidic acid produced by phospholipase D inhibits Fas-induced apoptosis via Bcl-2 upregulation in A20 murine B lymphoma cells. Cell Signaling, Luxembourg, 2002.

13 Lee, S. C., and Han, J. S. Involvement of Lyn, Src-family tyrosine kinase, in the activation of PLD by anti-Fas monoclonal antibody in A20 cells. The Korean Society of Medical Biochemistry and Molecular Biology, Korea, 2001.

14 Lee, S. C., Yoon, M. S., and Han, J. S. Charaterization of mechanism for anti-apoptotic effect against Jo2 by PLD2 in A20 murine B lynphoma cell lines. The Korean Society of Medical Biochemistry and Molecular Biology, Korea, 2001.

15 Lee, Y. W., Lee, S. C., J. S. and Lee, Y. M. In vivo biocompatibility of gelatin-chondroitin sulfate sponges. The Korean Society for Polymers, Korea, 2001.

16 Lee, Y. W., Lee, S. C., J. S. and Lee, Y. M. Effects by the increase of a sulfated glycosaminoglycan on the culture of rat chondrocytes on the gelatin based materials. The Korean Society for Biomaterials, Korea, 2001.

17 Lee, S. C., and Han, J. S. Involvement of PLD2 in suppression of Fas-mediated apoptosis in A20 cells. Experimental Biology Meeting, FL, USA, 2001.

18 Lee, S. C., and Han, J. S. Ras GTPase is essential for Fas-mediated activation of phospholipase D in A20 cells. International Union of Biochemistry and Molecular Biology, Birmingham, UK, 2000.

19 Lee, Y. W., Lee, S. C., J. S. and Lee, Y. M. Effects of IGF-I in chondrocytes culture and evaluation of gelatin/chondroitin sulfate as scaffolds. The Korean Society for Biomaterials, 2000.

20 Lee, S. C., and Han, J. S. Ras GTPase is essential for Fas-mediated activation of phospholipase D in A20 cells. Federation Meeting of Korean Basic Medical Scientists, Korea, 2000.

21 Lee, S. C., and Cho, B. H. Analysis of polyamines content by cold stress and ABA in spring radish and stem. The Korean Society of Analytical Sciences, Korea, 1998.

22 Lee, S. C., and Cho, B. H. Effect of free amino acids by ABS in spring radish and winter radish. The Korean Society of Analytical Sciences, Korea, 1997.

Summary of Dissertation

The gap-junctional channels that mediate intercellular communication are formed by head-to-head docking of two gap-junctional hemichannels from adjacent cells. The hemichannels are hexamers of connexins, proteins that have four transmembrane helices. The transmembrane helices that line the gap-junctional pore have not been consistently identified, and their identification was the main goal of my dissertation project. To accomplish this goal, I used a combination of molecular biology, biochemical and biophysical techniques that include poly-alanine helix scanning mutagenesis, the substituted cysteine accessibility method and luminescence resonance energy transfer. Using the latter methodology in particular, as well as a new method to produce purified hemichannels of controlled subunit composition, I was able to assign all helices in the available low-resolution cryoelectron microscopy structure published by others, where helices are named A through D, and generate the first model of gap-junctional channels and hemichannels based on experimental structural measurements. In this model, connexin transmembrane helices 1 through 4 correspond to helices A, B, C and D, respectively. Luminescence resonance energy transfer is a powerful method for structural studies of membrane proteins in their native bilayer environment. Taken advantage of this methodology, in combination with the generation of hemichannels of controlled subunit composition, I was also able to determine that PKC-mediated phosphorylation of Ser368 produces a partial closure of the Cx43 hemichannel pore, that this effects requires phosphorylation of all six Cx43 monomers in the hemichannel, and that the decrease in permeability is accompanied by significant conformational changes of the connexin molecules. These changes involve increases of the distances separating the C-terminal ends of the subunits and decreases in the distances separating the pore-lining helices; both changes in inter-subunit distances are of the order of several Angstroms. These results indicate that a simple ball-and-chain mechanism cannot explain the gating of Cx43 hemichannels by PKC-mediated phosphorylation and that a significant re-arrangement of pore helices takes place instead. In summary, my results allowed me to generate an experimentally-based model gap-junctional channels and hemichannels and to gain insight into the molecular mechanism of Cx43 regulation by PKC-mediated phosphorylation.

Permanent address: 2704 Genoa Ave. #GG4, Lubbock, TX 79407

This dissertation was typed by Sung-Chang Lee.